

## MAJOR NUTRIENT INFLUENCE ON *VERTICILLIUM DAHLIAE* INFECTIONS OF *ACER SACCHARUM*

by Larry D. Smith

**Abstract.** Little is known about the influence of N, P, and K on the growth and movement of *Verticillium* in the vascular systems of woody plants. Nutrient-depleted *Acer saccharum* trees were fertilized with six treatment preparations containing varying concentrations of N, P, and K. Following the response of the trees to the fertilization, the trees were inoculated with *Verticillium dahliae*. Growth response of the trees to fertilization and the extent of colonization of the trees by the pathogen were determined. Growth of the trees was correlated to the concentration of nitrogen in the fertilizer and was described by the regression equation  $Y = 76.71 + 1.16X$  ( $r = .306$ ). Phosphorus was not correlated to new growth of the trees. The effect of potassium was negatively correlated to growth and was described by the regression equation  $Y = 129.98 - 2.25X$  ( $r = -.456$ ). The concentration of potassium was significantly correlated to the colonization of inoculated trees with the relationship described by the regression equation  $Y = 100.7 - 2.141X$  ( $r = -.579$ ). Nitrogen and phosphorus concentrations were not correlated to pathogen colonization of inoculated trees.

*Verticillium dahliae* Kleb., a pathogen of Verticillium wilt, is extremely destructive to *Acer saccharum* Marsh. and numerous other tree species in landscape sites and nurseries. In nurseries, plants with symptoms are rogued out and destroyed, but symptoms frequently develop in infected plants after being transplanted into the landscape. Most homeowners and landscape managers prefer to treat infected trees rather than replace them. However, other than the cultural practices of watering, pruning and fertilizing, no methods of control have been effective in the treatment of *Verticillium*-infected trees. Even those methods have met with limited success. Fertilization to a balance of major nutrients (4) and

fertilization with urea to promote vigorous growth (3) have been successful under some circumstances.

Little is known about the growth and movement of *Verticillium* in the vascular systems of woody plants. Sinclair, et al. (5) determined the movement of *V. dahliae* in inoculated *A. rubrum* stems in relation to the recurrence of acute symptoms and the ability of the trees to compartmentalize the pathogen. Smith and Neely (6) determined the extent of linear colonization of inoculated twigs by *V. dahliae* to assess the relative susceptibility of several species to the pathogen.

Dwinell (2) has shown that major nutrients escalate the localization of *Verticillium* in infected trees. Sinclair, et al. (5) suggest that remission of acute symptoms in infected trees, following pruning and fertilization, results from the trees' enhanced compartmentalization response.

The objective of this study was to determine the effect of nitrogen, phosphorus, and potassium levels on the growth of *A. saccharum* trees and the resulting influence of the nutrient levels on the growth of *V. dahliae* in inoculated stems of differentially fertilized trees.

### Materials and Methods

Bare-root one-year old sugar maple (*A. saccharum*) trees were selected for this study. They were potted into three-gallon plastic containers using a 60-20-20 soilless mix of graded pine bark, sphagnum peat moss, and sand. Lime was added to the medium to adjust the pH to 6.8. The

trees were grown two years without fertilization. Each year they were maintained in a green-house and watered as necessary during the spring and summer, then moved outside to a cold frame to overwinter. The trees were pruned to produce a central leader and reduce side branching. At the beginning of the third growing season, the trees were moved into a greenhouse with 53% shade and watered with an automatic drip irrigation system to provide uniform watering of all the plants.

**Tree growth response.** Twenty trees were selected at random from a uniform group for inclusion in each of seven fertilizer treatments. The trees were fertilized with one of six liquid fertilizer preparations. Water served as a control treatment. The nutrient concentrations of the treatments were (1) 20-19-18, (2) 15-10-30, (3) 15-30-15, (4) 12-6-6, (5) 10-52-8, and (6) 33-6-6 (Table 1). The six fertilizer preparations were applied five times at 3-4 week intervals. Each application consisted of 14.2 grams of fertilizer dissolved in one liter of water and uniformly distributed over the surface of the media in each container. Treatments were applied in a completely randomized design. Following the fifth fertilization, the trees were measured for length of new growth of the central leader.

