Phytotoxicity of Fosetyl Al and Phosphonic Acid to Maize During Production of Vesicular-Arbuscular Mycorrhizal Inoculum

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ABSTRACT

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Research into the function of vesicular-arbuscular mycorrhizae (VAM) requires production of sufficient quantities of high-quality inoculum. Contamination of one of our Glomus cultures on maize with Pythium myriotylum prompted us to assess the effects of anti-oomycete fungicides on colonization of maize by strains of Glomus mosseae, G. macrocarpum, G. etunicatum, and G. microcarpum. Following mycorrhizal inoculation, partially sterilized soil in pots was treated with fungicides at rates recommended for Pythium control: 1) metalaxyl (0.05% Ridomil 250WP, 100 ml per pot as a soil drench, or 2.5 g of Ridomil 50G per 10 L of soil), 2) fosetyl Al (0.1% Aliette 740WP, 200 ml per pot as a soil drench), and 3) phosphonic acid (0.25% Foli-R-Fos 400, 100 ml per pot as a soil drench), all applied before planting the maize. Plants treated with fosetyl Al or phosphonic acid had stunted roots and tops that were rosetted and spindly with white variegated streaking of the leaves, symptomatic of induced zinc deficiency. Percent colonization of the roots was slightly decreased by fosetyl Al and phosphonic acid, but the total length of mycorrhizal root was markedly decreased. Metalaxyl had no phytotoxic effects on maize. Excellent mycorrhizal colonization of the Glomus-inoculated pots was obtained with the metalaxyl treatments, indicating that this fungicide did not adversely affect VAM cultures. Metalaxyl (granular form) is recommended for routine addition to open-pot cultures of VAM fungi on maize.

Research on the function of vesiculararbuscular mycorrhizae (VAM) requires production of high-quality, uncontaminated inoculum. We presently produce inoculum from single-spore cultures placed on the roots of host plants in open-pot culture, which are grown in glasshouse compartments in pots of sterilized soil watered by capillarity (9). In July 1988, some of the maize (Zea mays L.) plants inoculated with a particular culture of Glomus sp. were stunted, and examination of the primary root system showed rotting. A fungus was isolated and identified by the CAB International Mycological Institute as Pythium myriotylum Drechs., which is pathogenic to many plant species, including tomato (Lycopersicon esculentum Mill.), rye (Secale cereale L.), wheat (Triticum aestivum L.), oats (Avena sativa L.), sorghum (Sorghum bicolor (L.) Moench), maize (22), sugarcane (Saccharum officinarum L.), and cowpea (Vigna unguiculata (L.) Walp. subsp. unguiculata) (6). This pathogen has an optimum temperature of 37 C; our isolate was capable of causing stunting of inoculated sorghum seedlings (R. L. Dodman, personal communication).

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Pythium spp. may interfere with VAM colonization by reducing root growth, damaging roots, and altering root exudation; they also compete with VAM fungi during the first few weeks of plant growth, when initial mycorrhizal colonization occurs (1). Reduction in VAM colonization ultimately leads to reduced spore production, so that both quality and quantity of VAM inoculum are affected. The multiplication of a pathogen such as Pythium in VAM cultures poses problems in the use of VAM inoculum for research or commercial purposes.

The objective of this research was to examine the effects of metalaxyl, fosetyl Al, and phosphonic acid, used at rates recommended for Pythium control, on mycorrhizal colonization of the roots of maize by various Glomus species.

MATERIALS AND METHODS

Treatments. A pot experiment was initiated in December 1988 with five fungicide treatments in a factorial design with five different Glomus inoculum treatments and three replications. The fungicide treatments were as follows: 1) nil; 2) metalaxyl (100 ml of 0.05% Ridomil 250WP, i.e., 0.0125 g a.i. per pot of 1,300 g oven-dry soil-sand mix) as a soil drench; 3) metalaxyl (2.5 g of Ridomil 50G per 10 L of soil, i.e., 0.0188 g a.i. per pot) as granules; 4) fosetyl Al (200 ml of 0.1% Aliette 740WP, i.e., 0.148 g a.i. per pot) as a soil drench; and 5) phosphonic acid (100 ml of 0.25% Foli-R-Fos 400, i.e., 0.1 g a.i. per pot) (H_3PO_3) as a soil drench.

