Effect of Nitrogen Concentration in Juvenile Foliage of Rhododendron on Phytophthora Dieback Severity

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ABSTRACT

Hoitink, H. A. J., Watson, M. E., and Faber, W. R. 1986. Effect of nitrogen concentration in juvenile foliage of rhododendron on Phytophthora dieback severity. Plant Disease 70:292-294.

Nitrogen concentration in juvenile foliage of rhododendron cultivar Roseum Elegans was significantly and positively correlated with size and number of lesions caused by *Phytophthora cactorum*. Low numbers of tiny lesions were produced on nitrogen-deficient plants with concentrations of 0.7% in juvenile foliage. High numbers of large lesions that killed plants were produced on plants with nitrogen concentrations of 1.8–2.5% in juvenile foliage. Adding nitrogen to low-fertility plants on which small lesions had developed resulted in an increase in lesion size and eventual plant death.

During the 1960s, Phytophthora root rot of rhododendron was a serious disease in nurseries throughout the United States (8). *Phytophthora cinnamomi* Rands was the most important of eight *Phytophthora* spp. involved in the disease (8). Phytophthora dieback of rhododendron caused by *P. cactorum* (Leb. & Cohn) Schroet. and other *Phytophthora* spp. (2,8) was relatively unimportant. Epidemics were observed only in field plantings that were flooded during the growing season (8).

During the 1970s, bark from various tree species added to container media was found to suppress Phytophthora root rots (6,9,13,14). In the Midwest and some eastern states, media amended with composted hardwood bark were used (7,16). However, bark from hardwood trees in the Midwest is high in calcium and cellulose compared with pine bark (7). Producers of Ericaceae therefore replaced composted hardwood bark with pine bark to avoid negative effects of high pH caused by the high calcium content and nitrogen immobilization caused by high cellulose levels in composted hardwood bark. During the first growing season after this change in container medium composition, Phytophthora dieback epidemics developed on rhododendrons in several Ohio nurseries. Because these epidemics did not occur in nurseries that continued to use composted hardwood bark, the composition of the container medium appeared to affect dieback development.

This article presents data on effects of mineral nutrition on susceptibility of rhododendron to Phytophthora dieback. The relationship of this effect and container medium composition are discussed.

MATERIALS AND METHODS

Rooted cuttings of rhododendron cultivar Roseum Elegans were potted in 2.2-L (15-cm-tall) containers in a medium consisting of freshly milled pine bark (Kamlar Corp., Rocky Mount, NC), composted hardwood bark (Paygro, Inc., South Charleston, OH), and Canadian sphagnum peat (3:1:2, v/v, pH 5.5). The medium was amended with 0.367 kg of triple superphosphate $(0\text{-}46\text{-}0)/m^3$. Micronutrients were not added because composted hardwood bark releases adequate levels of these nutrients (7). Potted cuttings were placed in a greenhouse during May at day and night temperatures of 23–28 C and 20–22 C, respectively, without supplemental light. Cuttings were fertilized with solutions of ammonium nitrate and potassium nitrate that provided nitrogen and potassium at 0, 25, 50, 75, 125, or 200 $\mu g/ml$ each time the plants were watered. Fertility treatments were applied to three randomized complete blocks of 10 plants each.

After the first flush of new growth had

matured and as leaves of the second flush were expanding, half of all plants were inoculated (three replicates of five plants per fertility treatment). Inoculum of isolate 814 of *P. cactorum* (originally isolated from rhododendron) was cultured for 4 days on Difco lima bean broth at 22 C under continuous light. Cultures were then rinsed with distilled water at 4 C and incubated at 4 C for 30 min to induce zoospore release (15). Before inoculation, plants were incubated for 24 hr in a mist chamber (25 C, 225 μE m^{-2} sec^{-1}, 16 hr/day). A zoospore suspension (10^4/ml) was then sprayed onto foliage until runoff. Plants were incubated another 24 hr in the mist chamber and returned to the greenhouse. Lesions per plant were counted 72 hr after inoculation. In one experiment (with three replicates of 10 plants per treatment), lesion size also was measured at this time.

On the same day lesions were counted, juvenile leaves and mature leaves with attached stem tissues were removed separately from three replicates of five uninoculated plants for each fertility level, dried at 70 C to constant weight, and analyzed for concentrations of macronutrients and micronutrients. Nitrogen concentration was determined by automated Kjeldahl (Kel-Foss, Dickey-John Corp., Auburn, IL). Other elements were determined by an inductively coupled plasma spectrograph on tissue samples dry-ashed at 500 C. Total dry weight of juvenile leaves and mature leaves with attached stems of each plant also was measured. The numbers of lesions per plant for various fertility levels were regressed on the concentrations of nitrogen, phosphorus, potassium, calcium, magnesium, zinc, copper, iron, manganese, and boron in juvenile and mature rhododendron leaf tissues. A linear regression model was used in all cases. The experiment was repeated once.

