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CONTROL OF ROOTKNOT NEMATODES, *MELOIDOGYNE*  
*JAVANICA*, IN ROOTED STOCKS OF GRAPEVINE,  
*VITIS VINIFERA*, BY IMMERSION IN NEMATICIDE SOLUTIONS  
AT DIFFERENT TEMPERATURES AND IN HOT WATER

by

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The rootknot nematode *Meloidogyne javanica* (Treub) Chitwood is a major pest of grapevine, especially in sandy and sandy loam soils in the Murray Valley districts of Victoria, Australia. Rooted stocks of susceptible varieties are often planted in establishing new vineyards. They are produced in the Murray Valley area in nurseries where *M. javanica* has always been a problem. Despite soil fumigation in this area, either with DD or EDB, planting stocks produced have been found seriously infested. Since it was shown by Meagher (1960) that immersion in hot water (51.7°C for five minutes) as practiced in California (Lear and Lider, 1959) was effective in controlling rootknot nematodes, growers have refused rooted stocks that have not been treated. However, in recent years 10-20% of vines treated with hot water have been found to contain viable *M. javanica*, suggesting that the method may not be completely effective.

Since the introduction of systemic nematicides, there has been interest in the use of these chemicals as dip treatments to control nematodes in propagative stock. Literature since 1956, comprising about 50 reports and involving at least 20 crop species and 10 nematode species, gives accounts of concentration and exposure time as determinant factors for the effectiveness of chemical dips. Promising results with rootknot nematode infested propagative material of various crop species have been reported by many workers (Ahuja, 1978; Bolander and Santo, 1977; Dale, 1973; Jain and Bhatti, 1979; Overman, 1973). However, Walker and Wilson (1962) found that the efficacy of chemicals in controlling rootknot nematodes was improved by using dip treatments at temperatures above those commonly

encountered in propagating sheds, but that phytotoxicity was increased with increasing temperatures.

In view of the above, the current standard hot water treatment was re-evaluated and compared with immersion treatments in systemic nematicidal solutions at a range of exposure times and at temperatures that are likely to occur in nurseries.

### *Methods*

Two experiments were initiated in October 1977 with one year-old dormant rooted stocks of *Vitis vinifera* cv. Sauvignon Blanc heavily infested with *M. javanica*. Before treatment, plants with similar galling were selected; the shoots were trimmed to three buds and the rootsystems were trimmed to approximately 15 cm length, as is commonly done before planting in vineyards. In the first experiment, plants were totally immersed for 15, 30, or 60 minutes in 1000 ppm-solutions of fenamiphos, oxamyl or carbofuran. Two other treatments consisted of immersion in water at 51.7°C for five minutes in a thermostatically controlled water bath (HWT) or 30 minute-immersion in tap water. The temperature of the nematicide solutions and the tap water was 16°C. After treatment all stocks were immediately rinsed for 10-15 seconds in running water.

In the second experiment plants were immersed for 30 minutes in 1000 ppm-solutions of each of the same nematicides but the solutions were maintained at 5°C, 12°C, or 24°C. The control consisted of a 30-minute immersion in tap water at 24°C. The solutions were prepared within 18 hours of dipping. Before treatment, rootstocks were acclimatised at room temperatures similar to those of the root dip solutions.

There were twelve replicate vines in each treatment. The treated stocks were grown in pots containing phytonematode-free, pasteurized (60°C, 3 hours) potting mix and placed randomly in a heated glasshouse with an average temperature of 21.5°C. The vines were examined regularly for symptoms of phytotoxicity.

Fourteen weeks after potting, four two-week-old tomato (cv. Burnley Bounty) seedlings were planted as indicators around each vine. When the indicator plants were ten weeks old they were assayed for surviving nematodes using Daulton and Nusbaum's (1961) method modified as follows. The indicator plants were rated on a 0-8 scale

of infection classes (0 = no infection; 8 = massive invasion, no root development). However, instead of assigning arbitrary values to these classes, each class was given an index value based on the number of sessile nematodes per unit length of root. Root systems were stained with cottonblue-lactophenol and their lengths determined with an opto-electronic scanning machine (Richard *et al.*, 1979) <sup>(1)</sup>. As determined from six replicates, root systems of class 2 (10% coverage by small discrete galls) were found to harbour a mean of 2.0, class 4 (30% small discrete galls, 5% large discrete galls, 5% small fused galls) 10.1, and class 6 (30% small discrete galls, 10% large discrete galls, 30% small fused galls, 10% large fused galls) 19.3 sessile nematodes per 10 cm of root. The difference between all pairs of means of nematode numbers was significant ( $P = 0.05$ ) by Duncan's multiple range test. Classes 2, 4, and 6 were given index values 2, 10 and 20, respectively; classes 0, 1, 3, 5, 7, and 8 were given values 0, 1, 6, 15, 30, and 40, respectively. Experimental data are based on these index values.

### *Results*

The grapevines were not assessed because galls which formed after the treatments were not clearly distinguishable from pretreatment galls.

