Pathology and Nematology

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'No Spray' Rose Cultivars for the Mid South

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Key Words: disease resistance, black spot, Cercospora leaf spot, *Diplocarpon rosae*, *Cercospora rosicola*

Significance to Industry: In this study, 14 rose cultivars were found to be resistant to both black spot and Cercospora leaf spot. However, resistance claims in catalogs of companies selling roses were not substantiated for many rose cultivars. Caution should be used in purchasing roses based on these claims.

Nature of Work: The popularity of roses as an ornamental is tarnished because of foliar diseases such as black spot (*Diplocarpon rosae*). A large percentage of consumers will not conform to a spray routine or do not want pesticides in their gardens. This aversion to pesticide usage contributes to the popularity of disease resistant roses such as the 'Knock Out' series of shrub roses. Rose companies, recognizing the market potential for disease resistant roses have used colorful terms (remarkable disease resistance, care-free, trouble-free, resists the dreaded fungus, etc) to describe them. The number of roses with resistance claims has exploded since Hagan et al (2) screened 37 cultivars of shrub and grown cover roses for resistance to black spot and Cercospora leaf spot.

Sixty cultivars of roses, most having disease resistance claims associated with their descriptions in rose catalogs, were planted at the West Tennessee Research and Education Center in Spring, 2006. Roses were arranged in a completely random design (replications 4) with a spacing of 1.25 m (4 ft) in a double row with 3.75 m (12 ft) of grass between each double row of roses. After transplanting, plants were watered and mulched. A drip line was installed and plants were watered as needed during the summer. Plants were fertilized with a general 20-20-20 fertilizer at labeled rate once a month until late summer when plants were allowed to harden off. Pruning was only used to prevent one plant from overgrowing an adjacent plant. No fungicides were used in the study. The cultivar 'Peace' served as a control.

Plans were evaluated every two weeks from planting until frost for susceptibility to black spot and Cercospora leaf spot (*Cercospora rosicola*) using the following scale: 0 = no symptoms, 1 = < 2% of foliage diseased; 2 = < 10% of foliage diseased; 3 = < 25% of foliage diseased; 4 = < 50% of foliage diseased; 5 = > 50%, but <100% of foliage diseased; and 6 = 100% of diseased foliage. Defoliation was rated using the same scale. Data were analyzed by date and by year using the Proc GLM procedure of SAS. F values for black spot, Cercospora leaf spot and defoliation were considered significant at the 0.05 level. When a significant F-value was detected, cultivars were separated using a LSD means separation test (p=0.05).

Results and Discussion: Black spot was detected four weeks after planting and Cercospora leaf spot was detected at week six. Data for susceptibility to black spot and Cercospora leaf spot and tolerance to disease (defoliation) for the date August 31 are presented in Table 1. Many rose cultivars with disease resistance claims in industry catalogs were susceptible to black spot, Cercospora leaf spot or both. Cultivars 'Baby Love', 'Belinda's Dream', 'Carefree Sunshine', 'Fourth of July', 'Hansa', 'Homerun', 'Linda Campbell', 'Palmengarten Farnkfurt', 'Pink Knock Out', 'Red Knock Out', 'Sunsprite', 'Topaz Jewel', 'Wild Spice', and 'Wildberry Breeze' were resistant to both diseases. 'Linda Campbell' (1) and 'Red Knock Out' (2) were found to be susceptible to black spot in previous studies where both cultivars appeared to be resistant in the first year of the studies. In our study, roses will be evaluated for disease resistance again in 2007 and 2008.

As noted in previous studies (1, 2), resistance mechanisms for black spot and Cercospora leaf spot appear to be different. The cultivar 'Sea Foam' had no symptoms of black spot (Table 1) but was one of the most susceptible cultivars to Cercospora leaf spot. In contrast, the cultivar 'Be-Bop' was very susceptible to black spot, but was resistant to Cercospora leaf spot. Tolerance to both diseases was observed in the cultivar 'Disneyland Rose' which was statistically as susceptible to both diseases as any other cultivar, but had a relatively low level of defoliation.

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Table 1. Black spot, Cercospora leaf spot and defoliation scores for sixty cultivars of roses (table continued on next page).

Cultivar	Black spot	Cercospora leaf spot	Defoliation
About Face	2.0 defghi	0.25 jk	1.75 fghijklm
Baby Love	0.0 k	0.0 k	0.0 m
Ballerina	0.5 ijk	1.3 ghij	0.25 lm
Be-Bop	3.5 abcd	0.0 k	3.5 abcdef
Belinda's Dream	0.3 jk	0.5 ijk	1.0 ijkm
Bill Warriner	3.3 abcd	0.0 k	4.5 a
Bonica	2.3 cdefgh	0.8 hijk	2.8 abcdefghi
Bride Dream	2.5 bcdefg	0.0 k	2.0 defghijkl
Carefree Delight	0.0 k	2.5 cdef	2.0 defghijkl
Carefree Sunshine	0.0 k	0.8 hijk	0.0 m
Carefree Wonder	0.3 jk	2.5 cdef	2.8 abcdefghi
Cecile Brunner	0.8 hijk	1.8 efgh	1.0 ijklm
Crimson Bouget	3.3 abcde	0.3 jk	2.5 bcdefghij
Crystal Fairy	0.0 k	3.8 ab	1.8 fghijklm
Disneyland Rose	2.8 abcdef	1.8 ab	1.8 fghijklm
Fairy Queen	0.0 k	3.3 abcd	1.0 ijklm
Fourth of July	1.5 fghijk	0.3 jk	1.8 hijklm
Gold Medal	2.3 cdefgh	0.0 k	2.7 abcdefghi
Golden Zest	3.2 abcde	0.4 ijk	3.6 abcdef
Hansa	0.3 jk	0.3 jk	0.8 jklm
Homerun	0.0 k	0.3 jk	1.3 hijklm
Honor	3.7 abc	0.0 k	3.0 abcdefgh
Julia Child	4.0 ab	0.0 k	3.3 abcdefg
Linda Campbell	1.5 fghijk	0.8 hijk	1.3 hijklm
Love	2.0 defghi	0.5 ijk	2.0 defghijkl
Lovely Fairly	0.0 k	3.3 abcd	1.0 ijklm
Magic Blanket	0.0 k	2.5 cdef	0.5 klm
Magic Carousel	0.8 hijk	1.5 fghi	1.5 ghijklm
Memorial Day	3.3 abcde	0.0 k	3.3 abcdefg
Midas Touch	2.0 defghi	0.5 ijk	2.5 bcdefghij
Nearly Wild	0.8 hijk	3.5 abc	1.5 ghijklm
Olympiad	3.5 abcd	0.0 k	4.0 abc
Palmengarten Frankfurt	1.5 fghijk	0.5 ijk	3.4 abcdef
Pascali	2.6 bcdef	0.8 hijk	3.4 abcdef
Peace	4.3 a	0.0 k	4.0 abc
Pink Knock Out	0.0 k	0.3 k	2.0 defghijkl
Pretty Lady	0.0 k	1.0 hijk	0.8 jklm
Pristine	3.0 abcdef	0.0 k	0.8 jklm

Table 1 (continued). Black spot, Cercospora leaf spot and defoliation scores for sixty cultivars of roses.

Cultivar	Black spot	Cercospora leaf spot	Defoliation
Rainbow's End	3.3 abcde	0.5 ijk	3.0 abcdefgh
Red Knock Out	0.0 k	0.6 ijk	1.9 efgh
Red Ribbons	1.8 efghij	0.8 hijk	3.5 abcdef
Santa Claus	2.0 defghi	0.7 ijk	1.5 ghijklm
Scent From Above	2.8 abcdef	0.3 jk	3.0 abcdefgh
Scentimental	4.0 ab	0.0 k	3.8 abcdefgh
Sea Foam	0.0 k	4.0 a	1.5 ghijklm
Sexy Rexy	2.5 bcdefg	0.0 k	3.8 abcde
Snowcone	0.0 k	2.3 defg	0.5 k
Space Odyssey	2.8 abcdef	0.3 jk	4.0 abc
Starina	3.8 abc	0.5 ijk	3.0abcdefgh
Sunbright	4.3 a	0.3 jk	2.0 defghijkl
Sunsprite	1.5 fghijk	0.0 k	1.0 ijklm
Topaz Jewel	0.5 ijk	0.5 ijk	0.8 jklm
Tournament Roses	3.3 abcde	0.5 ijk	3.0 abcdefgh
White Dawn	0.0 k	1.8 efgh	0.8 jklm
Wild Spice	0.0 k	0.0 k	0.0 m
Wild Thing	0.9 ghijk	2.7 bcde	1.9 efghijkl
Wildberry Breeze	0.0 k	0.5 ijk	0.8 jklm
Winsome	2.8 abcdef	0.3 defg	2.0 defghijkl
Zephrine Drouhin	4.3 a	0.3 jk	4.3 ab

Grass Hosts of *Sclerotium rolfsii*, a Common Pathogen of Ornamental Plants

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Index Words: southern blight, *Sclerotium rolfsii*, tall fescue, bentgrass

Significance to Industry: *Sclerotium rolfsii,* the causal agent of southern blight, has been well documented as a pathogen of woody and herbaceous ornamental plants. This fungus may also be observed attacking amenity turfs such as creeping bentgrass and tall fescue. This is the first report of *S. rolfsii* on creeping bentgrass in Tennessee.

Nature of Work: In the Southeast, stem rots of ornamental plants caused by *S. rolfsii* are fairly common in landscape beds and nurseries during summer months. This fungus has a wide host range of several hundred plants including: acuba, ajuga, crabapple, forsythia, hellebores, hosta and Jacob's Ladder (1). *S. rolfsii* is well adapted to the warm climate of the South. In the Midwest, *S. rolfsii var. delphinii* may be found attacking herbaceous perennials (2). *Sclerotium rolfsii* is also a pathogen of amenity turf. It has been reported on bluegrass (4), creeping bentgrass (3), and tall fescue (1,4). On a creeping bentgrass green, symptoms can be very dramatic. Large patches of turf, several feet in diameter, may be killed. Dead bentgrass has a distinctive reddish-brown color. If the fungus is active, white mycelium is visible at the margin of the patch.

Sclerotium rolfsii and *S. rolfsii* var. *delphinii* may be separated on differential growth temperature, sclerotial morphology and with molecular techniques. *S. rolfsii* has two morphological characteristics that make identification fairly straight forward. First, it produces an abundance of white mycelium. The amount of mycelium may vary, depending on the host. Second, it produces spherical, tan, resting structures called sclerotia that may vary from less than a mm in diameter to several mm in diameter. Sclerotia are white when immature and tan to reddish-brown when mature. Microscopically, the fungus has primary hyphae which are several µm in diameter and have clamp connections; it also produces secondary hyphae which are of smaller diameter and have no clamp connections.