Juvenile foliage was collected during July 1984 from rhododendrons produced in pine bark and hardwood bark container media and in field soil in five Ohio nurseries. Triplicate samples were taken at each location and analyzed for macronutrient and micronutrient concentrations.

RESULTS

Lesions were most numerous on plants treated with highest fertility levels. Of all the macronutrient and micronutrient concentrations measured in juvenile leaf tissue, only nitrogen and iron were positively correlated at a significant level ($P=$ 0.01) with lesion number (Fig. 1). The range and mean concentrations of each nutrient level established by the six fertility levels in juvenile leaves are presented in Table 1. Sixty-four percent of the variation in the number of lesions could be explained on the basis of nitrogen concentration. The correlation between lesion number and nitrogen concentration in mature leaves was not significant.

Iron concentration in juvenile foliage also was significantly related to lesion number. Analysis of the ammonium nitrate and potassium nitrate fertilizers revealed that both contained low levels of iron (8.8 and 5.8 μg/g, respectively). Iron, therefore, was applied inadvertently in increasing concentrations with the nitrogen and potassium fertilizer treatments. Regression of number of lesions on iron concentration in juvenile leaves explained 63% of the variation in number of lesions.

Although potassium also was applied in increasing concentrations in the fertilizer treatments, the correlation between potassium concentration in juvenile or mature leaves and application rate of potassium was small.

No significant correlation was found among dry weights and nutrient levels applied during the 4-wk period of the experiment. The number of branches per rooted cutting used in this work ranged from two to five. The short growth period and the variation in initial size of rooted cuttings limited interpretation of growth effects.

The mean nitrogen concentration in juvenile leaves of plants produced in nurseries in pine bark media ranged from 1.8 to 2.1%. Mean nitrogen concentrations in juvenile leaves of plants produced in composted hardwood bark and field plants were 1.4 and 1.2%, respectively ($LSD_{0.05} = 0.3$).

Lesion size (72 hr after inoculation) varied widely among treatments. Small lesions were formed on chlorotic plants fertilized with 25 μg each of nitrogen and potassium per milliliter of irrigation water (Table 2). At the higher fertility levels (125 and 200 μg of nitrogen and potassium), lesions expanded until the entire plant died. Only plants produced under the lowest fertility level survived.

Eight weeks after inoculation, a high concentration of fertilizer (200 μg each of nitrogen and potassium per milliliter) was applied to nutrient-deficient plants previously fertilized with the low level. As nutrient stress symptoms were corrected, lesions increased in size and plants eventually died. Lesions on control plants

Table 1. Means and range of concentrations of plant nutrients in juvenile foliage of the rhododendron cultivar Roseum Elegans

	Concentration[a]	
Nutrient	Mean	Range
N	1.420	0.77–2.12
P	2,128.000	764.9–2,389
K	19,454.000	10,844–22,258
Ca	7,601.000	4,335–9,537
Mg	3,252.000	1,735–6,068
Mn	349.700	245.1–501.6
Fe	45.090	14.49–93.02
Zn	29.300	12.79–39.72
Al	9.502	4.989–24.06
Na	1,245.800	367.5–1,627.0
B	36.610	23.19–46.95
Cu	4.937	1.507–6.615

[a] N represents percent by dry weight and all other nutrients represent μg/g dry weight of tissue (based on three replicates of five plants for each of six fertility levels).
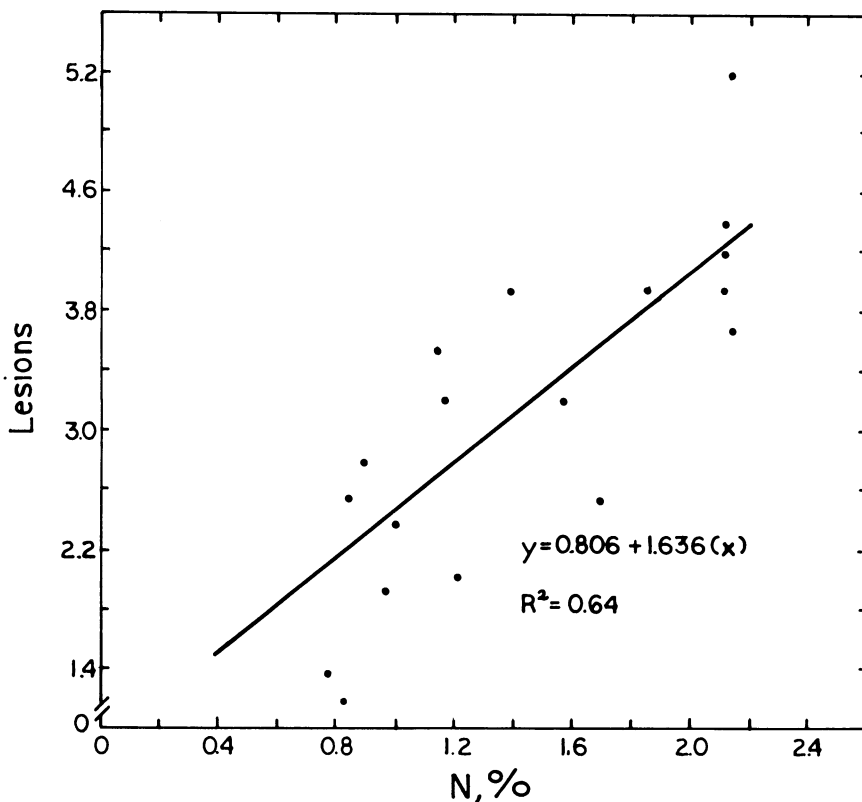
Fig. 1. Effect of nitrogen concentration in juvenile rhododendron leaf tissue on severity of Phytophthora dieback; lesion number was determined 72 hr after inoculation with zoospores.

