

Pathology and Nematology

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Daylily rust caused by *Puccinia hemerocallidis*: A new disease on daylily in the U.S.

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Index words: Rust, Daylily, Disease

Nature of Work: Few diseases affect daylily (*Hemerocallis* spp.). The most common are leaf streak (*Aureobasidium microstictum*), crown and root rot (*Sclerotium rolfsii*), soft rot (*Erwinia* spp.), and root rots (*Rhizoctonia* and *Pythium* sp.) (1). Although some of the diseases can kill plants, they are not considered to be major pests. Few other pests plague daylily other than spider mites, aphids, and thrips (1). For this reason, daylily is considered to be a relatively pest-free plant and is used heavily in residential, commercial, and municipal plantings across the country. In August 2000, a daylily sample from a commercial container nursery in Georgia was submitted to The University of Georgia Plant Disease Clinic. The submitted sample, cv. Pardon Me, exhibited symptoms of bright yellow spots and streaks on the leaves. Within the spots on both the upper and lower leaf surface, rust pustules containing urediniospores were evident. A rust disease on daylily was not known to occur in the United States (1), but had been identified in Asia and Europe (3).

Daylily rust, caused *Puccinia hemerocallidis* Thuem., is native to Asia (China, Japan, Korea, Taiwan, and Russia) (3), and may have been introduced into Georgia on infected plant material originating from Central America. The direct origin of the rust is unknown, however, since Central American producers also purchase and import plants from the United States for propagation and then sell divisions back to U.S. growers. Daylily divisions routinely enter the U.S. from Costa Rica, Guatemala, Honduras, Mexico, Bahamas, South Africa, and the Netherlands (4).

Results and Discussion: As of June 2001, daylily rust has been identified in ten U.S. states (Alabama, California, Florida, Georgia, Louisiana, Minnesota, Mississippi, Tennessee, Texas, and South Carolina). The spread of the disease has been primarily through the sale and trading of infected daylilies.

Identification of the rust pathogen found in Georgia as *Puccinia hemerocallidis* was based upon urediniospore and teliospore characteristics. Urediniospores were globose to ellipsoid and measured 19-30 x 17-22 μm (with an average size of 22 x 19 μm), corresponding to the previously reported description from Japan (3). Teliospores were initially absent from the cv. Pardon Me sample, but were found on rust-infected plants

cv. Star Struck beginning in October 2000. Teliospores differ from the published description in that many one-celled teliospores (i.e., mesospores) measuring 32-43 x 14-19 μm (with an average size of 38 x 16 μm) were produced in addition to two-celled teliospores measuring 41-53 x 16-21 μm (with an average size of 46 x 18 μm) (3). However, similar mesospores were present in a slide from an isotype specimen of *P. hemerocallidis* (US 72719) housed in the U.S. National Fungus Collection.

Daylily rust can be easily spread by wind or air currents. Daylily cv. Pardon Me was re-inoculated with the rust by shaking infected plants over uninfected plants and exposing plants to 100% humidity for 24 hours in a mist chamber. Symptoms of small yellow spots on the upper leaf surface developed within 3 to 7 days with new uredia containing urediniospores evident within 7 to 14 days after inoculation.

Daylily cultivars differ in their susceptibility to the disease. Infected plants are not killed, but infection makes the plants unmarketable. Also, repeated infection most likely will result in plant decline. Initial observations on experimentally infected daylily cultivars indicate that the cultivars Pardon Me, Lemon Yellow, Pandora's Box, Little Gypsy Vagabond, Karie Ann, Colonel Scarborough, Quannah, Ming Toy, Double Buttercup, Russian Rhapsody, Irish Ice, and Imperial Guard appear to be highly susceptible to the rust. Moderately susceptible cultivars include Happy Returns, Prelude to Love, Gertrude Condon, Stella D'Oro, Joan Senior, Butterflake, Wilson's Yellow, Star Struck, and Crystal Tide. Cultivars showing little rust infection were Mac the Knife, Yangtze, Holy Spirit, and Butterscotch Ruffles. These evaluations were conducted on donated plants in an inoculation trial in a greenhouse and do not reflect all the cultivars that can be infected with daylily rust. More evaluation trials are needed on the thousands of daylily cultivars currently grown.

Two symptom types were evident in the cultivar evaluation trials. Symptoms of rust infection were either bright yellow, discrete spots with sometimes a water-soaked border, and no necrosis such as exhibited on cv. Pardon Me or were small, water-soaked tan spots with a darker border, and some necrosis such as cv. Gertrude Condon. The necrosis in the latter symptom type may be due to a hypersensitive reaction to rust infection. Urediniospores are produced on plants with either symptom type, but production appears to be greater on plants having yellow spotting with no necrosis.

Not much is known about daylily rust. From published reports, the rust is heteroecious. Its alternate host is the herbaceous perennial, *Patrinia* spp., in the Valerianaceae family. Leaf rust has not been observed on

Patrinia spp. within the United States (2). Six species of *Patrinia* are grown in the U.S., with *P. scabiosifolia* cv. Nagoya being the most common. Studies are underway to experimentally infect *P. scabiosifolia* cv. Nagoya with daylily rust to confirm the rust's identity.

Hosta spp. are also listed in the literature as hosts of *P. hemerocallidis* (3). However, no *Hosta* plant has been infected by daylily rust in inoculation trials in Georgia, and at this time *Hosta* is not considered a host to daylily rust.

Also unknown about daylily rust is how or where the rust will survive from year to year. Daylily rust does not need its' alternate host to survive because the urediniospores produced on daylily are capable of re-infecting the same host plant leading to epidemics. In addition, there are dormant, semi-evergreen, and evergreen daylily cultivars. Theoretically, daylily rust could survive year-round on semi-evergreen and evergreen cultivars and re-infect dormant cultivars in the spring. More studies are needed to determine how the rust will survive, especially under a variety of environmental conditions.

Significance to Industry: Daylily rust is a major concern for daylily producers and gardeners. With the popularity of daylilies and the presence of daylily rust within container and field-grown nurseries, it is very likely that daylily rust will spread across the country, possibly becoming endemic in some areas. More research is needed on daylily rust's identity, genetics, survival, spread, and most importantly management, including host plant resistance and fungicide efficacy and timing.

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In Vitro Fungicide Sensitivity and Optimum Germination Temperature of the Daylily Rust Pathogen, *Puccinia hemerocallidis*

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Index words: Rust, Daylily, Fungicide, Germination

Nature of Work: A serious new rust disease on daylilies (*Hemerocallis* spp.) caused by the fungus *Puccinia hemerocallidis* has become an increasing problem for daylily growers. The rust was first identified on the variety 'Pardon Me' in Georgia in August 2000 and had not been previously reported on daylilies in the United States (1). *P. hemerocallidis* is native to Asia (China, Japan, Taiwan, USSR, and Ryukyu Islands) and may have been introduced into the United States from plant material originating from Central America (2). Daylily rust has since been identified in Florida, Alabama and South Carolina. Symptoms on the foliage range from bright yellow spots to streaks on variety 'Pardon Me', to smaller, water-soaked tan spots with darker borders on 'Gertrude Condon' (2, 3). Severe infection can kill the foliage and infected plants are not marketable.

At present very little is known about the biology of *P. hemerocallidis* on daylily. *P. hemerocallidis* is a heteroecious rust that needs two different hosts to complete its lifecycle. The uredinial/telial host is daylily in the Hemerocallidaceae family and the alternate (spermogonial/aecial) host is the herbaceous perennial, *Patrinia* spp. in the Valerianaceae family. However, the alternate host is not required for daylily infection. The uredinial or repeating spore stage on daylily can re-infect the same host plant leading to epidemics. Preliminary experiments indicate that daylily varieties differ greatly in susceptibility to the rust (3). Infection will occur on new leaves when exposed to inoculum and 100% relative humidity and leaf wetness for 24 hours. However, the precise temperature and leaf wetness required for infection are not known. Presently growers are advised to use fungicides labeled for rust on herbaceous perennials (e.g. propiconazole, azoxystrobin) but there are no data on the relative control of *P. hemerocallidis* on daylily by the different products.

As part of our preliminary studies into the biology and control of this new pathogen, our objectives of this research were to: 1) determine the optimum in vitro temperature required for germination of *P. hemerocallidis* urediniospores; and 2) determine the fungicide sensitivity of *P. hemerocallidis* urediniospores on fungicide-amended agar plates.

Rust was maintained on daylily variety 'Pardon Me' in a research greenhouse with average daily temperatures between 70 and 75 F. To encourage rust sporulation, plants were subjected to 4 minutes of misting every 60 minutes. For each experiment, urediniospores were gently scraped from new lesions and suspended in 0.5% Tween 20. Spore solutions were concentrated by centrifugation two times and the supernatant discarded. Spores were re-suspended in 0.1% Tween 20 solution at approximately 5×10^5 urediniospores ml^{-1} . For in vitro temperature experiments, four 50 μl aliquots of spore suspension were applied to a petri dish containing potato dextrose agar (PDA). Single dishes were incubated at 8 different temperatures for 18 hours and germination was then assessed by microscopic examination. Germination of a minimum 200 urediniospores were counted for each aliquot at each temperature. The affect of different fungicides on rust germination was assessed by plating urediniospores on PDA amended with varying concentrations of fungicides. Fungicides tested included: Cleary's 3336 (thiophanate-methyl), Prostar (flutaloni), Heritage (azoxystrobin), Banner Maxx (propiconazole), Touche (vinclozolin), Chipco 26019 (iprodione), Daconil Ultrex (chlorothalonil), Fore (mancozeb), and Eagle (myclobutanil). Data were analyzed by analysis of variance (ANOVA) with means separated by Fisher's Least Significant Difference (LSD) with $P = 0.05$. All experiments were repeated.

Results and Discussion: Rust urediniospores germinated between the temperatures of 50 F and 86 F on PDA (Fig. 1). Germination was not observed at the lowest (39 F) or highest (93 F) temperatures tested (Fig. 1). The inhibitory affect of high temperatures on germination suggests that the warmer temperatures associated with the summer growing season in the South will reduce the incidence of Daylily rust infection. Germination of *P. hemerocallidis* over a large range of temperatures also indicates that infection could occur over a similar range. However, this has to be verified with infection trials tested over a similar range of temperatures and by field studies.

In the fungicide experiments, *P. hemerocallidis* was most sensitive to azoxystrobin (Heritage) (Figure 2) and chlorothalonil (Daconil Ultrex) (Figure 3). Germination was only observed at the lowest fungicide concentrations tested for each fungicide. No effect on germination was observed with propiconazole (Banner Maxx), myclobutanil (Eagle), or thiophanate-methyl (Cleary's 3336). Azoxystrobin, chlorothalonil, propiconazole, and myclobutanil are all effective against rusts on various ornamentals (4) and suggested for use against *P. hemerocallidis* on daylily (3). Our preliminary data indicates that azoxystrobin and chlorothalonil could give good control of rust on Daylily. Fungicide trials are currently being conducted at the University of Georgia.

Significance to Industry: Control measures for daylily rust will increase production costs on a crop that has traditionally been identified as pest-free. Identifying the temperature ranges for germination and the fungicide resistance of *P. hemerocallidis* should help determine when threat of infection is highest and the best selection of fungicides for optimum disease control.

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Figure 1. Germination of *P. hemerocallidis* urediniospores at various temperatures

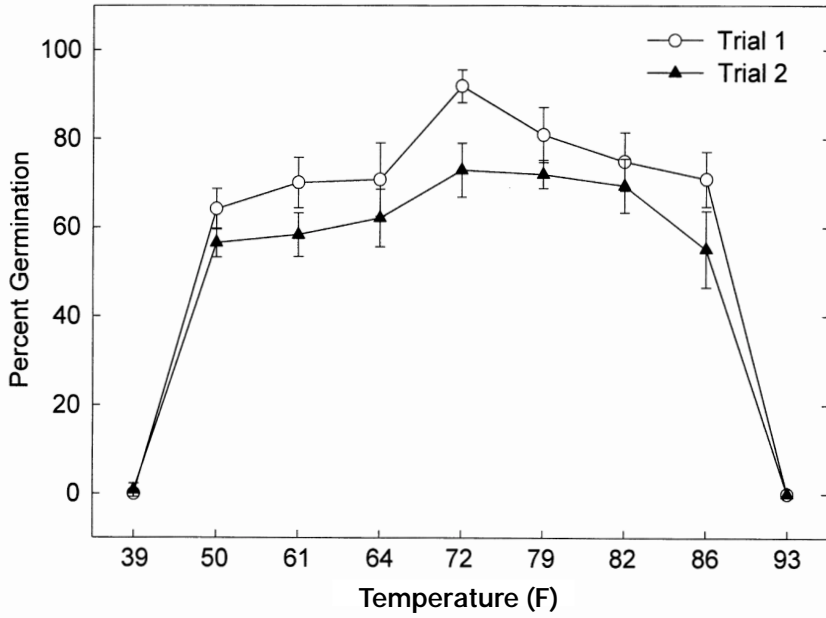


Figure 2. Germination of *P. hemerocallidis* urediniospores on fungicide-amended media

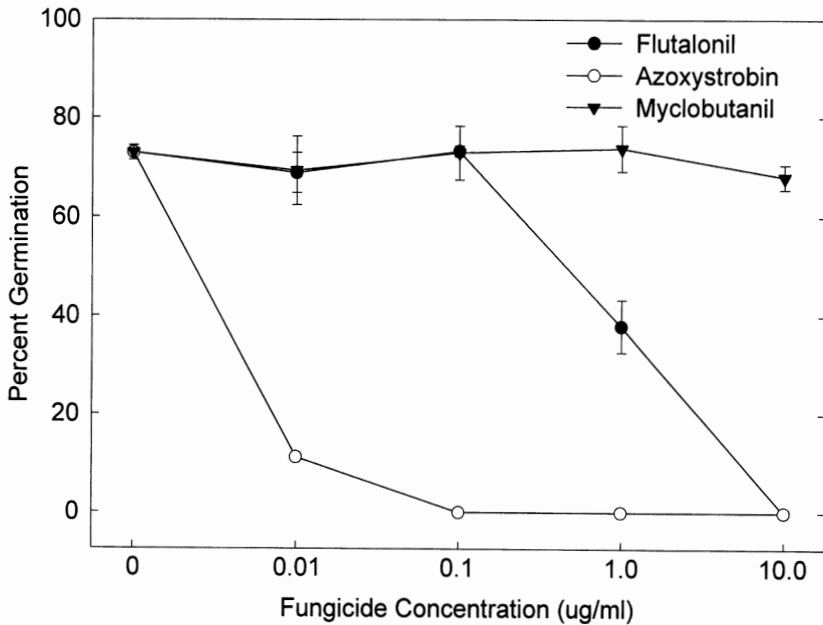
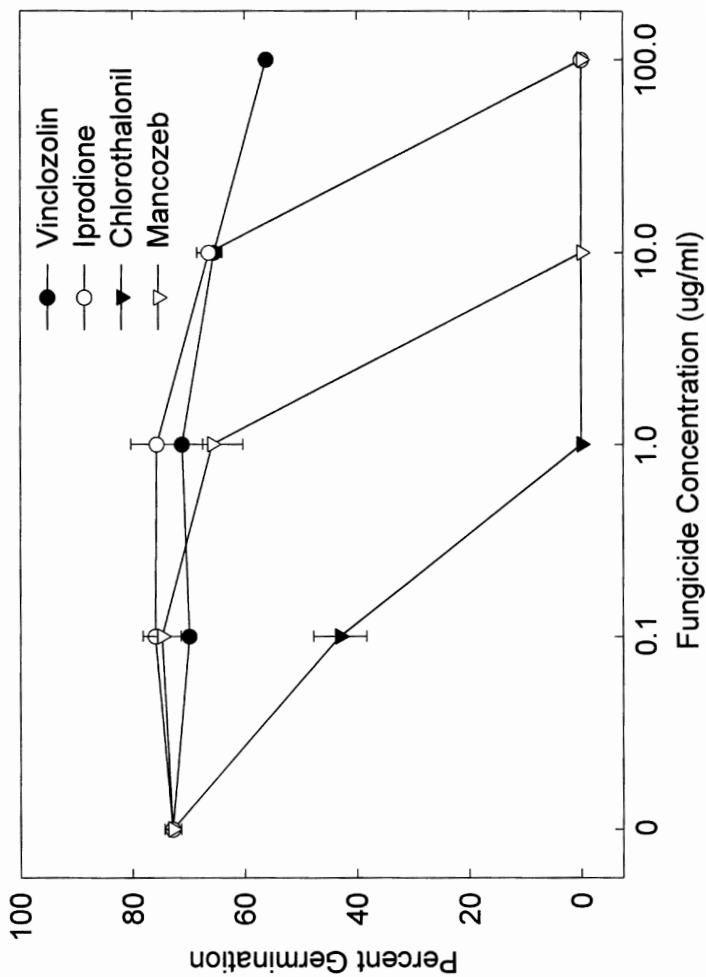


Figure 3. Germination of *P. hemerocallidis* urediniospores on fungicide-amended media



Resistance of Pansy and Viola Cultivars to Powdery Mildew and Cercospora Leaf Spot

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Nature of Work: A colorful winter and spring floral display has made pansy (*Viola x wittrockiana*) a landscape favorite across Alabama and the rest of the South. Some of the more heat tolerant selections may bloom well into the spring. Although a number of destructive diseases of pansy have been identified, particularly in production greenhouses, the identity and severity of diseases in landscape plantings has largely been ignored. This study was initiated to identify common diseases on pansy in the landscape, as well as to assess the susceptibility of pansy and viola cultivars to these diseases.

Approximately 400 pounds per acre of 13-13-13 or 5-10-15 fertilizer was broadcast and lightly incorporated immediately before transplanting pansy plugs. Flats of pansy and viola cultivars were obtained from commercial wholesale and retail outlets. The cultivars screened in all three years are listed in Table 1. Four plugs of each cultivar were planted on a 1-foot square in a Benndale sandy loam soil in a randomized complete block design consisting of 6 replications. Planting dates were October 22, 1996, November 15, 1999, and November 3, 2000. All studies were conducted at the Brewton Experiment field (USDA Zone 8a). A drip irrigation system was installed immediately after planting and the plants were watered as needed. In all three years, calcium nitrate at the rate of 10 pounds per acre was applied weekly through the drip irrigation system. The incidence of powdery mildew and/or *Cercospora* leaf spot were recorded on May 20, 1997, April 13, 2000, and May 9, 2001. In 1997, powdery mildew severity was visually rated on a scale of 1 to 5 where 1 = no disease, 2 = 0 to 25%, 3 = 25 to 50%, 4 = 50 to 75%, and 5 = 75 to 100% of the leaves colonized. For the 2000 and 2001 trials, the severity of this disease was assessed on a 1 to 12 Horsfall and Barratt rating scale where 1 = no disease, 2 = 0 to 3%, 3 = 3 to 6%, 4 = 6 to 12%, 5 = 12 to 25%, 6 = 25 to 50%, 7 = 50 to 75%, 8 = 75 to 87%, 9 = 87 to 94%, 10 = 94 to 97%, 11 = 97 to 100%, 12 = 100% of the leaves colonized by the powdery mildew fungus, *Sphaerotheca fuliginea*. In 1997, 2000, and 2001, *Cercospora* leaf spot damage was rated using a modified Florida peanut leaf spot scoring system where 1 = no disease, 2 = very few leaf spots, 3 = a few leaf spots in lower and upper canopy, 4 = some leaf spots with light defoliation, 5 = noticeable spotting of the leaves with some defoliation ($\geq 25\%$), 6 = spots numerous with significant defoliation ($\geq 50\%$), 7 = spots numerous with extensive defoliation ($\geq 75\%$), 8 = heavy spotting of the few remaining leaves, 9 = very few remaining leaves covered with spots, and 10 = plants defoliated (100%) or dead.

Results and Discussion: Over the three-year test period, *Cercospora* leaf spot was consistently the most common and damaging disease observed on pansy. Typically, the leaves colonized by the causal fungus *Cercospora viola* quickly yellowed, shriveled, and died. Leaf loss usually started at the base of the shoots and continued until all but the youngest leaves were killed. In all three years, the characteristic leaf spotting and premature leaf shed associated with this disease was seen on all pansy and viola selections. However, significant differences in the level of *Cercospora* leaf spot were observed in each year among the cultivars evaluated (Table 1). Overall, few differences in the level of *Cercospora* leaf spot pressure was noted among 1997, 2000, and 2001.

Of the pansy and viola cultivars evaluated in all three years, 'Maxim Sherbet' and 'Bingo Blue' suffered the least *Cercospora* leaf spot damage. Damage on both cultivars involved light leaf spotting and minor premature leaf loss (Table 1). The leaf spot ratings for 'Imperial Beaconsfield' were similar to those of the above cultivars in 2000 and 2001 but were higher in 1997. For the remaining cultivars screened in 1997, 2000, and 2001, moderate levels of leaf spotting and defoliation as indicated by disease ratings of 5.0 or above were recorded in a minimum of two years on 'Bingo Light Rose', 'Bingo White with Blotch', 'Crown Orange', 'Delta Pink Shades', and 'Majestic Giant Purple'. 'Bingo Red & Yellow', 'Bingo Red with Blotch', 'Crystal Bowl Purple', 'Crystal Bowl Supreme Yellow', 'Crystal Bowl Supreme True Blue', 'Dynamite Lavender' and 'Maxim Supreme Rose', which were evaluated for only one year, suffered relatively light damage.

The majority of cultivars of pansy screened proved to be susceptible to *Cercospora* leaf spot in at least one year indicating they would likely suffer heavy damage in landscape plantings. As indicated by a disease rating above 6.0 in one year, 'Bingo Light Rose', 'Crown Yellow Splash', 'Crown Scarlet', 'Crown White', 'Delta Red with Blotch', 'Delta Pure Rose', 'Delta Pure Yellow', 'Majestic Giant Blue Shades', and 'Penny Blue' were heavily damaged by *C. viola* (Table 1). Symptoms on these cultivars included a minimum of 50% defoliation, heavy spotting of the remaining leaves and a notable decline in plant quality and vigor. In other years, the *Cercospora* leaf spot ratings for these and many of the remaining pansy and viola selections often ranged from 5.0 to 5.8. At these disease ratings, defoliation levels typically were 25% to nearly 50% and overall plant condition and appearance was poor.