The Glomus treatments were: 1) nil, 2) Glomus mosseae (Nicol. & Gerd.) Gerd. & Trappe (strain Schmelzer 43), 3) G. macrocarpum Tul. & Tul. (strain Schmelzer 42), 4) G. etunicatum Becker & Gerd. (strain Emerald 7), and 5) G. microcarpum Tul. & Tul. (strain Emerald 8A).

Inoculum. All strains were previously isolated from clay soils (Vertisols) in the grain-producing regions of Queensland. Glomus inoculum was grown on roots of maize (cv. Pioneer 3906) from singlespore increases, collected on a 63-µm sieve, and cleaned by sucrose centrifugation (2). The spores were surface sterilized in a solution of chloramine-T (20 g L⁻¹) and streptomycin (200 mg L⁻¹) for 20 min, followed by thorough rinsing in sterile deionized water (17). To inoculate, the spores were added in suspension to each pot of soil-sand mix as 10-ml aliquots, and mixed thoroughly.

All fungicides were applied before planting. The granules of metalaxyl were mixed throughout the soil. All others were applied as drenches (100 ml) after pots were placed on the sand trays. One pregerminated maize seed (Pioneer cv. 3906) was planted in each pot.

Plant growth. The potting medium was a 50:50 (w/w) mix of coarse sand and a black earth (Vertisol) from Wellcamp, Queensland, containing 78% montmorillonitic clay, 20 mg kg⁻¹ of extractable P (5), pH 8.2. It was partially sterilized to kill native VAM fungi by aerated-steam treatment at 60 C for 30 min (21). Each pot of 14 cm diameter contained the equivalent of 1,300 g of oven-dry mix. A basal fertilizer treatment of 2 g of slow-release Osmocote (Sierra Australia Pty. Ltd.) was mixed through the soil medium so that each pot received 0.17 g NO₃-N, 0.19 g NH₄-N, 0.05 g P, 0.20 g K, 0.01 g Ca, and 0.08 g S. Pots were embedded in sand in trays in a bottom-watering system similar to that devised at Rothamsted Experimental Station (B. Mosse and C. Clarke, personal communication). The sand (packed into 1-m-long PVC trays) was wetted via foam wicks at each end of the trays dipping into gutters of water. The water was drawn up into the sand and then into the soil by capillary action.

Plant height was measured 13, 22, 30, 37, 44, and 51 days after planting. Stem diameter, fresh and dry weights, and phosphorus (P), zinc (Zn), and nitrogen (N) contents of tops were determined at 51 days. Uptakes of P, Zn, and N were calculated by multiplying dry weight of the tops by nutrient concentration. All roots were extracted from the soil by wet-

sieving through a 425-µm mesh screen (7), and total weight (less prop roots) was recorded.

From each pot, a subsample (0.5 g) of undried roots was cleared and stained with trypan blue (19) to determine percent colonization of the roots by Glomus spp., total root length per plant, and Glomus-colonized root length using the grid intersect method (10). Percent colonization data and root length data

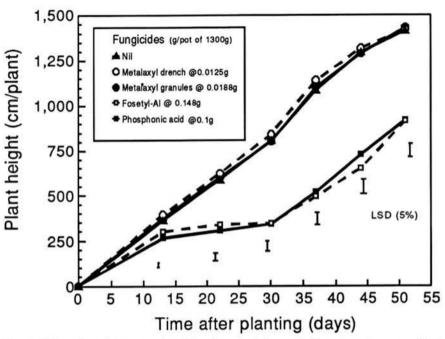


Fig. 1. Effect of anti-oomycete fungicides (mean of five vesicular-arbuscular mycorrhizal inoculum treatments) on height of maize plants. Fungicide rates (g/pot of 1,300 g soil-sand mix): metalaxyl drench, 0.0125 g; metalaxyl granules, 0.0188 g; fosetyl Al, 0.148 g; and phosphonic acid, 0.1 g.

