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Evaluation of Nutrient Phosphite for the Control of Phytophthora Shoot Blight on Annual Vinca[1]

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Abstract

Annual vinca (*Catharanthus roseus*) were grown in containers in a greenhouse and irrigated with a complete nutrient solution containing phosphite (0, 0.1, 0.3 mM), phosphate (0, 0.3, 0.5 mM), or combinations of these two compounds as a source of phosphorus. After 2 weeks, the plants were sprayed with a *Phytophthora nicotianae* zoospore inoculum to evaluate the potential for phosphite to protect annual vinca from Phytophthora shoot blight. To determine the extent and duration of protection from shoot blight provided by phosphite foliar applications, the plants were treated with phosphite foliar sprays (0.5 mM) at various intervals, and then inoculated. Application of phosphite to the soil/roots provided no protection from Phytophthora shoot blight. However, foliar applications of phosphite at a concentration of 0.5 mM at three to six day intervals gave protection similar to foliar applications of Aliette fungicide at 3 g/liter (2.5 lb/100 gal) applied at 14 day intervals.

Index words: *Phytophthora nicotianae*, *Phytophthora parasitica*, disease control, phosphonate.

Species used in this study: annual vinca (*Catharanthus roseus* (L.) G. Don) 'Little Bright Eye' and 'Pacifica Red'.

Significance to the Nursery Industry

Potassium phosphonate or phosphite is marketed as a phosphorus nutrient supplement. This product also has fungicidal properties against oomycetous fungi such as *Phytophthora* spp. Phytophthora shoot blight causes serious losses in landscape plantings of annual vinca across the southeastern United States. In nurseries, the causal pathogen, *P. nicotianae*, can be spread through recycled irrigation water. In this study, we determined that foliar applications of a commercial phosphite foliar nutrient applied at 0.125 ml/liter (1.6 oz/100 gal) at intervals of three to six days provided protection similar to that of Aliette applied at 3 g/liter (2.5 lb/100 gal) at 14 day intervals. This suggests regular applications of a phosphite nutrient through an overhead fertigation system could suppress Phytophthora-incited foliar diseases at a relatively low cost.

Introduction

Due to its almost summer-long display of colorful flowers, as well as heat and drought tolerance, annual vinca (*Catharanthus roseus* (L.) G. Don) remains a popular bedding plant throughout the southern United States. However, it is very susceptible to Phytophthora shoot blight, which is caused primarily by *Phytophthora nicotianae* Breda de Haan (synonym = *P. parasitica* Dastur). This disease is very destructive in landscape plantings throughout the Southeast (5) and has been found in production greenhouses (4) and nurseries. In greenhouses and nurseries, recycled irrigation water is a potential source of disease inoculum for waterborne pathogens such as *P. nicotianae* (6). McDonald et al. (8) showed that irrigation of nursery crops with recycled water could result in contamination of container crops with pythiaceous fungal species, including *Phytophthora* spp. Hong et al. (6) demonstrated Phytophthora shoot blight infection of annual vinca by irrigating with recycled reservoir water, but not with well water. *Phytophthora* spp. cause serious root and shoot diseases on many other important nursery crops in addition to annual vinca (1).

Potassium phosphonate or phosphite, the potassium salt of phosphorous acid, has been shown to be effective in controlling diseases caused by several *Phytophthora* species (9). Phosphite applied as a root drench protected seedlings of lupin, tobacco, and paw-paw from *P. cinnamomi*, *P. nicotianae*, and *P. palmivora*, respectively (11). Potassium phosphite applied as a foliar spray or trunk injection controlled Phytophthora root rot of avocado (2). Lovatt (7) also found that phosphorus-deficient avocado appeared to recover from this mineral deficiency when potassium phosphite was applied either as a soil drench or as a foliar spray. Pepper plants inoculated with *P. capsici* had significantly less Phytophthora crown rot when grown in a hydroponic system supplied with phosphite as opposed to phosphate; however, plant growth was reduced (3). Potassium phosphite is commercially available as Nutri-Grow® PK (Biagro Western Sales, Inc., Visalia, CA), a fertilizer supplement. In this study, we first evaluated the potential for using phosphite supplied during irrigation in a complete nutrient solution for the control of Phytophthora shoot blight of annual vinca. We then evaluated foliar applications of low concentrations of phosphite for control of Phytophthora shoot blight.

