Effects of Three Nematicides on the Physiology of Rose Infected with Meloidogyne hapla

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ABSTRACT

The nematicides DBCP, Mocap, and Temik reduced considerably the number of root-knot nematode galls (*Meloidogyne hapla*) on roses. Rates of 0.5 and 1 liter/hectare of DBCP and of 1 kg/hectare of Mocap or Temik reduced photosynthesis and transpiration and increased leaf resistance to CO_2 uptake and water vapor loss. Photosynthesis was not reduced significantly per mg of chlorophyll, but it was reduced more than the calculated reduction due to stomatal closure, which suggests an increase in mesophyll resistance. Reduction in net photosynthesis was followed by reduction in flower yield after treatment with the higher rate of DBCP and Mocap. The method described offers the means of an early detection of toxic symptoms due to nematicide application.

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Additional key words: Rosa multiflora, ventilated diffusion porometer, phytotoxicity, CO_2 and water vapor exchange, diffusion resistance, mesophyll resistance.

The root-knot nematode, *Meloidogyne hapla* Chitwood 1949, is a serious pest of roses in greenhouses mainly because of the perennial nature of the crop and the favorable conditions in the greenhouse. Symptoms include reduction of vigor of the plants, chlorosis, dwarfing of leaves and shoots, leaf drop, and reduced flower yield (6). Once greenhouse roses are infected, treatment with a nematicide is necessary. Although most of the post-plant nematicides can be applied alongside growing plants, there are reports of varying degrees of phytotoxicity, depending upon the plant and the conditions (7). Little is known about the direct effect of these nematicides on the physiology of the treated plants and, to the best of our knowledge, no method of early detection has previously been reported.

In this work we investigated the effect of three nematicides on the yield, photosynthesis, and transpiration of greenhouse-grown roses.

MATERIALS AND METHODS.–Rose plants, *Rosa multiflora* Thunb, were grown in 50-liter plastic containers, filled with a mixture of peat and soil (1:2, v/v) in a polyethylene greenhouse. The roses were inoculated with a suspension of chopped tomato roots infected with *M. hapla*. The efficacy of the nematicides was determined by planting tomato plants near the roses and checking the tomato roots for gall development 1 month after planting. A "gall index" was used as an estimate of the relative amount of galling; this ranged from 0=no galls, to 5=all roots with many galls. The number of flowers per plant was counted daily for 25 weeks, starting after the first treatment.

Nematicides tested were: Mocap (O-ethyl S,S-dipropylphorodithioate) (450 grams/liter emulsified concentrate), at a total rate of 4 liter/hectare; Temik [2-methyl-2-(methylthio)-propionaldehyde O-(methylcarbamoyl) -oxime], (10% granular) at a total rate of 4 kg/hectare; and DBCP (1,2-dibromo-3-chloropropane) (52% emulsified concentrate) at a total rate of 4 liter/hectare. All the nematicides were applied four times, at the time of irrigation, at 14-day intervals. Carbon dioxide and water exchange were measured 2 weeks after the last treatment with a ventilated diffusion porometer (3). Before each measurement the porometer was flushed with 300 liter/liter $^{14}CO_2/^{12}CO_2$ mixture with a specific activity of 24 $\times 10^{-3}$. The leaf CO_2 uptake was calculated by dividing the CO_2 uptake by the specific activity. Transpiration was measured by subjecting a portion of leaf to the internal atmosphere of the porometer in which an average relative humidity of 30% was maintained at an average temperature of 25 C. Leaf temperature was measured by a thermistor mounted at the top of a 24-gauge hypodermic needle. The leaf portions measured were subjected to the internal atmosphere of the porometer for 30-50 seconds. Leaf resistance to water vapor loss was calculated from transpiration and leaf temperature data using Gaastra's method (4). Each of the five treatments were replicated eight times in a randomized block design.

To test lower concentrations of DBCP, a large-scale experiment was conducted in a commercial greenhouse. At weekly intervals over a 5-week period, DBCP was applied in 100 liters of water at rates of 0.5 and 0.2 liter/hectare to beds of roses, using a low-pressure pump. The roses were irrigated immediately after treatment. Soil samples were taken before each treatment and tomatoes were planted in each sample to check for viable *M. hapla* larvae. Carbon dioxide and water exchange were measured 2 weeks after the last treatment, as described above. Flowers were counted twice a day for 1 month, starting 2 months after the last treatment.

RESULTS.–Leaf temperature, transpiration, leaf resistance, and number of flowers, of treated and

TABLE 1. The effect of three nematicides on temperature, leaf resistance to diffusion of water vapor, transpiration of rose leaves and flower yield

Treatment	Leaf temp (C)	Transpiration ($mg\ H_2O\ dm^{-2}\ hr^{-1}$)	Transpiration (% of control)	Leaf resistance ($sec\ cm^{-1}$)	Flower yield (number)	Infection[a]
No nematodes no treatment	25.9	357.39	100	4.08	69	
Nematodes no treatment	26.5	305.37	85.5	6.70	73	3
Mocap 1 liter/hectare	26.4	280.26	79.0	10.70	50	0
Temik 1 kg/hectare	27.6	226.20	63.5	12.60	63	0
DBCP 1 liter/hectare	25.0	223.39	62.6	15.60	40	0
F-value	2.08	2.44		2.07	10.46	
Standard error	0.67	36.10		2.60	0.79	

[a] Average gall index of eight replicates: 0 = no galls; 5 = heavy gall development.