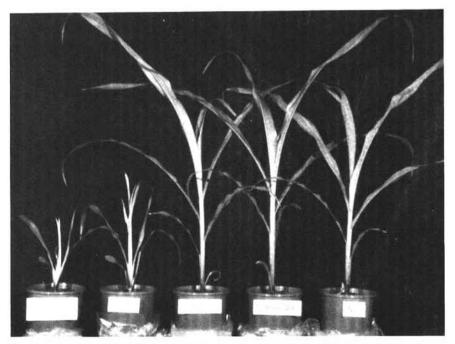


Fig. 2. Maize plants grown with phosphonate fungicides (phosphonic [phosphorous] acid, 0.1 g/pot, or Aliette, 0.148 g of fosetyl Al/pot) were stunted, rosetted, and spindly, with chlorotic striping on the leaves, compared with those grown with metalaxyl (Ridomil 50G, 0.0188 g metalaxyl/pot, or Ridomil 250WP, 0.0125 g metalaxyl/pot) or no fungicide.

were transformed by arcsine $\sqrt{\%}$ and $\ln(x+1)$, respectively; and equivalent means were calculated.

The tops and roots of an extra set of 18 plants (six treatments × three replications) were analyzed for phosphonic acid (13). The six treatments comprised the factorial combinations of two inoculum treatments (nil inoculum or G. mosseae inoculum) × three fungicide treatments (nil, fosetyl Al, and phosphonic acid). The three replicates of each treatment were bulked to obtain enough material for the analysis (>20 g fresh weight).

RESULTS

Plant growth. The effects of the fungicide treatments on plant height are shown in Figure 1. By 13 days after planting, the pregerminated seeds, all plants treated with either fosetyl Al or phosphonic acid, were significantly shorter than those receiving no fungicide or metalaxyl (either as drench or granules). These plants did not increase in height between 13 and 30 days, after which height increased again. Affected plants were stunted and also rosetted and spindly, with white variegated striping of the leaves (Fig. 2). This effect was consistent across all inoculum treatments.

There was no significant effect of inoculum treatment on plant height until day 30 (Fig. 3). At day 44, the plants without Glomus were significantly shorter than those with other inocula except G. microcarpum (Fig. 3). By day 51, those plants receiving G. mosseae or G. macrocarpum were, on average, reduced in height by 22% by fosetyl Al or phosphonic acid application; whereas plants not inoculated with Glomus or inoculated with G. etunicatum or G. microcarpum were reduced in height by 45% by application of the phosphonate fungicides (as shown by a significant inoculum × fungicide interaction in the statistical analysis, Table 1). Maize plants inoculated with Glomus had significantly greater dry weights than did uninoculated plants, and when averaged across all fungicide treatments, dry weights of G. macrocarpum-inoculated plants were the greatest (Table 1 and Fig. 4). The mean dry weights of plant tops in treatments receiving fosetyl Al or phosphonic acid were one-fifth of those receiving either of the metalaxyl treatments or no fungicide (Fig. 5). Plants that received no fungicide or either of the two metalaxyl treatments had significantly greater root weight and length than those receiving fosetyl Al or phosphonic acid (Table 1). Plants inoculated with G. mosseae had the highest root weight compared to other inoculum treatments. A highly significant (P < 0.01) inoculum × fungicide interaction indicated that the magnitude of the difference in root length between fungicide treatments was influenced by the type of inoculum received; i.e., plants with G. etunicatum and G. microcarpum were more severely affected by the application of either phosphonate fungicide than were those with G. mosseae, G. macrocarpum, or no VAM inoculum.

Inoculation with Glomus spp. (except G. macrocarpum) significantly increased phosphate concentration and uptake by plants. There was also a significant effect of fungicides on P concentration. Plants receiving fosetyl Al or phosphonic acid had twice the P content of those that received metalaxyl or no fungicide. However, when P uptake was calculated, plants with the greatest P concentration had the least amount of P per plant. Plants inoculated with G. microcarpum had the highest Zn concentration. All Glomus-inoculated plants had a greater uptake of Zn than did uninoculated plants. Fungicide treatments affected Zn nutrition in the same manner as P nutrition. Plants grown in soil treated with fosetyl Al or phosphonic acid had the highest Zn concentrations in the tops, but Zn uptake by these plants was sig-

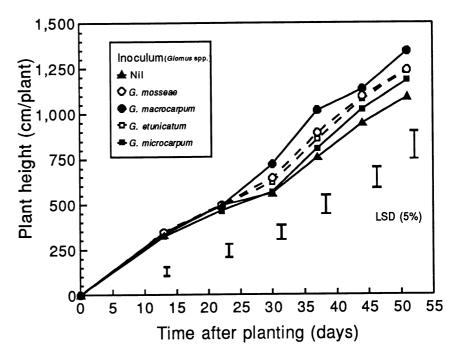


Fig. 3. Effect of inoculum treatments of various *Glomus* species (mean of five fungicide treatments) on height of maize plants.