Powdery mildew was observed in 1997 and 2000 but not in 2001. Overall, disease levels were higher in 1997 than in 2000. Powdery mildew appeared to have less of an impact on plant aesthetics and vigor than did *Cercospora* leaf spot.

On the majority of pansy cultivars examined in 1997 and 2000, the severity of powdery mildew was very low and only a few colonies of *S.*

fuliginea were seen on the leaves and petioles. However, significant differences in the level of leaf and petiole infection were seen in both years (Table 1). Among the pansy cultivars evaluated in 1997 and 2000, none were free of powdery mildew in both years. Cultivars with low mildew ratings for both years include 'Bingo Blue with Blotch', 'Bingo Deep Purple', 'Delta Pink Shades', 'Maxim Orange', and 'Maxim Sherbet'. Of the cultivars screened in 2000 only, *S. fuliginea* colonization was not observed on 'Bingo Yellow', 'Bingo Yellow with Blotch', 'Imperial Antique Shades', 'Penny Blue', 'Penny Mix', and 'Purple Rain'. Although light, unobtrusive colonization was noted in 2000 on an additional 19 selections, their disease ratings did not differ significantly from those of cultivars that were free of powdery mildew.

Of the cultivars evaluated in 1997 and 2000, the highest incidence of powdery mildew was seen on 'Bingo Light Rose' and 'Imperial Beaconsville'. Noticeable powdery mildew infections were seen in one of two years on 3 additional cultivars of pansy. Of the cultivars screened in 2000 alone, 'Bingo Clear Azure', 'Imperial Silver Bell', and 'Maxim Marina' had the highest incidence of powdery mildew.

In summary, *Cercospora* leaf spot was the predominant disease found in this simulated landscape planting of pansy and viola cultivars. In all three years, extensive leaf spotting and premature leaf shed were seen on nearly all of cultivars. Rapid disease spread in March and April may account for the rapid decline in plant aesthetics and vigor typically seen in the spring in landscape plantings of pansy and viola. The occurrence of powdery mildew was much more sporadic than that of *Cercospora* leaf spot. This disease occurred on fewer cultivars in two of three years and caused considerably less damage to pansy. In fact, only a handful of pansy cultivars, such as 'Bingo Light Rose' and 'Imperial Beaconsfield' appeared to be susceptible to this disease.

Of the cultivars screened over the three-year test period, 'Bingo Blue with Blotch' and 'Maxim Sherbet' suffered the least damage from the combination of *Cercospora* leaf spot and powdery mildew. Low levels of leaf spotting and premature defoliation due to *Cercospora* leaf spot were recorded for a number of pansy cultivars, particularly several members of the Crystal Bowl series, evaluated in only the 2001-growing season.

Significance to Industry: Pansy is the premier winter annual in the South. Study results show that diseases, especially *Cercospora* leaf spot, have a significant impact on the health and appearance of landscape plantings of pansy. In areas that receive heavy late winter and spring rains, *Cercospora* leaf spot, and to a lesser extent powdery mildew, may be as responsible as increasing temperatures for the rapid decline in plantings of pansy often seen in April and early May. Several cultivars of pansy were identified with partial resistance to *Cercospora* leaf spot and powdery mildew. Use of these cultivars in residential and commercial plantings should enhance the aesthetics and useful life span of pansy.

Table 1. Reaction of cultivars of pansy and viola to powdery mildew and Cercospora leaf spot at the Brewton Experiment Field in 1997, 2000, and 2001.

	Powdery Mildew ¹		Cercospora Leaf Spot ²		
	1997	2000	1997	2000	2001
Accord White Blotch Imperial	---	---	---	---	4.8 bcdefgh
Accord Yellow Blotch	---	---	---	---	5.3 abcde
Bingo Blue with Blotch	1.8 fghi ⁴	1.4 de	4.5 de	4.0 ghij	4.5 cdefgh
Bingo Light Rose	3.0 abcde	2.7 cde	5.3 bcde	6.5 a	4.3 defghi
Bingo Deep Purple	1.0 i	1.5 de	5.0 cde	4.3 efghi	
Bingo White with Blotch	2.0 efghi	1.0 e	5.3 bcde	5.7 abcd	4.2 dfghi
Bingo Yellow	---	1.0 e	---	5.3 abcdef	5.8 ab
Bingo Yellow with Blotch	---	1.0 e	---	5.5 abcde	
Bingo Red & Yellow	---	---	---	---	3.8 ghi
Bingo Clear Yellow	---	---	---	---	4.6 bcdefgh
Bingo Red with Blotch	---	---	---	---	4.0 fghi
Bingo Clear Azure	---	4.8 a	---	4.7 cdefghi	---
Clear Sky Primrose	2.0 efghi	---	5.5 abcd	---	---
Clear Sky Yellow	---	1.4 de	---	5.2 bcdefg	---
Crown Golden	2.8 bcdef	---	4.8 cde	---	---
Crown Blue	3.0 abcde	---	6.0 abc	---	---
Crown Orange	1.7 hi	3.2 abcd	5.5 abc	5.0 bcdefgh	4.7 bcdefgh
Crown Cream	2.0 efghi	1.0 e	6.0 abc	4.7 cdefghi	---
Crown Rose	2.0 efghi	1.0 e	5.8 abcd	5.7 abcd	---
Crown Purple	2.8 bcdef	---	5.8 abcd	---	5.2 abcdef
Crown Yellow	3.3 abcd	---	5.7 abcd	---	---
Crown Yellow Splash	1.5 ghi	---	6.8 a	---	---
Crown Scarlet	3.3 abcd	---	6.0 abc	---	---
Crown White	2.8 bcde	---	6.0 abc	---	---
Crown Azure	---	---	---	---	5.3 abcde
Crystal Bowl Yellow	---	---	---	---	4.8 bcdefgh
Crystal Bowl Supreme Lavender Shades	---	---	---	---	5.0 bcdefg
Crystal Bowl Supreme Purple	---	---	---	---	4.0 fghi
Crystal Bowl Supreme Yellow	---	---	---	---	4.0 fghi
Crystal Bowl Supreme True Blue	---	---	---	---	4.0 fghi
Crystal Bowl Supreme White	---	---	---	---	4.3 defghi
Crystal Bowl Mix	---	---	---	---	4.5 cdefghi
Delta Pink Shades	1.5 ghi	1.0 e	5.5 abcd	4.0 ghij	5.2 abcdef
Delta Blue with Blotch	3.5 abc	---	5.5 abcd	---	---
Delta Pure Yellow	1.5 ghi	---	6.0 abc	---	---
Delta White/Rose Wing	1.8 fgh	---	5.5 abcd	---	---
Delta Red with Blotch	3.0 abcde	---	6.5 ab	---	---
Delta Pure Violet	3.0 abcde	---	5.5 abcd	---	---
Delta Pure Rose	1.0 I	---	6.8 a	---	---
Delta Pure Primrose	2.5 cdefg	---	5.5 abcd	---	---
Delta White with Blotch	---	---	---	---	5.3 abcde
Dynamite Purple	---	---	---	---	4.5 cdefghi
Dynamite Scarlet	---	---	---	---	5.2 abcdef
Dynamite Lavender	---	---	---	---	3.7 hi
Fama See Me	---	---	---	---	4.2 efghi
Holloween	---	1.3 e	---	5.5 abcde	---
Imperial Pink Shades	---	1.7 de	---	5.0 bcdefgh	4.8 cdefgh

Imperial Antique Shades	---	1.0 e	---	5.8 abcd	5.5 abcd
Imperial Silver Blue	---	4.7 ab	---	4.5 defghi	---
Imperial Beaconsfield	4.0 a	3.0 bcde	5.5 abcd	3.7 ij	4.5 cdefgh
Majestic Giant White	---	1.2 de	---	4.8 bcdefghi	---
Majestic Giant Purple	3.8 ab	2.4 cde	5.7 abcd	5.4 abcdef	5.8 ab
Majestic Giant Blue Shades	---	2.3 cde	---	5.5 abcde	6.3 a
Majestic Giant Red & Yellow	2.5 cdefg	---	5.3 bcde	---	3.7 hi
Majestic Giant Red and Rose Shades	---	1.7 de	---	5.0 bcdefgh	5.3 abcde
Majestic Giant Rose Shades	---	---	---	---	5.2 abcdef
Majestic Giant Scarlet & Bronze	2.0 efghi	2.3 cde	5.0 cde	4.8 bcdefghi	---
Majestic Giant Blue with Blotch	3.8 ab	---	5.5 abcd	---	---
Majestic Giant Yellow with Blotch	2.3 cdefgh	---	5.5 abcd	---	5.7 abc
Majestic Giant White with Blotch	1.5 ghi	---	5.3 bcde	---	4.2 efghi
Majestic Giant Yellow	---	---	---	---	4.4 defgh
Maxim Orange	1.3 hi	1.8 de	5.5 abcd	5.7 abcd	---
Maxim Red & Yellow	---	1.5 de	---	3.8 hij	---
Maxim Sherbet	1.8 fghi	1.7 de	4.0 e	4.2 hij	3.2 i
Maxim Marina	---	3.8 abc	---	5.0 bcdefgh	---
Maxim Sunset	---	1.0 e	---	5.0 bcdefgh	4.2 efghi
Maxim Supreme Rose	---	---	---	---	4.0 fghi
Maxim Yellow & Blue	2.8 bcdef	2.3 cde	5.5 abc	4.5 defghi	---
Melody Purple & Orange	---	1.2 de	---	5.3 defghi	---
Penny Yellow	---	1.2 de	---	3.0 j	5.2 abcdef
Penny Blue	---	1.0 e	---	6.0 ab	---
Penny Mix	---	1.0 e	---	4.0 ghij	5.3 abcde
Penny Violet Flare	---	---	---	---	4.8 bcdefgh
Purple Rain	---	1.0 e	---	4.5 defghi	---
Skyline Blue	2.0 efghi	---	4.8 cde	---	---
Sorbet Plum Violet	---	2.0 cde	---	4.3 efghi	---
Trick or Treat	---	2.0 cde	---	5.8 abc	---
Ultima Silhouette Mix	---	1.8 de	---	5.2 bcdefg	4.3 defghi

¹In 1997, powdery mildew incidence was visually rated on a scale of 1 to 5 where 1 = no disease, and 5 = 80 to 100% of the leaves colonized. Two years later, the incidence of this disease was assessed on a 1 to 12 scale where 1 = no disease, 2 = 0 to 3%, 3 = 3 to 6%, 4 = 6 to 12%, 5 = 12 to 25%, 6 = 25 to 50%, 7 = 50 to 75%, 8 = 75 to 87%, 9 = 87 to 94%, 10 = 94 to 97%, 11 = 97 to 100%, 12 = 100% of the leaves colonized by *S. fuliginea*.

²*Cercospora* leaf spot damage was rated in 1997, 2000, and 2001 using a scoring system where 1 = no disease, 2 = very few leaf spots, 3 = a few leaf spots in lower and upper canopy, 4 = some leaf spots with light defoliation, 5 = noticeable spotting of the leaves with some defoliation ($\geq 25\%$), 6 = spots numerous with significant defoliation ($\geq 50\%$), 7 = spots numerous with extensive defoliation ($\geq 75\%$), 8 = heavy spotting of the few remaining leaves, 9 = very few remaining leaves covered with spots, and 10 = plants defoliated (100%) or dead.

³--- = no data, cultivar not evaluated in that year.

⁴Means within a column followed by the same letter do not significantly differ, Duncan's Multiple Range Test ($P=0.05$).

Occurrence of Cercospora Leaf Spot on Indian Hawthorn

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Nature of Work: Entomosporium has long been recognized as the predominate leaf spot disease in plantings of Indian hawthorn (1). In Alabama, the characteristic leaf spots associated with this disease typically appear as tiny deep red or maroon spots on leaves in late winter to early spring. Over a period of a few weeks, the usually circular spots enlarge to about 1/4 inches in diameter and have a distinct deep red to purple halo. The center of these spots turns gray and may contain one to several black fruiting bodies of the causal fungus *Entomosporium mespili*. Heavy spotted leaves are usually shed. Following lengthy periods of wet, warm weather in late winter or early spring, highly susceptible cultivars such as 'Harbinger of Spring' may be completely defoliated (3).

Beginning in the summer 2000, an unfamiliar leaf spot disease was noted in a large planting of Indian hawthorn at the Brewton Experiment Field on cultivars that previously were identified as resistant to Entomosporium leaf spot (4). Initially, the symptoms of this unknown disease were so similar to Entomosporium leaf spot that the two were easily confused. Subsequent microscopic examination of the leaves confirmed that the fruiting bodies of *E. mespili* did not develop on the surface of these leaf spots. However, the fruiting structures (sporodochia) and spores (conidia) of a fungus in the genus *Cercospora* were found on the lower and upper surface of the spots of leaves collected from several Indian hawthorn cultivars. In Florida, *Cercospora ariae* and *C. violamaculans* have previously been reported as causal agents of a leaf spot on Indian hawthorn (2).

The objective of this study was to describe the symptoms of this disease and to assess its occurrence in an established planting of selected cultivars of Indian hawthorn.

Observations were made in a simulated landscape planting of Indian hawthorn (*Rhaphiolepis umbellata*), which was established at the Brewton Experiment Field in Brewton, AL (USDA Hardiness Zone 8a) in 1994. At that time, 15 commercial selections were established in a randomized complete block with 6 three-plant replications. Three additional cultivars of Indian hawthorn were added to this study in the late winter or spring 1995 and 1996. A drip irrigation system was installed immediately after

planting and the plants were watered as needed. Beds were mulched each year with 1 to 2 inches of aged pine bark. Twice each spring, approximately 3 ounces of 16-4-8 fertilizer was uniformly distributed around each plant. Directed applications of 1 pound per acre of Gallery DF and 2 quarts per acre of Surflan T/O were made early each spring down the row center to control annual weeds. Hand weeding and spot applications of recommended rates of the herbicides Roundup or 912 Herbicide 6S (MSMA) was used to control escape weeds and invading centipedegrass. The centipedegrass alleys behind the rows were mown periodically.

On March 14, 2001, 5 to 10 symptomatic leaves were collected in one replication from each cultivar of Indian hawthorn. The leaf spots on each of the leaves were examined at 20x with a stereoscopic microscope for the presence of the characteristic fruiting structures and conidia of *Entomosporium mespili* or a *Cercospora* sp. On the same day the leaf samples were collected, the incidence of *Cercospora* leaf spot and *Entomosporium* leaf spot was rated using the 1 to 12 Horsfall and Barratt rating scale where 1 = 0%, 2 = 0 to 3%, 3 = 3 to 6%, 4 = 6 to 12%, 5 = 12 to 25%, 6 = 25 to 50%, 7 = 50 to 75%, 8 = 75 to 87%, 9 = 87 to 94%, 10 = 94 to 97%, 11 = 97 to 100%, and 12 = 100% of with characteristic symptoms of each disease. Significance of cultivar on the severity of each disease was tested by analysis of variance and means were compared using Duncan's Multiple Range Test at a level of significance of $P=0.05$.

Results and Discussion: At the Brewton Experiment Field, rainfall totals for December 2000 through April 2001 were at- to well above the historical average. While December and January were unusually cold, temperatures through the remainder of the winter and early spring were near normal levels. During this time frame, the combination of mild temperatures and frequent showers greatly favored the onset and spread of *Cercospora* leaf spot and *Entomosporium* leaf spot.

Tiny, nondescript red to maroon spots, which are similar to those associated with *Entomosporium* leaf spot, were the first symptoms of *Cercospora* leaf spot to appear on the leaves of Indian hawthorn. On the upright Indian hawthorns Majestic Beauty® and Rosalinda®, these randomly scattered spots darkened to a deep maroon and expand to approximately 3/8 inches in diameter. Initially, the smaller spots on the leaves of these two cultivars were circular. When the larger leaf veins slowed or stopped lesion expansion, the spots became more irregular or angular in shape. Often, several spots coalesced to form large purple blotches on the upper leaf surface. Once spot expansion slowed, slightly sunken and irregularly shaped, reddish-brown dead (necrotic) area

formed in the center of the purple spot. Because little or no purpling occurred on the lower leaf surface, these dead areas, which were light tan with a brown border, were much easier to see on the underside of the leaves. Diseased leaves occurred year-round on both of these cultivars. During periods of wet winter and spring weather, the tiny, clear fruiting bodies (sporodochia) and conidia could be seen at 20x to 40x magnification scattered over the surface of the dead areas in the spots on the lower and upper leaf surfaces. Eventually, the heavily spotted leaves of Majestic Beauty® and Rosalinda® turned bright red in color and fell to the ground around the base of the plant.

As indicated by March disease ratings of 6.3 and 7.0, Rosalinda® and Majestic Beauty®, respectively, proved to be susceptible to *Cercospora* leaf spot (Table 1). Heavy spotting of the leaves and premature defoliation were noted on both cultivars of Indian hawthorn. Rosalinda®, which put on new foliage in the early spring, appeared to recover faster from *Cercospora* leaf spot than did Majestic Beauty® (data not shown).

Symptoms of this disease on the dwarf type Indian hawthorn differed slightly in appearance. While the early symptoms were similar to those seen on Majestic Beauty® and Rosalinda®, the larger spots on the leaves of the dwarf type Indian hawthorn, which are two to three times the size of those associated with *Entomosporium* leaf spot, were more circular. On some cultivars, the upper leaf surface in the area of the spots was a mosaic of light brown to maroon patches of color and these areas on the more susceptible selections often coalesced into large multi-colored blotches. Dead, slightly sunken areas formed on the lower leaf surface directly under the discolored patches on the upper leaf surface. Again, these dead spots had a tan to light brown center and a reddish brown border. Under favorable weather patterns, the causal fungus sporulated in the dead areas on the lower leaf surface. As was the case with the upright cultivars of Indian hawthorn, the diseased plant eventually shed the heavily spotted leaves.

Cercospora leaf spot was the primary leaf spot disease on several dwarf Indian hawthorn selections. With *Cercospora* leaf spot ratings of 5.5 and 5.7, Indian Princess® and Gulf Green™ ('Dwarf Yedda'), respectively, suffered from heavy spotting of the foliage and considerable leaf shed (Table 1). Canopy coverage on Indian Princess® improved considerably between March and May 2001 (data not shown). Noticeable *Cercospora*-caused spotting of the leaves but with little or no leaf shed was also recorded for Eleanor Tabor® and Olivia®. A low level of *Cercospora* leaf spot was noted primarily on the older leaves of 'Heather', Harbinger of Spring®, and 'Pinkie' Indian hawthorn.

Entomosporium leaf spot was the predominate disease found on the leaves of Springtime®, Spring Rapture®, White Enchantress®, 'Snow

White', Enchantress®, 'Clara', Harbinger of Spring®, 'Pinkie', Bay Breeze®, and Becky Lynn® Indian hawthorn. While the defoliation level for 'Clara' and Becky Lynn, was in the range of 20%, the remaining Entomosporium leaf spot susceptible cultivars suffered at least 50% defoliation and heavy spotting of many of the remaining leaves. The one to several characteristic fruiting bodies of *E. mespili*, were easy to see with a hand lens in the numerous spots found on the upper surface of leaves of the above cultivars of Indian hawthorn.

Cercospora leaf spot is not a new disease of Indian hawthorn. This disease has been previously reported on Indian hawthorn in Florida (2). Due to the symptom similarity between this disease and Entomosporium leaf spot, the occurrence of Cercospora leaf spot on Indian hawthorn probably has been underreported. The highest incidence of this disease was recorded primarily on those cultivars of Indian hawthorn, such as Majestic Beauty®, Indian Princess®, Eleanor Tabor®, Olivia®, and Gulf Green™, which were previously shown in this same planting to be resistant to Entomosporium leaf spot (3,4). Of the above cultivars, Majestic Beauty®, Indian Princess®, Gulf Green™ as well as Rosalinda, suffered from heavy spotting of the leaves and noticeable defoliation. Damage to Olivia, and Eleanor Tabor, Indian hawthorn, which was largely limited to spotting of the foliage in the lower canopy, did not have a detrimental effect on the aesthetics or health of the plant. As was previously reported, both of the above cultivars were highly resistant to Entomosporium leaf spot (3).

Significance to Nursery Industry: Cercospora leaf spot caused significant damage on selected cultivars of Indian hawthorn in a simulated landscape planting at the Brewton Experiment Field. Since this disease has not been diagnosed in production blocks of Indian hawthorn, it is possible that the occurrence of Cercospora leaf spot will largely be limited to landscape plantings. Cultivars of Indian hawthorn significantly differ in their sensitivity to this disease. Some of cultivars such as Indian Princess®, which were previously shown to have a high level of resistance to Entomosporium leaf spot, proved to be susceptible to Cercospora leaf spot.

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Table 1. Ratings for Cercospora leaf spot and Entomosporium leaf spot on selected cultivars of Indian hawthorn at the Brewton Experiment Field, March 2001.

Cultivar	Leaf Spot Rating ¹		Cultivar	Leaf Spot Rating ¹	
	Cercospora	Entomosporium		Cercospora	Entomosporium
Majestic Beauty [®]	7.0	---	Springtime [®]	---	7.7
Spring Rapture [®]	---	8.8	White Enchantress [®]	---	8.0
Indian Princess [®]	5.5	---	Gulf Green [™]	5.7	---
Olivia [®]	3.2	---	Eleanor Tabor [®]	2.8	---
Rosalinda [®]	6.3	---	'Snow White'	---	5.0
'Heather'	---	7.8	Enchantress [®]	---	6.8
Becky Lynn [®]	---	8.2	'Clara'	---	5.8
Harbinger of Spring [®]	---	7.6	'Pinkie'	---	8.0
Bay Breeze [®]	---	5.6			

¹The incidence of Cercospora leaf spot and Entomosporium leaf spot was rated using a 1 to 12 Barratt and Horsfall rating scale where 1 = 0%, 2 = 0 to 3%, 3 = 3 to 6%, 4 = 6 to 12%, 5 = 12 to 25%, 6 = 25 to 50%, 7 = 50 to 75%, 8 = 75 to 87%, 9 = 87 to 94%, 10 = 94 to 97%, 11 = 97 to 100%, and 12 = 100% of the leaves damaged or prematurely shed.