Materials and Methods

Experiment 1. Effect of forms of phosphorus applied as a soil drench. Catharanthus roseus 'Little Bright Eye' were started from seed in peat plugs, then transplanted into 1.89 liter (2 qt) containers in a pine bark medium with 2 plants per pot. The plants were grown in a greenhouse and were irrigated three times per week with nutrient solutions made with distilled water and the following compounds: 3.5 mM $Ca(NO_3)_2 \cdot 4H_2O$, 1.5 mM $MgSO_4 \cdot 7H_2O$, 1.5 mM KCl, 0.05 mM EDTA-Fe, 25 M H_3BO_3, 2 M $MnSO_4 \cdot H_2O$, 2 M $ZnSO_4 \cdot 7H_2O$, 0.5 M $CuSO_4 \cdot 5H_2O$, and 0.5 M $Na_2MoO_4 \cdot 2H_2O$. In addition to the basic nutrient solution described above, the nutrient solution treatments contained

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combinations of phosphate (H_3PO_4) at 0, 0.3, or 0.5 mM, and phosphite (H_3PO_3) at 0, 0.1, or 0.3 mM, or 0.3 mM provided with a commercial phosphite nutrient supplement (Nutri-Grow® PK, 0–28–26, Biagro Western Sales, Inc., Visalia, CA) (Table 1). All solutions were adjusted to pH 6.2 with 0.5N KOH. The solutions were applied by hand to supply 250 ml per container at each irrigation.

An isolate of *Phytophthora nicotianae*, originating from a *Catharanthus roseus* plant located in Virginia, was subcultured on V8 agar for 2 weeks to allow sporangia production. A zoospore suspension was produced by incubating pieces of the *P. nicotianae* culture in 1% sterile soil water extract (SSWE) for 4 hr. The zoospore suspension was filtered through 8 layers of cheese cloth to remove the agar blocks, large particles, and mycelium fragments, and then adjusted to desired concentrations with the SSWE.

Two weeks after starting the nutrient treatments previously described, the zoospore suspension inoculum containing 3300 zoospores per ml was applied to the foliage with a hand mister to provide 5 ml per container (2 plants). Immediately after inoculation, the plants were placed in a humidity tent overnight. Five days later, disease damage was evaluated by counting the number of diseased leaves and the number of diseased shoot tips. This experiment was repeated using *C. roseus* 'Pacifica Red'.

The experiment was analyzed as a 3 × 4 factorial using analysis of variance (SAS ANOVA), with mean separations, where appropriate, by LSD, P = 0.05. Five replications were utilized, with 2 pots per treatment (2 plants per pot), arranged in a randomized complete block design.

Experiment 2. Phosphite foliar sprays vs. phosphate drench concentrations. Seedlings of *C. roseus* 'Little Bright Eye' were potted into 1.89 liter (2 qt) containers in a pine bark medium with1 plant per pot. Initially, the plants were fertilized with soluble 24N–3.6P–10K fertilizer (Peter's 24–12–12, Scott's, Marysville, OH) applied to provide 100 ppm N. After one week the previously described nutrient solution, modified to provide phosphorus as phosphate (H_3PO_4) at 0, 0.1, 0.3 or 0.5 mM, 150 ml per container, was applied 3 days per week. The pH of all solutions was adjusted to 6.2 with 0.5N potassium hydroxide (KOH). Phosphite foliar spray treatments were also started at this time and applied every seven days to provide phosphorus as technical phosphorus acid (H_3PO_3) or as commercial phosphite at 0.5 mM. A distilled water spray treatment was included as a control. The pH of the phosphorous acid spray was adjusted to 6.2 with KOH. Applications were made with a CO_2-pressurized sprayer at a rate of 3 ml per container. After four phosphite applications (four weeks), plants were inoculated with a SSWE suspension that contained 1000 *P. nicotianae* zoospores per ml at a rate of 4 ml per container with a hand mister. Inoculated plants were maintained under a humidity tent overnight, then placed on greenhouse benches. This experiment was repeated using 12.7 cm (5 in) square, 15.2 cm (6 in) deep pots, in a pine bark medium, with four plants per pot. At 3 and 6 days after inoculation, the plants were evaluated for disease. Data collected for the first trial only included number of diseased shoot tips and diseased stems. For the repeat experiment, number of diseased leaves and percentage of diseased plants were included in addition to diseased shoot tips and stems.

The experiment was set up as a 3 × 4 factorial arranged in a randomized complete block design. The first time the experiment was done, 5 replications were used, with 2 pots (2 plants) per replication. For the repeat experiment, 4 replications were employed, with 4 pots per replication and 4 plants per pot (16 plants per replication per treatment). Data were analyzed using ANOVA with mean separations, where appropriate, by LSD, P = 0.05.