**Pathogen growth response.** Twelve trees from each treatment were selected for inclusion in an inoculation study to determine the effect of differential fertilization on the growth of *V. dahliae*. Trees were selected that exhibited the greatest response to fertilization and appeared to be the

healthiest in each treatment. The trees were inoculated with *V. dahliae* three days after the last fertilization treatment. A hole was drilled through the central leader of each tree approximately ten cm above the soil line with a 5/32 in. (3.96 mm) bit. Two drops of inoculum suspension were placed into the hole, and the hole was covered with Parafilm to prevent desiccation. The inoculum suspension was a mixture of conidia and mycelial fragments that was produced by comminution of three isolates of *V. dahliae* in 500 ml of distilled water in a Waring blender. The isolates were grown on potato dextrose agar at 24C. The inoculated plants were arranged in a completely randomized design on benches in the greenhouse.

The linear distal growth of the pathogen in the central leader of each inoculated tree was determined using a two-stage isolation procedure 40 days after inoculation. The trees were excised at the point of inoculation and the main leaders were marked at 5 cm intervals. In the first stage of the procedure, isolations were attempted from the stems at 15 cm intervals. The stems were dipped in 95% alcohol and flamed. The bark was stripped away using a flamed knife. Cross-sections of the stems were aseptically made with flamed pruning shears and placed in Petri dishes containing a medium selective for *Verticillium* (1). The culture dishes were incubated at 18C for 12-14 days. They were observed with a dissecting microscope to determine the presence or absence of *Verticillium*. Immediately following the first stage isolation the stems were cut into five cm sections, marked for reference, and stored at 4C for use in the second stage isolation procedure.

The first stage isolation located the extent of the pathogen growth within a 15 cm interval. When this location had been determined, isolations were attempted on sites five cm proximal and five and ten cm distal to the most distal first stage location. The isolation technique described above was used.

## Results and Discussion

**Tree growth response.** Growth response of *A. saccharum* to the fertilizer treatments was determined by the length of new growth of the central leader of each plant in each treatment.

**Table 1. Fertilizer treatments applied to *Acer saccharum* trees at 3-4 week intervals.**

Treatment No.	Fertilizer	Analysis
1	Peter's Water Soluble Fertilizer	20-19-18*
2	Peter's Water Soluble Fertilizer	15-10-30
3	Stern's Miracle Gro	15-30-15
4	Ortho-Gro Liquid Plant Food	12-6-6
5	Warsaw Plant Starter Fertilizer	10-52-8
6	Ortho-Gro Liquid Plant Food plus Ammonium sulfate	12-6-6 21-0-0

\* All nitrogen supplied as nitrate, urea, or ammoniacal N; phosphorus supplied as available P<sub>2</sub>O<sub>5</sub>; potassium supplied as soluble K<sub>2</sub>O.

Measurements of new central leader growth lengths were subjected to analysis of variance.

Trees receiving treatment number six, which contained the highest concentration of nitrogen (33-6-6), had the greatest mean new growth, 132.3 cm (Fig. 1). However, the new growth mean was not statistically different from those of trees receiving treatments five (10-52-8) and three (15-30-15). Trees receiving the control treatment, number seven, had the smallest mean new growth, 29.0 cm, but this was not significantly different from the mean new growth of trees in treatment two (15-10-30). The new growth means for all treatments formed an overlapping array with no isolated values.

A simple analysis of the influence of the individual nutrients on new growth was conducted, ignoring the effect of the other nutrients or the interaction of the three. The extent of new growth was correlated (.05 level) to the nitrogen concentration in the treatments ( $r = .306$ ) and was described by the regression equation  $Y = 76.71 + 1.16X$  (Fig. 2). Phosphorus concentration was not correlated to new growth. However, potassium concentration was correlated (.05 level) to new growth ( $r = -.456$ ) and was described by the regression equation  $Y = 129.98 - 2.25X$  (Fig. 3). A multiple regression analysis of the effects of all nutrients combined did not provide more information about the growth of the trees in this study. Increasing nitrogen concentrations resulted in corresponding increases in growth of the fertilized trees. The negative correlation of potassium concentration on growth can

Figure 1. Growth response of *A. saccharum* to fertilization. Plants were fertilized five times at 3-4 week intervals.