Disease Resistance of Shrub and Groundcover Roses and the Impact of Fungicide Inputs on those Diseases

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Nature of Work: In recent years, the nursery industry has focused on bringing colorful, hardy roses that have lower maintenance requirements and more versatility than the classic hybrid tea. In addition, 30 or more weekly fungicide applications are often needed to protect hybrid tea and similar roses from black spot. The result has been a renewed interest in the use of easy-care shrub and ground cover roses in residential landscapes. Although claims of disease resistance have been made for many of these roses, their reaction to black spot and other foliar diseases has not been well documented, especially under hot, humid conditions found across the Deep South.

A simulated landscape planting of selected ground cover and shrub roses was established at the Brewton Experiment Field to assess their susceptibility to black spot and other diseases, as well as their overall adaptability to the climatic conditions in the Deep South. Bare-root roses were potted in a pine bark/peat (3:1 v/v) medium amended with 12 pounds of 17-7-12 Osmocote, 6 pounds of dolomitic limestone, 2 pounds of gypsum, and 1.5 pounds of Micromax per cubic yard at the Ornamental Horticulture Substation in Mobile, AL. On January 30 and March 19, 1998, the roses were transplanted into raised beds at the Brewton Experiment Field in Brewton, AL. Fertility and pH were adjusted according to the results of a soil assay. The beds were then mulched with aged pine bark. A drip irrigation system was installed at the time of plot establishment and the plants were watered as needed. The butterfly rose (*Rosa mutabilis*) was planted on June 4, 1998 and 'Double Delight', 'Carefree Wonder', 'Hansa', and 'Pink Grootendorst' were established on February 11, 1999. In 2000, 'Kent', 'Knock Out', 'Fire Meidiland', 'Ice Meidiland', 'Therese Bugnet', 'Raven' and 'Sweet Chariot' were substituted for 'Nearly Wild', 'Betty Prior', 'Royal Bonica', 'Magic Carpet', 'Bonica', and 'Double Delight'.

The study consisted of a split plot design in a randomized complete block design consisting of 5 replications with cultivars as the main plot and fungicide treatment as the split-plot. Selected shrub, ground cover, and in 1999 only the hybrid tea rose 'Double Delight' were included in this study. A total of 25, 29, and 30 cultivars were screened in 1998, 1999, and 2000, respectively. Fungicide treatments were: 1) an unsprayed control, 2) Daconil 2787 @ 2 pints per 100 gallons of spray volume applied at 2-week intervals, and 3) Daconil 2787 @ 2 pints per 100

gallons of spray volume applied at 4-week intervals. Fungicides were applied to run-off with a CO₂ pressurized backpack in from March 23 to October 25, 1998 and March 22 to November 2, 1999 and with a tractor-mounted sprayer from April 5 until October 19, 2000.

Beginning in March or April, the incidence of diseases was visually evaluated in 1998, 1999, and 2000 at 6 to 8 week intervals until late November or early December. Simultaneously, plants were examined for the characteristic symptoms and signs of powdery mildew, downy mildew, and *Cercospora* leaf spot. Leaf samples were collected periodically in 1999 and 2000 to confirm the occurrence of black spot (BS) and *Cercospora* leaf spot (CLS). A modified Florida peanut leaf spot rating scale where 1 = no disease, 2 = very few spots in lower canopy, 3 = light spotting lower and upper canopy, 4 = some spots in lower and upper canopy with light defoliation, 5 = spots noticeable with moderate defoliation ($\geq 25\%$), 6 = spots numerous with significant defoliation ($\geq 50\%$), 7 = spots numerous with severe defoliation ($\geq 75\%$), 8 = most remaining leaves spotted with excessive defoliation ($\geq 90\%$), 9 = very few remaining leaves covered with spots, and 10 = plants defoliated was used to assess the severity of black spot and *Cercospora* leaf spot. In 1999, ratings for black spot and *Cercospora* leaf spot were recorded on March 23, May 6, June 24, August 30, October 7, and November 11. During the 2000-growing season, disease ratings were logged on April 12, May 23, June 27, September 11, September 29, and November 10. The average disease rating for black spot and *Cercospora* leaf spot was calculated for 1999 and 2000 by adding the disease ratings collected for each treatment at each assessment date and then dividing by the total number (6) of disease observations taken in that year.

Results and Discussion: Although none of the roses proved immune to either black spot or *Cercospora* leaf spot, significant differences in the severity of both diseases were noted in 1998. Of the two diseases, BS was the by far the most common observed in 1998 (Table 1). With disease ratings of 5.6 and 5.0, 'Petite Pink Scotch' and 'The Fairy', respectively, were the two cultivars that suffered significant *Cercospora* leaf spot-related leaf spotting and early leaf shed. In 1998, 'Flower Carpet', 'Ralph's Creeper', 'Magic Carpet', *R. wichuraiana*, *R. mutabilis*, 'Happy Trails', and 'Red Cascade' suffered light to moderate leaf spotting, as well as little or no defoliation. As indicated by BS ratings of 5.2 to 6.6, unacceptable levels of leaf spotting and defoliation were recorded on the majority of the roses screened in 1998.

In 1999, significant differences in the severity of BS and CLS among 29 rose cultivars were seen again. Many of the 29 rose cultivars proved highly susceptible to BS or CLS but not to both diseases. Black spot was noted on 21 of the 29 cultivars. With BS ratings less than 4.0, *R. wichuraiana*, 'Red Cascade', 'Mystic Meidiland', 'Nozomi', and the

rugosa roses 'Hansa' and 'Pink Grootendorst' had the least leaf spotting and defoliation (Table 1). Moderate to heavy spotting of the leaves and moderate defoliation was seen on the 16 black spot-damaged roses. On 'Fushia Meidiland', 'Carefree Delight', 'White Flower Carpet', 'Happy Trails', 'Flower Carpet' and 'The Fairy', CLS was the cause of noticeable spotting of the leaves and light to moderate defoliation.

For the 2000-growing season, BS was noted on 24 cultivars while CLS damaged the remaining 6 rose cultivars. As indicated by season-long BS ratings less than 4.0, light to moderate spotting of the leaves along with limited defoliation was seen on 'Mystic Meidiland', 'Pink Grootendorst', 'Hansa', 'Knock Out', 'Ice Meidiland', 'Therese Bugnet', *R. wichuraiana*, and 'Red Cascade' (Table 1). In excess of 50% defoliation was seen on the cultivars 'Sevilliana', 'Cherry Meidiland', 'Livin' Easy' and 'Jeeper's Creeper'. As was seen in 1999, significant CLS-related leaf spotting and moderate defoliation was recorded on 'Fushia Meidiland', 'The Fairy', 'Flower Carpet', 'Happy Trails', and 'White Flower Carpet'. Light CLS-related leaf spotting and defoliation was observed on 'Petite Pink Scotch'.

Although significant reductions in BS and CLS were obtained in 1999 with Daconil 2787 at 2 and 4 week intervals, the effectiveness of this fungicide in controlling these diseases varied considerably among the rose cultivars. As expected, Daconil 2787 gave better control of BS when applied at 2 than at 4-week intervals. When Daconil 2787 was applied every 2 weeks, little if any BS-related defoliation, as indicated by disease ratings of 3.0 or below, was noted on some of the cultivars. Only on the highly BS-susceptible cultivars 'Betty Prior', 'Easy Livin', 'Double Delight', and 'Carefree Wonder' suffer noticeable defoliation when treated with Daconil 2787 at two-week intervals. With the exception of 'Hansa', 'Carefree Wonder', and *R. wichuraiana*, the severity of black spot in 1999 was significantly lower with the 2 week rather than the 4 week treatment interval (Table 2). On 'Hansa', 'Pearl Sevillana', 'Royal Bonica', 'Red Cascade', and 'Nozomi' in 1999, the BS disease ratings noted on the plants treated with Daconil 2787 every 4 weeks and the unsprayed control were similar.

On the cultivars susceptible to CLS, the severity of this disease was significantly lower with Daconil 2787 applied at 2- rather than 4-week intervals. With the exception of 'Carefree Delight', CLS damage on the roses treated at 2-week intervals was limited to light spotting throughout the canopy, which was unobtrusive (Table 3). In 1999, CLS severity on the unsprayed roses was higher on those treated with Daconil 2787 applied at 4 week intervals except for 'Carefree Delight', 'Flower Carpet', and 'Happy Trails'.

In 2000, significant reductions in the severity of BS and CLS were obtained with Daconil 2787 applied at 2- rather than 4-week intervals on nearly all cultivars (Tables 2 and 3). Typically, better control of BS was

obtained with Daconil 2787 applied at 2- week than at 4-week intervals. With the exception of a few cultivars like 'Livin' Easy', the level of defoliation on the roses treated every 2 weeks with Daconil 2787 was minimal. As indicated by BS ratings below 2.0, spotting of the leaves was confined to the lower canopy of 'Mystic Meidiland', 'Red Cascade', and '*R. wichuriana*'. For 'Knock Out', 'Fire Meidiland', 'Nozomi', 'Carefree Wonder', 'Hansa', *R. wichuriana*, 'Ice Meidiland', and 'Red Cascade', no significant differences in BS ratings were noted on the plants treated with Daconil 2787 at 2- and 4-week intervals. However, the BS ratings for several of the above cultivars, most notably *R. wichuriana*, 'Hansa', 'Mystic Meidiland', and 'Ice Meidiland' were quite low.

When compared with the untreated control, the severity of CLS was significantly reduced in 2000 by Daconil 2787 applied at 2- and 4-week intervals on 4 of the 6 cultivars susceptible to this disease (Table 3). For 'Flower Carpet', no differences in CLS severity was noted between the roses treated with Daconil 2787 and the untreated control. On 'Fushia Meidiland', 'Carefree Delight', and 'The Fairy' the severity of CLS was lower on the plants treated at 2- rather than 4-week intervals but not on 'White Flower Carpet' and 'Happy Trails'. As indicated by disease ratings ranging between 2.3 and 2.9, *Cercospora* leaf spot damage was limited on five of the six cultivars treated at two-week intervals to unobtrusive spotting of the leaves.

Overall, black spot was the most common disease on the majority of rose cultivars. In 1999 and 2000, *Cercospora* leaf spot, however, extensively damaged 6 of the ground cover and shrub-type roses. The level of leaf spotting and premature defoliation on the *Cercospora* leaf spot-damaged roses was similar to that seen on cultivars that were susceptible to black spot. Typically, either black spot or *Cercospora* leaf spot was the predominate disease on a particular rose and mixed outbreaks of the two diseases were not apparent. Among the ground cover and shrub rose cultivars, several including *R. wichuriana* and 'Red Cascade' demonstrated a high level of resistance to both diseases. 'Mystic Meidiland', which in 2 of 3 years suffered only light to moderate spotting of the leaves and low levels of defoliation, as well as 'Hansa' and 'Pink Grootendorst' also appeared to be partially resistant to black spot and *Cercospora* leaf spot. In 2000, low levels of black spot were also seen on in the first year on 'Ice Meidiland'.

Significance to Nursery Industry: Historically, black spot and other diseases have heavily damaged cultivated roses, particularly in the hot, humid South. In addition, the intensive fungicide regimes typically required to control black spot and other diseases has discouraged the installation of roses in residential and commercial landscapes. The introduction of more disease-resistant roses, such as those described in this report, has the potential to greatly broaden the market for these attractive, versatile and fragrant plants across the South.

Table 1. Sensitivity of rose cultivars to *Cercospora* leaf spot and blackspot.

Cultivars	Black Spot ¹			Cercospora Leaf Spot ¹		
	1998	1999	2000	1998	1999	2000
Shrub Roses						
Betty Prior ²	6.6	5.4	---	---	1.0	---
Royal Bonica ²	6.6	5.2	---	---	1.0	---
Sevillana	6.4	5.3	6.1	---	1.0	1.0
First Light	4.9	4.7	5.0	---	1.0	1.0
Carefree Delight ²	6.2	1.0	---	---	5.4	---
Bonica ²	6.1	5.2	---	---	1.0	---
Cherry Meidiland	6.0	5.4	6.3	---	1.0	1.0
Newly Wild ²	5.8	5.2	---	---	1.0	---
Pearl Sevillana	5.6	4.6	5.0	---	1.0	1.0
Easy Livin'	5.2	4.9	6.0	---	1.0	1.0
Sea Foam	5.2	4.5	4.4	---	1.0	1.0
Mystic Meidiland	5.2	3.6	3.2	---	1.0	1.0
Nozomi	4.4	3.8	4.5	---	1.0	1.0
White Flower Carpet	4.2	1.0	1.0	---	5.2	4.6
<i>R. mutabilis</i>	3.8	4.5	4.9	---	1.0	1.0
Flower Carpet	2.6	1.0	1.0	---	4.5	3.9
Petite Pink Scotch	1.0	1.0	1.0	5.6	ND	3.2
The Fairy	1.0	1.0	R	5.0	4.8	4.9
Carefree Wonder ³	---	4.5	4.1	---	1.0	1.0
Pink Grootendorst ²	---	3.6	3.9	---	1.0	1.0
Hansa ³	---	3.0	3.2	---	1.0	1.0
Kent ⁴	---	---	4.4	---	---	1.0
Knock Out ⁴	---	---	3.6	---	---	1.0
Fire Meidiland ⁴	---	---	4.2	---	---	1.0
Ice Meidiland ⁴	---	---	2.6	---	---	1.0
Therese Bugnet ⁴	---	---	3.4	---	---	1.0
Raven ⁴	---	---	5.1	---	---	1.0
Sweet Chariot ⁴	---	---	5.9	---	---	1.0
Ground Cover Roses						
Red Cascade	3.8	2.7	2.7	---	1.0	1.0
<i>R. wichuraiana</i>	3.6	2.4	2.5	---	1.0	1.0
Happy Trails	2.6	1.0	1.0	---	4.0	5.1
Ralph's Creeper	3.0	4.5	5.6	---	1.0	1.0
Magic Carpet	3.0	ND ⁵	---	---	ND	---
Fushia Meidiland	4.8	1.0	1.0	---	4.5	4.3
Jeeper's Creeper	5.6	5.4	6.3	---	1.0	1.0
Hybrid Tea						
Double Delight ³	---	4.9	---	---	1.0	---
LSD (P=0.05)						

¹Black spot and *Cercospora* leaf spot were rated on a 1 to 10 scale using a modified Florida peanut leaf spot rating system where 1 = no disease, 2 = very few spots in lower canopy, 3 = light spotting lower and upper canopy, 4 = some spots in lower and upper canopy with light defoliation, 5 = spots noticeable with moderate defoliation ($\geq 25\%$), 6 = spots numerous with significant defoliation ($\geq 50\%$), 7 = spots numerous with severe defoliation ($\geq 75\%$), 8 = most remaining leaves spotted with excessive defoliation ($\geq 90\%$), 9 = very few remaining leaves covered with spots, and 10 = plants defoliated.

²R = cultivar removed in 2000.

³Cultivar added in 1999.

⁴Cultivar added in 2000.

⁵The unsprayed 'Magic Carpet' roses died.

Table 2. The effect of fungicide treatments on the average severity of black spot on selected cultivars of shrub and ground cover roses at the Brewton Experiment Field, 1999 and 2000¹.

Cultivar	1999				2000			
	Spray Interval				Spray Interval			
	2 wk	4 wk	UTC	LSD ⁵	2 wk	4 wk	UTC	LSD
Nearly Wild ²	3.1 ⁴	4.0	5.2	0.5	---	---	---	
Kent ³	---	---	---		2.5	3.6	4.4	
Betty Prior ²	4.1	4.5	5.4	0.4	---	---	---	
Knock Out ³	---	---	---		3.2	3.2	3.6	NS
First Light	2.7	3.8	4.7	0.4	3.0	3.5	5.0	
Mystic Meidiland	1.9	3.0	3.6	0.6	1.8	2.6	3.2	
Pearl Seviliana	3.3	4.2	4.6	0.6	3.1	4.2	5.0	
Sevilliana	3.3	4.3	5.3	0.3	3.4	4.6	6.1	
Livin' Easy	4.2	5.0	4.9	NS ⁶	3.9	5.6	6.0	
Cherry Meidiland	3.1	4.5	5.4	0.3	2.8	5.2	6.3	
Royal Bonica ²	3.7	4.9	5.2	0.7	---	---	---	
Fire Meidiland ³	---	---	---		3.0	3.2	4.2	
Red Cascade	1.3	2.2	2.7	0.6	1.8	2.1	2.7	
Nozomi	2.5	3.2	3.8	0.6	3.1	3.5	4.1	
Sea Foam	2.5	3.8	4.5	0.7	2.5	3.4	4.4	
<i>R. wichuraiana</i>	1.5	1.5	2.4	0.5	1.8	1.8	2.5	
Ralph's Creeper	2.5	3.7	4.5	0.5	3.1	4.0	5.6	
Ice Meidiland ³	---	---	---		2.1	2.3	2.6	NS
Jeeper's Creepers	3.6	4.4	5.4	0.4	3.3	4.2	6.3	
Bonica ²	2.8	4.1	5.2	0.5				
Therese Bugnet ³	---	---	---		2.6	3.6	3.4	
<i>R. mutabilis</i>	2.6	3.5	4.5	0.2	2.6	3.3	4.9	
Double Delight ²	3.9	3.9	4.9	0.6				
Raven ³	---	---	---		2.7	4.2	5.1	
Carefree Wonder	3.4	3.7	4.5	0.7	3.3	3.5	4.1	
Hansa	2.5	2.6	3.0	0.4	2.6	2.8	3.2	
Pink Grootendorst	1.9	2.9	3.6	0.4	2.1	2.8	3.9	
Sweet Chariot ³	---	---	---		3.5	4.7	5.9	

¹The average disease rating for black spot was calculated by adding the disease ratings for each cultivar and dividing by the number of observations recorded in that year.

²Cultivar was removed in early 2000.

³Cultivar was added to study in late winter 2000.

⁴Black spot was rated on a 1 to 10 scale using a modified Florida peanut leaf spot rating system where 1 = no disease, 2 = very few spots in lower canopy, 3 = light spotting lower and upper canopy, 4 = some spots in lower and upper canopy with light defoliation, 5 = spots noticeable with moderate defoliation ($\geq 25\%$), 6 = spots numerous with significant defoliation ($\geq 50\%$), 7 = spots numerous with severe defoliation ($\geq 75\%$), 8 = most remaining leaves spotted with excessive defoliation ($\geq 90\%$), 9 = very few remaining leaves covered with spots, and 10 = plants defoliated.

⁵Mean separation within rows was tested according to Fisher's Protected Least Significance (LSD) test ($P=0.05$).

⁶NS = not significant.

Table 3. The effect of fungicide treatments on the average severity of Cercospora leaf spot on selected cultivars of shrub and ground cover roses at the Brewton Experiment Field, 1999 and 2000^{1,2}.

Cultivar	1999				2000			
	Spray Interval			LSD ³	Spray Interval			LSD
	2 wk	4 wk	UTC		2 wk	4 wk	UTC	
Fushia Meidiland	2.0 ³	3.6	4.5	0.7	2.3	3.2	4.3	
Carefree Delight	3.4	4.7	5.4	0.7	2.9	4.7	5.6	
The Fairy	2.8	4.0	4.8	0.4	2.5	3.7	4.9	
Flower Carpet	2.9	3.8	4.5	1.0	3.4	3.6	3.9	
Happy Trails	2.2	3.6	4.0	0.5	2.7	3.6	5.1	
White Flower Carpet	2.9	3.9	5.2	0.4	2.7	3.5	5.1	
Petite Pink Scotch					1.7	2.1	3.2	

¹The average disease rating for Cercospora leaf spot was calculated by adding the disease ratings for each cultivar and dividing by the number of observations recorded in that year.

²Cercospora leaf spot was rated on a 1 to 10 scale using a modified Florida peanut leaf spot rating system where 1 = no disease, 2 = very few spots in lower canopy, 3 = light spotting lower and upper canopy, 4 = some spots in lower and upper canopy with light defoliation, 5 = spots noticeable with moderate defoliation ($\geq 25\%$), 6 = spots numerous with significant defoliation ($\geq 50\%$), 7 = spots numerous with severe defoliation ($\geq 75\%$), 8 = most remaining leaves spotted with excessive defoliation ($\geq 90\%$), 9 = very few remaining leaves covered with spots, and 10 = plants defoliated.

³Mean separation within rows was tested according to Fisher's Protected Least Significance (LSD) test ($P=0.05$).

Evaluation of powdery mildew resistant flowering dogwood selections in Middle Tennessee

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Index Words: Dogwood, *Cornus florida*, powdery mildew

Nature of Work: Since 1994, powdery mildew caused by *Microsphaera pulchra* has been a constant problem on the flowering dogwood, *Cornus florida* L. in nurseries and landscapes (1). Powdery mildew appears as white, fungal growth on the leaves of flowering dogwoods. Infected leaves may be strap-shaped and curled. Powdery mildew may stunt the growth of flowering dogwood during periods of drought.

Production costs for flowering dogwood have increased due to dogwood anthracnose and powdery mildew. While dogwood anthracnose is uncommon in field nurseries, powdery mildew can be found in most nurseries and landscapes throughout the range of flowering dogwood. Fungicides can be used to successfully manage powdery mildew, however, resistance is the preferred management strategy when available (2). Three cultivars of flowering dogwood, that are resistant to powdery mildew, have been released by the Tennessee Agricultural Experiment Station. Other selections continue to be evaluated for potential release to the nursery industry.

In June 1999, eighteen selections of rooted dogwood liners were planted into beds in a clay-loam soil at the Plant and Pest Diagnostic Center in Nashville. Three replications of each were planted using a randomized complete block designed. The beds were mulched with hardwood bark and irrigated via drip irrigation. The three dogwood cultivars that have been released were included as standards. The trees were evaluated for powdery mildew during late summer and fall of 2000 and flower buds were enumerated in October, 2000. The rating scale was: 0 = no disease, 1 < 2% of foliage affected, 2 < 10%, 3 < 25%, 4 < 50%, 5 > 50%, 6 = 100% of the foliage with powdery mildew.