In July 2007, a golf course in Southeast Tennessee submitted a bentgrass specimen to the UT Soil, Plant and Pest Center for diagnosis. The plug was from an oval patch ca. 50cm in diameter on a creeping bentgrass green. An abundance of white mycelium was visible at the periphery of the patch. Immature, white sclerotia and tan, mature sclerotia 1mm in diameter were visible within the turf canopy. Primary hyphae were hyaline, septate, 3-8µm in diameter with clamp connections. The causal agent was identified as *S. rolfsii*. In late August and early September of 2007, small patches of

dead grass ca. 15cm in diameter where observed in turf-type tall fescue plots at the MS State University North Farm. White mycelium was present, but not in abundance. Tan sclerotia .5-1mm were observed on infected leaf blades. Hyphae were hyaline, septate and clamp connections were present. Again, *S. rolfsii* was identified as the causal agent.

Results and Discussion: *S. rolfsii* is well known as a pathogen on dicots, especially ornamental plants. It is not readily recognized as a pathogen of turfgrass. On creeping bentgrass, the symptoms of southern blight on a golf green are dramatic. On tall fescue, the symptoms of southern blight could easily be mistaken for brown patch caused by *Rhizoctonia solani*. One thing that is clear, is that on turfgrass unless a specimen is closely examined, southern blight may be mistaken for other diseases. This may complicate management strategies if the proper chemical control is not selected. To our knowledge, this is the first report of southern blight caused by *Sclerotium rolfsii* on creeping bentgrass, *Agrostis stolonifera*, in Tennessee.

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Freeze Injury to Ornamental Plants in Tennessee in 2007

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Index Words: ornamentals, freeze injury

Significance to Industry: In 2007, a devastating spring freeze severely damaged ornamental plants in landscapes, container and field nurseries in Tennessee. This is a review of the weather patterns that led to the damage and examples of the freeze injury that was observed on woody plants.

Nature of Work: Spring 2007 in Tennessee was drier than normal, but otherwise plants in nurseries and landscapes were in very good shape. According to the Normalized Difference Vegetation Index (NDVI), a measure of the greening of the canopy of the forest collected by NOAA (1), at the Walker Branch Watershed in Oak Ridge, TN, the green-up of vegetation was three weeks earlier than 2006 and was due to higher than normal temperatures. In most regions of the state, temperatures had reached the high 70's to low 80's by late March. During the first week of April, flowering dogwood (*Cornus florida*) was in full bloom.
Weather conditions changed drastically. In Murfreesboro, TN, the geographic center of Tennessee, the daily high on April 4 was 80F with a low of 50F. By April 6, the daily high was 50F with a low of 30 F. On Easter morning, April 8, the low had dropped to 19F, and on April 9 the low was again 19F. Record low temperatures were reported from nearly all reporting stations in TN. In Crossville, TN on the Cumberland Plateau at nearly 2,000ft in elevation, the record low was 13F. In many of the areas where nursery stock is grown, lows ranged from the high teens to low twenties. In addition to record lows, other factors played a role in plant damage. First, the Mid-South region was already in a moderate to severe drought. Second, the duration of the cold temperatures undoubtedly added to the intensity of the damage. At Crossville, TN, from April 4-10, temperatures below 32F were recorded for 70 hrs.

On Easter morning, a common sight on crape myrtle was ribbons of ice being extruded from twigs and branches. Once temperatures warmed, there was widespread damage to trees and shrubs in landscapes and nurseries. Foliar symptoms ranged from wilted leaves on sugar maple to scorched foliage on Japanese maple and boxwood. New shoots on plants such as nandina, mahonia, and some holly species were killed by the freeze. Some of the more severe damage such as bark splitting on the trunk or main stem occurred on crape myrtle, azalea, juniper and arborvitae. Many mature crapemyrtle were killed to the ground. Bark splitting on the lower trunk of some shade trees such as oak was also observed. Ginkgo and zelkova were especially hard hit by

the freeze. Small specimens of each species were killed to the ground. Even mature ginkgo that lost its foliage to the freeze did not refoliate until 5-6 weeks after the damage had occurred, whereas most tree species began producing shoots 3-4 weeks after the freeze. Fallout from the freeze continued in May, as many ornamental cherry that had appeared to be spared, began exhibiting shoot and branch dieback. A quick look at the cambial layer of the branches and trunks showed discolored tissue, evidence of freeze injury. As of May 3, NOAA reported that CO_2 levels in the Mid-South were above normal as the forest canopy had not yet recovered.

Damage to flowering dogwood was especially significant. Dogwood seed planted in fields in the fall had germinated and seedlings were growing well prior to the freeze. Nearly all of the seedlings that were unprotected were killed by the freeze. These would have been used for budding in late summer. Some growers were able to protect seedlings by covering them with sawdust or wheat straw. Also, many of the dogwoods that were budded in 2006 were killed by the freeze. To make matters worse, dogwood fruit set prior to the freeze was killed, making it more difficult to procure seed for planting during fall 2007.

A second blow to the nursery and landscape industry was the drought of 2007. As the drought intensified in early summer, damage due to the freeze compounded the stress observed on certain plants. *Taxus* spp. in landscapes in Middle Tennessee showed widespread branch dieback. Wounds created by the freeze were colonized by opportunistic plant pathogens. Leyland cypress and other conifers showed branch dieback due to canker causing fungi such as *Botryosphaeria* and *Seiridium*. These fungi often live on their host plants as harmless endophytes and cause little damage unless the host is exposed to severe stress.

Results and Discussion: The Spring Freeze of 2007 will no doubt play a significant role in the short term in the nursery industry in the Mid-South. Plants that had to be cut back to remove branches killed by the freeze will be smaller and of a lower grade. No doubt, some budded dogwood cultivars will be more scarce and harder to find. Growers had to network to find dogwood fruit (seed) to plant for the 2008 crop. Losses of liners and small trees may be a final blow to some growers that may not be able to withstand such a catastrophic financial setback.

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Detection of *Botryosphaeria dothidea* as the Pathogen of Dogwood Leaf Blight by PCR-Based Markers

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Additional index words: Dogwood, Cornus florida, Botryosphaeria dothidea, PCR.

Significant to Industry: *Botryosphaeria dothidea* can infect trees, fruit and ornamental crops and causes leaf blight, stem canker, and fruit rot (1-6). The fungus *B. dothidea* was isolated from dogwood leaves and stems that had leaf blight symptoms; pathogenicity tests confirmed that *B. dothidea* caused foliage blight in flowering dogwood (*Cornus florida)*. To confirm the identification of *B. dothidea* as the causal agent of leaf blight in dogwood, a PCR-based DNA analysis was done and a bioassay technique was developed for the detection and identification of *B. dothidea*. The information will allow early detection and facilitate disease management.

Nature of Work: Cultures of *Botryosphaeria dothidea* were isolated from infected dogwood leaves and stems. Genomic DNA was extracted from conidia and mycelium using a DNeasy Plant Mini Kit (Qiagen Inc, Valencia, CA). The PCR amplification was performed in a DNA thermal cycler following standard PCR procedures with minor modifications. Each 50 µl PCR reaction mixture consisted of 36 µl sterile ddH_2O, 5 µl 10X PCR buffer, 3 µl $MgCl_2$ (25 mM), 1.5 µl dNTP (10 mM total, 2.5 mM each), 1.5 µl primer each (20 ng/µl), 0.2 µl *Taq* polymerase (Promega) (5 U/µl), and 1.3 µl template DNA (20 ng/µl). PCR cycles consisted of an initial denaturation step at 94 ^{0}C for 4 min followed by 42 cycles of 1 min at 93 ^{0}C (denaturation), 1 min at 40 to 60 ^{0}C (annealing), and 2 min at 72 ^{0}C (extension). The annealing temperature was set based on the primer Tm and usually a five degree less than the lower primer Tm (Tm – 5) was used as the annealing temperature for the PCR reaction. An extension cycle at 72 ^{0}C for 5 min was used to terminate the reaction and finally at 4 ^{0}C soak. The PCR products were visualized in 1.5% agarose gel in 1X TBE, stained with ethidium bromide. Three universal primers, ITS1, ITS1-F and ITS4 (Table 1), were used to amplify the ITS region in the pathogen by a using standard PCR procedures. The two PCR products amplified from the two primer pairs, ITS1/ITS4 and ITS1-F/ITS4, were sequenced by the Davis Sequencing Inc. at Davis, CA (http://www.davissequencing.com). Before sequencing of PCR products, PCR products were purified by use of QIA quick PCR Purification Kit (Qiagen Inc, Valencia, CA) (http://www.qiagen.com). The two sequences obtained were analyzed and compared with all sequences of ITS region in GenBank by using BLAST (http://www.ncbi.nlm.nih.gov/BLAST/) to determine the pathogen of leaf blight in dogwood. PCR primers for the ITS region of *B. dothidea* were designed by using the Software Primer 3 (http://frodo.wi.mit.edu/cgi-bin/primer3/primer3_www.cgi). Five

primers including two forward primers and three reversal primers were designed for the ITS region of *B. dothidea* (Table 1). Six primer pairs (2 x 3) were tested in this research.

Results and Discussion: The PCR product was 621 bp amplified from the primer pair ITS1-F/ITS4 and 583 bp amplified from ITS1/ITS4 (Fig. 1) (Table 2). The sequence of the PCR product amplified from the primer pair ITS1/ITS4 was 99.5% (581/583) matching to the accession AY259092, and 100% (517/517) to accession AY236950 of *B. dothidea* in GenBank. Other close matching ones in GenBank are 97.9% (509/521) to AF243397 of *B. cortices*, 98% (522/543) to AF246930 of *B. mamane*, 94% (425/448) to AY259091 of *B. lutea*, and 94% (424/450) to AY259098 of *B. parva*. The results indicated the pathogen of leaf blight in dogwood is *B. dothidea.*

Six primer pairs in combination of three forward primers bd-f1, bd-f2, bd-f3 and two reversal primers bd-r1 and bd-r2 showed specific band only for *B. dothidea* in all tested samples including *Alternaria alternata, Acremonium alternatum, Botrytis cinerea, Cladosporium sp., Colletotrichum acutatum, C. gloeosporioides*, and *Phomopsisi eucommicola*. The polymorphic bands and their sizes amplified from the six primer pairs are showed in Table 2 and Fig. 2. The sequences of the six PCR products matched those in corresponding location of ITS region of *B. dothidea.* These results verified that the *B. dothidea* is the pathogen of dogwood leaf blight. The species-specific primers can be used as molecular markers for the detection and identification of *B. dothidea* of dogwood.

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Table 1. The primer name, sequence, size, Tm, and specificity.

Primer Name*, **	Sequence (5' to 3')	Size	Tm °C	Specific
ITS1-F	cttggtcatttagaggaagtaa	22	60.00	universal
ITS1	tccgtaggtgaacctgcgg	19	64.48	universal
ITS4	tcctccgcttattgatatgc	20	58.35	universal
bd-f1	ggaccatcaaactccagtcag	21	62.57	For Bd***
bd-f2	gccagaggaccatcaaactc	20	62.45	For Bd
bd-f3	cccacccttttgtgtacctacc	21	64.52	For Bd
bd-r1	gctccgaagcgagatgtatg	20	62.45	For Bd
bd-r2	aaaggacggtgcccaatac	19	60.16	For Bd

* All primers were made from Qiagen.com
** bd-f1, bd-f2 and bd-f3 were forward primers; bd-r1 and bd-r2 reverse primers.
***Bd = *Botryosphaeria dothidea*

Table 2. The polymorphic band size amplified from eight ITS primer pairs for the dogwood leaf blight pathogen, *Botryosphaeria dothidea*.