$$y = 0.806 + 1.636(x)$$
$$R^2 = 0.64$$

Table 2. Effect of nitrogen concentration in juvenile foliage of Rhododendron cultivar Roseum Elegans on sizes of lesions caused by *Phytophthora cactorum*

Nitrogen concentration (%)	Lesion diameter[a] (mm)
0.68	23.2
1.80	73.7
2.53	74.0
$LSD_{0.05} = 41.0$	

[a] Measured 72 hr after spraying with a suspension of 10^4 zoospores per milliliter (based on three replicates of 10 plants each).

maintained under low fertility levels did not increase in size and plants did not die.

DISCUSSION

The effect of substrate fertility on severity of *Phytophthora* diseases was reviewed recently (12). High fertility increased severity of tobacco black shank, soybean root rot, eucalyptus dieback, and Ohia decline. The review established that contradictory results have been published frequently for effects of chemical factors on *Phytophthora* disease severity. This could be due in part to the lack of accepted standards for "high" vs. "low" levels of nutrition for crops produced under various conditions. Furthermore, effects of salinity caused by high levels of fertilizer on *Phytophthora* disease severity (11) and the contribution of individual or interacting cations and anions supplied with the fertilizer constituents in soil often have not been identified. Finally, the actual nutrient concentrations in tissues invaded by the *Phytophthora* spp. were not established; rather, the amounts of fertilizer supplied to plants were compared (12).

In this study, a significant linear correlation was found between nitrogen concentration in juvenile leaf tissue of rhododendron cultivar Roseum Elegans and susceptibility to Phytophthora dieback. Although a positive relationship between iron concentration and disease severity was found as well, we are unable to interpret the significance of this. At low (deficient) nitrogen concentrations (0.68%), lesions remained tiny and the number of visible lesions produced was low. At a nitrogen concentration of 1.8%, lesions expanded, numbers of lesions were high, and plants died. At very high

nitrogen concentrations (2.53%), lesions were not significantly larger than at the 1.8% nitrogen level, but the number of visible lesions increased and the plants also died. This suggests that rhododendrons possibly could be produced at low nitrogen levels to control Phytophthora dieback. This in fact may account for the low incidence of Phytophthora dieback in field-grown rhododendrons and in the landscape where soil nitrogen concentrations often are low. The mean leaf nitrogen concentration on such plants in nurseries in 1984 was 1.2%. The significant positive correlation between nitrogen concentration and susceptiblity may also explain the low incidence of dieback in nurseries where composted hardwood bark was the principal organic component in container media (1.4% nitrogen in juvenile foliage). Composted hardwood bark immobilizes nitrogen, resulting in lower amounts available for uptake by plants (4). Pine bark, whether fresh or composted, does not have this severe nitrogen-immobilizing effect (3).

The recommended concentration of nitrogen for production of rhododendron varies with cultivar (17). The recommended leaf nitrogen concentration for cultivar Roseum Elegans ranges from 1.7 to 1.9% (17). The nitrogen concentration maintained in plants of this cultivar produced in pine bark media in Ohio nurseries in 1984 ranged from 1.8 to 2.1%. This is within the range associated with high susceptibility to *P. cactorum* (Fig. 1). Excessive nitrogen, therefore, indeed may have accounted for the increased disease incidence observed in nurseries after composted hardwood bark was replaced with pine bark in media.

Plants produced under conditions of low nitrogen in the field or in composted hardwood bark may appear free of dieback but in fact have small lesions. Tiny lesions, as shown in this work, expanded as higher foliar nitrogen levels were established in infected plants; therefore, control of dieback by production of plants under low levels of nitrogen may not be an acceptable practice. Protective fungicide programs and cultural control procedures developed specifically for *Phytophthora* diseases (1,5,10) must be used in nurseries, regardless of the nutritional levels maintained during production, to avoid losses after outplanting in the landscape.

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