Results and Discussion: Most of the flowering dogwood selections evaluated were highly resistant to powdery mildew during a year of intense disease pressure. Several also produced numerous flower buds two seasons after propagation. Most of the selections were equivalent to the three cultivars that have been released in disease resistance (Table 1). However due to poor flowering characteristics, it's doubtful that they will be released. Selections 95-24 (Figure 1) and 95-25 were not only highly resistant to powdery mildew, but also had attractive flowers with stiff, white bracts.

Significance to Industry: If the demand for powdery mildew resistant flowering dogwood grows, there are additional trees that merit release to the nursery industry.

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Table 1. Reaction of *Cornus florida* selections to powdery mildew and flower bud production.

Selection	August 22	September 20	October 10	Flower Bud/Mean/Tree
94-5	0.0	1.0	1.66	1.66
94-48	1.0	2.0	4.0	4.0
94-49	2.0	2.0	3.0	0
94-60	1.33	1.7	2.33	.33
94-67	0.0	0.0	0.0	5.66
94-83	0.66	0.66	0.66	2.33
95-1	2.0	2.0	2.33	3.66
95-3	0.33	0.0	0.66	2.33
95-4	2.0	1.3	1.33	8.66
95-8	1.0	2.0	3.0	0
95-9	0.0	0.0	0.0	1.33
95-10	0.0	0.0	0.33	6.0
95-11	2.0	2.3	3.0	9.33
95-12	0.66	1.0	0.66	3.66
95-17	0.0	0.33	0.66	0.33
95-24	0.0	0.0	0.0	9.33
95-25	0.0	0.0	0.33	4.33
95-28	2.3	2.0	2.66	0.33

Rating Scale: 0=healthy, 1<2%, 2<10%, 3<25%, 4<50%, 5>50%, 6=100%

Flower buds were counted on October 10.

94-67 (Jean's Appalachian Snow)

95-12 (Karen's Appalachian Blush)

95-10 (Kay's Appalachian Mist)

Isolation of *Seiridium* and *Botryosphaeria* spp. from Diseased Leyland Cypress

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Index Words: *x Cupressocyparis leylandii* (A.B. Jacks. & Dallim.)Dallim.,
Seiridium canker, Botryosphaeria canker

Nature of Work: Leyland cypress trees are popular ornamental evergreens in Tennessee and are used for Christmas trees, quick growing screens, groupings, and hedges (3). A hybrid of Monterey cypress and Alaska cedar, Leyland cypress trees may not be considered a long-term investment (10-20 years) due to frequent problems with canker diseases. *Seiridium* canker is the most common and damaging causal agent of Leyland cypress (4). *Botryosphaeria* canker has also been attributed with deadly canker diseases. Our objective of this study was to determine the predominance and identity of the pathogenic agents present in seventeen sites across Tennessee.

Seventeen diseased Leyland cypress sites (Table 1) were chosen to investigate the causal agent of canker formation on trees. Site conditions such as slope, height, spacing, available water, soil pH, sun exposure, diameter at breast height (DBH), and overall appearance of trees were recorded at each location. Cankers were collected from twigs and placed in moisture chambers to induce sporulation. Spores were collected and identified by using microscopy techniques.

Results and Discussion: *Seiridium* sp. conidia were isolated from sixteen of the seventeen sites. On site 1, *Seiridium* sp. spores were isolated from 92% of the trees sampled. *Botryosphaeria* conidia were identified from five of the seventeen sites included in the experiment. *Botryosphaeria* sp. spores were isolated from 40% of trees sampled from site 10. We found *Seiridium* canker to be the more prominent pathogen causing damage to Leyland cypress in Tennessee.

Parameters for each site varied tremendously. Tree spacing among sites ranged from 0.6 m to 4.3 m, average DBH per site ranged from 2 to 15 cm, tree height per site ranged from 2 to 9 m, and average site pH ranged from 4.1 to 7.3. All variables were correlated with isolation frequency of *Seiridium* and *Botryosphaeria* using the CORR Procedure in SAS. In the first test, only pH was found to be positively correlated with presence of specific pathogen found. No other variables were found to

be positively correlated with the specific pathogen found. In test two, DBH and tree height was found to be positively correlated with specific pathogen found. No other variables were found to be positively correlated with the specific pathogen found.

Due to the large ranges in tree size and age of the sites chosen for this experiment, we conclude that *Seiridium* and *Botryosphaeria* sp. can infect any size or age of plant. The smaller trees (2m) had similar disease percentages per site as the larger trees (9m). Our data did not support previous conclusions (1,2) that canker diseases are more associated with smaller and younger trees.

Significance to Industry: It has been suggested that all sizes and ages of Leyland cypress may be infected with *Seiridium* or *Botryosphaeria* canker (4). Infected trees observed in this study ranged in height from 2-9 m and averaged 2 to 15 cm in diameter. All ages and sizes of Leyland cypress trees are susceptible to canker diseases.

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Table 1: Sites, parameters, and frequency of isolation.

Site	Location	Seiridium % per site	Botry % per site	Average DBH cm	Average height m	Average pH	Spacing m
1.	Lovell View Dr. Knoxville, TN	92%	0	15	9	5.86	1.5
2.	Nursery Production Area, Campus, Knoxville, TN	90%	0	15	8	6.55	1.5
3.	Neyland Dr., Knoxville, TN	75%	0	15	9	7.3	4.3
4.	Downtown Park, Maryville, TN	33%	0	10	7	6.8	1.5
5.	Centennial High School, Franklin, TN	33%	17%	22	2	6.4	2.5
6.	Jackson District Office, Jackson, TN	42%	0	14	7	4.1	1.4
7.	Rhodes College, Memphis, TN	70%	0	8	2	5.14	2.5
8.	Memphis City Zoo, Memphis, TN	50%	0	7	3	6.96	NA
9.	Montaque Park, Chattanooga, TN	17%	17%	6	5	6.78	1.9
10.	Warner Park, Chattanooga, TN	20%	40%	10	8	7.26	.6
11.	Bailey St. Chattanooga, TN	50%	33%	7	5	6.86	NA
12.	Off 24-E, Chattanooga, TN	47%	13%	13	7	7.24	.9
13.	Grady St., Johnson City, TN	50%	0	6	8	5.3	1.2
14.	Hideaway Farms Rd., Piney Flats, TN	0	0	4	5	6.85	3
15.	Old Fort Gold Course, Murfreesboro, TN	92%	0	13	8	5.58	1.9
16.	Franks Rd, McMinnville, TN	83%	0	4	3	5.4	2.2
17.	Mars Plant, Cleveland, TN	83%	8%	14	9	7.15	2.5

Seasonal and Genomic Differences of *Discula destructiva* and *Discula* species

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Index Words: *Discula destructiva*, *Discula* species, Seasonal Occurrence, Genomic Analysis

Nature of Work: Two pathogens of dogwood anthracnose, *Discula destructiva* and an undescribed species of *Discula*, were isolated from dogwood trees at 13 locations in five southeastern states (Table 1). Environmental and geographical research was conducted on both fungi, but primarily on *D. destructiva* (Windham, 1989). In this study, the occurrence of dogwood anthracnose was correlated to slope position, elevation, proximity to water, diameter breast height (DBH), temperature and humidity. One aspect of the host/pathogen relationship that has not been studied is the differences in apparent seasonal presence of *D. destructiva* and *Discula* species.

Trigiano and co-workers (1995) completed a genomic study of 28 isolates of *D. destructiva* and 3 isolates of *Discula* sp. both fungi using DNA amplification fingerprinting (DAF: Catano-Anollés et al., 1991). Results indicated that the genome of *D. destructiva* was conserved, whereas *Discula* sp. isolates exhibited greater variability.

The objectives of the present study were to determine when *D. destructiva* and *Discula* sp. could be isolated from symptomatic foliage and determine genomic differences between 18 isolates of *Discula* sp. and 2 isolates of *D. destructiva*.

Discula destructiva and *Discula* species were isolated from dogwoods at 13 sites in five southeastern forested sites in all four seasons. Dogwood foliage and twigs were collected and diameter breast height (DBH), elevation, proximity to water and slope were noted at each site. Symptomatic leaves and twigs were cut up into small 4-inch pieces and placed into moisture chambers. After 2 days, plant materials were examined for acervuli, which are characteristic of *Discula* sp. Multisporic isolates were obtained by inserting a sterilized inoculation needle into an acervulus and then inserting the needle into potato dextrose agar (PDA) that had been amended with 20 mg/L each of streptomycin and chlortetracycline (PDA+). Culture plates were sealed with Parafilm and incubated at 20° C. Colonies were confirmed as *Discula* via microscopically examination and then placed in vials of PDA+ for storage. Gallic acid medium was used to identify and differentiate the two species of *Discula* (Trigiano et al., 1991).

Genomic DNA was extracted (Yoon et al., 1991") and amplified using DAF. Amplification reactions were primed with ten octomer primers. Genetic analysis of data was completed with Numerical Taxonomy and Multivariate Analysis System (Ntsys) version 2.0 9Exeter Software, Setauket, New York). Cluster analysis and Principal Coordinate Analysis confirmed that *Discula* sp. was genetically diverse whereas *D. destructiva* was genetically conserved.

Results and Discussion: Thirty-four isolates of *D. destructiva* and 28 isolates of *Discula* sp. were collected in the spring and summer months. One isolate of *D. destructiva* was collected in the fall, and no isolates were isolated in the winter. Our study revealed a positive correlation (*D. destructiva* -0.24322 and *Discula* sp.-0.21837) between the ability to recover both *Discula* species and the seasonal presence of symptomatic tissues.

Environmental and geographical aspects play a role in the epidemiology of dogwood anthracnose (Windham et al., 1995). Since both *Discula* species prefer cool temperatures with high humidity, spring and early summer months should be the optimum time for isolation from host tissues. In the fall, *Discula* can be difficult to isolate because the hottest part of summer may cause the fungus to go dormant, and secondary organisms can invade senescent foliage making isolation difficult. In winter the *D. destructiva* and *Discula* sp. are usually not isolated because dogwood trees are dormant and extremely cold temperatures are not conducive for growth.

DAF was used to genetically profile isolates of *Discula* sp. and *D. destructiva* (Trigiano et al., 1995). *Discula* sp. exhibited relatively variable genome (97.8% polymorphic loci) whereas *D. destructiva* was more conserved (8.9% polymorphic loci). Our results are consistent with previous finding that *Discula* sp. has a high variable genome (Trigiano et al., 1995) and supports the concept that the fungus is native to North America. The data also supports the conclusion that *D. destructiva* is most likely an exotic or introduced fungus.

Significance to Industry: Diagnosis of dogwood anthracnose may only be verified by isolation of the pathogens during late winter, spring and early summer. Confidence in diagnosis of dogwood anthracnose in late summer and fall may be problematic since isolation of *D. destructiva* and *Discula* sp. is not possible.

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Table 1. Isolates found at 13 sites in all four seasons.

Location	2000				2001			
	March – April		May – July		Sept. - Oct.		Dec. – Feb.	
	D.d.*	D.s.	D.d.	D.s.	D.d.	D.s.	D.d.	D.s.
Oak Ridge TN	9	4	3	5	0	0	0	0
Sewanee TN	16	13	5	10	0	0	0	0
Lookout Mountain TN	1	0	1	2	1	0	0	0
Russell Cave AL	1	0	1	2	0	0	0	0
Lookout Mountain GA	2	2	0	0	0	0	0	0
Oswald Dome, TN	5	5	2	4	0	0	0	0
Hiwassee Dam NC	1	1	0	0	0	0	0	0
Appalachian Trail NC	3	0	1	0	0	0	0	0
Bent Creek NC	11	6	11	3	0	0	0	0
Ozone TN	3	0	2	3	0	0	0	0
Blue Ridge Tuggle Gap VA 1	3	1	4	3	0	0	0	0
Blue Ridge Tuggle Gap VA 2	7	3	7	4	0	0	0	0
Virginia Polytechnical Institute and State Univ. VA	0	0	0	0	0	0	0	0

*Number of isolates obtained of *Discula destructiva* (D.d.) and *Discula species* (D.s.)

Searching for Fire blight Resistance in Flowering Pears (*Pyrus* spp.)

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Index Words: *Erwinia amylovora*, host plant resistance, pears

Nature of Work: The genus *Pyrus*, a member of the Rosaceae subfamily Maloideae, consists of approximately 22 species native to Europe, North Africa, and Asia. Pears rank second to apples as the most important deciduous tree fruit crop in the world (5). The popularity and use of flowering pears as an ornamental has increased significantly in recent decades. Callery pear (*Pyrus calleryana*) is one of the most important flowering trees used in landscape horticulture and there are many opportunities for further breeding of improved hybrids.

Fire blight, caused by the bacterium *Erwinia amylovora*, is one of the most significant diseases of rosaceous plants. This disease can be especially problematic with pears, particularly in regions where environmental conditions for the pathogen are favorable. Susceptible plants can be severely damaged and killed by fire blight in both nursery and landscape plantings. Research on fire blight resistance among flowering pears has been limited and has primarily been based on observations of natural infection (4,6). Research utilizing a rigorous approach of controlled inoculations, with specific pathogen strains, to screen cultivars for resistance to fire blight under "worst case" conditions provides an effective means for identifying resistant plants (1,2).

The considerable genetic diversity and interspecific crossability within pears creates specific opportunities for developing improved hybrids. Species vary in flowering time, heat and cold tolerance, leaf shape, tree form, and disease resistance. The ornamental pear breeding program at the Landscape Plant Development Center (Chanhassen, MN) has been successful in obtaining many hybrid plants (5). Approximately 40 promising plants have been identified based on growth, flower, fruit, and foliage data. While desirable ornamental traits from parent plants have been maintained in hybrid offspring, their resistance to fire blight still needs to be documented.

Twenty-seven taxa of containerized pears, arranged in a randomized complete block experimental design with 4-12 replications, were screened for fire blight resistance using controlled inoculations at the

Mountain Horticultural Crops Research Station, Fletcher, N.C. The inoculum for this study was *Erwinia amylovora* strain 2002A obtained from Cornell University. One to two actively growing shoots (subsamples) per tree were inoculated on 11 May 2000. The two youngest leaves were bisected with a pair of scissors that had been dipped into the inoculum at a concentration of $\sim 1.53 \times 10^7$ cfu/ml prior to each cut. Lesion length and total length of the current season's growth were measured. The severity of infection was expressed as the length of the fire blight lesion as a percentage of overall shoot length. All data were subjected to analysis of variance.

Results and Discussion: The taxa included in this study showed considerable variation in resistance to fire blight with the severity of infection ranging from 1 – 100% of the current season's shoot growth (Table 1). Nine taxa were highly susceptible with extensive infection that did not differ significantly from 100%. These susceptible taxa included specific clones of *P. fauriei*, *P. elaeagrifolia*, *P. pyrifolia*, *P. nivalis*, and *P. salicifolia* as well as a number of hybrid cultivars. At the other extreme, two taxa, *Pyrus ussuriensis* 'Prairie Gem' and *Pyrus* 950104, a hybrid clone derived from open pollination of a *Pyrus calleryana* x *Pyrus betulifolia*, were highly resistant with minimal infection that was not significantly different from 0%. Fifteen other taxa were intermediate with lesion length ranging from 16 to 81% of the annual shoot growth.

Pyrus ussuriensis and *P. calleryana* have been reported as being two of the most fire blight resistant species of pears. However, seedling populations of all species are known to show considerable variation in levels of resistance. The study reported here included clonal selections of a variety of species. *P. ussuriensis* 'Prairie Gem' was extremely resistant. Comparisons among cultivars of *P. calleryana* showed significant differences with *P. calleryana* 'Chanticleer' being significantly more resistant than *P. calleryana* 'Bradford', 'Whitehouse', 'Aristocrat', and 'Red Spire'. Our results were in general agreement with previous studies which the cultivars 'Bradford', 'Fauriei', and 'Whitehouse' showed greater resistance than 'Aristocrat' and 'Redspire' under natural conditions, though in our study 'Whitehouse' was not significantly more resistant than either 'Aristocrat' or 'Red Spire' (4,6). We also found *P. calleryana* 'Chanticleer' (syn. 'Cleveland Select') to be significantly more resistant than 'Bradford'. *Pyrus betulifolia* 'Dancer', a clone of *P. regelii*, and the clone 93-70-2 were all as resistant as *P. calleryana* 'Chanticleer'.

Significance to Industry: Cultivars of *Pyrus calleryana* are among the most widely planted flowering trees in the Southeastern United States. Despite their popularity, these plants decline significantly with age. Some cultivars are very weak wooded and susceptible to fire blight.

Genetic diversity and interspecific crossability in pears has led to the development of many new hybrid plants. The success of new hybrids is dependent on their resistance to fire blight. Fire blight resistant pears will lower production costs and greatly enhance the appearance and life of the tree as an ornamental. This research used a rigorous approach to screen for fire blight resistance and the information will serve as a basis for the selection of new improved hybrid cultivars of flowering pears.

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Table 1: Fire blight ratings for controlled inoculations of flowering pears (Pyrus spp.)

Pyrus taxa	Severity of infection (% lesion length)
950104 ^y (<i>P. calleryana</i> x <i>P. betulifolia</i>)	1 ^z
<i>P. ussuriensis</i> 'Prairie Gem'	3
<i>P. betulifolia</i> 'Dancer'	16
<i>P. regelii</i>	22
93-70-2 ^y (<i>P. calleryana</i> 'Chanticleer' x <i>P. elaeagrifolia</i>)	22
<i>P. calleryana</i> 'Chanticleer'	31
93-61-1 ^y (<i>P. amygdaliformis</i> x <i>P. calleryana</i> 'Chanticleer')	32
91-42-1 ^y (<i>P. amygdaliformis</i> x <i>P. regelii</i>)	38
911014 ^y (<i>P. ussurensis</i> x <i>P. regelii</i>)	42
93-15-1 ^y (<i>P. elaeagrifolia</i> x <i>P. ussuriensis</i>)	44
<i>P. calleryana</i> 'Fauriei'	46
<i>P. calleryana</i> 'Bradford'	50
<i>P. calleryana</i> 'Whitehouse'	62
91-53-1 ^y (<i>P. calleryana</i> 'Chanticleer' x <i>P. betulifolia</i>)	63
<i>P. calleryana</i> 'Aristocrat'	65
<i>P. calleryana</i> 'Red Spire'	69
93-17-3 ^y (<i>P. elaeagrifolia</i> x <i>P. amygdaliformis</i>)	81
93-2-2 ^y ((<i>P. calleryana</i> x <i>P. fauriei</i>) x <i>P. nivalis</i>)	87
<i>P. fauriei</i> 'Korean Sun'	89
<i>P. elaeagrifolia</i> 'Turkish Mist'	91
911010 ^y (<i>P. ussuriensis</i> x <i>P. nivalis</i>)	92
93-32-4 ^y (<i>P. salicifolia</i> 'Pendula' x <i>P. ussuriensis</i>)	94
<i>P. pyrifolia</i>	95
<i>P. nivalis</i> 808	97
93-8-5 ^y (<i>P. fauriei</i> x <i>P. salicifolia</i> 'Pendula')	98
<i>P. salicifolia</i> 'Pendula'	100
LSD _{0.05}	15

^z% of total shoot length infected

^y interspecific hybrid taxa

Evaluation of phosphite as an alternative phosphorus nutrient and control for Phytophthora disease

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Index words: Annual vinca, *Catharanthus roseus*, *Phytophthora nicotianae*

Nature of work: Phosphorus fertilizers are generally supplied to plants as salts of phosphoric acid (H_3PO_4). When phosphoric acid is neutralized by potassium hydroxide (KOH) it forms the phosphate fertilizer, potassium phosphate (KH_2PO_4). However, if phosphorous acid (H_3PO_3) is neutralized with KOH, it forms the salt of phosphorous acid, potassium phosphite (KH_2PO_3), also known as phosphite or phosphonate. Phosphite fertilizers are available commercially and are labeled as phosphorus supplements for foliar or soil treatment to nursery, turf, and landscape crops. However, some plants are sensitive to phosphite and may become chlorotic or stunted if phosphite is used as the only source of phosphorus.

Phytophthora spp. are fungi that cause some of the most widespread and serious root rot and foliar blight diseases on plants. Phosphite or phosphonate salts have been effective in controlling *Phytophthora* diseases when applied as fungicides (1, 3). Phosphite utilized as a phosphorus nutrient in hydroponic culture also reduced the incidence of *Phytophthora* root and crown rot of tomato and pepper (2). The objective of this study was to determine if phosphite fertilization could provide acceptable growth of ornamental plants in a standard pine bark medium and, at the same time, inhibit the development of *Phytophthora* disease. *Phytophthora* foliar blight of annual vinca (*Catharanthus roseus*) was used for this study.

In the first experiment, annual vinca seedlings were potted into 2 quart containers in a pine bark medium, 2 plants per pot, and placed on greenhouse benches. The plants were irrigated individually with a complete hydroponic nutrient solution containing varying amounts of phosphorus supplied as phosphite or phosphate or combinations of the two (Tables 1 and 2). The pH of all solutions was adjusted to 6.2 with 0.5 N potassium hydroxide (KOH). Plants were measured after 21 and 31 days, and given a visual appearance rating 31 days after initiation of the nutrient treatments. After 63 days, shoot and root dry weights were obtained in addition to plant measurements and appearance ratings.

In the second experiment, vinca plants were potted and grown as in the first experiment and irrigated as before with nutrient solutions containing selected proportions of phosphite or phosphate (Tables 3 & 4). A

commercial phosphite nutrient [NutriGrow PK (0-28-26), Biagro Western Sales, Inc., Visalia, CA] in amounts equivalent to the technical phosphite was also evaluated at this time. Two weeks after initiation of the nutrient treatments, the plants were inoculated by misting them with a water suspension of *Phytophthora nicotianae* zoospores containing 3.3×10^3 zoospores per milliliter (approximately 5 ml/pot). The plants were evaluated for disease 5 and 12 days after inoculation. For both experiments a randomized complete block design was used with 5 replication, and 2 plants per treatment. Data were analyzed with analysis of variance (ANOVA) with mean separations by LSD, $P=0.05$.

Results and Discussion: In the first experiment, plants grown with phosphite alone as a phosphorus source were significantly smaller than plants grown with equivalent amounts of phosphate or combinations of phosphite or phosphate. Visual ratings and shoot and root dry weights for the phosphite only plants were also significantly lower than for the other treatments (Tables 1 & 2). However, plants receiving 9 or 15 ppm phosphorus as phosphate in combination with 3 or 9 ppm phosphorus as phosphite were comparable in size and appearance to those receiving 15 ppm phosphorus as phosphate alone.