Table 1. Effect of Glomus inoculation and anti-oomycete fungicides on mycorrhizal colonization and growth of maize at 51 days

Treatments		Plant height	Dry weight	VAM	Root length ^c	VAM-colonized root length
Inoculum	Fungicide	(cm/plant)	(g/plant)	(%) ^b	(m/plant)	(m/plant) ^c
None	FI	135.2	8.3	0	5.8	0
Tronc	F2	121.3	7.6	0	5.8	0
	F3	136.2	9.1	0	5.8	0
	F4	77.3	1.9	0	3.4	0
	F5	76.1	2.8	0	3.9	0
G. mosseae	F1	139.2	15.3	50.8	6.1	5.6
G. Mosseuc	F2	143.3	17.1	55.2	5.9	5.5
	F3	128.0	12.4	51.7	5.6	5.1
	F4	108.0	4.4	45.4	4.5	3.8
	F5	102.0	2.6	44.2	3.8	3.1
G. macrocarpum	F1	140.5	15.6	61.5	5.9	5.6
	F2	145.7	18.9	58.9	5.9	5.6
	F3	153.2	15.7	61.7	5.5	5.3
	F4	118.2	3.9	59.0	3.7	3.4
	F5	115.0	5.1	56.3	4.2	3.8
G. etunicatum	F1	152.7	14.6	51.9	5.8	5.3
G. C.	F2	152.8	17.0	54.1	5.8	5.4
	F3	153.5	15.2	48.7	5.6	5.0
	F4	73.2	1.7	30.9	3.2	1.9
	F5	86.5	1.6	41.3	2.9	2.2
G. microcarpum	F1	140.3	13.2	53.4	5.5	5.1
	F2	146.7	17.3	55.8	6.0	5.6
	F3	145.0	13.3	55.7	5.5	5.1
	F4	80.3	2.1	43.6	3.3	2.6
	F5	80.0	2.2	45.8	3.3	2.6
LSD $(P = 0.05)$		24.1	4.4	8.6	0.6	0.5
Significance of factoria	al effects					
(F test)		*d	**	**	*	**
Inoculum Europiaido		**	**	**	**	**
Fungicide		*	. •		*	**
Inoculum $ imes$ fungicide						

^aF1 = no fungicide, F2 = metalaxyl (0.0125 g a.i./pot as soil drench of 100 ml), F3 = metalaxyl (0.0188 g a.i./pot as granules), F4 = fosetyl Al (0.148 g a.i./pot as soil drench of 200 ml), and F5 = phosphonic acid (0.1 g a.i./pot as soil drench of 100 ml).

^bMeans of arcsine $\sqrt{\%}$ transformations with equivalent means in parentheses.

^{**}Weans of $\ln(x+1)$ transformations with equivalent means in parentheses; VAM = vesicular-arbuscular mycorrhiza.

 $^{^{}d*}$ = Significant at P < 0.05; ** = significant at P < 0.01.

nificantly less than for those with metalaxyl or no fungicide treatments (Table 2).

Nitrogen concentration in the tops was highest in the plants without *Glomus* inoculum and lowest in those inoculated with *G. mosseae* and *G. macrocarpum*. Uptake of N, however, was greatest in the *G. mosseae* and *G. macrocarpum* treatments. Plants treated with fosetyl Al or phosphonic acid had greater percent N in the tops than those treated with

metalaxyl or no fungicide. However, the application of fosetyl Al or phosphonic acid decreased N uptake (Table 2).

Root colonization. The percentage of root length colonized by mycorrhizal fungi was influenced by the inoculation treatment and fungicide treatment, but there was no significant interaction between the two (Table 1). Without Glomus inoculation, percent colonization was nil. Greatest colonization was seen in plants receiving G. macrocarpum,

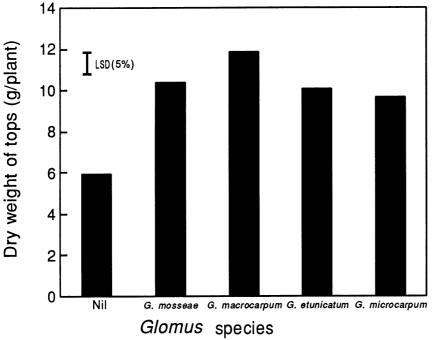


Fig. 4. Effect of *Glomus* species (mean of five fungicide treatments) on dry weight of maize tops at 51 days.