Primer pair	Band size (bp)	Lane in Figure 1 or 2
ITS1-F/ITS4	621	Lane 1 & 2 in Fig. 1
ITS1/ITS4	583	Lane 3 & 4 in Fig. 1
bd-f1/bd-r1	317	Lane 1, 2 in Fig. 2
bd-f1/bd-r2	243	Lane 3, 4 in Fig. 2
bd-f2/bd-r1	333	Lane 5, 6 in Fig. 2
bd-f2/bd-r2	259	Lane 7, 8 in Fig. 2
bd-f3/bd-r1	420	Lane 9, 10 in Fig. 2
bd-f3/bd-r2	346	Lane 11, 12 in Fig. 2

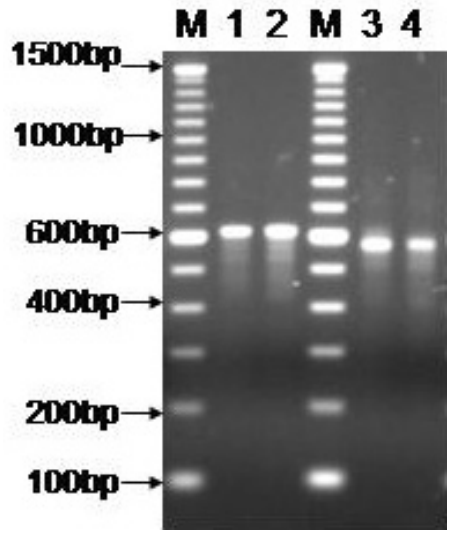

Fig. 1. Amplification pattern of DNA detected by two primer pairs in *Botryosphaeria dothidea*: (1) ITS1-F/ITS4 (Lanes 1 & 2) and (2) ITS1/ITS4 (Lanes 3 & 4). Lane M is a 100 bp molecular-weight marker.

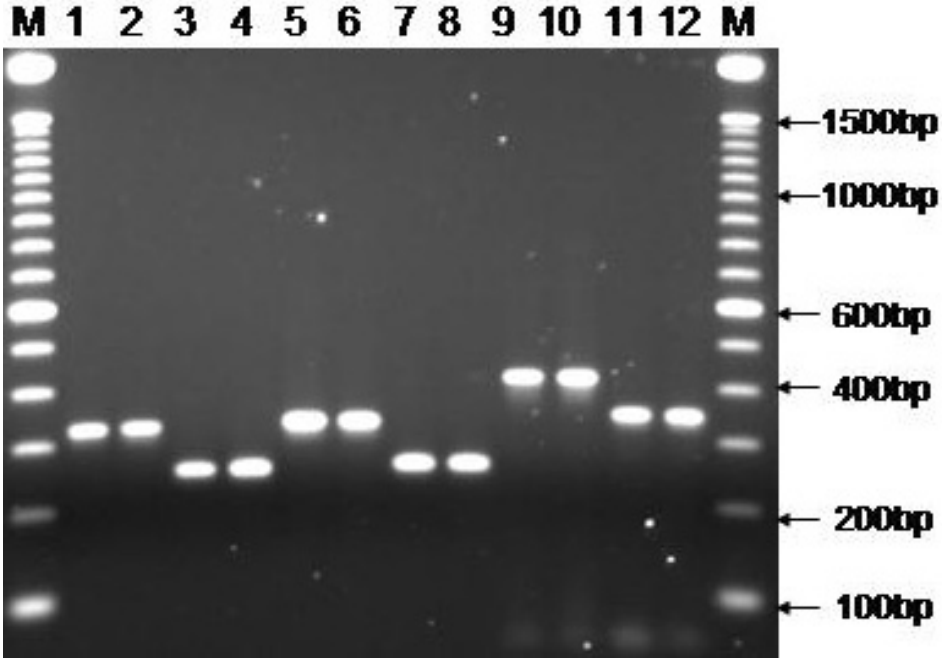

Fig. 2. DNA pattern amplified from six primer pairs: (1) bd-f1/bd-r1 (lane 1 & 2), (2) bd-f1/bd-r2 (lanes 3 & 4), (3) bd-f2/bd-r1 (lanes 5 & 6), (4) bd-f2/bd-r2 (lane 7 & 8), (5) bd-f3/bd-r1 (lane 9 & 10), and (6) bd-f3/bd-r2 (lane 11 & 12) in two DNA samples: (1) *Botryosphaeria dothidea* infected dogwood leaf (lane 1, 3, 5, 7, 9, and 11) and (2) *B. dothidea* conidia (lane 2, 4, 6, 8, 10 and 12). Lane M is a 100 bp molecular-weight marker.

Identification of NBS-LRR Type Disease Resistance Gene Analogs in Dogwood (*Cornus florida*)

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Additional index words: Dogwood, *Cornus florida*, disease resistance gene, resistant gene analogues

Significant to Industry: Flowering dogwood (*Cornus florida* L.) is one of the most popular ornamental trees in the eastern United States and its popularity is threatened by disease problems. Host resistance is recognized as the best method for disease management. Disease resistance genes (R-genes) have been cloned from a number of plant species and can be categorized into four classes on the basis of the conserved amino acid sequences of their protein products (5). The objective of this research was to identify NBS-LRR gene analogs for disease resistance and conduct phylogenetic analysis of the NBS-LRR motifs in *C.florida*. Information on disease resistance will provide a better understanding of disease resistance in flowering dogwood and benefit disease management.

Nature of Work: The sequence comparison of cloned genes for disease resistance in plants has shown structural homology regardless of whether the resistance was to fungal, bacterial, nematode or viral pathogens. The classes include: the NBS-LRR genes with a nucleotide-binding site (NBS) and a leucine-rich repeat (LRR) motif; extracellular LRR genes; protein kinase genes; and, receptor kinase genes (1, 5). The NBS-LRR genes can be divided into two classes, TIR and non-TIR, according to whether they code for a TIR domain which contains an amino terminus with homology to the Drosophila Tol protein and mammalian interleukin-1-receptor (1, 5). The non-TIR group often contains a coiled-coil motif with a subset of these coding for a leucine zipper structure (1, 2, 5). Both TIR and non-TIR, groups contain some conserved amino acid motifs, most notably the P-loop, kinas-2, and GLPL motifs (1, 5). These conserved motifs have enabled rapid isolation of the NBS-LRR genes or resistance gene analogs (RGAs) from different plant species by using a PCR-based approach with degenerate oligonucleotide primers designed from these domains (1-6).

Previously identified powdery mildew resistant accessions of *C. florida* (MI 8, MI 9, MI 17, WR 19 and 'Cherokee Brave'), were used in this study (7, 8). Genomic DNA was extracted from fresh dogwood leaves of these plants by using DNeasy Plant Mini Kit

(Qiagen Inc, Valencia, CA) and protocols from the company. A variety of degenerate primers for PCR amplification of RGAs that had been successfully used in isolation of RGAs and R-gene in *Arachis*, cotton, and grape (Table 1) (2-4, 6) were selected for this study. The primers were previously designed from P-loop, and GLPL (2-4, 6). PCR amplification was performed in a DNA thermal cycler following standard PCR procedures with minor modifications. Each 50 µl PCR reaction mixture consisted of 36 µl sterile ddH_2O, 5 µl 10X PCR buffer, 3 µl $MgCl_2$ (25 mM), 1.5 µl dNTP (10 mM total, 2.5 mM each), 1.5 µl primer each (20 ng/µl), 0.2 µl *Taq* polymerase (Promega) (5 U/µl), and 1.3 µl template DNA (20 ng/µl). PCR cycles consisted of an initial denaturation step at 94 ^{o}C for 4 min followed by 42 cycles of 1 min at 93 ^{0}C (denaturation), 1 min at 40 to 60 ^{o}C (annealing) and 2 min at 72 ^{0}C (extension). The annealing temperature was set based on the primer Tm and usually a five degree less than the lower primer Tm (Tm – 5 ^{o}C) was used as the annealing temperature for the PCR reaction. An extension cycle at 72 ^{o}C for 5 min was used to terminate the reaction and finally at 4 ^{o}C soak. PCR products were run on a 1% low-melting-point agarose gel for visualization. Bands of the appropriate size for the specific PCR reaction were excised from the gel and purified using a QIAquick gel extraction kit (QIAGEN, Valencia, Calif.). Each purified DNA band was cloned into a plasmid vector using TOPO T/A Cloning kit (Invitrogen, Carlsbad, Calif.). Clones were sequenced by a sequencer such as Applied Biosystems model 377 PRISM automated sequencer. The RGAs DNA sequences were translated to amino acid sequences by Translate tool (ExPASy) and were compared to protein sequences in the GenBank database using Blastx. Phylogenetic trees were constructed for RGAs in dogwood by using neighbor-joining (NJ) by ClustalX viewed by TreeView.

Results and Discussion: Out of 42 primer pairs, seven produced single bands. Figure 1 is an example of the PCR products with about 510 bp fragments amplified from the primer pair P1a-fwd/P3d-rev in three dogwood accessions MI 9, WR 19 and Cherokee Brave. Amplification with some primer pairs resulted in multiple PCR products in different dogwood accessions. Eleven translated sequences were similar to the NBS-LRR proteins in the GenBank. Amino acid identity between the 11 PCR products and the GenBank sequences ranged from 38% to 64%. The conserved motifs in the 11 translated sequences are (fv)L(ilv)(iv)LDD(iv)(adw) in Kinase II, TTR in RNBS-B, and GLPL in GLPL-motif regions. The presence of the Kinase II, RNBS-B, and GLPL motifs indicated that the PCR amplified sequences are from the NBS region of the RGAs.

Phylogenic analysis of the identified RGAs indicated that the eleven RGAs from dogwood are highly diverged; some are similar but others are distantly linked to the four known resistance genes of flax M, soybean LM6, tomato I2C1 or *Arabidopsis* RPS2 (Fig. 2). This research is the first study to focus on the isolation and identification of resistance gene analogs in dogwood and will be useful in efforts to clone disease RGAs and map RGAs associated with resistance loci for specific diseases like powdery mildew in dogwood.

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Table 1. Degenerate PCR primers used for RGAs isolation in dogwood (*Cornus florida*)

Primer	Conserved amino acid motif	Primer sequence (5' to 3')
P1a-fwd	P-loop	GGIATGGGIGGIIIIGGIAAGACIAC
P3d-rev	GLPL	AIITCIGIICIAGIGGTAAICC
F1a	NBS	GGTATGGGAGGTGTCGGTAAGAC
R1a	NBS	ACCTTGAATGCCAATGGCAAGCC
F1	NBS	GGAATGGGAGGTGTAGGCAAAAC
R1	NBS	ACTTTGAACGCTAATGGCAATCC
RG1-f1	P-loop	GAATTCGGAGTCGGTAAGACCAC
RG1-f2	P-loop	GAATTCGGAGTAGGTAAGACGAC
RG1-f3	P-loop	GAATTCGGTGTAGGCAAGACAAC
RG1-f4	P-loop	GAATTCGGTGTGGGAAAAACCAC
RG2-r1	GLPL	GTCGACAATGCTAGTGGCAGACC
RG2-r2	GLPL	GTCGACAGTGCGAGTGGAAGACC
RG2-r3	GLPL	GTCGACAACGCAAACGGTAGACC
RG2-r4	GLPL	GTCGACAGAGCTAGAGGCAACCC

*(I, inosine)

P1a-fwd and P3d-rev were designed from *Arachis* (Bertioli et al. 2003); F1a, R1a, f1, and r1 from cotton (He et al. 2004); RG1-f1, RG1-f2, RG1-f3, RG1-f4, RG2-r1, RG2-r2, RG2-r3, and RG2-r4 from grape (Gaspero & Cipriani, 2002).