In the second experiment, by 12 days after inoculation, inoculated plants fertilized with solutions containing 3 or 9 ppm phosphorus as phosphite, either technical or commercial, had significantly fewer diseased leaves and shoot tips than plants fertilized with equal amounts of phosphate (Table 4). However, plants receiving combinations of phosphite and phosphate had amount of disease that was not significantly different from the plants getting phosphate alone. These results suggest that phosphite applied as part of a nutrient solution provides some control of phytophthora foliar blight of annual vinca. However, this control appears to be negated if phosphate is applied simultaneously to improve plant growth.

Significance to the Nursery Industry: Although phosphite applied in a nutrient solution provided significant control of phytophthora blight on annual vinca, it cannot be recommended as a substitute for phosphate fertilization at this time. When phosphite was used as the only source of phosphorus, poor growth of vinca resulted. Additional research is needed on application methods or timing of application.

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Table 1. Effect of varying proportions of phosphate and phosphite phosphorus (P) fertilization on growth and appearance of *Catharanthus* 'Little Bright Eye'.

ppm P as Phosphate	ppm P as Phosphite	21 Days ²	31 Days	
		Growth Index (cm)	Growth Index (cm)	Visual ³ Rating
0	0	18.4	22.8bc ^x	2.1d
3	0	17.6	22.8bc	3.1c
9	0	18.4	22.9bc	3.1c
15	0	17.9	23.8ab	3.6ab
0	3	18.1	22.1c	2.2d
3	3	18.4	23.9ab	3.4bc
9	3	18.0	23.5ab	4.0a
15	3	18.9	24.3a	3.3bc
0	9	16.8	19.4d	1.9d
3	9	18.8	23.6ab	3.2bc
9	9	18.2	24.0ab	3.9a
15	9	17.9	24.5a	3.9a
		NS		

² Days after start of nutrient treatments.

³ Appearance rating based on plant fullness, color, amount of leaf drop, and flower size and shape.

^x Mean separation by LSD, (P = 0.05). NS = no significant difference.

Table 2. Effect of varying proportions of phosphate and phosphite phosphorus (P) fertilization on growth, appearance, and shoot and root dry weights, 63 days after initiation of nutrient treatments.

ppm P as Phosphate	ppm P as Phosphite	Growth Index (cm)	Visual ² Rating	Shoot Dry Wt (g)	Root Dry Wt (g)
0	0	30.0e ^y	1.0c	5.7d	4.0de
3	0	34.1cd	3.3a	14.7c	5.8a
9	0	37.1ab	3.3a	16.3b	4.4cd
15	0	36.1abc	3.7a	18.3a	5.1ab
0	3	24.6f	1.0c	6.8d	1.4f
3	3	34.9bc	2.7b	17.2ab	3.2e
9	3	37.0ab	3.7a	18.4a	5.0abc
15	3	37.8a	3.4a	17.7ab	5.6a
0	9	23.6f	1.2c	6.6d	1.0f
3	9	31.9de	2.3b	14.6c	4.2cd
9	9	36.0abc	3.3a	17.3ab	3.8de
15	9	37.9a	3.3a	17.4ab	3.8de

² Appearance rating based on plant fullness, color, amount of leaf drop, flower size and shape.

^y Mean separation by LSD (P = 0.05).

Table 3. Effect of varying levels of phosphate and phosphite phosphorus (P) fertilization on disease development on *Catharanthus 'Little Bright Eye'* 5 days after inoculation with *Phytophthora nicotianae*.

ppm P as Phosphate	ppm P as Phosphite	Number of diseased plants	Number of diseased leaves	Number of diseased shoot tips
9	0	1.6ab ^z	3.8ab	0.6
15	0	2.0a	5.4a	0.4
0	3	1.6ab	4.6ab	0.6
9	3	2.0a	4.0ab	0.4
0	9	0.4cd	0.8cd	0.0
9	9	1.6ab	4.6ab	0.4
15	9	1.8a	4.4ab	1.0
0	9 commercial ^y	1.0bc	2.6bc	0.8
9	9 commercial	1.8a	4.4ab	0.4
9	0 non-inoculated	0.0d	0.0d	0.0
0	9 non-inoculated	0.0d	0.0d	0.0
				NS

^z Mean separation by LSD, P = 0.05.

^y Provided as NutriGrow PK (0-28-26) commercial nutrient.

Table 4. Effect of varying levels of phosphate and phosphite phosphorus (P) fertilization on disease development on *Catharanthus 'Little Bright Eye'* 12 days after inoculation with *Phytophthora nicotianae*.

ppm P as Phosphate	ppm P as Phosphite	Number of diseased plants	Number of diseased leaves	Number of diseased shoot tips
9	0	1.8a ^z	11.6abc	1.8a
15	0	2.0a	13.2ab	2.0a
0	3	1.8a	7.8cd	1.0bc
9	3	2.0a	12.8ab	2.0a
0	9	0.4c	2.4ef	0.4cd
9	9	1.8a	10.6abcd	1.6ab
15	9	1.8a	15.0a	1.6ab
0	9 commercial ^y	1.2b	6.2de	1.0bc
9	9 commercial	1.8a	10.2bcd	1.4ab
9	0 non-inoculated	0c	0f	0d
0	9 non-inoculated	0c	0f	0d

^z Mean separation by LSD, P = 0.05.

^y Provided as NutriGrow PK (0-28-26) commercial nutrient.

Improved Control of Black Spot Disease on Roses with Winter Fungicide Treatments

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Index Words: *Rosa* sp., *Diplocarpon rosae*, black spot

Nature of Work: Black spot disease, caused by *Diplocarpon rosae* F.A. Wolf, is the most important disease of cultivated roses (*Rosa* spp.) in the southeastern U.S. (3, 6). This disease is characterized by black spots on foliage and premature defoliation of plants. Once established on plants, black spot is difficult to control despite a combination of practices including sanitation measures and fungicide applications (1).

The protectant fungicide chlorothalonil, when applied on 14-day intervals through the growing season, provides better control of black spot than several other fungicides including myclobutanil and triforine (4, 5). While best control of black spot is achieved when chlorothalonil is applied on 7-day intervals, longer intervals between applications are desirable in order to reduce fungicide use. In addition to fungicides applied during the growing season, lime-sulfur applications to dormant plants are also recommended for control of this disease (1). In a recent study, various fungicidal compounds, applied to dormant plants in the winter, were evaluated for their effectiveness in reducing black spot severity (2). Winter applications of Funginex and cyproconazole delayed disease initiation in the spring, and decreased disease development and improved plant vigor through subsequent growing seasons, but only cyproconazole had a significant effect on disease and plant vigor (2).

Cyproconazole is no longer available for use in the U.S., yet there are other fungicides of similar chemistry on the market that may be labeled for use on roses. The objectives of this study were to evaluate systemic fungicides as winter treatments for control of black spot disease and to determine if such winter treatment can allow reduction of fungicide applications during the subsequent growing season.

This study was initiated in the fall of 1999 using rose plants established at the E.V Smith Research Station near Shorter, Alabama. Three hybrid tea rose cultivars, 'Cary Grant,' 'Dolly Parton,' and 'Princess of Monaco,' comprise each plot and treatment plots are replicated three times in a randomized complete block design. Four winter treatments were combined with each of five foliar treatments applied through the growing season for a total of twenty fungicidal treatments. Winter treatments were: an

untreated control, tetraconazole (Eminent[®] 125 ME, 0.35 g A.I./L), Funginex[®] (triforine, 0.26 g A.I./L), and Eagle[®] WSP (myclobutanil, 0.12 g A.I./L). The initial winter application was planned within 1 week of the first frost, with subsequent applications following 30 consecutive days > 28 F. Winter applications were done 3 Dec 1999 and 8 Mar 2000 in the first study year, and 28 Nov 2000 in the second. Foliar treatments were applied on 2 week intervals except for Daconil[®] Ultrex (chlorothalonil, 1.3 g A.I./L) treatment applied weekly as a control. Foliar treatments began about 1 May and were: Daconil Ultrex, tetraconazole, Funginex, and Eagle WSP, each at rates previously stated. Rose plants were rated every 14 days for disease severity, vigor and flower production starting mid-April and continuing through Sept. Black spot ratings were based on a scale of 0-5 where 0=no disease, 1=black spot on approximately 20% of foliage, 2=black spot on approximately 40% of foliage, 3=black spot on 60% of foliage, 4=black spot on 80% of foliage, and 5=black spot on 100% of foliage. Vigor was rated on a scale of 1-5 with 1=poorly formed plant with little or no new growth and 5=well-developed plant with abundant new growth. Flower production was the sum of buds showing color, blooms, and spent flowers. Root sprouts, diseased or dead canes, and spent flowers were regularly removed with pruning. Disease as well as vigor ratings and flower production were averaged over each growing season (May 1 through Sept).

Results and Discussion: Ratings done in early May of each of the two study years indicated that tetraconazole reduced disease severity early in the growing season, although this effect was not significant in 2001 (Table 1). There were no significant effects on vigor due to winter treatments in early May of each year (Table 1). Among winter treatments, the tetraconazole treatment contributed to minimizing disease through the entire 2000 season better than winter treatment with Funginex or Eagle (Table 2). Plants sprayed weekly with Daconil through the growing season, as well as plants treated with tetraconazole in the winter plus either Daconil or tetraconazole on 14 day intervals, had the lowest disease levels in 2000 (Table 2). Average vigor and flower production over the growing season were not affected by treatment in 2000. Data collected through May 2001 indicate that treatment effects are consistent with those of the first study year. No phytotoxicity was observed on any plants in the study. These results indicate that winter fungicide treatments can contribute to black spot minimization on roses and may allow a decrease in numbers of fungicide applications through the subsequent growing season.

Significance to the Nursery Industry: Reduction of fungicide use, without a loss in disease control, is desirable for several economic and environmental reasons. With chlorothalonil becoming more limited in availability, alternatives to this efficacious product will need to be found. Our results indicate that there are alternatives to chlorothalonil for control

of black spot on roses. In addition, treatment of roses during their dormancy, with certain systemic products, can contribute to black spot disease management.

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Table 1. Average black spot disease and vigor ratings of hybrid tea roses after treatment during the winter.

Date	Winter Treatment	Disease	Vigor
2 May 2000	None	2.81ab	3.81
	tetraconazole	2.04b	3.88
	Funginex	2.89a	4.11
	Eagle	2.67ab	3.50
4 May 2001	None	2.84	3.96
	tetraconazole	2.80	4.05
	Funginex	2.99	3.97
	Eagle	3.04	3.88

Data in columns are means of three cultivars and three replications of each treatment. Letters following means within a column for either date, when different, indicate a significant difference according to Fisher's protected least significance difference test at $P = 0.05$.

Table 2. Average black spot disease ratings of hybrid tea roses through the 2000 growing season when treated with products during the winter and May through September.

Winter Treatment ^b	Foliar Treatment ^a						Winter Trt Average
	weekly Daconil Ultrex	Daconil Ultrex	tetraconazole	Funginex	Eagle		
None	0.53 a	1.55 de	0.88 abc	1.58 e	1.25 cd		1.16 AB
Tetraconazole	0.64 a	0.67a	0.67 a	1.27 cd	1.58 de		0.98 A
Triforine	0.64 a	1.23 cd	0.79 ab	1.81 e	1.85 e		1.29 B
Myclobutanil	0.65 a	0.96 bc	0.91 abc	1.61 de	1.89 e		1.20 B
Foliar Trt Average	0.61 A	1.10 B	0.81 A	1.58 C	1.69 C		

^aFoliar treatments applied May through September on 14-day intervals except for the weekly chlorothalonil which was applied on 7-day intervals.

^bWinter treatments were applied 3 December 1999 and 8 March 2000.

Data are means of three cultivars and three replications of each treatment. Noncapitalized letters following means, and capitalized letters within a column or across a row, when different, indicate a significant difference according to Fisher's protected least significance difference test at $P = 0.05$.

Fungicide Movement in Runoff Water at a Container Plant Nursery

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Index Words: Integrated pest management; Cleary's 3336; Daconil 2787; Subdue GR.

Nature of Work: Container plants are managed to produce a quality product in a short period of time. Production conditions that are optimal for plant growth are equally optimal for proliferation of plant pathogens, and fungicides are routinely used in container nurseries to manage and prevent pest infestations. Fungicides have been detected in runoff water from a container nursery (1). Concern over the possibility of contamination of aquatic ecosystems warrants investigation of management practices that will reduce pesticide levels in runoff water. A basic tenet of integrated pest management (IPM) is the reduction of quantities of applied pesticides through a holistic management strategy. This study investigated the impact of IPM based pesticide applications on container plant health and fungicide amounts transported in runoff water at a container nursery.

During the summer of 1998, a field study was conducted at Gilbert's Nursery, near Chesnee, SC. Eight contiguous production beds were divided into two treatments: preventative pesticide applications in which fungicides were applied on a monthly basis; and, IPM based pesticide applications in which fungicides were applied only to affected plant species when signs of fungal pathogens were detected during weekly scoutings. Three applications of fungicides were applied. In July, Cleary's 3336 (50% thiophanate-methyl) and Daconil 2787 (82.5% chlorothalonil) were spray applied to the preventative treatment only. On August 4, 1998, Cleary's 3336 and Daconil 2787 were applied to the preventative treatment and to Nikko blue bigleaf hydrangea in the IPM treatment. Application rates were 0.6 lbs a.i. A⁻¹ Cleary's 3336, and 0.7 lbs a.i. A⁻¹ Daconil 2787 for both applications. Pulse irrigation consisting of three 30-min cycles with a 90-min rest between cycles was applied on the day after application (DAA) in July and August and runoff samples were collected at 15 min intervals for all pulse cycles.

In September, Subdue GR (1% metalaxyl) was applied to all plant species in the preventative treatment and to four plant species in the IPM treatment that were manifesting symptoms of *Phytophthora* diseases. Application rate was 1.8 lbs a.i. A⁻¹. Pulse irrigation was applied in two 30-min cycles with a 90-min rest between cycles. Runoff samples were taken at 15 min intervals of runoff flow from the first pulse cycle and one

runoff sample was taken from the second cycle at approximately 15 min of runoff flow. Pesticides were extracted from runoff water onto C₁₈ solid phase extraction columns and analysis was by high pressure liquid chromatography.

To document the impact of the two management strategies on plant health, the number of culls (unsalable containers) of individual plant species in treatments was counted in November 1998. Containers were designated as culls if defoliation had occurred or if pathogen presence was observable.

Results and Discussion: In July, Cleary's 3336 was detected in all runoff samples in the preventative treatment. Greatest concentrations were found in the first pulse irrigation cycle (Table 1). Highest concentration was 1.36 $\mu\text{g ml}^{-1}$, below the maximum water solubility of Cleary's 3336. Amounts of the fungicide detected in runoff were 3.6, 4.1 and 0.3 g from pulse cycles A, B and C, respectively (LSD=1.4), and total amount detected was 7% of the applied amount. Daconil 2787 was also detected in all runoff samples (Table 1). Greatest concentration found was 0.95 $\mu\text{g ml}^{-1}$ which is slightly above reported maximum solubility indicating some transport of dispersed chemical not in solution. As a percent of applied, 4% of Daconil 2787 was detected in runoff water.

In August, Cleary's 3336 and Daconil 2787 were detected at much greater concentrations from the preventative treatment than the IPM treatment, as expected (Table 2). In both treatments, Cleary's 3336 was only found in runoff from pulse cycles A and B. Total amounts of Cleary's 3336 detected as leaving application site in runoff water were 2% of applied amount from the preventative treatment and 37% from the IPM treatment. Total amounts of Daconil detected as leaving application site in runoff water were 4% of applied amount from the preventative treatment and 12% from the IPM treatment. Differences in percent of applied amounts may be the result of application method. Wind transported spray solution from the mist application to the preventative treatment may have resulted in contamination of IPM beds.

In September, Subdue GR was detected at higher concentrations and amounts from the preventative treatment than the IPM treatment in all runoff samples (Table 3). Highest level detected was 61 $\mu\text{g ml}^{-1}$ for the preventative treatment and 30 $\mu\text{g ml}^{-1}$ for the IPM treatment. As a percent of applied amount, 25% was noted in runoff water in the preventative treatment and 30% from the IPM treatment. Amounts detected indicate that the potential for Subdue GR to leave application site in runoff water is great. Though concentrations detected in runoff water were below levels toxic to fish (3), broadcast applications should not be made at

container nurseries. Subdue GR is stable in aqueous solutions for long periods (2) and repeat applications could result in a build-up to toxic concentrations.

IPM based pesticide applications will only be adopted by the container nursery industry if plant health is not compromised. In this study, plant health was evaluated by assessing marketability of species at study end. There were no differences between treatments in number of unsalable plants.

Significance to Industry: Implementation of IPM strategies effectively reduced amounts of fungicides transported in runoff water, without compromising container plant health and marketability. Total amounts of Cleary's 3336, Daconil 2787 and Subdue GR detected in runoff water were 12.5, 12.8 and 83.4 g for the preventative treatment and 1.5, 1.2 and 20.6 g from the IPM treatment. Plant quality was similar between the treatments. Greatest concentrations detected for all fungicides were below levels toxic to aquatic organisms, and risks to aquatic ecosystems associated with the use of these pesticides appear to be minimal. However, repeat applications of Subdue GR and Daconil 2787 may result in a build-up of contaminants in surface water based on reported half-life values. Implementation of IPM pesticide application strategies would result in smaller amounts of pesticides transported in runoff water.

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Table 1. Concentrations ($\mu\text{g ml}^{-1}$) and amounts (g) of fungicides detected in runoff water from preventative treatment beds in July 1998. Means are of $n = 4$ replications.

Irrigation (Pulse - Sample)	Cleary's 3336		Daconil 2787	
	$\mu\text{g ml}^{-1}$	g	$\mu\text{g ml}^{-1}$	g
A-1	1.01	0.4	0.95	0.4
A-2	1.18	1.3	0.82	0.9
A-3	1.36	1.9	0.68	1.0
B-1	1.06	0.7	0.83	0.5
B-2	0.77	1.0	0.65	0.8
B-3	1.00	2.5	0.92	1.7
C-1	0.24	0.1	0.51	0.2
C-2	0.10	0.2	0.32	0.5
C-3	0.03	0.1	0.34	0.7
LSD ($P=0.05$)	0.49	0.7	0.46	0.7
Total		8.4		6.7
% of applied		7		4

Table 2. Cleary's 3336 and Daconil 2787 amounts (g) from the preventative and IPM treatments for pulse cycles A, B, and C, from the August application. Asterisks indicate a significant difference between treatments at $\alpha = 0.05$ (*) and 0.01 (**).

Pulse	Cleary's 3336		Daconil 2787	
	Preventative	IPM	Preventative	IPM
	-----		-----	
	$\mu\text{g ml}^{-1}$		g	
A	2.5**	1.0	1.2**	0.5
B	1.6*	0.5	3.5**	0.5
C	nd ¹	nd	1.4*	0.2
LSD ($P=0.05$)	0.5	0.4	0.8	0.4
Total	4.1**	1.5	6.1**	1.2
% of applied	2	37	4	12

¹ Not detected.

Table 3. Subdue GR concentrations ($\mu\text{g ml}^{-1}$) and amounts (g) in runoff water detected in the preventative and IPM treatments. Asterisks indicate a significant difference between treatments at $\alpha = 0.05$ (*) and 0.01 (**).

Runoff Sample	Preventative		IPM	
	$\mu\text{g ml}^{-1}$	g	$\mu\text{g ml}^{-1}$	g
A-1	61.0*	30.1	6.4*	3.2
A-2	40.1*	7.7	29.1*	5.6
A-3	29.1**	3.1	14.8**	1.6
B	16.6*	5.1	33.1*	10.3
LSD ($P=0.05$)	19.8	18.4	16.1	7.1
Total			83.4**	20.6
% of applied			25	30

Influence of select inorganic elements and pH on the fungicidal activity of chlorine dioxide in water.

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Index Words: chlorine dioxide, disinfectant, water treatment, suspension test

Nature of Work: Chlorine dioxide (ClO_2) is gaining market share as a disinfectant used to kill bacteria and/or fungi in treatment of public and private drinking water, fish, poultry, red meat, fruits, potatoes, and vegetables (1,4,6). Part of the market share is replacing use of hypochlorites, because ClO_2 is less affected by pH, less reactive to organic and inorganic materials (eg. ammonium, chloramines, bromines, etc.), removes phenolic tastes and odors, and produces fewer to no toxic by-products (eg. trihalomethanes and chlorophenols) (1,4). ClO_2 has a higher oxidative capacity than most disinfectants, 2 $\frac{1}{2}$ times higher than sodium hypochlorite (bleach), thus it has a higher biocidal activity at an equal concentration. While limited, research to date shows ClO_2 treated water should not damage many plant species at rates required to kill common plant pathogenic fungi (2, 3, 7, Copes unpublished).

One limitation is ClO_2 can not be purchased directly, but must be generated on site, which increases cost (1). In fruit and vegetable packing lines, the generating system is part of a water circulating system. In a return line, the water stream is shunted through a by-pass loop where ClO_2 concentration is measured with an oxidative-reduction potential (ORP) probe at a regular time interval (eg. every 5 minutes), and monitored by a small computer system. At a low-limit set point, sodium chlorite (NaClO_2) and an acid, such as hydrochloric acid (HCl), are proportionally metered into a holding tube. The two chemicals react and generate ClO_2 , which is injected into the water line that goes to a dump tank or spray nozzles over the grading line. This system could be adapted for ornamental plant production industries to treat irrigation water pumped from catchment ponds or in ebb-and-flow systems.

Most ornamental plant production watering systems do not continuously circulate water, so ClO_2 could not be regulated in the exact manner previously described. ClO_2 would probably need to be injected close to the water pump and rates would be precalculated by calculating for water flow rate and demand load of the local water source.

