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EFFECT OF MALEIC HYDRAZIDE, A PLANT GROWTH INHIBITOR, ON THE BIOLOGY OF ROOT-KNOT NEMATODE, *MELOIDOGYNE INCOGNITA*

by
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Increased levels of auxin were reported in root galls following invasion by various species of *Meloidogyne* (Bird, 1962, Balasubramanian and Rangaswami, 1962; Yu and Viglierchio, 1964; Setty and Wheeler, 1968). Giebel and Wilski (1969) and Giebel (1970) suggested that IAA released from 'complexes' in the root tissue caused nuclear division without cytokinesis resulting in the giant cell formation which is necessary for normal development of root-knot nematodes. Furthermore, the adverse effect on the reproduction of plant parasitic nematodes following foliar application of a growth inhibitor like maleic hydrazide indicates the necessity for phytohormones for their successful development (Nusbaum, 1958; Peacock, 1960; Davide and Triantaphyllou, 1968). Because little is known about the reasons for the adverse effect of growth inhibitors on the biology of nematodes, investigations have been made with maleic hydrazide (MH) and *Meloidogyne incognita* (Kofoid *et* White) Chitw.

Material and Methods

Cultures of *M. incognita* originating from a single egg-mass were reared on tomato (*Lycopersicon esculentum*) cv. Pusa Ruby. Single 4 week old tomato seedlings were inoculated with approximately 500 surface sterilized second-stage larvae either 24 hours before or 24 hours after foliar application of 12 mg/plant MH by atomizer. Care

was taken to avoid contamination of soil with the spray. Another set of plants was sprayed with water and left inoculated to serve as control. At pre-determined intervals, the plants were removed from the pots and the roots stained in boiling lactophenol acid fuchsin for two to three minutes and stored in clear lactophenol over-night for clearing. Then the nematodes were carefully dissected out of galls and mounted in freshly prepared lactophenol for microscopic examination. Galled root tissue from all the treatments was fixed in 4% formalin for hand sectioning and observations of histological changes.

Results

Irrespective of time of application, MH adversely affected the larval penetration (Table I). The percentage of penetration in the controls was almost double than in treated plants. The MH treatment affected the development of *M. incognita* by increasing the proportion of males and inducing rapid differentiation of adults (Table II). Abnormal females were encountered only in MH treated plants.

Twentyeight days after inoculation, the majority of larvae had developed into males compared with females only in control plants (Table III). Furthermore, male intersexes, female intersexes and abnormal males (Fig. 3 B) were observed at this interval of observation in treated plants. Female intersexes are those developing females which have undergone partial sex reversal and show some character-

Table I - *Larval penetration of* M. incognita *in tomato plants treated with MH (average of 4 replications per treatment).*

Treatment	Hours after inoculation					
	24		48		72	
	Mean	SEm	Mean	SEm	Mean	SEm
Control	74	± 8.23	126	± 6.13	180	± 10.89
MH sprayed 24 hours before inoculation	38	± 6.73	65	± 3.62	98	± 11.16
MH sprayed 24 hours after inoculation	46	± 6.64	91	± 11.51	103	± 10.34

Table II - *Effect of MH on the development of* M. incognita, *14 days after inoculation (average of three replications); number of nematodes found in one root system.*

Treatment	Sexually undifferentiated larvae		Sexually differentiated						Abnormal female	
			Developing female		Adult female		Developing male			
	Mean	SEm	Mean	SEm	Mean	SEm	Mean	SEm	Mean	SEm
Control	43	± 10.53	102	± 13	33	± 7.59	4	± 2.08		
MH sprayed 24 hours before inoculation	1.8	± 2.64	24	± 6	25	± 9.47	52	± 12.48	5	± 3
MH sprayed 24 hours after inoculation	10.33	± 3.05	15	± 5	11	± 1.52	100	± 21.08		

Table III - *Effect of MH on the development and reproduction of* M. incognita *28 days after inoculation (average of three replications); number of nematode found in one root system.*

Treatment	Adult male		Adult female		Developing male		Male intersexes		Female intersexes		Abnormal male		Number of egg masses	Number of eggs/ egg mass Average of five replica-tions
	Mean	SEm	Mean	SEm	Mean	SEm	Mean	SEm	Mean	SEm	Mean	SEm		
Control			170	± 45.82	—	—	—	—	—	—	—	—	104	303
MH sprayed 24 hours before inoculation	78	± 11.8	41	± 15.71	11	± 2.64	7	± 2.5	—	—	—	—	38	286
MH sprayed 24 hours after inoculation	80	± 26.45	19	± 7.54	13	± 5.2	14	± 3.6	5	± 3	17	± 6.12	26	258

istics of the male sex such as spicules etc. Male intersexes have undergone partial sex reversal and show some characteristics of the female sex such as vulva (l.c. Davide and Triantaphyllou, 1968).

MH-induced abnormal male were shorter (620-830 μm) than normal males recovered from untreated control plants (1135 μm). In MH-treated plant, the development of female gonads appeared to be retarded.

Reduced fecundity and fewer egg masses were found in MH-treated plant (Table III). The percentage of root infestation in MH-treated plant was reduced to 26-38% compared with 76% in the controls. The galls were very small on the MH-treated plants. Compared with the controls, there were fewer giant cells with lesser number of nuclei in MH-treated plants, presumably reflecting the effect on nematode feeding. Occasionally aborted giant cells were found in MH-treated plants.

Discussion

The results of the experiments failed to provide a conclusive explanation of the influence of the growth inhibitor MH on the biology of the invading *M. incognita*. Possibly, the growth inhibitor may interfere with the development of nematode indirectly by influencing the availability of the growth regulators required for its development and reproduction. Indeed a number of researchers (Sayre, 1958; Sandstedt and Schuster, 1966a and b; Webster, 1967; Kochba and Samish, 1971) demonstrated that many plant parasitic nematodes including *Meloidogyne* spp. require growth regulators for their development.

There is a link between the plant growth inhibitor, the reduction of feeding sites for developing females and retardation of the development of the nematode but elaborate experimentation would be needed to investigate this. Nevertheless, it is clear that exogenous application of the plant growth inhibitor directly or indirectly induced environmental stress, leading to sex reversal and development of many second-stage larvae into males, intersexes etc. This observation is consistent with that of Davide and Trantaphyllou (1968) who also reported the transformation of many larvae into male and intersexes. The present investigation demonstrated the potential use of MH as a possible control of root-knot nematodes.

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SUMMARY

Foliar application of Maleic hydrazide (MH) to tomato seedlings, adversely influenced the penetration, development and reproduction of the root-knot nematode, *Meloidogyne incognita*. Irrespective of time of application, MH increased the proportion of males, and increased male and female intersexes, abnormal males and females. There were fewer galls and fewer or aborted giant cells in MH treated plants. Male intersexes of *M. incognita* were obtained for the first time from plants treated with MH. The result of the present investigation demonstrates the potential of using MH for the control of root-knot nematodes.

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