Treatment No.	Fertilizer composition	Mean new growth of central leader (cm)
6	33-6-6	132.3 a*
5	10-52-8	114.8 ab
3	15-30-15	106.1 ab
1	20-19-18	93.5 b
4	12-6-6	90.1 b
2	15-10-30	56.8 c
7	water, 0-0-0	29.0 c

\* Means are from 20 plants. Means followed by the same letter are not significantly different at the .05 level by Duncan's New Multiple Range Test.

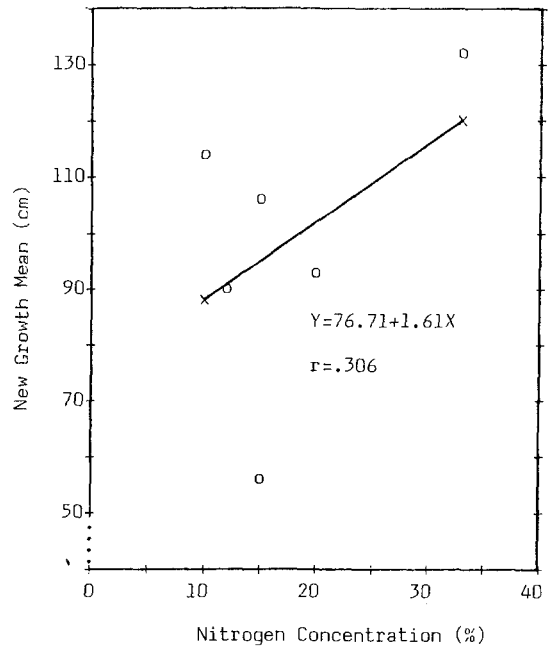


Figure 2. Effect of Nitrogen Concentration on the Growth of *A. saccharum*.

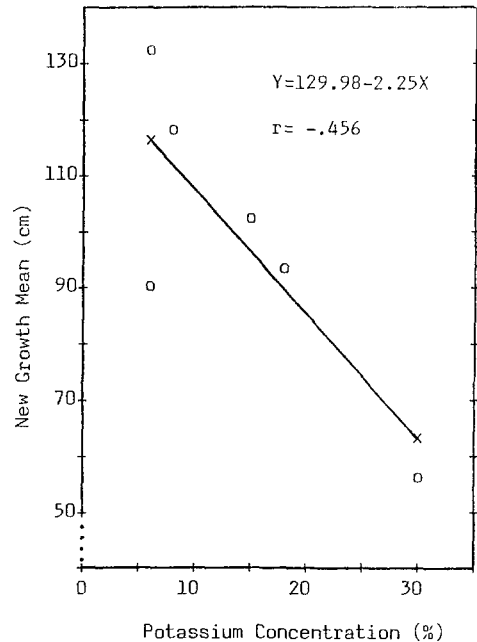


Figure 3. Effect of Potassium Concentration on the Growth of *A. saccharum*.

be understood from the standpoint that this nutrient affects metabolic processes not directly related to linear growth of the trees.

**Pathogen growth response.** The length of pathogen colonization of the central leaders of the inoculated trees was determined. The values were subjected to analysis of variance with significance stated at the .05 level.

*V. dahliae* colonized the trees in treatment number three (15-30-15) to a greater extent, 80.4 cm, than the trees receiving the other treatments (Fig. 4). However, the mean colonization distance was not significantly different from those for treatments one (20-19-18), six (33-6-6), five (10-52-8), or four (12-6-6). Colonization was least in the control treatment trees, 27.5 cm. The pathogen growth in the fertilized trees was significantly less in treatment two (15-10-30) which contained the highest potassium concentration. In all of the inoculated trees, the isolations of the pathogens were continuous along the stems. It was concluded that movement of the pathogen in each case resulted from linear growth.

The pathogen growth response in the fertilized trees, excluding the control treatment, was analyzed for linear regression relating to nutrient concentration. The concentration of potassium was significantly correlated to the growth of the pathogen ( $r = -.579$ ) with the relationship described by the regression line  $Y = 100.7 - 2.141X$  (Fig. 5). Similar analysis of the effect of nitrogen and phosphorus concentrations on pathogen growth resulted in nonsignificant correlation values. The control treatment was excluded from these analyses because the concentrations of the nutrients in the media were not determined. Increasing potassium concentrations suppressed the linear growth of the pathogen in inoculated trees.