A demand load comes from the presence and type of reactants, such as pH, nutrient leachates, organic matter, etc., that reduce the activity of a disinfectant (1,4,5). All disinfectants react quickly with various elements

and molecules (eg. hydrophilic acids, ionic solutes, phenols), although the type and rate of reactions vary with the disinfectant (1,4). The reactive nature of a disinfectant as well as degradation due to light and loss due to volatility are some of the reasons for a lack of residual protection similar to fungicides (1,4). Disinfectants must be added at a concentration that compensates for the demand load, so the remaining concentration will be high enough to kill problematic microorganisms.

Instrumentation exists to measure concentrations of disinfectants in water for the purpose of verifying that a concentration has been reached, however this is not likely to become a common practice. Rate guidelines that account for demand loads and pathogen differences do not exist. The objective of this research was to determine biocidal activity of various concentrations of ClO_2 at a set exposure period (30 sec) against several fungal species and types of spores after ClO_2 was exposed to different concentrations of ammonium (NH_4), nitrate (NO_3), copper (Cu), iron (Fe), manganese (Mn), zinc (Zn), and synthetic hard water (sHW = calcium and magnesium carbonates) and different pH levels that may be encountered in irrigation water.

Results and Discussion: Ionic water amendments and pH significantly affected ClO_2 activity similarly for all fungal species, but the lethal concentration of ClO_2 varied by fungal species and spore type. ClO_2 activity was unaffected by NH_4 , NO_3 , Zn; slightly reduced by Cu, sHW; moderately reduced by Fe; highly reduced by Mn; and severely reduced by pH 9 and 10. Biocidal activity of ClO_2 was higher at pH 4 then pH 8, but the influence was overcome by an rate increase of 2.5-5 ppm ClO_2 . Effects from ionic amendments were generally additive, while pH and sHW had significant interactions with ionic amendments.

A \geq 95% mortality of *Fusarium oxysporum* micro- and macro-conidia resulted from 1 part per million (ppm) ClO_2 (= 1 μg ClO_2 per 1 ml water) with pH 5 or 8 and no elements, from 5 ppm ClO_2 with pH 5 and 5 ppm micro-nutrients (a mixture of 5 ppm of each Cu, Fe, Mn, and Zn), and from 9 ppm ClO_2 with pH 8 and 5 ppm micro-nutrients. A \geq 95% mortality of *Thielaviopsis basicola* endoconidia resulted from 1 ppm ClO_2 with pH 5 and no elements, from 3 ppm ClO_2 with pH 8 and no elements, and from 9 ppm ClO_2 with pH 5 or 8 and 5 ppm micro-nutrients. A \geq 95% mortality of *Botrytis cinerea* conidia resulted from 5 ppm ClO_2 with pH 5 and no elements, from 10 ppm ClO_2 with pH 8 and no elements, and from 20 ppm ClO_2 with pH 5 or 8 and 5 ppm micro-nutrients. A \geq 90% mortality of *Thielaviopsis basicola* aleuriospores resulted from 34 ppm ClO_2 with pH 5 and no elements, from 58 ppm ClO_2 with pH 8 and no elements, and from 70 ppm ClO_2 with pH 5 or 8 and 5 ppm micro-nutrients.

Results show that a low concentration of ClO_2 can kill fungal spores, but fungal species, propagule type, and properties of water influence the rate, whereby a higher rate would be required. Activity of ClO_2 would also be affected by organic matter, temperature, and length of exposure to the ClO_2 , factors not included in this experiment. To disinfect irrigation water, rates of ClO_2 would have to be calibrated to water flow rates and time of exposure from the injection point to emission from sprinkler heads.

Significance to Industry: Chlorine dioxide is a disinfectant that has potential application in treating large quantities of irrigation water. In addition, ClO_2 may have application via irrigation water to treat production and plant surfaces. Data shows the activity of ClO_2 is reduced by water properties, such as manganese, iron, and alkaline pH to the point that higher rates would be required. This research provides partial definition of rates required to kill propagules of some fungal pathogens.

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Evaluation of Azoxystrobin (Heritage) for Control of Southern Blight of *Aucuba*

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Index Words: *Sclerotium rolfsii*, *Aucuba japonica*, azoxystrobin

Nature of Work: Southern blight (*Sclerotium rolfsii*) is a common and persistent fungal disease in the southeastern U.S. It attacks a number of ornamental hosts in container nurseries, field nurseries, and landscapes. The disease is favored by high day (86°F or above) and night (70°F or above) temperatures, and severe outbreaks often follow periods of heavy rain or over watering (1). Symptoms of the disease include sudden wilting of the entire plant, often accompanied by a white mass of fungal growth visible at the crown and on the medium surface. Numerous white to brown mustard seed sized sclerotia may be visible on the soil surface. Sclerotia can persist in the soil or potting media for years making the control of this disease difficult. Labeled fungicides for control of this disease are limited.

Recently potted *Aucuba japonica* liners in trade gallon pots were obtained from a commercial nursery. The potting medium was a 2:1 (v/v) pine bark:grit medium amended with 14 lbs of Osmocote 17-7-12 and 8 lbs of dolomitic limestone per yard³.

Treatments were applied on 5 September and 3 October 2000 as a drench (approximately 250 ml or 8.5 fl oz per plant) or as a heavy spray of the lower stem (approximately 60 ml or 2 fl oz per plant). Plants were inoculated 24 hours after the first treatment application by placing 6 to 10 infested oat seeds adjacent to the stem of each plant just below the soil surface. Infested oat seed inoculum was prepared as described previously (2). Plants were arranged in a randomized complete block design with 6 single plant replications and maintained in a plastic covered greenhouse. Plant mortality was assessed on 25 October 2000.

Results and Discussion: Disease pressure was high as indicated by 100 percent plant loss in the non-treated inoculated plants (Table 1). Heritage applied at 1 oz/100gal, either as a drench or heavy spray, did not provide satisfactory control of the disease. Both Moncut at 8 oz/100 gal and Heritage at 4 oz/100 gal applied as a drench provided excellent control. Similar results were observed with the 2 oz /100 gal Heritage drench, the 4 oz/100 gal Heritage heavy spray, and the Terraclor drench, although there was some plant loss in these three treatments.

Significance to Industry: Heritage (azoxystrobin), Moncut (flutalonil), and Terraclor (PCNB) can be effective in reducing plant death caused by southern blight. The effective drench rates for Heritage used in this test are somewhat higher than the label rates. Although Moncut is not labeled for ornamental use, there are ornamental fungicides available that contain flutalonil. Work has been initiated to refine both rate and method of application for azoxystrobin and flutalonil to develop control strategies that will meet label specifications for use on ornamentals. This work does not endorse or recommend the use of products outside their labeled crops or rates. Always follow the label directions when applying any pesticide.

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Table 1. Percent mortality of *Aucuba japonica* inoculated with *S. rolfsii* and treated with selected fungicides.

Treatment	Rate/100 gal	Treatment Method ¹	% Mortality
Inoculated Control	—	—	100a
Uninoculated Control	—	—	0c
Heritage 50W (azoxystrobin)	1 oz	Heavy Spray	100a
Heritage 50W	1 oz	Drench	83a
Heritage 50W	2 oz	Heavy Spray	83a
Heritage 50W	2 oz	Drench	17c
Heritage 50W	4 oz	Heavy Spray	33bc
Heritage 50W	4 oz	Drench	0c
Terraclor 75W(PCNB)	8 oz	Drench	33bc
Moncut 50W(flutalonil)	8 oz	Drench	0c

Mean separation according to Duncan's Multiple Range Test (P = 0.05).

¹Drench was applied to lower stem and medium surface in a volume of water equal to approximately 250 ml or 8.5 fl oz per gallon. Heavy spray was applied to lower stem in a volume of water equal to approximately 60 ml or 2 fl oz per gallon.

DNA Analysis of Powdery Mildew Pathogens of Dogwood

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Nature of the Study: *Phyllactinia guttata* and *Microsphaera pulchra* have been associated with the powdery mildew disease in flowering dogwood [*Cornus florida*]. DNA analysis of these fungi provides a helpful tool for the understanding of their genetic structure. Such information is useful in disease management especially with host genetic resistance. Since these fungi are obligate parasites, they cannot be grown in artificial media to produce a sufficient mass of fungal tissue for DNA extraction. The objective of this study was to develop a technique that uses a very small amount of fungal tissue to generate a larger amount of DNA sufficient for genetic analysis.

A technique previously used in genetic analysis of single sperm cells by using primer extension pre-amplification (PEP) primers and standard polymerase chain reaction (PCR) protocols was evaluated on the dogwood pathogens. The technique produced a larger amount of DNA and allowed subsequent PCR reactions involving ITS1 and ITS 4 primers from a very small concentration of initial DNA. Products from the ITS 1 and ITS 4 for *P. guttata* and *M. pulchra* using the PEP technique were of similar size and were tested with specific primers for *P. guttata* and *M. pulchra*. Results were similar to those generated with out using the PEP technique, but the quantity of the DNA was much larger. Information from this study will facilitate genetic studies of the powdery mildew pathogens and subsequently benefit efforts in breeding for disease resistance.

Introduction: Powdery mildew is one of the most common and easily recognizable diseases in dogwood. Two fungi, *P. guttata* and *M. pulchra* have been associated with this disease. These obligate parasites obtain their nutrients from plants by sending specialized feeding organs (haustoria) into the epidermal cells of the leaves, without invading the tissue itself. It causes a reduction of photosynthetic surface, increases the surface area for water loss and reduces plant growth and aesthetic value of the crop. Control of the disease has primarily been with fungicides. However, disease control with host resistance will benefit from a better understanding of the pathogen (1).

Although structurally different, the powdery mildew fungi can appear relatively similar during their conidial stage. Researchers are now able to use DNA primers of the conserved Intra transcribed spacer region (ITS) of rDNA as diagnostic tools, due to the primers' ability to produce distinct bands for each species. The ITS bands in *P. guttata* and *M. pulchra*

were analyzed and sequenced and specific primers were designed for *P. guttata* and *M. pulchra* (2). Even though the two fungi may be diagnosed at conidial stage by using the specific primers, population analysis for genetic variability is still a problem because it is difficult to generate a large mass of conidia for DNA analysis. The objective of this research was to develop a method for amplifying a small amount of DNA to a larger, more substantial quantity that can facilitate genetic analysis studies.

Materials and Methods: Infected leaves were obtained and fruiting structures (cleistothecia) of *P. guttata* and *M. pulchra* were harvested. One hundred cleistothecia of each species were collected and placed in microtubes. Cell lysis and DNA release from the cleistothecia was achieved by adding 10 μ l of lyse-N-Go™ PCR reagent (Pierce, Rockford IL) to each microtube. The cleistothecia were then manually crushed and homogenized to ensure that the DNA was released into the solution. The reagents were placed in a thermocycler and processed using a program designed for this type of crude lysing (Pierce, Rockford IL): 65°C for 30 sec., 8°C for 30 sec., 65°C for 90 sec., 97°C for 180 sec., 8°C for 60sec., 65°C for 180 sec., 97°C for 60 sec., and then held at 80°C if the samples were to be analyzed immediately; otherwise the samples were removed promptly and stored at -20°C until used.

Two types of master-mixes for PCR were prepared. The standard, traditional master-mix consisted of 6 μ l 50mMMgCL, 30 μ l 10X buffer (with 15mM MgCl₂), 3 μ l 10mM dATP, 3 μ l 10mM dCTP, 3 μ l 10mM dGTP, 3ml 10mM dTTP, 5 units Taq DNA polymerase, and 217 μ l double distilled H₂O. The PEP primers used in this study consisted of 6 base random oligonucleotides for SP 180 and 15 bases for SP200 (Operon Technologies, Inc. Alamada, CA). 2 μ l of DNA template (either *P. guttata* or *M. pulchra*) and 5 μ l of primer (either SP 180 or SP 200) was added to microtubes containing 54 μ l of the master-mix to make a total volume of 61 μ l in each microtube. The second master-mix utilized Ready-to-Go PCR beads (Amersham Pharmacia Biotech). The PCR bead method involved the addition of 20.92 μ l of double distilled water to a microtube with a bead containing the necessary PCR components. A 2 μ l DNA template from either fungus was added along with 2.1 ml PEP primer (either SP 180 or SP 200), totaling 25 μ l per reaction tube.

The thermocycler was programmed to perform the following cycles (PEP cycle program): 94°C (3 min.), 92°C (1 min.), 37°C (2 min.), a ramping step of 10 sec/degree to 55°C, and 55°C (4 min.). These cycles were repeated fifty times starting back with step 2 (92°C for 1 min.). Afterwards, they underwent 55°C (6 min.) and were held at 4°C until analyzed. These samples were termed PEP 1 and 20 μ l was used as DNA template in subsequent reactions using SP 180 primer. These amplification products

were termed PEP2. 3 μ l of PEP2 were used as DNA templates in microtubes with a PCR bead, and 2 μ l ITS 1, 2 μ l ITS 4. 18 ml of double distilled water was added to bring the reactions to 25 μ l per reaction tube. A positive control that contained a known DNA sample and negative control that did not have a DNA template were included in all reactions.

ITS amplification program included 94°C initial melting for 3 min, 32 cycles of 94°C (60sec.), 40°C (70sec.), 72°C (90sec.), final extension at 72°C (450sec.), hold at 4°C. Specific *P. guttata* primers and specific *M. pulchra* primers were then used to test the two pathogens. These reaction tubes were processed in the thermocycler using the ITS cyler program. All samples were then analyzed by agarose gel electrophoresis using a 2% agarose gel, and the products were stained with Ethidium Bromide, and viewed under UV light.

Results and Discussions: The lyse-N-Go™ PCR reagent was useful for isolating and extracting the DNA from the cleistothecia and from conidia. When the PEP primers were initially used, it was apparent that they would produce results different from other primers. Unlike other primers, DNA bands were not distinct, but DNA smears were discernible in the Ethidium Bromide stained gels. (Fig. 1) DNA amplification templates from PEP2 samples revealed characteristic ITS bands for *P. guttata* and *M. pulchra* when PEP2 samples amplified were from SP 200, but PEP2 samples from SP 180 failed to produce distinct ITS bands and PEP product remained in the wells. A series of dilutions were performed on the SP 180-PEP2. Characteristic ITS bands were visible when the SP 180-PEP2 was diluted by 1/10,000 for *M. pulchra* and 1/100 for *P. guttata* (Fig. 2). In our experimental conditions, SP 180 was found to be more efficient in amplifying DNA than SP 200 since it produced a more concentrated product. The need to perform serial dilutions supports the theory that PEP primers can amplify small amounts of DNA into a more useful, larger concentration. Further studies, however, need to be performed in order to research their proficiency in studies involving restriction enzymes.

The specific primers were successful in selectively amplifying the fungal DNA of the two pathogens. The specific primer for *P. guttata* amplified only *P. guttata* DNA and failed to amplify *M. pulchra* DNA and the specific *M. pulchra* primer amplified only *M. pulchra* DNA, and did not amplify *P. guttata* DNA (Fig. 3). With further modifications and experimental trials, the genomic DNA of these fungi could be amplified to at least 78% of its entirety using PEP primer (2). Such technique will be useful for DNA analysis studies of obligate parasites. This would enable researchers to gain a better understanding of their genetic makeup and that could benefit disease control.

Significance to the Industry: The techniques that were tested have potential in DNA analysis of obligate pathogens including powdery mildew fungi. Understanding of the genetic structure of the pathogens is beneficial in disease control especially with host resistance and this will be beneficial to the nursery industry.

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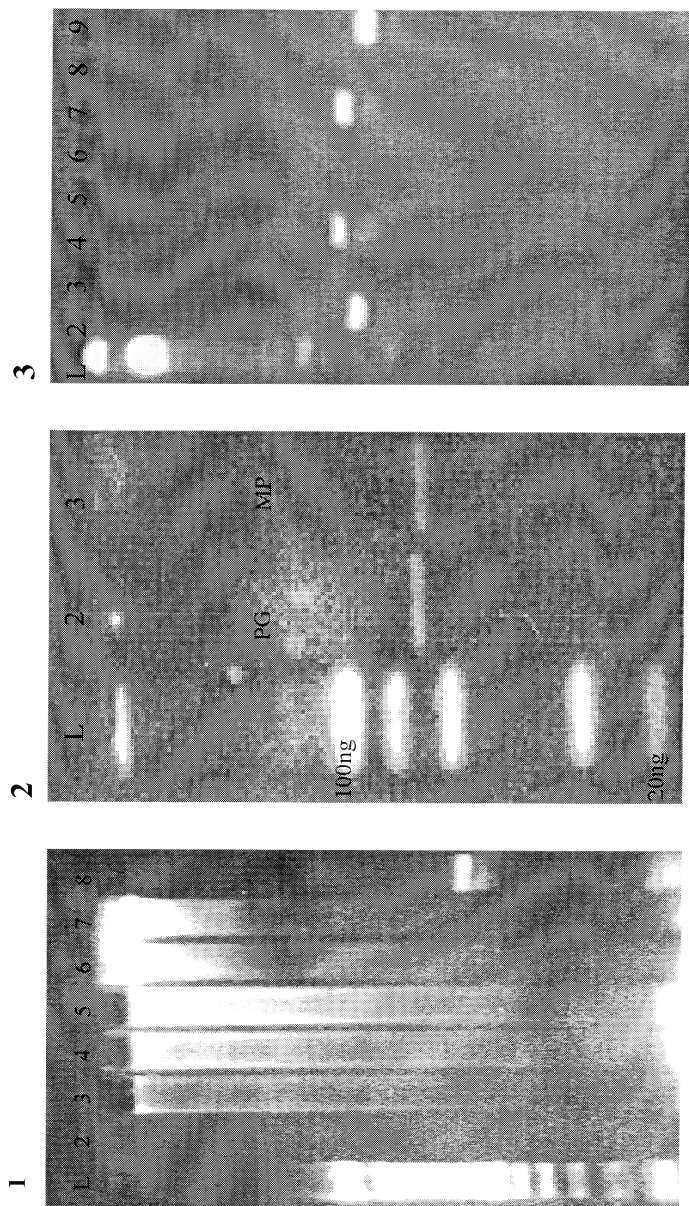


Fig. 1 - 3. DNA amplification products from minute amounts of *Microsphaera pulchra* (MP) and *Phyllostictia guttata* (PG) using primer extension pre-amplification (PEP) with 2 primers SP 180 (6 bp) and SP 200 (15 bp) and standard PCR technique.

- (1) Pre-amplification products shown by DNA smears (lane 3-5) from different volumes of PEP products, 2 μ l, 4 μ l, 6 μ l respectively. PEP products were used as DNA template and amplified with Intra transcribed spacer region (ITS) 1/4 primers (lane 6-8). Products from PEP primer SP 180 remained in the wells (lane 6,7) product from PEP primer SP 200 produced the characteristic ITS band (lane 8).
- (2) 1/10,000 dilutions of PEP product from SP 180 for *M. pulchra* and 1/100 for *P. guttata* amplified with ITS 1/4.
- (3) ITS primers designed to selectively amplify *P. guttata* and *M. pulchra* were specific. Lanes 2= PG with *P. guttata* specific primer, 3= PG with MP specific primer, 4= PG with ITS primers, 5= MP with PG specific primer, 6= MP with MP specific primer, 7= MP with ITS primers, 8= MP with PG specific primer, 9= MP with MP specific primers.

Host Resistance to *Microsphaera pulchra* in Dogwood

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Index Words: Powdery mildew, *Cornus* spp.

Nature of Study: Fungicide applications to control powdery mildew have become a routine practice in nursery production of dogwood in Tennessee; however, host resistance is recognized as the best method for disease management. Most of the *Cornus kousa* cultivars are resistant to powdery mildew (1, 2, 3, 4, 5), but the absence of high-level resistance within *C. florida* is a problem. In this study commercial cultivars and genetically diverse plants generated from natural out-crossing were evaluated for powdery mildew resistance. Commercial cultivars showed differences in susceptibility to powdery mildew and some plants generated from seed exhibited high level of powdery mildew resistance under high disease pressure. These results show clearly that powdery mildew resistance is available in *C. florida*. The diversity of the resistance displayed suggests a possibility of a new generation of diverse cultivars with powdery mildew resistance.

Germplasm evaluation. A total of 48 cultivars grown in the NCRS landscape evaluation plot were evaluated for powdery mildew reaction from 1997 to 2000. A randomized complete block design was used, with each cultivar replicated five times. Susceptible plants inoculated with powdery mildew were used as disease spreader plants. The pot-in-pot system was used to place the disease spreader plants in the field such that each test plant was adjacent to a spreader plant. This ensured ample availability of inoculum, facilitated infection uniformity and increased disease pressure on every test plant.

Single plant selections in plants generated from open pollinated *Cornus florida*. Seed produced from open-pollinated flowering dogwoods exhibit the genetic variation expected in an cross- pollinated crop. Observations of dogwood plants from thousands of plants grown from seed resulted in the selection of 187 plants that showed powdery mildew resistance under nursery or landscape environments. These were transplanted to the TSU Nursery Crop Research Station disease nursery where high disease pressure was promoted by using disease spreader plants as described above. The plants were evaluated for powdery mildew reaction for three years. Plants that exhibited high resistance to powdery mildew were selected for further evaluation.

Results and Discussion: **Germplasm evaluation.** Almost all of the *C. kousa* cultivars tested were resistant to powdery mildew with the exception of 'Bush Pink' which was moderately susceptible (Table 1). In 1999 disease pressure was low and ten cultivars showed resistance to powdery mildew (Table 2). However, since the weather was highly favorable to powdery mildew in 2000, disease pressure was high and some of the cultivars that had shown moderate resistance in 1999 had higher disease ratings in 2000. The hybrids 'Celestial', 'Stellar Pink', 'Constellation', and

'Aurora', remained resistant and *C. florida* 'Sterling Silver' moderately resistant in 2000 under high disease pressure (Table 2). Cultivars of *C. florida* 'Fragrant Cloud', 'Little Princess', 'Cherokee Brave' and 'Plena' and hybrid 'Ruth Ellen' were among the best cultivars for powdery mildew resistance in 1999, but the cultivars developed more disease symptoms under the 2000 high disease pressure and were categorized as moderately susceptible (Table 1). *C. florida* 'Barton' was moderately susceptible in both years (Table 2).