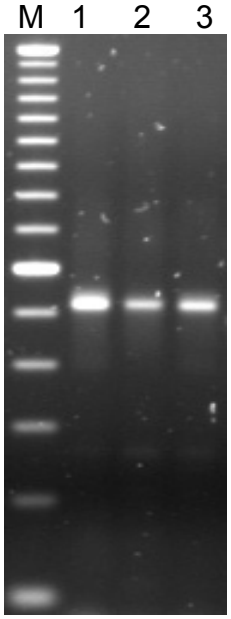

Fig. 1 The PCR product is about 510 bp amplified from the primer pair P1a-fwd/P3d-rev in three dogwood (*Cornus florida*) accessions: 1. MI 9, 2. WR 19 and 3. Cherokee Brave. Lane M is a 100 bp molecular-weight marker

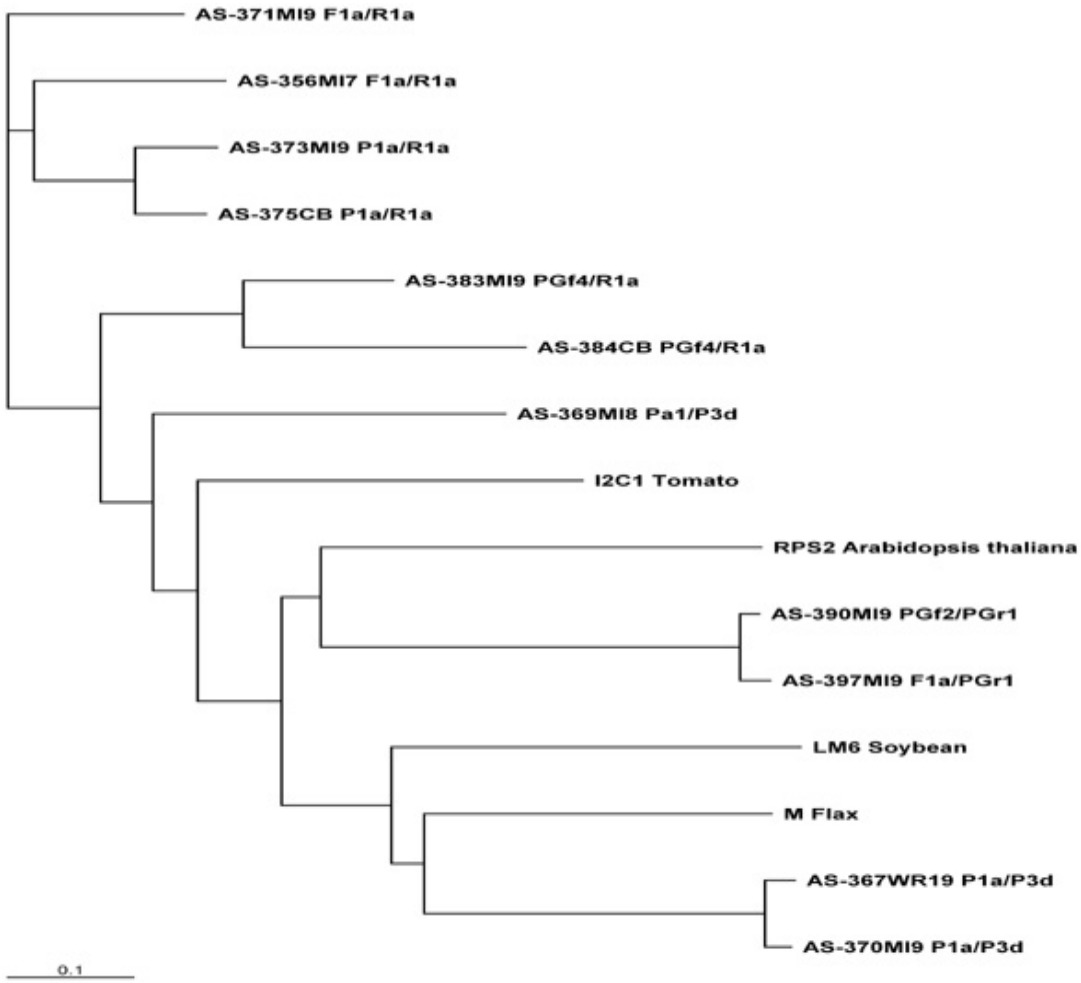

Fig. 2. Phylogenetic tree of the deduced amino acid sequences of flowering dogwood RGAs, using neighbor-joining (NJ) by ClustalX viewed by TreeView. Resistance genes: flax M, soybean LM6, tomato I2C1 and *Arabidopsis* RPS2.

A 2006 survey for Phytophthora and other pathogens causing Phytophthora-like symptoms in Tennessee nurseries

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Additional index words: Fusarium oxysporum, Fusarium solani/Nectria hematoccoca, Verticilium, Botyosphaera sp., root rots, vascular wilts

Significant to Industry: *Phytophthora* causes plant mortality, root rots and leaf blights in nurseries and the landscape. Phytophthora diseases are important in field and container-grown nursery plants. Disease symptoms caused by other soil-borne fungi such as *Fusarium, Verticilium, Botyosphaera* and *Rhizoctonia*, may superficially resemble those caused by *Phytophthora* spp., but fungicides effective on *Phytophthora* may not be effective on the other non-related fungi. A survey of Phytophthora species on different hosts, their incidence and symptoms they cause will identify Phytophthora spp. and other soil-borne pathogens that cause Phytophthora-like symptoms in TN nurseries. The information will be useful in the development of sustainable disease management system.

Nature of Work:
The genus *Phytophthora* includes many destructive pathogens that have a wide host range with only a few species having restricted hosts (Mithchell and Kannwischer-Mitchell 1993). *Phytophthora* is an important pathogen in container and field grown nursery plants in Tennessee and causes sporadic plant mortality in landscapes. In many ways, the control of *Phytophthora* spp. in nurseries would present far fewer obstacles than management of these pathogens in the landscape and forest plants. In addition to *Phytophthora*, other soil-borne pathogens may cause Phytophthora-like symptoms, but without proper identification and documentation, the plant mortality may be ascribed to Phytophthora. Chemical fungicides effective on *Phytophthora* may not be ineffective on the other non-related soil-borne pathogens. The objective of this study was to survey and identify Phytophthora species and other soil-borne pathogens associated with woody plants, their incidence and the threat they pose to trees and shrubs in Tennessee nurseries.

In 2006, a survey for Phytophthora in Tennessee nurseries was **c**onducted in Warren County (six locations), Cheatham County (one location), Davidson County (one location), and Rutherford County (one location). Direct isolation of *Phytophthora* spp. from plant material was conducted on *Phytophthora* semi-selective medium (PARPH) containing antibiotic amendments, pimaricin, ampicilin, rifampicin, pentachloronitrobenzene and hymexazole (Jeffers and Martin 1986). To improve the chance of recovering *P. ramorum*, direct isolation from very fresh

samples was done within 24 hours of sample collection. Plant tissue from stem, collar region, roots, and leaves were used for direct isolation of *Phytophthora*. All tools used in the isolation were surface sterilized to reduce contamination. Five specimens were plated in each Petri dish, sealed with parafilm, labeled and incubated in the dark at 20 - 23°C and observed over a 7-day period.

Soil collected from the rhizosphere of sampled plants was assayed for *Phytophthora* by using a leaf disc baiting technique. In order to detect a variety of *Phytophthora* species, several baits (pine needles, *Rhododedren* and *Pieris* leaf discs) were used simultaneously (Erwin and Ribeiro, 1996, Ferguson and Jeffers 1999). The soil samples were collected in Petri dishes and flooded with sterile distilled water. Three plates were used per sampled tree. Five baits were floated in each plate and incubated at 20-22 C for 48-72 hours. The baits were then removed, blotted dry on sterile paper towels and placed on *Phytophthora* semi-selective medium (PARPH). Cultures were incubated in the dark at 20 C. Observations were made over a 7-day period. Similar isolation was done from irrigation water including irrigation ponds and streams.
Morphological characterization based on sporangia, hyphae & chlamydospores etc**.** was done. In addition, PCR-based DNA analysis using universal primers ITS_1 and ITS_4 was conducted (Herion *et al.*1994). The PCR products were cleaned and sequenced and the sequence was compared with information available in the GenBank using a Blast search.

Results and Discussion:
A total of 540 samples were evaluated for *Phytophthora* (Table 1). *Phytophthora* was isolated from plant tissue including roots and collar region of symptomatic plants and from soil (Table 1). *Phytophthora* was isolated from flowering dogwood more often than from other plants suggesting its high susceptibility to *Phytophthora* (Table 2). Bold Cyprus Japanese Hollie and White Pine were also highly susceptibility to Phytophthora (Table 2). Based on morphological characteristics, the *Phytophthora* isolates seemed to represent different species. Out of 540 isolates collected, *Phytophthora* species identified included *P. nicotianae,* and *P. cactorum.* In addition to *Phytophthora*, other soil borne pathogens such as *Fusarium oxysporum, Nectria haematococca, Fomes radians, Verticillium sp.* and *Botryosphaeria* were also isolated from plants with symptoms indicative of Phytophthora infections. The incidence of *Phytophthora* and of the other pathogens, host plants sampled, disease symptoms and results from pathogenicity tests will be used to describe the role of *Phytophthora spp.* and of the other pathogens on plant mortality. The isolation of *Phytophthora* from soil was not always associated with isolation from the plant tissue. Since *Phytophthora* survives in the soil for a long time, its presence in the soil poses a threat to future plantings.

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Table 1. 2006 Summary on isolation of *Phytophthora* spp and other soil borne pathogens[1] on ornamentals

Location	Materials sampled for Phytophthora			Total no of samples.	Phytophthora detected in ELISA test
	Collar and root	Soil	Pond water		
TSU farm, Warren	85	82	20	175	+ve
MC Nursery, Warren	50	27	20	97	+ve
CW Nursery, Warren	33	38	20	91	+ve
SV Nursery, Warren	56	46	-	102	+ve
Cheatham County		36	-	36	+ve
Nashville, Davidson	No	27	-	27	+ve
SR Nursery, Rutherford	5	4	3	12	+ve
Total:				540	

[1]Other soil borne fungi isolated included: *Fusarium oxysporum*, *Fusarium solani*/*Nectria hematoccoca*, *Verticillium*, *Phoma spp and Formes sp.*

Table 2. Host plants sampled and tissues evaluated for Phytophthora in 2006.