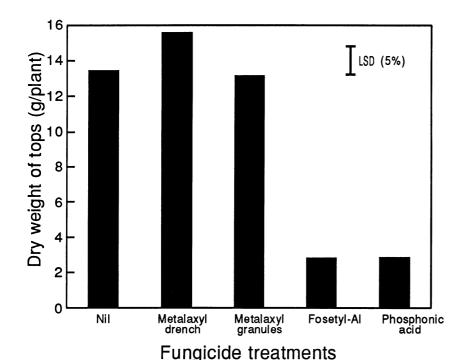


Fig. 5. Effect of anti-oomycete fungicides (mean of five inoculum treatments) on dry weight of maize tops at 51 days. Fungicide rates (grams per pot of 1,300 g of soil-sand mix): metalaxyl drench, 0.0125 g; metalaxyl granules, 0.0188 g; fosetyl Al, 0.148 g; and phosphonic acid, 0.1 g.

with an average of 74.2% (averaged across fungicide treatments, back-transformed to give the equivalent mean), which was significantly higher than in other inoculum treatments. Plants treated with fosetyl Al and phosphonic acid had significantly less colonization (equivalent means of 49.6 and 53.2%) than did the other three treatments (65.9, 68.7, and 66.0% for nil, metalaxyl drench, and metalaxyl granules, respectively).

By day 51, the number of extramatrical *Glomus* spores, expressed either on a per plant basis or per centimeter of root, was significantly lower where fosetyl Al or phosphonic acid was applied. The greatest number of spores was in plants inoculated with *G. mosseae*, and significantly fewer were seen on plants inoculated with *G. macrocarpum* and *G. etunicatum*.

Phosphite analyses. High concentrations of phosphonic acid were present in the tops and to a lesser extent in the roots of plants treated with phosphonic acid or fosetyl Al (Table 3). Virtually no phosphonic acid was detected in the tissues of plants not treated with fungicide. Of the plants receiving fungicides, those inoculated with *Glomus* had higher concentrations of phosphonic acid in the roots than did uninoculated plants (Table 3).

DISCUSSION

A major finding from this study was the reduced growth of maize by fosetyl Al and phosphonic acid applied at rates recommended for control of Pythium spp. Plants were stunted as early as 13 days after planting pregerminated seeds, and none recovered completely from this effect over the trial. The dry weight of the tops, fresh weight of roots, root length, and uptake of P, Zn, and N were all reduced significantly by both phosphonate fungicides. The phytotoxic effects we observed were very similar to symptoms described by Lucas et al (15) in maize treated with a liquid fertilizer (9% N, 8% P, 7.5% K) at rates of 47 L ha⁻¹. The P source was identified as phosphite (HPO₃)⁻², and the damaged plants showed white, variegated streaking of the leaves in mild cases and spindly, rolled, yellowish white leaves with severe toxicity, which was labeled phosphite (phosphonate) injury (15). MacIntire et al (16) concluded that concentrated phosphorous materials, e.g., phosphorous acid and calcium phosphite, were not suitable fertilizers because of phytotoxic effects (stunting and chlorosis of first true leaves) on red clover (Trifolium pratense L.), alfalfa (Medicago sativa L.), and ryegrass (Lolium rigidum Gaudin). These materials were, however, beneficial to a successive crop because of transformation of phosphite to phosphate in the soil over time.

The phytotoxic effects we observed appeared similar to Zn deficiency

symptoms described by Grundon (12). Avocado trees (Persea americana Mill.) treated with high rates of phosphonic acid showed phytotoxicity symptoms similar to Zn deficiency (3). Further research into the mode of action of the phosphonates in plant tissue is needed.

The influence of Glomus inoculation on plant growth was consistent across the fungicide treatments. All Glomusinoculated plants had greater dry weight, greater height from day 44, and much improved phosphorus nutrition than did nonmycorrhizal plants. Inoculation with the various Glomus spp. caused some differences in plant response to phosphonate fungicides. Plants inoculated with G. mosseae or G. macrocarpum were not as severely stunted as plants with G. microcarpum or G. etunicatum, or those not inoculated. The root length and VAM root length data showed a significant interaction between inoculum and fungicide treatments. The difference among the fungicide treatments was influenced by the type of inoculum and was not as great when G. mosseae and G. macrocarpum colonized the roots.