Single plant selections in plants generated from open pollinated Cornus florida. A total of 47 plants exhibited high level of resistance and 18 exhibited moderate resistance to powdery mildew in 1999; however, disease pressure was moderate that year. In 2000, when weather was highly favorable for powdery mildew and disease pressure was high, only 35 of the selected plants remained resistant and 10 remained moderately resistant. Out of the 35 resistant selections, a total of 14 plants have been selected as having superior resistance under high disease pressure. These display diverse horticultural characteristics identified as superior by growers and are being characterized as potential sources of new cultivars resistant to powdery mildew. All the resistant selections have been propagated for evaluation at multiple locations and are also being evaluated for spot anthracnose and other foliar diseases.

Significance to Industry: While it is important to eliminate susceptible cultivars from the production system, it is also desirable to maintain plant diversity. The resistant selections identified in this study can be used to develop new cultivars and add to the new generation of cultivars that have powdery mildew resistance. The development of the identified resistant selections into cultivars is important and beneficial to the nursery and landscape industry and complements resistant-breeding programs which normally take a long time.

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Table 1. Reactions of 48 *Cornus* taxa to powdery mildew in 2000.

Immune	Resistant (R) and moderately resistant (MR) ¹	Moderately susceptible	Susceptible
<i>C. kousa</i>	<i>C. kousa</i> (R) 'Spring Grove' 'Snow Flake' 'Milky Way'	<i>C. florida</i> 'Fragrant Cloud' 'Little Princess' 'Barton' 'Plena'	<i>C. mutalli</i> 'Boyd'
'Agate' 'Autumn Rose' 'Big Apple' 'Blue Shadow' 'China Girl' var. <i>chinensis</i> 'Dwarf Pink' 'Emerald Star' 'Gold Star' 'Lustgarten Weeping' 'Moonbeam' 'Satomi' 'Square Dance' 'Wolf Eyes'	<i>C. kousa</i> x <i>C. florida</i> (R) 'Aurora' 'Celestial' 'Constellation' 'Stellar Pink'	<i>C. kousa</i> 'Bush Pink' <i>C. kousa</i> x <i>C. florida</i> 'Ruth Ellen'	<i>C. florida</i> 'Cherokee Brave' 'Cherokee Chief' 'Cherokee Daybreak' 'Cherokee Sunset' 'Cloud 9' 'Ozark Spring' 'Pink Beauty' 'Purple Glory' 'Pygmy' Rainbow' 'Red Beauty' var. <i>rubra</i> 'Wonderberry', 'World's Fair'
<i>C. sericea</i> ² 'Cardinal' 'Isante'	<i>C. florida</i> (MR) 'Sterling Silver'		
<i>C. mas</i> 'Redstone' 'Golden Glory'			
<i>C. alternifolia</i> 'Pagoda'			

¹ A disease severity rating scale of 0-5 was used where 0= no symptom, 1=1-10%, 2=11-25%, 3=26-50%, 4=51-75 and 5=75-100% plant infection. Immune = 0, Resistant= >0-1; Moderate resistant = >1-2, Moderate susceptible =>2-3, Susceptible=>3-5.

² 'Cardinal' and 'Isante' were highly susceptible to Septoria leafspot.

Table 2. Commercial cultivars of *Cornus florida* or hybrids that exhibited least amount of powdery mildew disease in 1999& 2000

<i>Dogwood species</i>	Cultivar name	Disease Severity (0-5 scale) ¹	
		August 1999	August 2000
Hybrid (Rutdan)	Celestial	0.5	0.1
Hybrid (Rutgan)	Stellar Pink	0.3	0.1
Hybrid (Rutcan)	Constellation	0.5	0.5
Hybrid (Rutban)	Aurora	1.0	0.5
Hybrid (Rutlan)	Ruth Ellen	1.0	2.4
<i>C. florida</i>	Sterling Silver	1.4	1.8
<i>C. florida</i>	Little Princess	0.4	2.5
<i>C. florida</i>	Fragrant Cloud	0.4	2.5
<i>C. florida</i>	World's Fair	1.0	2.7
<i>C. florida</i>	Cherokee Brave	0.5	2.9
<i>C. florida</i>	Barton	2.3	2.4
<i>C. florida</i>	Plena	1.0	3.0

¹ Disease severity on a 0-5scale where 1=1-10%, 2=11-25%, 3=26-50% 4=51-75 and 5=75-100% plant infection. Statistical analysis of variance (SAS) compared all 48 cultivars. Least significant differences between the means (LSD) for 1999 =1.1 and for 2000 LSD = 0.9.

Ascocarp Formation and Powdery Mildew Disease Severity in *Cornus florida*

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Nature of work: Previous studies have demonstrated that ascocarps (cleistothecia) constitute the main mechanism of winter survival and source of primary inoculum of powdery mildew in Tennessee (Mmbaga, 2000). Ascocarps form late in the growing season between late September and early November and they have to reach some level of maturity to survive winter. Because time is a factor in the maturation of ascocarps, the timing of ascocarp formation can influence primary inoculum density in the following spring. Previous studies have indicated that cool temperatures influence the timing of ascocarp formation and day-length had no effect (Mmbaga & Sheng 1999).

The objective of this study was to confirm previous results on: (a) the effect of temperature on ascocarp formation and maturation and (b) effect of temperature on ascocarp survival.

i) Effect of temperature on ascocarp formation and maturation: Three experiments were conducted in growth chamber conditions and temperatures 18/10°C, 23/15°C, 26/18°C and 28/20°C (day/ night), with 80% relative humidity and 12h day-length were the main treatments. Seedlings of *C. florida* were infected with powdery mildew from air-borne spores prior to temperature treatments. Selection of these temperatures was based on weather conditions during a two-week period preceding ascocarp formation in McMinnville, TN (1996 -1998).

The presence of ascocarps was observed on intact leaves by using a dissecting microscope and ten ascocarps of each color were picked and observed under a compound microscope to assess their developmental stages. The number of plants that had ascocarps and the abundance of ascocarps at each developmental stage was assessed using a scale of 1-3, where 1 = 1<10 (few), 2 = 11<20 (moderate), and 3 = >20 (abundant). Since one leaf often contained ascocarps of different development stage, some cream to light yellow immature ascocarps were marked and their development followed over time to assess the rate their development to mature stage.

The first experiment was conducted in late summer, 1999 and one-year-old plants were used with a replication of ten plants per temperature treatment. The first observation for ascocarp presence was done 40 days after treatments started, and at this time, some ascocarps were already mature, but new ascocarp initials and partly developed intermediate stage were also present.

The second experiment was conducted during winter (February, 2000) and seedlings newly germinated at 24/20°C greenhouse conditions were exposed to air-borne inoculum from previously infected plants. When the

foliage had developed severe powdery mildew symptoms, they were separated into four groups of 10 plants each and placed at different temperatures. Observation for ascocarp formation was initiated 12 days after treatments started and continued at 3-6 days interval.

The third experiment was conducted in early summer (June, 2000) and one-year-old seedlings infected with powdery mildew in shade-house conditions where temperature was above 28°C were used. Observations for ascocarp formation were initiated 10 days after treatments started and continued at 3-4 days interval.

ii) Effect of temperature on ascocarp survival. Infected leaves were collected and observed under a dissecting microscope. Leaves that carried abundant ascocarps were cut into small pieces about 1x1 cm and separated into three groups: (a) leaf pieces with immature cream colored ascocarps, (b) leaf pieces with light brown partly developed ascocarps (intermediate), and (c) leaf pieces with dark-colored mature ascocarps. Each group was put in Petri dishes, approximately 1.5g leaf pieces per Petri dish and placed at the following temperatures: 24/20°C, 4°C, -10°C, and -20°C; with a replication of two Petri dishes per treatment. The physical appearance of ascocarps and ascospore viability were monitored over a four months period by using a dissecting microscope (35x), and a compound microscope (200-400x), respectively. During each evaluation, twenty leaf pieces and twenty ascocarps from each maturity group and temperature were observed.

Results and Discussion: (i) Effect of temperature on ascocarp formation and maturation: Ascocarp color was associated with their developmental stage as follows: a) Cream to light yellow ascocarps were immature and did not contain asci or ascospores; b) Dark-brown to black colored ascocarps with or without appendages, contained asci and ascospores and were mature and, c) Light-brown ascocarps were at intermediate stage and contained asci with few or no ascospores. These observations is in agreement with previous results (Mmbaga, 2000).

In all three experiments, 18/10°C and 23/15°C were best temperatures for ascocarp formation, ascocarp formation was delayed at 26/18°C and no ascocarps formed at 28/20°C. (Fig 1). By the time the first observation was made 40 days after temperature treatments started in experiment 1, some ascocarps had already matured. The number of leaves containing ascocarps increased over time until all leaves had ascocarps (Fig 1a). In experiment 2, ascocarps were observed at all temperatures on day 12, and at 18/10°C, a few leaves had moderately developed ascocarps. However the number of leaves containing ascocarps declined sharply at 26/18°C and at 28/20°C (Fig 1b). The immature ascocarps had shriveled and withered at these temperatures and thus caused ascocarp decline. New ascocarps formed later (day 27) at 26/18°C, the abundance was low; no ascocarps formed at 28/20°C (Fig 1b). It is suspected that when the plants were moved from 24°C to different temperatures, ascocarp

formation had already been initiated and influenced ascocarp formation at 26/18°C and 28/20°C. The assumption was confirmed in experiment 3 where ascocarps were observed in 12-14 days at 18/10°C and 23/15°C, but was much delayed (day 25) at 26/18°C and not formed at 28/20°C (Fig 1c). Ascocarp abundance was highest at 18/10°C (Fig 2). Ascocarp initials reached intermediate stage in about two weeks and matured in less than 30 days (data not shown). These results confirmed previous results that cool temperature trigger ascocarp formation and development.

ii) Effect of temperature on winter survival of ascocarps of different maturity stages. Immature cream-yellow ascocarp initials withered and became shriveled at all four temperatures evaluated, and are not expected to contribute to the primary inoculum density for the following season. Brown colored ascocarps that had not developed appendages, but contained asci and some ascospores maintained viable ascospores during the four months experiment (Fig 3a). More than 60% of the mature ascocarps had viable ascospores after four months storage at -10°C and -20°C and viability spores declined slightly over time (Fig. 3b). Maturation of ascocarps was important in their winter survival, thus, ascocarp formation in September versus November would have significant implications on the density of ascocarps that can survive winter and contribute to primary inoculum density in the following season. Ascocarps formed late in the season may be partly developed and have a lower survival rate and smaller inoculum density in the following season. Early frost when ascocarps are young would likely kill the ascocarps and reduce the number of ascocarps that survive winter especially if plants lose their leaves early. Since ascospores remain viable for several months at -10°C and -20°C, the pathogen can recycle locally and perpetuate the problem from season to season.

Significance to the industry: Information from this study will be useful in the development of disease management strategies that target the reduction of primary inoculum density and delay of disease outbreak. Weather information may be used to predict primary inoculum density for the following season and allow growers to prepare for early or delayed fungicide applications. Information from this study may be used to improve the timing of fungicide applications.

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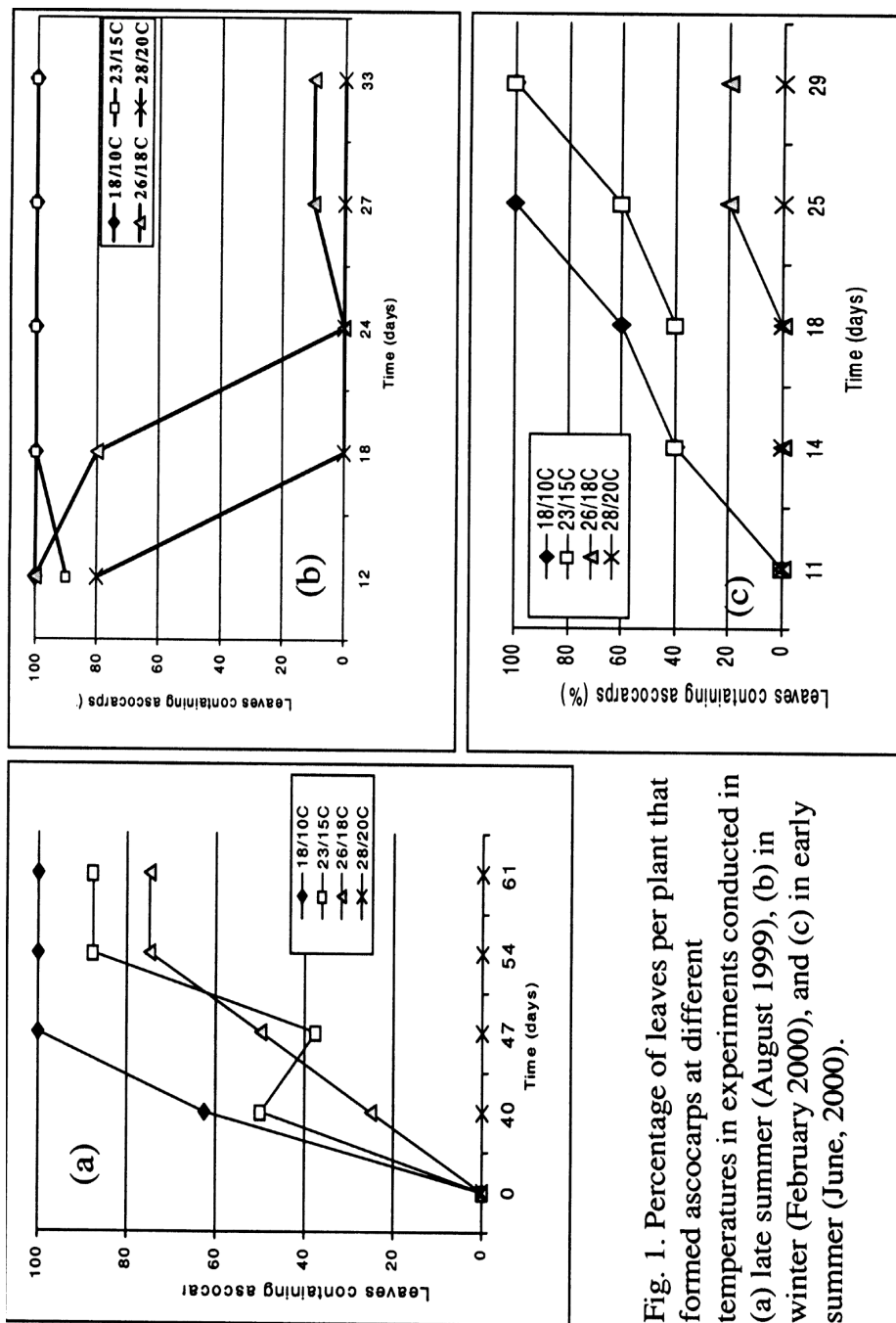


Fig. 1. Percentage of leaves per plant that formed ascocarps at different temperatures in experiments conducted in (a) late summer (August 1999), (b) in winter (February 2000), and (c) in early summer (June, 2000).

Fig 2. Abundance of ascocarps at different temperatures

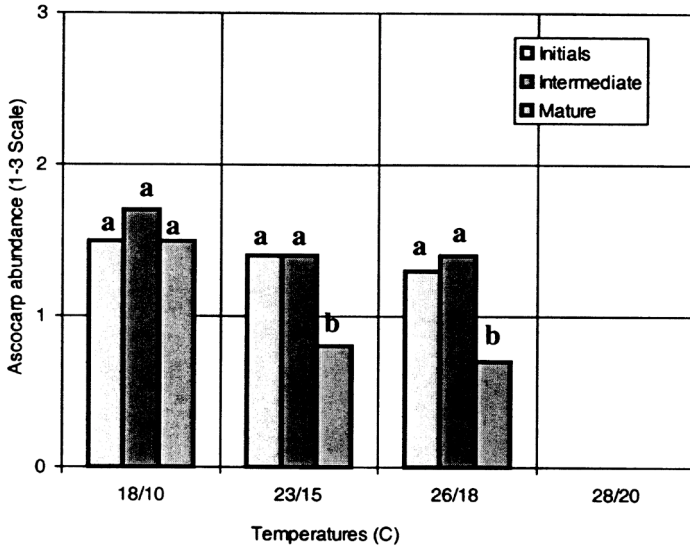
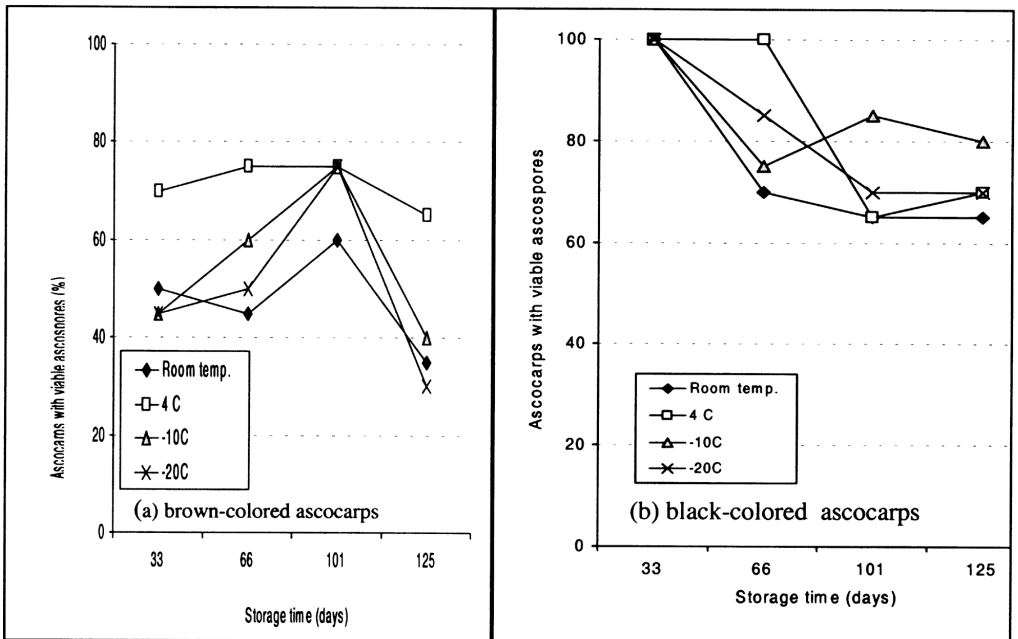


Fig 3. Percentage of ascocarps with viable ascospores at different storage temperatures.



Effects of Late-Season Hardening on Development of Resistance to *Phytophthora* Root Rot in a Woody Perennial

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Index Words: *Phytophthora* Root Rot, Cold hardiness, *Rhododendron*

Nature of Work: Inoculum was supplied by USDA ARS laboratory, Corvallis. The inoculum was spread on the bottom of the container and a standard media was placed over the inoculum. Two-inch liners of *Rhododendron* were upshifted to four-inch containers that contained the inoculum and standard mix. Three different fertilizer regimes were investigated: 1) a high nitrogen treatment represented by a preplant incorporation of Osmocote, 16-10-10, type 40 applied at 5lbs/yd; 2) a balanced fertilizer treatment represented by a preplant incorporation of Apex 20-10-10, 12-15 month formulation applied at 14 lbs./yd; and, 3) a low fertility program of preplant incorporation of K_2SO_4 at 6.5 lbs./yd and phosphorous, 0-46-0 at 2 lbs./yd. Plants were allowed to cold acclimate under natural temperatures and photoperiod.

Four evaluation dates were investigated: October 30, 1998; November 30, 1998; December 30, 1998; and, January 30, 1999. On these four dates, cold hardiness evaluations were conducted in an Ultra-Low Freezer at OSU. Temperatures were dropped at a rate of 2°C per hour until reaching the first setpoint temperature, -2°C. After holding at this temperature for 30 minutes, four replicates and three sub-samples per species were removed and placed in a 3°C greenhouse. The freezer was then dropped to the next setpoint temperature and the process was repeated until completing all the temperature treatments (-2, -5, -8, -11 and -14°C). After 24 hours the plants were moved from the 3°C greenhouse to another greenhouse set at 15°C.

Six weeks after freezing, plants were rated for *Phytophthora* infection, survival and salability. *Phytophthora* infection was recorded on a scale of 1 to 10, where 1 is 10%, and 10 is 91-100% of the crown discolored. Shoot dry weights were recorded after drying at 70°C for 24 hours. Drying of samples occurred after salability and survival ratings were determined. Species that retained shoot tissue were evaluated for regrowth only. In both cases, plants were qualitatively rated on a scale of 1 to 5 as follows: 1 = no regrowth and/or up to 100% dieback, and 5 = vigorous regrowth and/or no dieback. Plants that failed to produce shoot tissue were considered dead. A random sample of inoculated plants was tested for positive identification of *Phytophthora* infection. The objectives

were: 1) determine the affects of late applications of controlled-release and slow-release fertilizers on cold hardening and resistance to *Phytophthora* in a woody perennial; 2) determine the affects of potassium and phosphorous applications on the cessation of growth and susceptibility to *Phytophthora*; and, 3) determine the affects of high level of fertility regimes during the growing season on cold hardening and susceptibility to *Phytophthora*.

Results and Discussion: The two-way interactions of *Phytophthora* X fertilizer, temperature X date and fertilizer X temperature were significant. The influence of fertility on temperature is shown in Figure 1, indicating a balanced fertilizer program resulted in less cold injury. The three-way interaction of *Phytophthora* X fertilizer X temperature was also significant (Table 1). The influence of fertility on *Phytophthora* infection is represented with the interaction of *Phytophthora* X fertility X temperature, in the rating of infection data, at the two lowest temperature evaluations, control, -2 and -5°C (Table 2). The more nitrogen that was available, the lower the rating of infection with the *Phytophthora* inoculated plants. If inoculum is not present, plants receiving no nitrogen suffered significantly and the Apex fertilizer was the best treatment, control, -2 and -8°C (Table 2). This relationship of less injury with higher nitrogen was also true of the browning data. Again, if no inoculum was present, Apex fertilizer was the better treatment, control, -2 and -8°C (Table 1).

If plants were immediately exposed to freezing temperatures in October without acclimation, aboveground injury recorded as browning was at its worse. However, in the examination of root infections, date was not significant. This indicates, based on high root infections scores at all dates, that infection had taken place before the acclimation occurred, and that acclimation had no effect on root infections. This indicates that it is necessary to examine both aboveground and belowground factors over time. By utilizing the principal of disease progress through area under the disease progress curve (AUDPC) it will be possible to examine whether plants inoculated with *P. cinnamomi* are less hardy when exposed to poor fertility and/or freezing temperatures.