TABLE 2. The effect of three nematicides on photosynthesis and chlorophyll content of rose leaves

Treatment	Photosynthesis ($mg\ CO_2\ dm^{-2}\ hr^{-1}$)	Photosynthesis (% of control)	Chlorophyll content ($mg\text{-}dm^{-2}$)	Photosynthesis ($mg\ CO_2\ mg^{-1}$ chlorophyll)
No nematodes no treatment	3.62	100	8.80	1.41
Nematodes no treatment	3.29	96	9.01	1.20
Mocap 1 liter/hectare	2.32	63.7	6.56	1.14
Temik 1 kg/hectare	2.05	56.8	6.78	0.97
DBCP 1 liter/hectare	1.96	54.0	5.89	1.07
F-value	668		4.78	0.78
Standard error	0.242		0.644	0.187

TABLE 3. The effect of DBCP on transpiration, leaf resistance to water vapor diffusion, photosynthesis, and chlorophyll content of rose leaves, and on rose yield in a commercial greenhouse

Treatment	Transpiration ($mg\ H_2O\ dm^{-2}\ hr^{-1}$)	Leaf resistance ($sec\ cm^{-1}$)	Photosynthesis ($mg\ CO_2\ dm^{-2}\ hr^{-1}$)	Chlorophyll ($mg\ dm^{-2}$)	Number of flowers
Nematodes no treatment	440	33	6.6	7.14	1,843
DBCP 0.2 liter/hectare	414	36	6.8	7.14	1,824
DBCP 0.51 liter/hectare	385	4.5	5.5	7.27	1,502
F-value	1.43	1.80	1.52	1.01	6.29
Standard error	22.7	0.8	5.6	0.5	88.2

control plants, are shown in Table 1. Leaf temperature was about 1-2 C above the surrounding air temperature, with no variation among treatments. Transpiration was reduced slightly in the nontreated, inoculated plants, and considerably (38%) in plants treated with Temik and DBCP. Leaf resistance to water vapor loss increased in reaction to the treatments. An increase of about 38% in resistance to water loss was observed in the infected, nontreated plants. It is apparent that the nematicides increased leaf resistance to water loss in the order Mocap<Temik<DBCP; the last caused an almost fourfold increase in resistance to water loss than the control. Photosynthesis per unit leaf area was almost the same in the inoculated and the control plants. However, a greater reduction occurred following the use of chemicals, with Temik and DBCP giving the lowest values (Table 2). No significant changes were noticed in nematode-infested, nontreated plants.

Chlorophyll content was reduced by all chemicals, whereas no difference was seen between inoculated and noninoculated plants (Table 2). Chlorophyll content per unit leaf area decreased in the order Mocap>Temik>DBCP. The reduction with DBCP amounted to about 50%. No significant differences were observed in the photosynthesis as calculated per unit of chlorophyll, although the values obtained in the treated plants were slightly lower. It is apparent that the three nematicides tested decreased photosynthesis relatively more than transpiration. The reduction of photosynthesis amounted to about 50% with DBCP compared to 38% in transpiration.

The yield of flowers was reduced by all three nematicides, especially by DBCP (52%) and Mocap (28%) (Table 1).

In the large-scale experiment it was found that 0.5 liter/hectare DBCP reduced the number of flowers about 20% and 0.2 liter/100 l/hectare DBCP did not reduce the number at all, as compared with the control (Table 3). Photosynthesis and transpiration were reduced, and leaf resistance to water loss was increased in the 0.5 liter/hectare treatment. Due to variability in the commercial field, differences were not significant.

DISCUSSION.–Our observations that the nematode infestation did not affect photosynthesis or chlorophyll content are consistent with those of Bergeson (1). On the other hand, high rates of the nematicides caused acute damage; i.e., reduction in transpiration and photosynthesis followed by yellowing of the leaves and leaf drop. The reduction in transpiration and increase in leaf resistance to water loss indicates the stomata were closing. However, photosynthesis was reduced by the chemicals to a greater extent than might have been expected from stomatal closure, since partial closure of stomata should decrease transpiration relatively more than photosynthesis (4, 9). Since leaf temperature did not vary among treatments in our experiment, greater decrease in photosynthesis indicates an effect on the so-called "mesophyll resistance" (4).

Further support for a nematicide effect on mesophyll resistance is obtained from calculations made either by Gaastra's method (4), in which the equation

$$P = \frac{[CO_2]\ air}{r_m + r_s}$$

is used, or by the equation

$$P = \frac{[CO_2]\ air\text{-}r}{r_m + r_s}$$

[which has been used by others (2, 5, 8)] where P is net photosynthesis, r_m is mesophyll resistance, r_s is stomatal resistance and r is CO_2 compensation point (50 μl/liter for roses). The values obtained by the later method were 38.5 sec-cm^{-1} for the control and 43.2, 52.5, and 67.7 sec-cm^{-1} for Mocap, Temik, and DBCP, respectively. These values are inversely related to the net photosynthesis rate.

Thus, it may be concluded that three factors are involved in the reduction of photosynthesis; i.e., stomatal resistance, mesophyll resistance, and chlorophyll content. Since net photosynthesis per mg of chlorophyll did not vary significantly, it might be that the increase in mesophyll resistance was due mainly to reduction in chlorophyll content. At the present time we have no explanation for the mode of action of bromine and the other active ingredients in the nematicides tested on chlorophyll production or destruction in the leaves.

The results of the experiment in the commercial greenhouse showed that a concentration of 0.5 liter/hectare DBCP was still phytotoxic to roses and reduced yield without any visible toxic symptoms. Thus a method of early detection is important in evaluating nematicide action on plants.

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