Experiment 3. Cumulative effects of phosphite applications and application/inoculation intervals. Seedlings of *C. roseus* 'Little Bright Eye' were potted as described for the repeat of Experiment 2. All plants were hand-irrigated three times per week with the basic nutrient solution described in experiment 1 that was amended with phosphorus as phosphate (H_3PO_4) at 0.1 mM. Prior to inoculation with *P. nicotianae*, randomly-selected plants were sprayed with a commercial phosphite nutrient solution providing phosphorus acid at 0.5 mM, from one to four times at one week intervals. A distilled water spray was also included as a control. The sprays were applied as described for experiment 2. Plants receiving zero to three phosphite applications were inoculated one day after the last phosphite application by spraying them with SSWE inoculum containing 1000 *P. nicotianae* zoospores per ml, as described for experiment 2. Plants receiving four phosphite applications were inoculated one, ten, or 17 days after the last phosphite application. Regardless of application number, the start of phosphite applications was timed so that all plants were inoculated at the same time. Immediately after inoculation, plants were placed in a humidity tent and maintained overnight, then placed on greenhouse benches. This experiment was repeated with some modification. For the repeat experiment, the plants receiving four phosphite applications were inoculated one, seven and 14 days after the last treatment, and a treatment was added to provide an inoculation 7 days after a single phosphite application. A randomized complete block design was utilized with 5 replications of 4 pots per treatment and 4 plants per pot. Five days after inoculation the numbers of diseased plants, leaves, and shoot

Table 1. Phosphate and phosphite nutrient combinations applied to the root zone of *Catharanthus roseus* 'Little Bright Eye' and 'Pacifica Red' and evaluated for control of phytophthora shoot blight.

		'Little Bright Eye'		'Pacifica Red'	
Phosphate mM	**Phosphite mM**	**Diseased leaves**	**Diseased shoot tips**	**Diseased leaves**	**Diseased shoot tips**
0.3	0	11.6[z]	1.2	8.1	0.9
0.5	0	13.2	2.0	6.9	0.5
0.0	0.1	7.8	1.0	5.6	0.6
0.3	0.1	12.8	2.0	5.4	0.5
0.0	0.3	2.4	0.4	7.2	0.4
0.3	0.3	10.6	1.6	3.4	0.7
0.5	0.3	15.0	1.6	7.7	0.3
0.0	0.3 com.[y]	6.2	1.0	3.6	0.2
0.3	0.3 com.	10.2	1.4	1.3	0.2
Phosphate		***[x]	***	NS	NS
Phosphite		NS	NS	NS	NS
Phosphate × phosphite		NS	NS	NS	NS

[z]Values for each treatment are a mean of 5 replications with 2 containers per replication.

[y]Commercial phosphite applied as Nutri-Grow® PK foliar nutrient.

[x]Statistical significance of the main effects of and interactions among phosphate and phosphite treatments. NS = not significant, *** = significant at P ≤ .001.

tips were counted. Data were analyzed with ANOVA, with mean separation by LSD, P = 0.05.

Experiment 4. Impact of phosphite foliar application interval on disease control efficacy. Seedlings of *C. roseus* 'Little Bright Eye' were potted as described for experiment 2. The plants were irrigated as needed and fertilized once a week with Peter's 24N–3.6P–10K diluted to provide 200 ppm nitrogen. Beginning one week after potting, foliar applications of a phosphite nutrient solution that provided phosphorus acid at 0.5 mM were made at 1, 2, 3, 4, 5, 6 or 7 day intervals. For comparison, Aliette® WDG (Bayer Environmental Science, Montvale, NJ), an aluminum phosphonate fungicide commonly recommended for Phytophthora control, was applied at intervals of 7 or 14 days at the bedding plant label rate of 3g product/liter (2.5 lb/100 gal). Untreated control plants also were included. Thirty days later, plants were inoculated by spraying with 5 ml per container of a SSWE suspension that contained 2000 zoospores per ml. The initiation and timing of the phosphite treatments were such that the inoculation of all treatments occurred at the same time but provided an interval from the last phosphite treatment to inoculation that was equal to the phosphite application intervals for each treatment. The experiment was arranged in a randomized complete block design with 6 replications of 4 pots per treatment with 4 plants per pot (16 plants per treatment per replication). Five days after inoculation the numbers of diseased plants, leaves, stems, and stem tips were counted. The data were analyzed with ANOVA, with mean separations by LSD, P = 0.05.