Significance to Industry: Container production has become increasingly popular as an alternative to field production (3) but it is not without drawbacks (5). Roots and crowns in overwintering containers are often exposed to lower temperatures relative to plants overwintered in the ground and roots are the most susceptible organs to cold injury (6). As a result, containerized plants are more susceptible to winter kill, especially if excess nitrogen has been applied (4). Another major difficulty in container production is disease control. It is estimate that growers can lose up to 20 - 80% of their crop annually due to disease (Hoitink, personal communication) with *Phytophthora* causing the majority of losses.

Interactions between low temperatures and plant diseases have long been reported in crop plants (2, 7); however, these interactions have not been well investigated in ornamentals. The more nitrogen that was available, the lower the rating of infection with the *Phytophthora* inoculated plants. This relationship of less injury with higher nitrogen was also true of the browning data and again if no inoculum was present the Apex fertilizer was the better treatment (Table 1). Some authors have reported that high nitrogen feeding is a way of managing *Phytophthora* infection. It appears that if *Phytophthora* is present high nitrogen can reduce the severity of symptoms. However, to prevent infection a balanced fertilizer program is superior. *Phytophthora* root rots are increasing in incidence in container culture. One reason is because of the increased use of recirculated water in container nurseries. Understanding the relationships between nutrition and potential root rot control is becoming increasingly important.

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Table 1. Three way interaction of temperature, *Phytophthora cinnamomi* inoculation and fertilizer treatment for browning.

	Control	Temperature (C)				
		-2	-5	-8	-11	-14
<i>Phytophthora</i>						
Osmocote	1.83c	3.00c	2.82c	3.75b	4.00b	2.06c
Apex	4.04b	3.67b	6.64a	4.36b	3.10c	6.07a
60ppm K, 4ppm P	3.78b	4.58b	4.94a	4.58b	4.42b	4.68b
No <i>Phytophthora</i>						
Osmocote	5.46a	3.33b	2.78c	4.00b	3.03c	5.43a
Apex	1.00d	2.25c	4.49b	3.13c	3.72b	4.64b
60ppm K, 4ppm P	4.07b	4.28b	4.58b	4.79b	4.36b	4.54b

Table 2. Three way interaction of temperature, *Phytophthora cinnamomi* inoculation and fertilizer treatment for root infection

	Control	Temperature (C)				
		-2	-5	-8	-11	-14
<i>Phytophthora</i>						
Osmocote	2.65c	3.00c	2.82c	3.75b	4.00b	3.96a
Apex	3.55b	3.67b	6.64a	4.36b	3.10c	4.35a
60ppm K, 4ppm P	3.31b	4.58b	4.94a	4.58b	4.42b	3.90a
No <i>Phytophthora</i>						
Osmocote	3.38b	3.33b	2.78c	4.00b	3.03c	4.26a
Apex	2.03d	2.25c	4.49b	3.13c	3.72b	4.08a
60ppm K, 4ppm P	3.69b	4.28b	4.58b	4.79b	4.36b	4.21a

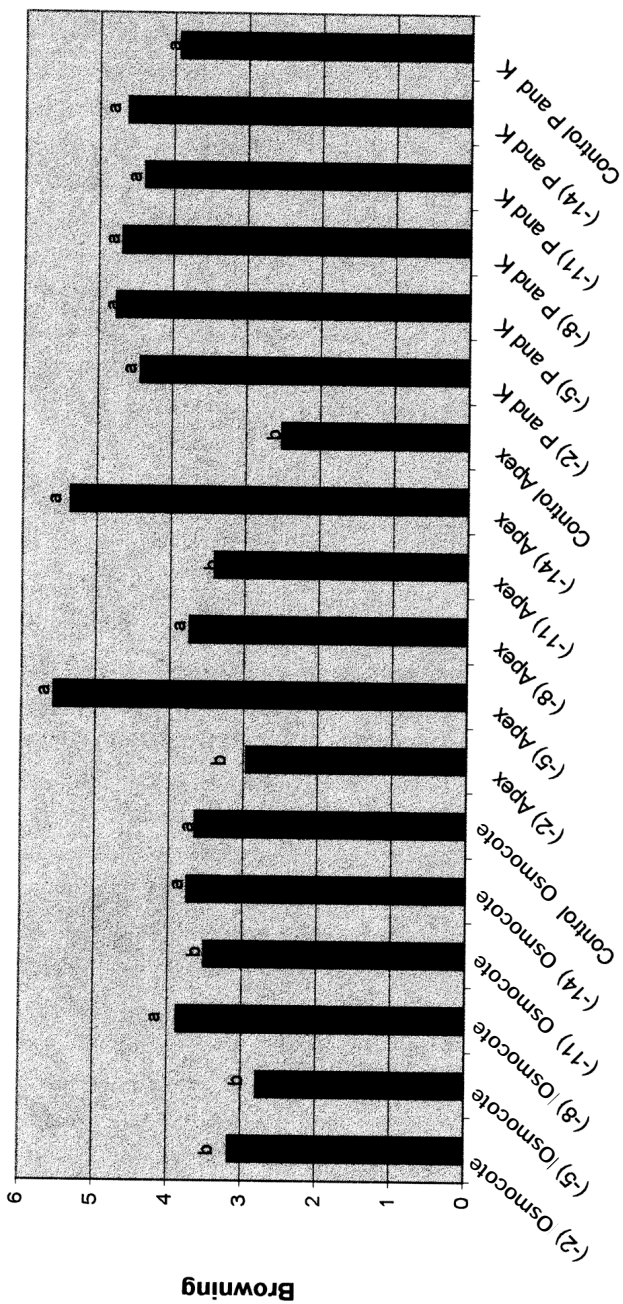


Figure 1. Interaction of freeze temperatures and fertilizer treatments for browning

Development of Bacterial Soft Rot on *Hosta* in Cold Storage

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Index Words: *Hosta* sp. (Tratt.), Cold Storage, *Erwinia carotovora* subsp. *carotovora*, Bacterial Soft Rot

Nature of Work: *Hosta* spp. Tratt., commonly known as plantain lilies, are herbaceous perennial plants native to the temperate regions of China and Japan (1). Since the introduction of hostas into the United States in 1790 (3), they have become one of the most popular perennials across the country (6).

To keep up with consumer demand, nurseries in warmer regions of the United States, USDA Plant Hardiness Zones 8B and below, are producing hostas to take advantage of the long growing season. Unfortunately, hostas have a dormancy requirement that must be achieved by a period of low temperature chilling before bud break can occur, and southern production areas cannot supply these cold temperatures. As a result, hostas produced in southern nurseries are often slower to emerge in the spring and exhibit poor growth in comparison to plants grown in cooler climates (5). The use of cold storage, provided by the use of coolers or refrigerated trailers, enables growers in warmer climates to supply an artificial chilling period and complete the dormancy requirement of the plant.

Many diseases can develop under conditions of cold storage. While there has been extensive postharvest research conducted on fruits and vegetables in cold storage (4), diseases of ornamental plants have not been studied. A soft rot on hosta caused by the bacterial pathogen *Erwinia carotovora* subsp. *carotovora* (ECC), was observed at a large wholesale nursery in South Carolina in 1999. This disease was causing maceration of the rhizomatous tissue following storage in a chilling facility. The epidermal tissue covering the fleshy roots remained intact, while the parenchymatous tissue dissolved into a watery rot. Aboveground symptoms exhibited were yellow, wilted leaves with water-soaked petioles that eventually collapsed at the soil line. Infected plants had a distinctive malodorous aroma once the rhizome began to rot. Bacterial soft rot was known to occur on hostas, but ECC had not been previously reported as the pathogen.

The objectives of this study were to 1.) distinguish the role that cold storage has on the development of bacterial soft rot on hosta and 2.) identify temperatures that satisfy chilling requirements without causing disease.

Tissue culture plugs of hosta produced in 72-cell flats were sprayed with ECC at three concentrations. The bacteria were prepared by growing isolates on nutrient yeast dextrose agar (NYDA) for 24 hours at 30°C. Plates were flooded with sterile distilled water and diluted to concentrations of 10², 10⁴ and 10⁸ colony-forming units per milliliter (cfu/ml). Using a chromatographic sprayer, the bacterial suspensions were applied to the plants until runoff occurred (~2 ml/plant). One 72-cell flat was sprayed with sterile distilled water as a control. The foliage was allowed to dry 24 hours to allow for possible epiphytic colony establishment. Each flat of 72 plants, representing a different bacterial concentration, was divided into three treatments for placement at 0, 2 or 4°C. Temperatures were chosen because they are actual temperatures used by hosta producers for chilling purposes. Each treatment of plants was placed in a sterile plastic bag prior to cold storage to prevent contamination of the plants. All treatments of plants were held at their respective temperatures of 0, 2 or 4°C for 24 hours in separate Percival coolers. Upon removal of the plants from cold storage, the plants were potted into 0.35 L containers filled with sterile potting media (Pro-Mix BX, Premier Horticulture Co., Red Hill, PA) and placed in the greenhouse on the University of Georgia campus in Athens, Georgia. Plants were arranged in a randomized block design with 18 replications per treatment. Irrigation occurred daily and day temperatures were held at 27°C while night temperatures were maintained at 16°C. Plants were observed for two weeks for possible soft rot development. Suspected soft rot infected tissues were cultured for the presence of *E. carotovora* subsp. *carotovora*. The experiment was conducted twice from October to November, 2000.

Results and Discussion: Regardless of bacterial concentration applied to the hosta plants, no disease occurred unless the plants were held at 0°C for 24 hours. Inoculated plants stored at 2 and 4°C showed no decline or soft rot symptoms. No soft rot symptoms developed on any non-inoculated plants, but water-soaking of the foliar tissue that was indicative of freeze injury was observed on control plants held at 0°C. Bacterial concentrations on the frozen plants had an effect on the percentage of plants with soft rot, but did not affect the severity of symptoms. Forty-six percent of plants sprayed with 10² cfu/ml of inoculum developed bacterial soft rot while 77% of plants inoculated with 10⁴ cfu/ml and 83% of plants inoculated with 10⁸ cfu/ml of bacteria became infected. Since none of the plants held at temperatures above 0°C were infected with ECC, it is to be assumed that storage temperature has a role in disease development. Freeze injury, due to cold damage sustained by the hostas, provided an entryway for ECC, an opportunistic pathogen. Also, the solutes that were released by the damaged plant cells, especially after the plants were exposed to warmer temperatures in the greenhouse, encouraged bacterial populations to increase to levels that allow disease to develop (2).

From this study, it has been determined that bacterial soft rot of hosta caused by *Erwinia carotovora* subsp. *carotovora*, can be prevented by storing plants at temperatures above freezing. Even when epiphytic bacterial populations are relatively high, bacterial soft rot does not occur until plant injury has occurred. Chilling hostas at 4°C satisfies dormancy requirements of hosta (5), and will reduce the likelihood of developing bacterial soft rot disease.

Significance to Industry: *Hosta* sp. grown in the southeastern United States benefit from an artificial chilling period. As the use of cold storage expands in perennial plant production, it will be important to understand the effect of cold temperatures in the development of disease in dormant plants. This study established that storage of dormant rhizomes at 4°C prevents disease development while allowing the plant to complete dormancy requirements.

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Animal Manure Amendments to Container Mixes for Suppression of Root Pathogens

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Index Words: Azalea, *Rhododendron obtusum*, Phytophthora root rot, *Phytophthora cinnamomi*, swine manure, poultry compost, *Pythium ultimum*

Nature of Work: Phytophthora root rot caused by *Phytophthora cinnamomi* attacks many important nursery crops in container production. Although pine bark-based container mixes are suppressive to root pathogens, in general, disease can still develop when environmental conditions exceed the capacity of the mix to suppress the pathogen (4, 5). Previous work suggests that animal manures may be suppressive to root rot pathogens in part due to the high microbial population associated with manures (2,3). Development of value-added products based on manures generated by the expanding poultry and swine production in the North Carolina could be beneficial both to the animal and nursery industry.

Two types of experiments were conducted to test suppression of manures in potting mixes. General suppression of the manures was tested in a cucumber bioassay with *Pythium ultimum* (1). Specific suppression was tested with containerized azaleas and *P. cinnamomi* at a research nursery. The animal manures used were a poultry compost consisting of composted waste and bird mortality (NCSU, Animal Waste Management Center), and swine waste collected from a lagoon, dried, and pelletized (BION Soil, BION, Inc. Smithfield, NC).

In the cucumber bioassay, manures were amended to Fafard no. 2 mix at rates of 4, 8, or 12% (v/v). The amended mix in 4" pots was planted with eight cucumber seeds, irrigated heavily to remove salts, and then placed in a growth chamber with 16 hr of light/day at a constant temperature of 20 °C. After 10 days, each pot was examined and the cucumbers rated for disease as follows 1= healthy seedling, 2=emerged but diseased (stunted or chlorotic), 3=post-emergence damping-off, and 4=pre-emergence damping off. Fafard mix without manure was used as a control.

In the nursery, Hinodegiri azaleas from 2.5" pots were transplanted May 31, 2000, to 3/4 gallo pots in pine bark mix amended with the manures at rates of 4, 8, or 16% (v/v). Pots were placed in a randomized complete block design under a shade cloth and irrigated daily (0.9") by sprinklers. On June 9, one half the plants in the pots were inoculated with rice grains colonized by *P. cinnamomi*. Plants in control treatments without manures

received 3.2 g N per pot of 16-5-10 Wilbro fertilizer. Foliar symptoms of disease were rated regularly over the summer. The VTEM 'pour through' method was used to determine soluble salts and pH levels in the pots during the season. On September 19, 2000, plant top weight was determined and the roots of each plant were rated for extent of *Phytophthora* root rot where 1= healthy roots, 5 = roots completely rotten and plant dead.

Results and Discussion: Both swine waste and poultry compost incorporated into Fafard were suppressive to *Pythium* root rot in two different trials of the cucumber bioassay (Table 1). The best suppression of *Pythium* root rot was found at incorporation rates of 8 or 12% in trial 1. Thus, swine waste and poultry compost are generally suppressive but disease was not suppressed completely as ratings of 2 meant that cucumber seedlings emerged but were stunted or chlorotic compared to seedlings in the controls.

In the nursery, soluble salt levels were high initially ranging up to 10 mS/cm with the 16% poultry compost treatment, but fell to values near 0.2 mS/cm in all treatments within 30 days. Over the experiment, pH ranged from 6.6 to 7.4 across all treatments. Azaleas in mixes infested with *P. cinnamomi* developed foliar symptoms of *Phytophthora* root rot beginning in July that progressed over the summer. *Phytophthora* root rot was not suppressed in any pine bark mix incorporated with the animal manures at rates of 4 to 16% compared to pine bark mix only (Table 2). Plant top weights in all mixes infested with *P. cinnamomi* were much less than in comparable mixes without *P. cinnamomi* (Table 2). Microbial activity as measured by CO₂ evolution was very high in both swine waste and poultry compost treatments for the first 5 days after potting and was maintained at levels well above the activity in the pine bark + sand mix (data not presented). Apparently, higher levels of general microbial activity do not directly relate to suppression of *Phytophthora* root rot. In the absence of *Phytophthora*, azaleas grown in the mix amended with poultry compost were generally smaller than those in the swine waste amended pine bark.

Significance to Industry: Animal manure amendments were suppressive to *Pythium* in a peat-based mix but not *Phytophthora* in a pine bark mix. More research is needed before manure amendments can be recommended for disease suppression. In the absence of *Phytophthora*, azalea growth in 4% swine waste may be comparable to that in pine bark mix with inorganic fertilization.

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Table 1. Suppression of *Pythium ultimum* in a cucumber bioassay with Fafard Mix #2 amended at various rates of either poultry compost or swine pellets.

Treatment and (Rate)	<i>Pythium</i>	Disease severity (1-4)	
		Trial 1	Trial 2
Fafard no 2	Yes	3.3 ab	3.9 a
Fafard no 2 + swine pellets (4%)	Yes	3.4 a	3.7 ab
Fafard no 2 + swine pellets (8%)	Yes	2.5 c	3.3 bc
Fafard no 2 + swine pellets (12%)	Yes	2.4 c	2.8 d
Fafard no 2 + poultry compost (4%)	Yes	2.6 bc	3.7 ab
Fafard no 2 + poultry compost (8%)	Yes	2.6 bc	3.2 cd
Fafard no 2 + poultry compost (12%)	Yes	2.3 c	3.0 cd
Fafard no 2	No	1.1 d	1.0 e
Fafard no 2 + swine pellets (12%)	No	1.0 d	1.0 e
Fafard no 2 + poultry compost (12%)	No	1.0 d	1.1 e

Means within a column followed by the same letter are not different according to the Waller-Duncan k ratio, k=100, p=0.05.

Table 2. Development of Phytophthora root rot in azaleas grown in pine bark mix amended with either swine waste or poultry compost.

Treatment and (Rate)	Root rot rating (1-5)		Top weight (g)	
	Inoc.	Control	Inoc.	Control
Pine bark + Sand (6:1)	3.0 bc	1.1 d	14 g	44 ab
Pine bark + swine pellets (4%)	2.8 c	1.0 d	18 efg	37 bc
Pine bark + swine pellets (8%)	2.9 c	1.2 d	16 fg	35 bc
Pine bark + swine pellets (16%)	3.1 abc	1.2 d	16 fg	38 bc
Pine bark + poultry compost (4%)	3.5 a	1.4 d	15 g	26 de
Pine bark + poultry compost (8%)	2.7 c	1.1 d	12 g	24 ef
Pine bark + poultry compost (16%)	2.8 c	1.4 d	10 g	33 cd
Pine bark (100%)	3.4 ab	1.0 d	17 efg	48 a

Means within the inoculate and control column followed by the same letter are not different according to the Waller-Duncan k ratio, k=100, $p=0.05$.

Effect of chlorine concentration and contact time on zoospore survival of *Phytophthora nicotianae*

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Index Words: *Phytophthora nicotianae*, chlorination, disease management

Nature of Work: *Phytophthora* species were isolated from recycled water in California (MacDonald, et al, 1994), North Carolina (Lauderdale and Jones, 1997), and Virginia (Hong, et al, unpublished). The recovery levels of some *Phytophthora* species were well above pathogen thresholds in irrigation water for nursery crops (Hong and Epelman, 2000). These previous investigations help explain why many nurseries have experienced increased disease pressure after starting recycling irrigation runoff. They also provide justification for disinfestation of recycled water prior to reuse. The objective of this research was to determine the effect of chlorine concentration and contact time on the survival of *Phytophthora nicotianae* using zoospores, the principal fungal structure in irrigation water, as a model system. Chlorination was initially developed for purification of drinking water and had not been assessed previously to disinfest recycled irrigation water for use in the nursery industry.

Five chlorine concentrations (0.25, 0.5, 1.0, 2.0, and 4.0 ppm) and five contact times (15, 30, 60, 120, 240 seconds) plus a water control were included in the test. Zoospore suspension of *P. nicotianae* was prepared by incubating mycelial plugs in 1% nonsterile soil water extract and diluted to 3300 spores/ml. Chlorine treatments were accomplished by mixing an equal amount of zoospore suspension and chlorine solutions in Eppendorf tubes then adding sodium thiosulfate to instantly remove residual chlorine after the required contact time was met. An aliquot (100 μ L) of treated zoospore suspension was then spread onto a Petri dish containing 20 ml of PARP-V8 medium (Ferguson and Jeffers, 1999). The dishes were incubated at 23°C for 2 days before colonies of *P. nicotianae* were counted. The test was repeated once with six Petri dishes per treatment. A reduction in zoospore survival (% control) was calculated by comparing the number of colonies in treated Petri dishes with that in the water control.

Results and Discussion: Control of *P. nicotianae* increased linearly from 50% to more than 99% as free chlorine concentration increased from 0.25 ppm to 2 ppm. No *Phytophthora* zoospores survived at 4 ppm of free chlorine in our test (Figure 1). Contact time was not a significant factor at the chlorine concentrations evaluated in this research.

Significance to Industry: This research suggests that 2 ppm of free chlorine at discharge points is required to achieve good control of *P. nicotianae* in irrigation water. This information can be used to evaluate the chlorination protocols in use and to develop new protocols, as *Phytophthora* diseases are considered the number one disease problem and *P. nicotianae* is among the most common pathogens affecting a variety of annual plants in many nursery operations.

Recycling irrigation is of critical importance to the nursery industry in ensuring the availability of quality irrigation water in the wake of global water scarcity and in meeting the EPA requirements for nitrate and pesticide levels in runoff. But recycling irrigation systems may serve as the primary source of *Phytophthora* species and as a powerful vehicle for spreading the diseases in nursery production. As more and more nurseries start recycling irrigation runoff, waterborne pathogens will be of increasing importance to the industry. Additional chlorine assays are needed to include other fungal structures such as mycelium and sporangia of *P. nicotianae* and other major waterborne plant pathogens, such as *Pythium* spp. Factors affecting free chlorine concentration in recycling irrigation systems will also have to be considered.

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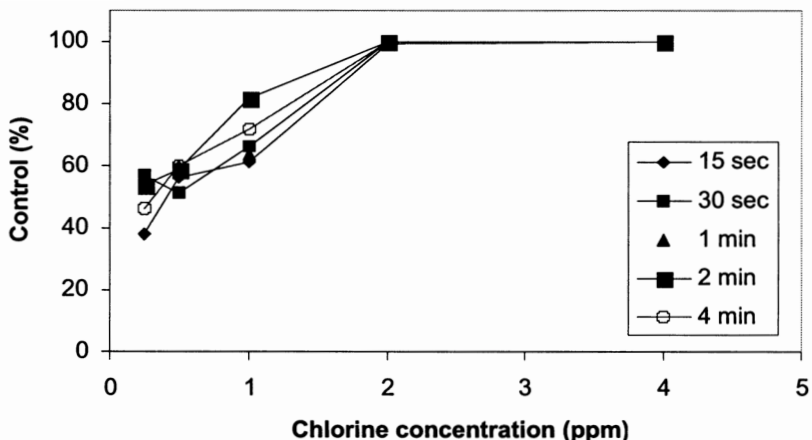


Figure 1. Effect of chlorine concentration and contact time on zoospores survival of *Phytophthora nicotianae*