Host plant	Plant tissues sampled and *Phytophthora* recovered[1]			
	Leaves	Roots	Stem/Collar	Soil
Flowering dogwoods	Yes	Yes	Yes	Yes
Lilac	No	No	No	Yes
Crape myrtle	No	No	No	Yes
Oakleaf Hydrangea	No	Yes	Yes	Yes
Bold Cyprus	No	Yes	Yes	Yes
Japanese Holly	No	Yes	Yes	Yes
White Pine	No	Yes	Yes	Yes
Oak	No	Yes	No	Yes
Maple	No	Yes	No	No
Forsythia	No	No	No	Yes
Red twig dogwood	No	Yes	Yes	Yes

[1]Other soil borne fungi isolated included: *Fusarium oxysporum*, *Fusarium solani*/*Nectria hematoccoca*, *Verticillium*, *Phom app and Formes sp.*

First Report of Downy Mildew in Lilac in Tennessee

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***Additional index words*:** lilac, downy mildew, leaf spots

Significant to Industry: Lilac is a common flowering shrub credited for its beautiful and fragrant bloom in spring. Several foliage diseases plague lilac in Tennessee. The most common disease is powdery mildew that makes the plants look unsightly due to foliage looking more white than green. Disease resistant cultivars are available, among which is *Syringa* 'Old Glory'. 'Old Glory' has displayed high level disease resistance to powdery mildew, bacterial blight and other foliage diseases in McMinnville, TN. However, in summer 2005 and in 2006, symptoms that appeared like common leaf spot were observed on Old Glory' and close observations on the underside of the leaf revealed fungus mycelia characteristic of downy mildew. This was the first time downy mildew was observed in Lilac and to our knowledge, this is the first report of downy mildew in lilac in Tennessee.

Nature of Work:
Syringa 'Old Glory' is a product of the lilac hybridization program at the U.S. National Arboretum; a selection from a controlled hybridization between *Syringa* 'Sweet Charity' and *Syringa* x *hyacinthflora* 'Pocahontas' made by Don Egolf in 1978 and released in March 2006. 'Old Glory' was selected for its abundant fragrant bluish-purple flowers, rounded growth habit, and disease tolerant foliage (Fig 1). 'Old Glory' is mildew-resistant and it has good field tolerance to Cercospora blight and *Pseudomonas syringae* problematic in warmer climates (USDA Zones 5-7). The new hybrid was field-grown for observation at the USDA National arboretum germplasm evaluation plots in McMinnville TSU Otis L. Floyd Nursery Research Center. Disease symptoms consisting of leaf necrotic lesions were first observed in 2005, and the symptoms intensified in 2006 making the plant unsightly (Fig.2). The foliar symptoms appeared like common leaf spots, similar to those associated with bacterial blight infections, but abaxial leaf surfaces had aerial, grayish mycelia which were suggestive of downy mildew. The downy mildew pathogen was characterized under a compound microscope in summer 2006.

Results and Discussion:
Symptoms that appeared like leaf spot symptoms (Fig.2) were associated with signs of aerial mycelia restricted to the abaxial leaf surfaces (Fig 3). Fungal colonies began as white, aerial mycelia that turned gray with age. On the adaxial leaf surface, initial

symptoms were chlorotic lesions that later turned brown and necrotic (Fig 2). Leaf lesions were circular or irregular and developed between the veins appearing angular in shape. Lesions often coalesced and formed large lesions. Disease symptoms persisted through the summer and caused defoliation. Microscopic observations revealed abundant sporangiospores borne on branched sporangiophores. Sporangiophores had monopodial branching with branches arranged at right angles to the supporting branch and the tips were distinctly obtuse. The sporangia were small, hyaline and ovoid in shape with dimensions of approximately 19.5 to 22 µm × 14 to 17 µm. Oospores were not observed. On the basis of symptoms and morphology of the organism, the pathogen was identified as *Plasmopara* sp.

Downy mildew of lilac has not been previously observed in McMinnville, TN and the inoculum source for this disease is unknown. Downy mildew fungi can overwinter as oospores in the soil, in colonized roots and host debris for many years. Infection may occur soon after transplanting a susceptible host into an infested area or after infested plant is transplanted in an area with favorable environmental conditions. It is probable that the infection may have started from infested plant material that had not shown disease symptoms due to unfavorable environmental conditions. Ideal conditions for downy mildew are cool night temperatures of 8-16 C and day temperatures less than 24 C with rain drizzle or fog resulting in wet plants until midmorning for at least 4 days in a row. Once infection occurs, a new crop of sporangia can be produced in 4 to 5 days. The sporangia are disseminated by rain and wind and they can germinate within 4 hours. As seasonal temperature rises, plants tend to outgrow the disease. However, the established disease symptoms remain throughout the season. New infections can also occur in the autumn as seasonal temperatures begin to fall. According to the McMinnville weather, infection may have started in early spring when cool temperatures and Spring rains are normally favorable to Downy mildew infections.

Studies on the management of this disease were not done, but recommendations for the management of the downy mildew in other crops include avoiding planting lilac in infested areas, use of resistant plants and chemical fungicides with active ingredient that specifically targets oomycete fungi (Cortner, 1930; www.ces.ncsu.edu/depts/pp/cucurbit/). Copper fungicides are affective on downy mildew (Horst, 1990). Other fungicides including metalaxyl plus chlorothalonil (Ridomil/Bravo®81W Mobile (systemic, translaminar), mefenoxam (Ridomil Gold Bravo, Ridomil Gold Copper, Group 4) are highly effective on downy mildew and the choice will depend on label recommendation for ornamentals.

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3. North Carolina State University web site (www.ces.ncsu.edu/depts/pp/cucurbit/)

Fig. 1. *Syringa* 'Old Glory' (NA 62974; P1641803) US National Arboretum Plant introductions, Floral and Nursery Plants Research Unit. http://www.usna.usda.gov/Research/Lilacs_Release.html).

Fig 2. Brown necrotic lesions caused by downy mildew in lilac 'Old Glory'.

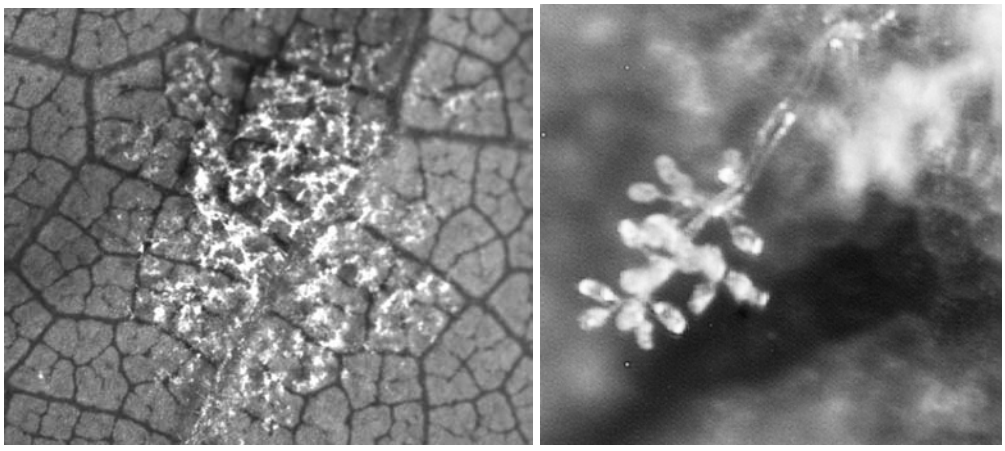

Fig 3. Growth of the downy mildew fungus on the underside of the leaf at x10 and x100 magnification

Phytophthora species identified in Tennessee nurseries in 2007

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Additional index words: Soil-borne pathogens, commercial nurseries, *Phytophthora citricola, P. megasperma, P. cinnamommi, P. cryptogea, P. nicotianae*, unclassified *Phytophthora* species.

Significant to Industry: *Phytophthora* species concern Tennessee growers because they cause economic losses in many species of field and container-grown trees. Identification of *Phytophthora* spp. associated with plant decline and mortality in commercial nurseries will allow early detection of new Phytophthora diseases and facilitate disease management and containment of destructive species.

Nature of Work:
Phytophthora species include very destructive pathogens that have a wide host range with only a few species being restricted by hosts (3, 8). Several *Phytophthora* species have been associated with diseases in Tennessee nursery production systems (9), but a comprehensive survey of *Phytophthora* species in commercial nurseries has not been done. The only reports on sudden oak death (SOD) reported in Tennessee nurseries were on plants that were traced back to the west coast and none of the plants had been in Tennessee for a long time. However, more than 60 % of SOD reported hosts such as *Viburnum* species (Viburnum), *Kalmia latifolia* (Mountain laurel); *Rhododendron* species (Ornamental rhododendron); *Lithocarpus densiflora* (Tanoak), *Camellia* species (Camellia), and *Acer macrophyllum* (big leaf maple), are grown in Tennessee nurseries. In addition, most of Tennessee is included in the high-risk area for *P. ramorum*. Early detection of destructive *Phytophthora* spp. will facilitate pathogen containment and minimize impact on Tennessee nursery industry, state economy and other states receiving plants from Tennessee nurseries.

Plant movement is an important mechanism for long distance spread of *Phytophthora* spp. Visibly healthy plants may play a significant role in spreading Phytophthora diseases to new areas. Tennessee commercial nurseries purchase and sell plants to diverse geographic areas including regions where *P. ramorum* has been reported. The control of *Phytophthora* spp. in nurseries would be less problematic than management in landscape plants. The objective of this study was to identify Phytophthora diseases in woody ornamentals and document species affecting Tennessee nurseries.

In summer 2007, a field survey was conducted in six Tennessee counties covering approximately three nurseries per county and collecting more than 100 samples per nursery. The survey included sites with poorly growing plants and plant mortalities in field-grown and container grown trees. Nurseries with irrigation facilities and others with

no irrigation system were included in the survey. Plants were closely examined for abnormal symptoms including cankers, wilting, dieback, and leaf blights and symptoms associated with *P. ramorum* infections such as bleeding, necrosis, leaf spots, leaf blotches and leaf tip necrosis. Sample collection from nurseries included plant tissues and rhizosphere soil.

Direct isolation of *Phytophthora* from plant tissue was done within 24 hours of sample collection on a selective growth media for *Phytophthora* (4, 6). The media contained pimaricin-ampicillin-rifampicin- PCNB (PARP) at concentrations of 10:250:10:100 μg/ml respectively (4, 6,). Hymexazole was also added to the media (PARPH) to control the growth of *Pythium* spp. Cultures were incubated in the dark at 20-22°C for about 7 days. *Phytophthora* species were isolated from rhizosphere soil using a bating bioassay technique described by Ferguson and Jeffers (5). Several baits (leaf discs of host plants *Pieris, Rhododendron* and intact pine needles) were used simultaneously, to detect a variety of *Phytophthora* species in rhizosphere soil (5). Baits were floated in each soil sample for 72 hours, at 21-23°C and then plated onto PARPH media. A bating bioassay technique was also used to detect *Phytophthora* in water sources used for nursery irrigation, including streams and ponds. Baits (leaf discs, pears and apple fruits) were placed in field environment using a floating device and after 72-hours the baits were retrieved and transported to the laboratory for isolation of *Phytophthora* species. Leaf discs and pieces of fruit baits were plated onto PARPH media and incubated for 7 days in the dark at 21-23°C. Isolates of *Phytophthora* were transferred to fresh PARPH-V8 medium to obtain pure cultures for species characterization and identification. Fungal isolates were observed for morphological characteristics. Based on colony characteristics and fungal morphological features such as chlamydospores, sporangia, and hyphal swellings, isolates were grouped into different morphological types. Each morphological type was farther characterized using DNA analysis of the internal transcribed spacer (ITS) region using universal primer pair ITS1 /ITS4 and standard protocols. The ITS sequences were compared with GenBank information using a BLAST search (2).