Both fosetyl Al and phosphonic acid decreased percent colonization by Glomus spp., total root length, and Glomus-colonized root length. Consequently, spore production may be

reduced by these fungicides, which the spore counts indicated, although the experiment was terminated before maximum spore production could occur. The modest decrease in colonization by Glomus spp. caused by the phosphonates was probably an indirect effect due to phytotoxicity, producing poorer plants unable to support as high a level of colonization. Clarke (4) found that neither drenching soil inoculated with G. microcarpum and other strains of VAM fungi nor immersing this inoculum in fosetyl Al at 49 g a.i. L-1 for 24 hr adversely affected the infectivity of the inoculum; i.e., the fungi were not directly

The manner in which a fungicide is applied may influence the activity of VAM fungi. Spokes et al (20) found that the mode of application of fungicides such as benomyl and captan affected the level of colonization by G. caledonium (Nicol. & Gerd.) Trappe & Gerd. in crops such as onion (Allium cepa L.) and sweet corn. When applied as seed coatings, the fungicides had no effect on colonization of the seedlings; but as soil drenches, they greatly inhibited colonization. Foliar applications of fosetyl Al at 0.3, 1.0, and 3.0 g a.i. L⁻¹ per plant per pot increased mycorrhizal colonization of leek (Allium porrum L.) roots by Glomus intraradices

Schenck & Smith by 17% and dry weight of tops by 46% at the highest rate of fungicide (14), and at 49 g a.i. L^{-1} had no reduction of mycorrhizal colonization of lettuce (Lactuca sp.) seedlings (4). Fosetyl Al was applied in our experiment at a considerably lower rate (0.74 g a.i. L⁻¹) than that used by Clarke (4) but was added to the soil as a drench. This suggests that there was possibly a greater intake of the chemical via the roots than the leaves, or that maize is more sensitive to the presence of phosphonate. Apparently, small maize plants cannot regulate the uptake of phosphonate, resulting in extremely high levels of phosphonic acid in young plants (K. G. Pegg, personal communication). Walker (23) found that the growth of young Mandarin (Citrus reticulata Blanco cv. Imperial) trees could be adversely affected by foliar sprays or soil drenching with phosphonic acid when the fungicide was used at the rate recommended for established trees (2 L a.i./100 L of water).

Our results indicate that high concentrations of phosphonic acid in maize tissues lead to phytotoxicity. Lower concentrations of phosphonic acid in the roots than in the tops indicate efficient translocation of the chemical. Furthermore, of those receiving phosphonate fungicides, Glomus-inoculated plants

Table 2. Effect of various Glomus species and anti-oomycete fungicides on mineral content of maize at 51 days

Treatments		P conc	P uptake	Zn conc	Zn uptake	N conc	N uptake
Inoculum	Fungicide ^a	(%)	(mg/plant)	(mg/kg)	$(\mu g/plant)$	(%)	(mg/plant)
None	F1	0.11	8.5	8.4	66	1.6	124
	F2	0.13	9.9	8.2	61	1.9	140
	F3	0.11	9.9	9.9	86	1.8	157
	F4	0.30	5.7	18.5	34	3.2	59
	F5	0.26	6.4	18.9	47	3.1	76
G. mosseae	F1	0.13	20.1	8.2	125	0.9	142
	F2	0.15	24.8	8.1	139	1.0	176
	F3	0.17	20.2	11.2	143	1.0	130
	F4	0.38	15.8	14.6	69	2.5	107
	F5	0.32	8.4	19.1	51	2.8	74
G. macrocarpum	F1	0.14	21.8	10.2	158	1.1	166
	F2	0.11	20.6	10.3	192	0.9	165
	F3	0.14	21.8	10.5	164	1.1	164
	F4	0.31	12.1	19.2	74	2.5	98
	F5	0.29	13.9	17.6	87	2.4	115
G. etunicatum	F1	0.16	23.6	10.5	151	1.2	177
	F2	0.15	24.8	9.1	155	1.1	190
	F3	0.15	21.9	9.2	137	1.1	172
	F4	0.39	6.9	23.6	41	3.3	57
	F5	0.33	5.2	24.5	38	3.2	49
G. microcarpum	F1	0.17	21.6	12.4	157	1.4	171
	F2	0.14	23.1	9.6	160	1.0	175
	F3	0.17	21.5	13.3	181	1.3	171
	F4	0.37	7.8	25.0	51	3.1	65
	F5	0.36	7.8	24.6	52	3.2	71
LSD ($P = 0.05$)		0.06	5.2	4.4	55	0.5	41
Significance of factor (F test)	orial effects						
Inoculum		** ^b	**	**	**	**	**
Fungicide		**	**	**	**	**	*
Inoculum × fungici	de		**	**			