The results of the first experiment (Table I) indicate that all the nematicide treatments suppressed nematode populations in the rooted stocks. Best results were obtained with fenamiphos which eradicated *M. javanica* after immersion for 60 minutes. Although differences between results for the eradicator fenamiphos treatment and HWT were not statistically significant, control by HWT was incomplete; each indicator plant from one pot with a hotwater-treated vine supported 8 small galls. Indicator plants associated with vines treated with fenamiphos for 30 minutes showed 2-3 small galls. Gallings of Class 8 were found only in pots with water-immersed control vines.

In the second experiment, in the range of temperatures studied,

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<sup>(1)</sup> Using this apparatus the length of a rootsystem can be estimated with a high degree of accuracy in about three minutes. The machine is available in revised form (Comair Root Length Scanner) from Commonwealth Aircraft Corporation Limited, 304 Lorimer Street, Port Melbourne, Victoria, Australia 3207.

Table I - Effect of nematicidal solutions of 16°C and standard hot water treatment on *M. javanica* in rooted stocks of grapevine.

Chemical	Exposure time (minutes)	No. of pots where <i>M. javanica</i> detected (out of 12)	Mean index value of indicator plants	
Fenamiphos 1000 ppm	15	8	0.83	f
	30	2	0.17	f
	60	0	0.00	a
Oxamyl 1000 ppm	15	12	4.58	d
	30	12	10.25	c
	60	12	2.33	e
Carbofuran 1000 ppm	15	12	7.83	cd
	30	12	5.67	cd
	60	12	7.25	cd
Hot water 51.7°C	5	1	0.08	a
Control, water 16°C	30	12	24.17	b

Means followed by no letter in common are significantly different ( $P = 0.05$ ) by Duncan's multiple range test.

all nematicide solutions gave control of rootknot nematodes in vines, with fenamiphos showing a high level of control and eradication (Table II). Infected indicator plants from pots with fenamiphos-treated vines exhibited 2-5 small galls in each rootsystem.

Five hundred ml of soil was processed (Seinhorst, 1956) from each pot in the fenamiphos treatments that gave infection-free indicator plants; microscopic examination failed to detect infective *Meioidogyne* larvae. No evidence of phytotoxicity was found in any of the treatments.

### Discussion

The results for the standard HWT tend to confirm impressions that it does not eradicate *M. javanica*. Eradicant temperature-time combinations can be determined, but the critical nature of these formulations may remain a serious limitation to its routine use under nursery conditions.

Microscopic examination of hot-water-treated rootlings indicated

Table II - Efficacy of nematicidal dips at different temperatures.

Chemical treatment (1000 ppm, 30 minutes)	Temperature (°C) of solution	No. of pots where <i>M. javanica</i> detected (out of 12)	Mean index value of indicator plants
Fenamiphos	5	5	0.21 e
	12	1	0.04 a
	24	0	0.00 a
Oxamyl	5	12	4.92 cd
	12	12	2.67 d
	24	12	4.00 cd
Carbofuran	5	12	7.00 c
	12	12	5.29 cd
	24	12	7.54 c
Control, water	24	12	15.83 b

Means followed by no letter in common are significantly different ( $P = 0.05$ ) by Duncan's multiple range test.

that juvenile *M. javanica* were embedded in small, young galls consisting of tightly packed cells and the nematodes may thus survive the treatment. Large galls, especially those with tissues disrupted under the pressure of exuded egg masses, provided no protection for the nematodes. Lear and Lider (1959) and Meagher (1960) used a bioassay method which consisted of incorporating chopped roots of the treated stock into soil in which indicator plants were then grown. It is possible that this method did not detect surviving larvae because within clipped roots life may not have been sustained long enough for larvae to develop into mature, egg laying females. The method is suitable to detect motile, infective larvae, which are normally present together with mature females and eggs in large galls.

The results with fenamiphos show that, within the range tested, the effectiveness of the solution was increased with increasing temperature. The results appear to agree with those of Walker and Wilson (1962) who, although working with heated solutions, also found that nematicide solutions were better when used at higher temperatures. McKenry and Thomason (1974) showed that, as temperature increases from 5° to 25°C, the toxicity of 1,3-D and EDB to *M. javanica* increases. They suggested that temperature influences the metabolic activity of the nematode and that, as the metabolic activity

is increased, the amount of chemical required to kill the nematode decreases. High levels of metabolic activity, particularly in moulting nematodes, result in greater susceptibility to toxicants (Evans and Thomason, 1971).

The method of systemic nematicidal immersion may have the advantage that the exposure time is less critical, and that the chemical acts over a relatively long time within the plant after treatment. A chart may be needed for nursery use, showing eradicant temperature-time combinations of treatment with suitable systemic compounds at effective rates.

#### S U M M A R Y

Immersion treatment in water at 51.7°C for five minutes (HWT) or 1000 ppm solutions of fenamiphos, oxamyl and carbofuran were evaluated for control of *Meloidogyne javanica* in rooted grapevine cuttings. The effect of varying times of chemical treatment (15 to 60 minutes) and temperature (5° to 24°C) was also tested. A bioassay showed that immersion in fenamiphos for 60 minutes at 16°C or 30 minutes at 24°C eradicated *M. javanica*. The difference between data obtained from HWT and the eradicant fenamiphos treatment at 16°C was not statistically significant, but control by HWT was incomplete. None of the chemical treatments was phytotoxic.

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