Results and Discussion:

The baiting technique using leaf discs on rhizosphere soil was the most successful method for recovering *Phytophthora* in nurseries. Isolates of *Phytophthora* from rhizosphere soil were identified as *P. cinnamommi, P. cryptogea, P. nicotianae* and several isolates were not identified. *Phytophthora* isolated from irrigation water was *P. megasperma,* and some species did not match those in the GeneBank and are unidentified/ unclassified. Direct isolation of Phytophthora from plant tissue including samples from roots and collar region of trees such as dogwood, juniper, white pine, arborvitae, yew and maples was unsuccessful and no *Phytophthora* was detected. However, *Phytophthora citricola* was isolated from dogwood (*Cornus florida*) leaves that had leaf blight symptoms. Since *Phytophthora* was isolated from soil and irrigation water, but not from plant tissues (with one exception), it is possible that sampled plants were not good hosts for the species in the soil or irrigation water. The recovery of *Phytophthora* from plant tissues may have also been affected by the extraordinarily high

temperatures that persisted between July to October 2007, when the survey was conducted.

The detection of *Phytophthora* species in rhizosphere soil and irrigation water indicates that *Phytophthora* is likely to become a problem when susceptible trees are planted and the environment is favorable for disease development. Contaminated water can be a primary source of *Phytophthora* inoculum in nurseries, recycling of irrigation water and water run-off can easily spread Phytophthora in the whole nursery (7). The presence of several *Phytophthora* species in one nursery increases the chance for disease outbreaks in the nursery. The *Phytophthora* species isolated in this study have a wide host range including ornamental plants (8) and pose a potential problem to Tennessee nurseries. Growers should be vigilant toward early detection of Phytophthora problems and avoid planting susceptible plant cultivars. Comprehensive identification of *Phytophthora* species in commercial nurseries will allow growers to make informed decisions on the choice of plants they grow to minimize Phytophthora diseases.

Our previous studies on *Phytophthora* species associated with dogwood leaf blight (9) identified *P. cactorum* based on morphological features (Fig 1). However, current studies on isolates from dogwood foliage blight showed closest DNA sequence similarity with *P. citricola*. Previous reports by Alfierri et al (1) associated twig and foliage blight of dogwood with *P. parasitica* (syn. *P. nicotiana*). The question on whether two *Phytophthora* species may be associated with dogwood leaf blight is currently being investigated. This report represents a very small section of the survey, but five species detected in this study have been previously reported in ornamental plants in the southeastern region (5). The unclassified *Phytophthora* detected in our survey may represent new species.

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Acknowledgements:
The effort of Ms Bethany Wormack in the survey, identification and characterization of the isolates is deeply appreciated. The project has been funded by USDA/CSREES project no:2004-38814-1049 CRIS Number 0201101

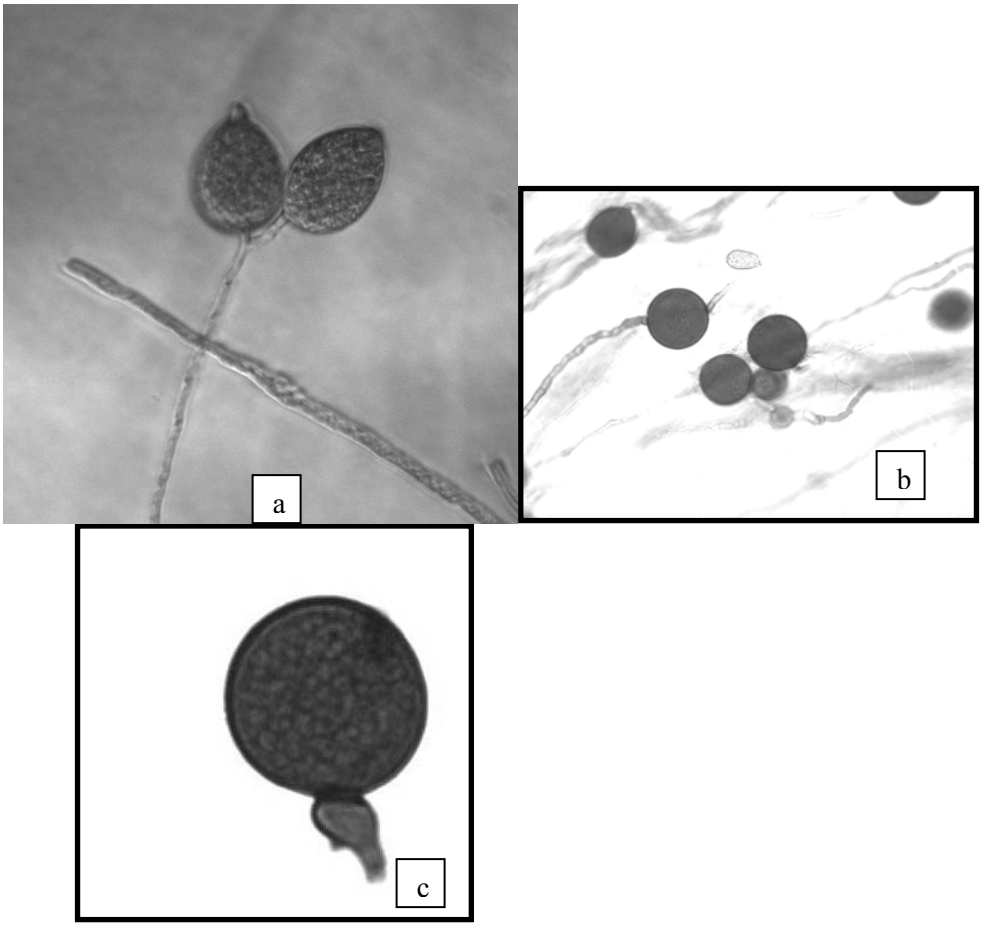

Fig 1. Zoosporangia(a), chlamydospores (b) and paragynous oogonium (c) of *Phytophthora citricola* causal agent of leaf blight disease.

The Effect of *Pythium* and Cold Storage on the Root Growth Potential and Root Collar Diameter of Longleaf Pine (*Pinus palustris*) Seedlings

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Significance to Industry: *Pythium dimorphum* and *Pythium irregulare* are commonly found in nursery soils throughout the southern United States. Standard operational lifting procedures used by nurseries might allow these fungi to infect pine seedlings and be placed in cold storage. These conditions might be favorable for fungal growth. Both *Pythium* species caused longleaf seedlings to lose RCD. Decreases in RCD could result in reduced quality and a potential increase in pine seedling mortality.

Nature of Work: Forest tree nursery managers in the southern United States routinely cold store (≈ 1-4°C) pine seedlings for days, weeks, or even months after they are lifted from nursery beds. Bareroot pine seedlings lifted and cold stored before mid-December often die after planting if stored for long periods of time (Venator 1984). If bareroot seedlings are lifted in the fall, planting should be done no longer than a few days following lifting. During lifting, roots of bareroot seedlings can be injured and wounded, especially in unfavorable soil conditions (Carlson 1991). Soil-borne pathogens (particularly, *Pythium* species) have been speculated to use wounded roots as pathways for infection, which could reduce seedling viability as storage time increases. *Pythium* spp are generally considered opportunistic pathogens that cause gradual deterioration and death by destroying feeder roots that are important for nutrient absorption (Hendrix and Campbell 1973). Higher percentages of *Pythium* have been isolated from longleaf pine roots with longer durations of storage (Jones et al. 1992). The cold, moist storage conditions might favor *Pythium* growth and the chance for root infection that could lead to poor seedling survival.

Root growth potential (RGP) is a common measurement used to detect the physiological vigor of a seedling based on the amount of new root growth. The presence of new roots gives the seedling the ability to absorb water and nutrients to become established in soil, thereby reducing transplant shock and increasing survival. Root collar diameter (RCD) is one of the best indicators of seedling quality based on seedling morphology. A larger RCD usually indicates a healthier seedling and has been correlated with higher survival and growth after outplanting (South et al. 1985). The objective of this experiment was to determine if exposure to *Pythium* species during cold storage affected bareroot longleaf pine seedling quality based on root growth potential and root collar diameter measurements. Understanding the interaction between *Pythium* presence on roots during cold storage would be helpful for nurseries so they can continue to produce high quality pine seedlings.

Methods: Three 0.5 cm disks of *Pythium dimorphum* and *Pythium irregulare* were transferred from stock cultures to oatmeal agar plates to use as the inoculum source. Prior to inoculation, 1,190 g of Quaker oats and 400 ml distilled water were added to two autoclavable bags, mixed thoroughly, and autoclaved for 20 minutes. The sterilized oats were allowed to cool for 24 hrs and one bag of oatmeal received three oatmeal agar plates of *P. dimorphum* and the other bag received three plates of *P. irregulare*. The oatmeal-*Pythium* inoculum was mixed every 12 hrs and stored at room temperature for 11 days prior to longleaf pine root inoculations.

Bareroot longleaf pine seedlings were acquired from a south Alabama nursery on December 7, 2007 and paced in cold storage (≈ 4-5ºC). Root inoculation treatments consisted of 50 g and 200 g of *P. dimorphum* oatmeal inoculum, 50 g and 200 g of *P. irregulare* oatmeal inoculum, and controls dipped in 11 L of water. On January 19, 2008, seedling roots were dipped into a bucket of the water/oatmeal mixture containing the *Pythium* treatment. Thirty seedlings were used for each treatment and placed in separate plastic bags. The bucket was emptied, rinsed, and filled with a fresh mixture of inoculum after each bundle of seedlings was inoculated. Three replications (each with 30 seedlings from each of the five treatments) were randomly placed in cold storage (≈ 4-5ºC). Three weeks after inoculation, seedlings were removed from the cooler and placed in an aerated hydroponic system, which utilized aquariums (38 L) with a cover that allowed each seedling's root system to be suspended in the water. Five aquariums were used for each of the three treatment replications (15 total), set out in a completely randomized design on three greenhouse tables. As seedlings were placed in the aquariums, the RCD (mm) of each seedling was measured. After 60 days in the hydroponic system, RCD was re-measured and the number of new roots > 0.5 cm was counted on each seedling.

Results: Root collar diameter was reduced for all seedlings inoculated with *Pythium* (Table 1). There was no difference in the amount of RCD reduction for either of the *Pythium* species or inoculation levels (Table 2). However, as the level of *Pythium* increased, the effect on RCD increased. In contrast, non-treated seedlings increased in RCD, which was significantly greater than the RCD's of seedlings treated with either *P. dimorphum* or *P. irregulare* (Table 2).