The results of this study indicate that treatment of *Verticillium*-infected trees should include fertilization with a formulation containing abundant potassium. This study indicates that increased potassium concentrations result in a reduction of host growth and a suppression of pathogen growth. Suppression of pathogen growth in inoculated trees was not related to a reduction of nitrogen concentration since pathogen growth

Figure 4. Linear Colonization of *A. saccharum* by *Verticillium dahliae* in response to fertilization.

Treatment No.	Fertilizer composition	Mean linear colonization (cm)
3	15-30-15	80.4 a*
1	20-19-18	75.4 a
6	33-6-6	72.9 a
5	10-52-8	71.9 a
4	12-6-6	67.9 a
2	15-10-30	47.9 b
7	water, 0-0-0	27.5 c

\* Means are from 12 plants. Means followed by the same letter are not significantly different at the .05 level by Duncan's New Multiple Range Test.

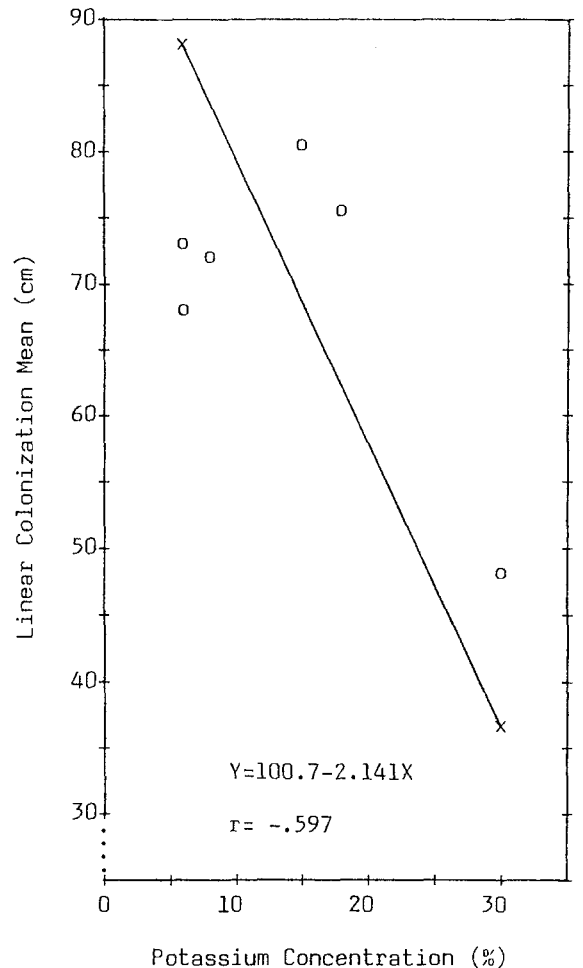


Figure 5. Effect of Potassium Concentration on the Colonization of *A. saccharum* by *Verticillium dahliae*.

was suppressed in the treatments containing high nitrogen concentrations. The mechanism of pathogen suppression is not evident from this study but it seems that pathogen suppression is desirable, even with an accompanying reduction in tree growth.

Compartmentalization of the pathogen was not examined in this study. However, an increased compartmentalization response would account for the pathogen growth reduction observed. The potassium concentrations used in this study were not sufficiently high to be toxic or inhibitory to the pathogen. More study is necessary to determine the extent of pathogen growth suppression by potassium and the mechanism involved in the suppression.

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## ABSTRACT

ANDERSON, R.G. and J.R. HARTMAN. 1983. **Symptoms of Phomopsis twig blight on weeping figs indoors.** *Interscape* 5(37): 38.

Leaf loss and twig dieback on weeping figs indoors can be caused by a disease as well as insufficient care and lack of acclimatization. *Phomopsis cinerescens* has been identified as a pathogen that caused decline of *Ficus benjamina* specimen plants in New York and Ohio. Phomopsis twig blight disease caused significant damage to weeping figs in a commercial plantscape in Kentucky and has been noted on other plantscape and homeowner weeping fig specimens. Weeping figs infected with *Phomopsis* had leaf loss and twig dieback symptoms similar to plants that lacked acclimatization or received insufficient water. Interior plantscape professionals must consider Phomopsis twig blight as a cause for continued leaf loss when cultural problems have been remedied.