^aF1 = no fungicide, F2 = metalaxyl (0.0125 g a.i./pot as soil drench of 100 ml), F3 = metalaxyl (0.0188 g a.i./pot as granules), F4 = fosetyl Al (0.148 g a.i./pot as soil drench of 200 ml), and F5 = phosphonic acid (0.1 g a.i./pot as soil drench of 100 ml). b* = Significant at P < 0.05; ** = significant at P < 0.01.

Table 3. Effect of phosphonate fungicides and vesicular-arbuscular mycorrhiza (VAM)^a inoculation on phosphonic acid concentration in maize

	VAM	Plant	tops	Roots		
Fungicide		Fresh weight (g/plant)	H ₃ PO ₃ (mg/plant) ^b	Fresh weight (g/plant)	H ₃ PO ₃ (mg/plant) ^b	
Nil	_	96.8	0.1	24.2	0.0	
	+	136.7	0.0	37.0	0.0	
Fosetyl Al	_	31.8	196.0	5.8	13.9	
•	+	49.0	150.1	7.8	51.0	
Phosphonic acid	_	25.0	138.6	4.3	13.2	
•	+	30.8	133.4	4.1	79.4	
LSD ($P = 0.05$)		29.6		10.0		

 $^{^{}a}VAM = Glomus\ mosseae.$

had higher concentrations and overall uptakes of phosphonic acid in the roots and tops than did those not inoculated. Therefore, *Glomus* may mediate the uptake of phosphonic acid as well as phosphate. Despatie et al (8) found that in onions treated with successive foliar applications of fosetyl Al, roots from mycorrhizal plants contained more phosphonic acid than did roots from nonmycorrhizal plants.

Metalaxyl at rates up to 9.0 kg ha⁻¹ applied to the soil before planting had no adverse effect on mycorrhizal colonization or growth of sour orange (Citrus aurantium L.) inoculated with G. etunicatum (18). The addition of metalaxyl at 0.92 and 2.4 mg kg⁻¹ caused respective increases of 11 and 9% in VAM colonization of soybean (Glycine max (L.) Merr.) inoculated with G. mosseae, and there was an increase from 57 to 72\% in colonization of roots of maize grown in an unsterilized soil-sand mix with metalaxyl added at 2.9 mg kg^{-1} (11). In nonfumigated soil, cotton (Gossypium hirsutum L.), onion, and pepper (Capsicum frutescens L.) inoculated with G. intraradices and grown in soil drenched with 100 ml of 0.1 mg of metalaxyl per liter had 2.4 to 3.4 times higher percent mycorrhizal colonization than did plants not treated with metalaxyl; but in fumigated soil, metalaxyl did not increase VAM colonization (1). However, Jabaji-Hare and Kendrick (14) in fact found that rates of 0.5, 1.0, and 2.0 mg of metalaxyl per plant as a soil drench reduced colonization of leek roots by G. intraradices by two-thirds and halved their shoot dry weights.

In the present experiment, plants grown in partially sterilized metalaxyltreated soil grew similarly to those with no fungicide, and colonization by Glomus spp. and root length were neither reduced nor significantly increased. This indicates that metalaxyl can be used safely during production of Glomus inoculum on maize to protect against chance contamination by oomycete pathogens. Since this may not be so for all hosts, further research is warranted for other plant species.