Inoculating longleaf pine seedlings with *Pythium* did not reduce RGP (Table 1). Seedlings treated with *P. irregulare* (50 g and 200 g) had more seedlings with new roots, followed by the control and both levels of *P. dimorphum*, respectively. *P. dimorphum* seemed to have the most negative effect on root growth.

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Table 1. Mean root growth potential, root collar diameter (mm) before and after exposure to the hydroponic system, and difference in root collar diameter growth between Pythium inoculation treatments.

Treatment	Rep	New roots >0.5 cm	RCD before	RCD after	Difference in RCD growth
Control	1	8	9.68	9.68	0.00
	2	44	9.98	10.31	0.33
	3	5	10.15	10.39	0.24
	Mean	20	9.94	10.12	0.19
P. dimorphum 50	1	12	9.01	8.49	-0.52
	2	21	9.81	9.81	0.00
	3	3	9.01	9.02	0.01
	Mean	13	9.28	9.11	-0.17
P. dimorphum 200	1	17	10.13	9.49	-0.65
	2	14	9.35	9.10	-0.25
	3	9	9.72	9.76	0.04
	Mean	14	9.74	9.45	-0.29
P. irregulare 50	1	19	8.51	8.14	-0.37
	2	35	8.27	8.30	0.04
	3	35	8.12	8.45	0.31
	Mean	30	8.30	8.29	-0.01
P. irregulare 200	1	11	8.61	8.31	-0.30
	2	16	8.28	7.95	-0.33
	3	29	9.66	9.53	-0.13
	Mean	19	8.85	8.60	-0.25

Table 2. Probability (p-value) results testing for significance differences in mean root growth potential and root collar diameter among *Pythium* treatments. Values less than 0.05 are significantly different.

Contrast	New roots	RCD before	RCD after	RCD difference
Control vs *P. dimorphum*	20 vs 13 (0.4488)	9.94 vs 9.51 (0.2425)	10.12 vs 9.28 (0.0400)	0.19 vs -0.23 (0.0042)
Control vs *P. irregulare*	20 vs 24 (0.5646)	9.94 vs 8.57 (0.0040)	10.12 vs 8.45 (0.0013)	0.19 vs -0.13 (0.0169)
P. dimorphum vs *P. irregulare*	13 vs 24 (0.1254)	9.51 vs 8.57 (0.0102)	9.28 vs 8.45 (0.0184)	-0.23 vs -0.13 (0.2831)

Effects of Shading on Cercospora Leaf Spot in Bigleaf Hydrangea

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Index Words: *Cercospora hydrangeae*, disease resistance

Significance to Industry: Shading densities significantly affected disease severities of Cercospora leaf spot on bigleaf hydrangeas. In general, lower disease severities were associated with higher shading densities. However, significantly differences in disease severities among cultivars could not be detected in higher shading densities with 90% and 60% shade. These results suggest that planting bigleaf hydrangeas under shade could be an effective component in integrated management of the disease. Screening of bigleaf cultivars for leaf spot resistance needs to be conducted under full-sun or low shading density to avoid false conclusions.

Nature of Work: Cercospora leaf spot, caused by *Cercospora hydrangeae* Ellis & Everh., is a common disease of bigleaf hydrangea (*Hydrangea macrophylla* (Thunb.) Ser.) in ornamental nurseries and gardens in late summer and fall (2, 3). Some degree of shade is favorable to the growth of bigleaf hydrangeas although they grow normally in full sun (3). While the effects of light on development of diseases caused by *Cercospora* species have been studied in coffee, sugar beet and banana (1), the effect of shading on leaf spot of hydrangea is unclear. In order to develop screening methods for disease resistance and integrated disease management, effects of shading densities on disease development of *Cercospora* leaf spot were investigated in six bigleaf hydrangea cultivars.

A field experiment was conducted at the Plateau Research and Education Center at Crossville, TN. The experiment was established as a split plot design, with four shading treatments, 0% (full sun), 30%, 60% and 90% shade, and six bigleaf hydrangea cultivars, ‘Blue Deckle’, ‘Fasan’, ‘Lilacina’, ‘Miranda’, ‘Pretty Maiden’ and ‘Sister Theresa’. The cultivar factor was treated as subplots within each of four main plots of shading treatments that were randomized in complete blocks. Each plot was 24 ft long and 6 ft wide. Plots were spaced sufficiently so that shading from one plot would not affect light intensity of another plot. Two year-old plants of six bigleaf hydrangea cultivars were purchased from Bell Family Nursery, Aurora, OR and transplanted in plots with two plants for each cultivar in each plot in May, 2007. Plants were mulched

and irrigated using drip irrigation. Disease indices of leaf spot were assessed on each plant using a 0-5 index scale (0: healthy; 1: less than 5% of leaves with lesions; 2: 5-10% leaves with lesions; 3: 11-30% leaves with lesions; 4: 31-60% of leaves with lesions; 5: over 60% leaves with lesions) on September 17, 2007. Data were analyzed using a split plot design as a randomized complete block design.

Results and Discussion: Shading treatments significantly (P = 0.0003) affected disease severity of Cercospora leaf spot on bigleaf hydrangeas. In general, increasing shade densities decreased disease severity of leaf spot. In the 60% (P = 0.2828) and 90% (P = 0.4348) shade treatments, disease indices were not significantly different among cultivars. However, significant differences in disease indices were detected among cultivars in 30% shade (P = 0.0053) and full-sun (P = 0.0001) treatments. 'Fasan', 'Blue Deckle' and 'Lilacina' had significantly lower disease severities than 'Miranda' and 'Pretty Maiden', whereas 'Sister Theresa' was intermediate in both 30 and 0% shade.

Shaded environments favor growth of bigleaf hydrangea (3). In this study, hydrangea plants grew more vigorously under higher shade density than under less shade (data not shown). Disease severity in full-sun and a low degree of shading could be associated with stress associated with less vigorously growing plants in high light intensity conditions. Lower disease severities under higher shade densities suggest that evaluating disease resistance or selecting resistant cultivars should be performed in full sun to low shade environments. The present results provide knowledge in the disease management and resistance screening for hydrangea leaf spot.

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Impact of nitrogen application rate on diseases and growth of crapemyrtle

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Index Words: *Lagerstroemia indica* 'Carolina Beauty', Heritage 50W.

Significance to Industry: While powdery mildew and Cercospora leaf spot often have a detrimental impact on crapemyrtle aesthetics, nothing is know on the influence of nitrogen rate on the severity of these diseases in a production nursery or landscape setting. From 2002 until 2005, nitrogen in the form of ammonium nitrate was annually applied to 'Carolina Beauty' crapemyrtle at 18.8, 37.5, 75, 150, 300, and 600 pounds of actual nitrogen per acre**.** One of two trees in each plot was treated with the fungicide Heritage 50W. Data for disease intensity and tree dimensions were collected in 2003, 2004, and 2005. Intensity of Cercospora leaf spot declined with increasing N rates in 2004 and 2005 on the non-fungicide treated and in all three years on the fungicide-treated trees. Enhanced leaf retention and fall color in late October to early November was noticeable on the non-fungicide treated trees maintained at the higher compared with the lower N rates. Heritage 50W suppressed Cercospora leaf spot development regardless of N rate, which resulted in superb fall color display on the fungicide-treated trees. Despite considerable premature defoliation, Cercospora leaf spot did not have a detrimental impact on crapemyrtle growth. Since the occurrence of powdery mildew was very sporadic, no relationship between N rate and the incidence of this disease was established. Raising N rates above the recommended 44 to 130 lb N/A for landscape plantings should result in a reduction in the intensity of Cercospora leaf spot.

Nature of Work: Crapemyrtle are among the premier flowering trees for southern landscapes. While powdery mildew (caused by *Erysiphe lagerstroemia*) is often considered the most common and damaging disease on crapemyrtle, heavy leaf shed associated with the disease Cercospora leaf spot (caused by *Cercospora lythracearum*) has been shown to greatly detract from the fall color display of crapemyrtle (6). Influence of nitrogen (N) fertilization rate on the development of diseases of shrubs and trees has not been extensively studied. Increased N rates has been associated with a reduction in the severity of anthracnose on walnut (9), Cercospora leaf spot on flowering dogwood (5), and several fungal and bacterial-incited diseases of tropical herbaceous and woody plants (2). Also, the influence on N rate on the growth of crapemyrtle in a landscape setting is not well documented. Currently, N rate recommendations for landscape trees are 44 to 130 lb N/A and 87 to 174 lb/A for quick- and slow-release forms of nitrogen (12). For field grown nursery stock, optimum annual N rate is 250 lb N/A (8). Influence of N rate on the development of powdery mildew and Cercospora leaf spot on the disease susceptible crapemyrtle cultivar 'Carolina Beauty' was studies in a simulated landscape setting. The relationship between the above diseases and tree growth was also evaluated.

Crapemyrtle 'Carolina Beauty' (*Lagerstroemia indica*) was transplanted from #3 containers into a Benndale (A) fine sandy loam (< 1% organic matter) at the Brewton Agricultural Research Unit in Brewton, AL (USDA Plant Hardiness Zone 8a) on February 5, 2002. An 8 ft square (64 ft^2) area around each tree was mulched with one inch of aged pine bark. Trees were pruned each in January of each study year. Separate applications of 3.6 oz of murate of potash (0-0-60 K_2O) and super-phosphate (0-46-0 P_20_4) were made over the mulched area on March 3, 2003 and March 9, 2004. Directed applications of 1 lb/A of Gallery® DF (isoxaben, Dow AgroSciences LLC, Indianapolis, IN) plus 2 qt/A of Surflan® T/O (oryzalin, United Phosphorus, Trenton, NJ) were made on March 5, 2003, November 3, 2003, April 22, 2004, and March 18, 2005 for pre-emergent weed control. Hand weeding and spot applications of Finale® 1E (glufosinate-ammonium, Bayer Environmental Science, Kansas City, MO) at 2 fl oz/gal were used to control escaped weeds. Centipedegrass alleys that separated each tree row was periodically mowed but not fertilized during the study period. A drip irrigation system with a single emitter per tree was installed and the trees were watered as needed.

A split plot design with 72 plants in 6 replications with nitrogen (N) rate as the main plot and fungicide treatment as the sub-plot. Ammonium nitrate (33N-0P_2O_5-0K_2O) was applied at 18.3, 37.5, 75, 150, 300, and 600 pounds of actual N per acre per year. One quarter (25%) each nitrogen rate was evenly distributed over the mulched area at monthly intervals from March to June. Heritage 50W (azoxystrobin, Syngenta Professional Products, Greensboro, NC) at a rate of 4 oz/100 gal of spray volume was applied to one of two 'Carolina Beauty' crapemyrtle, which were planted on 12 ft centers, in each plot. Fungicide applications, which were made with a CO_2-pressurized sprayer, were scheduled at two-week intervals from June 1 to July 10, 2002; May 2 to July 10, 2003; May 5 to July 14, 2004; and May 4 to July 29, 2005.