Results and Discussion

Experiment 1. Effect of source of phosphorus applied as a soil drench. With *C. roseus* 'Little Bright Eye' there were no interactive effects between phosphorus supplied as phosphate or phosphite on numbers of diseased leaves or shoot tips. There were also no significant differences in disease due to phosphite levels or source. However, there were significant main effects due to levels of phosphate nutrition (Table 1). Plants that received no phosphate in the nutrient solution had fewer diseased leaves and shoot tips than those receiving 0.3 or 0.5 mM phosphate. When this experiment was repeated with *C. roseus* 'Pacifica Red', there were no interaction effects, and no significant differences in disease due to phosphate or phosphite levels (Table 1). These results show that phosphite as a soil/root application provided no significant control of *Phytophthora* shoot blight on annual vinca. This differs from the results of Förster et al. (3), who obtained about 90% control of *P. capsici*-incited root and crown rot of peppers when phosphite was the phosphorus source in a hydroponic solution, and an intermediate level of control with phosphate/phosphite combinations. The difference may be due to different *Phytophthora* species and plant species involved, direct availability of phosphite to potentially infected tissues (roots) via the hydroponic solution, or direct toxicity of phosphite to *P. capsici*. In our experiments, applications were made to the roots for the control of a foliar disease. Phosphite is translocated in both the xylem and the phloem (10); however, it may be that phosphite translocation to the shoots and leaves was insufficient to provide disease control.

Experiment 2. Phosphite foliar sprays vs. phosphate drench concentrations. The first time this experiment was conducted there were no significant interaction effects between the soil-applied phosphate levels and the phosphite spray applications; however, there were significant main effects due to both phosphate levels and phosphite application. Plants that received technical and commercial phosphite sprays had less disease than the water-sprayed controls (Table 2). Also, plants that had no phosphate in the soil-applied nutrient solution had slightly but significantly less disease than the plants receiving phosphate in the nutrient solution. When the experiment was repeated, no interaction between soil-applied phosphate and phosphite spray applications was noted. The phosphite foliar sprays resulted in a reduction in numbers of diseased leaves and shoot tips, and in percentage of diseased plants (Table 3) but there was no effect due to phosphate concentrations applied to the potting medium (data not shown). The commercial phosphite product gave somewhat better disease control than did technical phosphite.

Table 2. Main effects of phosphite foliar sprays and phosphate nutrient levels on severity of Phytophthora shoot blight of *Catharanthus roseus* 'Little Bright Eye.' There were no significant phosphate × phosphite interaction effects.

Phosphite (mM)	Number of diseased shoot tips	Number of diseased stems
0.0	4.6a[z]	4.4a
0.5	0.5b	0.5b
0.5 com.[y]	1.1b	1.1b
Phosphate (mM)		
0.0	1.0b	1.4b
0.1	2.5a	1.5ab
0.3	1.9ab	2.7a
0.5	2.9a	2.3ab

[z]Means within a column followed by the same letter are not significantly different as determined by LSD, P ≤ 0.05.

[y]Commercial phosphite applied as Nutri-Grow® PK foliar nutrient.

Experiment 3. Cumulative effects of phosphite applications and application/inoculation intervals. Plants that were inoculated one day after receiving the last phosphite application had less disease than the water-treated control plants. However, there was no difference in disease due to the total number of phosphite applications received (Table 4), and plants that were inoculated 10 or 17 days after the last phosphite application had essentially the same amount of disease as the water-treated controls. When the experiment was repeated, a reduction in disease again was seen if the phosphite application and inocu-

Table 3. Main effects of phosphite foliar sprays on Phytophthora shoot blight of *Catharanthus roseus* 'Little Bright Eye.' There were no significant phosphate × phosphite interactions or phosphate main effects.

Phosphite mM	Percent diseased plants	Number of diseased leaves	Number of diseased shoot tips
0	75a[z]	39.4a	13.8a
0.5	52b	20.9b	8.4b
0.5 com.[y]	33c	11.8c	3.9c

[z]Means within a column followed by the same letter are not significantly different as determined by LSD, P ≤ 0.05.

[y]Commercial phosphite applied as Nutri-Grow® PK foliar nutrient.

Table 4. Phytophthora shoot blight development on *Catharanthus roseus* 'Little Bright Eye' treated with phosphite[z] foliar sprays at weekly intervals 0, 1, 2, 3, or 4 times, and inoculated 1, 10, or 17 days after the last treatment.