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LITERATURE CITED

- Afek, U., Menge, J. A., and Johnson, E. L. V. 1990. Effect of *Pythium ultimum* and metalaxyl treatments on root length and mycorrhizal colonization of cotton, onion, and pepper. Plant Dis. 74:117-120.
- Allen, M. F., Moore, T. S., Jr., Christensen, M., and Stanton, N. 1979. Growth of vesicular-arbuscular mycorrhizal and non-mycorrhizal Bouteloua gracilis in a defined medium. Mycologia 71:666-669.
- Broadley, R. H., ed. 1991. Avocado pests and disorders. Page 35 in: Inf. Ser. Q190013 -Queensl. Dep. Primary Ind.
- Clarke, C. A. 1978. Effects of pesticides on VA mycorrhiza. Pages 236-237 in: Rothamsted Exp. Sta. Rep. 1978. Part 1.
- Colwell, J. D. 1963. The estimation of phosphorus fertiliser requirements of wheat in southern New South Wales by soil analysis. Aust. J. Exp. Agric. Anim. Husb. 3:190-197.
- 6. Croft, B. J. 1988. Root rot of cowpeas caused

- by Pythium myriotylum in northern Queensland. Australas. Plant Pathol. 18:8-9.
- Daniels, B. A., and Skipper, H. D. 1982. Methods for the recovery and quantitative estimation of propagules from the soil. Pages 29-35 in: Methods and Principles of Mycorrhizal Research. N. C. Schenck, ed. American Phytopathological Society, St. Paul, MN.
- Despatie, S., Furlan, V., and Fortin, J. A. 1989. Effects of successive applications of fosetyl-Al on growth of Allium cepa L. associated with endomycorrhizal fungi. Plant Soil 113:175-180.
- Fiske, M. L., and Thompson, J. P. 1988. Single endophyte pot-culture at the Queensland Wheat Research Institute. Page 40 in: Symposium on Vesicular-arbuscular Mycorrhizae (VAM) in Agriculture and Horticulture, Toowoomba. (Abstr.) Aust. Inst. Agric. Sci., Darling Downs Sub-Branch.
- Giovanetti, M., and Mosse, B. 1980. An evaluation of techniques for measuring vesiculararbuscular mycorrhizal infection in roots. New Phytol. 84:489-500.
- Groth, D. E., and Martinson, C. A. 1983. Increased endomycorrhizal infection of maize and soybeans after soil treatment and metalaxyl. Plant Dis. 67:1377-1378.
- Grundon, N. J. 1987. Hungry crops: A guide to nutrient deficiencies in field crops. Queensl. Dep. Primary Ind.
- Hargreaves, P. A., and Ruddle, L. J. 1990. Analysis of residues of phosphonate in plant material. Australas. Plant Pathol. 19:137.
- 14. Jabaji-Hare, S. H., and Kendrick, W. B. 1987. Response of an endomycorrhizal fungus in Allium porrum L. to different concentrations of the systemic fungicides metalaxyl (Ridomil®) and fosetyl-Al (Aliette®). Soil Biol. Biochem. 19:95-99.
- Lucas, R. E., Warncke, D. D., and Thorpe, V. A. 1979. Phosphite injury to corn. Agron. J. 71:1063-1065.
- MacIntire, W. H., Winterberg, S. H., Hardin, L. J., Sterges, A. J., and Clements, L. B. 1950. Fertilizer evaluation of certain phosphorus, phosphorous, and phosphoric materials by means of pot cultures. Agron. J. 42:543-549.
- Mosse, B., and Hepper, C. 1975. Vesiculararbuscular mycorrhizal infections in root organ cultures. Physiol. Plant Pathol. 5:215-223.
- Nemec, S. 1980. Effects of 11 fungicides on endomycorrhizal development in sour orange. Can. J. Bot. 58:522-526.
- Phillips, J. M., and Hayman, D. S. 1970. Improved procedures for clearing roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection.
 Trans. Br. Mycol. Soc. 55:158-162.
- Spokes, J. R., Hayman, D. S., and Kandasamy, D. 1989. The effects of fungicide-coated seeds on the establishment of VA mycorrhizal infection. Ann. Appl. Biol. 115:237-241.
- Thompson, J. P. 1990. Soil sterilisation methods to show VA-mycorrhizae aid P and Zn nutrition of wheat in vertisols. Soil Biol. Biochem. 22:229-240.
- van der Plaats-Niterink, A. J. 1981. Monograph of the Genus *Pythium*. Pages 106-108 in: Studies Mycol. No. 21.
- 23. Walker, G. E. 1989. Phytotoxicity in mandarins caused by phosphorous acid. Australas. Plant Pathol. 18:57-59.

^bValues are from analysis of a composite of three replicate plants.