Number of applications	Days between last application and inoculation	Percent diseased plants	Number of diseased leaves	Number of diseased shoot tips
0	—	99a[y]	58a	6a
1	1	58b	14c	1c
2	1	59b	11c	2bc
3	1	53b	10c	1c
4	1	61b	14c	2bc
4	10	99a	56a	5a
4	17	94a	48b	4ab

[z]Applied as Nutri-Grow® PK foliar nutrient at the equivalent of 0.5 mM phosphorous acid.

[y]Mean separation by LSD, P ≤ 0.05.

lation interval was only one day but, in most cases, not if this interval was lengthened to 7 or 14 days (Table 5). When the application/inoculation interval was 7 days, however, greater reductions in diseased plants and leaves were obtained with four phosphite applications than with only one. These results suggest a possible cumulative protection or resistance effect from repeated phosphite applications.

Experiment 4. Impact of phosphite foliar application interval on disease control efficacy. The amount of Phytophthora shoot blight decreased as the interval between phosphite applications was reduced (Fig. 1). A one to two day application interval provided the best disease control. However, application intervals of three to six days provided control similar to Aliette® WDG applied at 3g/liter (2.5 lb/100 gal) at 14 day intervals, which is a mid-range label rate for control of Phytophthora diseases on bedding plants. The 0.5 mM phosphite concentration used in this experiment was obtained using 0.125 ml/liter (1.6 oz/100 gal) of the commercial phosphite nutrient. This concentration is much lower than the recommended 2.5 lb/100 gal rate for Aliette® WDG, and product

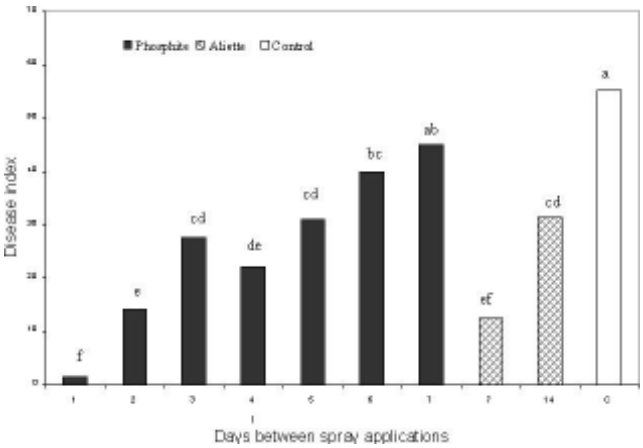

Fig. 1. Effect of phosphite foliar application frequency and corresponding Phytophthora inoculation interval on severity of Phytophthora shoot blight of annual vinca. Phosphite applied as Nutri-Grow® PK at the equivalent of 0.05 mM phosphorous acid. Aliette® WDG comparison treatments applied at 2.5 lb product per 100 gal. Disease index = (No. of diseased leaves + no. of diseased shoots + no. of diseased stems) × percentage of plants with disease symptoms. Mean separations among treatments by LSD, P ≤ 0.05.

Table 5. Phytophthora shoot blight development on *Catharanthus roseus* 'Little Bright Eye' treated with phosphite[z] foliar sprays at weekly intervals 0, 1, 2, 3, or 4 times, and inoculated 1, 7, or 14 days after the last treatment.

Number of applications	Days between last application and inoculation	Percent diseased plants	Number of diseased leaves	Number of diseased shoot tips
0	—	50ab[y]	33.2a	17.8a
1	1	24cd	12.6bc	7.2cd
2	1	20cd	13.0bc	7.4cd
3	1	18d	10.0c	4.8d
4	1	15d	8.8c	3.6d
1	7	56a	30.6a	12.0abc
4	7	36bc	10.0c	14.6ab
4	14	49ab	24.8a	11.4bc

[z]Applied as Nutri-Grow® PK foliar nutrient at the equivalent of 0.5 mM phosphorous acid.

[y]Mean separation by LSD, P ≤ 0.05.

cost is accordingly lower. For example, 100 gal of Aliette applied at 2.5 lb would have an approximate retail cost of $56.50 while 100 gal of the 0.5 mM phosphite nutrient would use approximately 76 cents worth of product. While spray applications at three to six day intervals would not be practical in a nursery or greenhouse due to excessive labor costs, low concentrations of phosphite delivered through an overhead irrigation system may be practical and could enhance control of Phytophthora-incited root rot and shoot blight in an overall Phytophthora control program in addition to providing a supplemental source of phosphorus nutrition.

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