Leaf spot intensity to Cercospora leaf spot was rated using the 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 diseased and/or prematurely shed leaves. Ratings were recorded on July 16, August 19, September 13, October 2, October 15, October 28, and November 15, 2003; July 29, August 24, September 30, October 27, and November 9, 2004; and June 27, July 27, August 24, October 10, October 25, November 5, and November 16, 2005. Disease ratings were not recorded in 2002.
Tree height and canopy diameter on two axes were recorded on January 14, 2004; January 31, 2005; and January 12, 2006. The growth index (GI) was calculated using the following formula: GI = (height + width 1 + width 2)/3. Within one to two weeks after dimensions were recorded, each tree was pruned.

In each year, area under the disease progress curve was calculated for Cercospora leaf spot intensity (AUDPCI) (11). The AUDPCI values were subjected to analysis of variance (ANOVA) according to a split plot design arrangement of treatments using the GLM procedure in SAS (10). The main factor in this arrangement was N rate and the split plot was the fungicide treatment. Significance of interactions was first evaluated. Means were separated using Fisher's protected least significant difference at P=0.05.

Results and Discussion: While Cercospora leaf spot intensity (AUDPCI) for the non-fungicide treated trees were similar for all N rates in 2003, AUDPCI values for the fungicide-treated crapemyrtle were lower at 600 lb N/A than 150 lb N/A (Table 1). In contrast to 2003, AUDPCI values for the non-fungicide treated crapemyrtle in 2004 declined as N rate increased (Table 1). On the non-fungicide treated trees, AUDPCI values at 300 and 600 lb N/A were lower than at 18.8 lb N/A, while values at 37.5, 75, and 150 lb N/A were intermediate. For the fungicide-treated trees, AUDPCI values, which were higher at 18.8 than 600 lb N/A, were intermediate at the remaining N rates. In 2005, Cercospora leaf spot intensity on the non-fungicide treated trees declined with increasing N rates (Table 1). For the non-fungicide treated crapemyrtle, AUPDCI values were higher at 18.8 lb N/A than at 300 and 600 lb N/A. Intermediate N rates had AUDPCI values that were similar to those for the highest and lowest N rates. On the fungicide-treated crapemyrtle in 2005, N rate had a significant impact on Cercospora leaf spot intensity (Table 1). The AUDPCI values were lower at the two higher than two lower N rates but were similar at the four higher N rates.

Occurrence of powdery mildew was sporadic during the study period. While colonization of the buds and flower blooms was occasionally seen, disease signs were not found on the leaves.

Generally, Cercospora leaf spot intensity on crapemyrtle declined as N rates increased. Previously, increasing N rates have been associated with a reduction in the incidence of Cercospora leaf spot on flowering dogwood (5), walnut anthracnose (9) as well as Alternaria and bacteria-incited diseases of tropical herbaceous and woody foliage plants (2). For non-fungicide treated crapemyrtle, N rate was inversely proportional to disease intensity (AUDPCI) in two of three years. When N rate impacted Cercospora leaf spot development on the non-fungicide treated trees, AUDPCI values, which usually were highest at 18.8 lb N/A, steadily declined as N rate increased to 600 lb N/A. By late October and November, enhanced leaf retention on the non-fungicide treated trees at the higher than lower N rates was noticeable, particularly in 2005 at the three highest compared with three lowest N rates. In addition, a similar decline in AUDPCI values for Cercospora leaf spot with rising N rates was seen in all three years on the fungicide-treated trees.

While the treatment schedule for Heritage 50W was originally designed to control powdery mildew, Cercospora leaf spot-related leaf spotting and premature leaf shed was suppressed into November (Table 2). Overall, Heritage 50W reduced Cercospora leaf spot intensity by 51%, 49%, and 60% in 2003, 2004, and 2005, respectively. Despite sizable reductions in the level of disease-related leaf spotting and premature leaf loss obtained with Heritage 50W, tree growth as defined by the annual Growth Index (GI) for the non-fungicide and fungicide treated trees was similar in 2004, 2005, and 2006 (Table 3). Although the control of Cercospora leaf spot with Heritage 50W did not result in an increase in tree growth, the fungicide treated trees had a denser leaf canopy and vivid orange-yellow fall color compared with the non-fungicide treated crapemyrtle.

While Heritage 50W applications ceased at the time of Cercospora leaf spot onset in mid- to late July, this fungicide delayed disease development until late October to mid-November. Previously, Hagan and Akridge (4) noted that Heritage 50W gave effective control of Cercospora leaf spot on field-grown 'Wonderful White' crapemyrtle. Despite the superior fall color display on the fungicide-treated crapemyrtle, Cercospora leaf spot control was not reflected in tree growth. In all three years, GI for the non-treated and fungicide-treated trees was similar. In contrast, damaging outbreaks of Cercospora leaf spot on flowering dogwood were correlated with reduced tree height and trunk diameter (5). Increased growth and flower bud counts are associated with control of black spot and Cercospora leaf spot on rose (1,7).

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Table 1. Influence of N rate on the intensity of Cercospora leaf spot on crapemyrtle.

Nitrogen rate	AUDPCI[z]					
	Non-Fungicide Treated Trees			Fungicide Treated Trees[x]		
lb/A	2003	2004	2005	2003	2004	2005
18.8	590	715[w]	442	242	393	306
37.5	529	704	435	240	355	288
75.0	549	685	405	244	337	271
150.0	515	674	397	286	334	256
300.0	605	641	389	254	332	253
600.0	546	638	388	230	319	243
LSD (P=0.05)	NS	68	49	52	77	33

[z]Season-long Cercospora leaf spot intensity is represented by AUDPCI.
[x]Treated with Heritage 50W fungicide.
[w]Mean separation was according to Fisher's protected least significant difference (LSD) test P=0.05.

Table 2. Suppression of Cercospora leaf spot on crapemyrtle with the fungicide Heritage 50W.

	AUDPCI Values[z]		
Treatment	2003	2004	2005
Non-fungicide treated	556 a[y]	676 a	409 a
Fungicide treated[x]	250 b	344 b	164 b
LSD (P=0.05)	28	35	35

[z]Season-long Cercospora leaf spot intensity is represented by AUDPCI.
[y]Mean separation was according to Fisher's protected least significant difference (LSD) test P=0.05.
[x]Heritage 50W fungicide was applied at approximately 2-week intervals between May 2 to July 10, 2003; May 5 to July 14, 2004; and May 4 to July 29, 2005.

Table 3. Impact of Heritage 50W fungicide on the growth of 'Carolina Beauty' crapemyrtle.

	Growth Index (GI)[z]		
Treatment	2004	2005	2006
Non- fungicide treated	172[y]	196	202
Fungicide treated	167	197	200
LSD (P=0.05)	NS	NS	NS

[z]Growth Index = (height + width 1 + width 2)/3.
[y]Mean separation was according to Fisher's protected least significant difference (LSD) test P=0.05.

Drench and foliar fungicides for Entomosporium leaf spot control on photinia

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Index Words: *Photinia* x *fraseri* 'Birmingham', 'All-In-One', Bayer Disease Control, Daconil Ultrex, Immunox, Ortho Rose Pride, tebuonazole, chlorothalonil, myclobutanil, triforine.

Significance to Industry: Typically, weekly to bimonthy foliar applications of a fungicide are needed to control diseases in landscape plantings of herbaceous and woody plants. Bayer Advanced All-In-One, which contains a soluble N-P-K source, the insecticide imidaclorprid (Merit®), and fungicide tebuconazole, is formulated to be applied as drench treatment at 6-week intervals to the root zone individual plants to control leaf spot and blight diseases. Over the two-year study period, All-In-One failed to control of Entomosporium leaf spot on photinia. Bi-monthly applications of the retail fungicide formulations Bayer Advanced Disease Control, which contains the same tebuconazole a.i. as All-In-One, as well as Spectracide Immunox, and Ortho Rose Pride proved equally effective in controlling this disease as the commercial standard Bravo Ultrex.

Nature of Work: Entomosporium leaf spot, which is caused by the fungus *Entomosporium mespili*, remains a common and damaging disease in landscape plantings of red tip photinia (*Photinia* x *fraseri* 'Birmingham') across the South. Indian hawthorn, flowering pear, loquat, and other photinia species such as *P. serrulata* and *P. glabra* are among the other common targets of this disease (3). Indian hawthorn but not photinia selections with a high level of leaf spot resistance have been released (6). Protective fungicide treatments are not only required to produce but also maintain the health and beauty of photinia in landscape plantings. Effective control of Entomosporium leaf spot can be maintained with weekly to bimonthly foliar applications of fungicides such as Zyban (thiophanate-methyl + mancozeb), Daconil Weather Stik 6F (chlorothalonil), and Eagle 40W (myclobutanil) on photinia (1,2). In a simulated landscape planting of Indian hawthorn, Daconil Weather Stik gave better Entomosporium leaf spot control when applied on 2 than at 4-week intervals (7). While a nursery has the personnel and equipment required to maintain a preventative foliar fungicide program required for disease control, homeowners are looking for a less time-consuming means of controlling this and other shrub diseases. The retail product 'All-In-One', which is distributed through retail outlets by Bayer Advanced, contains the systemic triazole fungicide tebuconazole and is formulated to be applied as a soil drench rather than a foliar spray for the control of leaf spots and blights of flowers, shrubs, and trees. Previously, foliar applications of this fungicide have been shown to control black spot on rose and Entomosporium leaf spot on photinia (1). Soil drenches of a systemic fungicide are a possible alternative to foliar-applied fungicides for

controlling leaf spots and blights. When applied as a soil drench to established roses, tebuconazole gave good control of black spot (4). However, foliar applications of Daconil Ultrex later proved far superior to most rates of tebuconazole for black spot control on rose (5). At rates where black spot control was obtained, noticeable leaf yellowing and shortening of the shoot internodes of tebuconazole-treated roses was seen (Hagan, personal observation). Performance of drench treatments of 'All-In-One' against Entomosporium leaf spot of photinia has not been determined. The objective of this study was to compare the efficacy of drench treatments of 'All-In-One' with bimonthly foliar applications of the home retail fungicide products Bayer Advanced Disease Control (tebuconazole), Spectracide Immunox (myclobutanil), and Ortho Rose Pride (triforine) as well as the commercial standard Bravo Ultrex for the control of Entomosporium leaf spot in a simulated landscape planting of red-tip photinia

In February 2004, red-tip photinia in #1 containers were transplanted into a Benndale fine sandy loam on 6 foot centers with 10 feet between rows. A drip irrigation system was installed at planting and the plants were watered as needed. Beds were mulched with aged pine bark. Plants were pruned on in January 2006 and 2007. In February 2006 and 2007, pre-emergent weed control was obtained with a tank mixture of 2 qt/A of Surflan + 1.0 lb/A of Gallery. Escape weeds were removed by hand or with a hoe. Foliar fungicide applications were made at 2-week intervals from 4 January to 5 July 2006 and 12 January to 11 July 2007 to drip with a CO2-pressurized sprayer. Monthly drench treatments of approximately 1 qt of a suspension of 'All-In-One' were scheduled from 4 January to 5 July 2006 and 12 January to 27 June 2007. Entomosporium leaf spot intensity was rated on 17 May 2006 and 16 May 2007 using a modified 1 to 10 Florida peanut leaf spot rating scale where 1 = no disease, 2 = very few lesions in canopy, 3 = few lesions noticed in lower and upper canopy, 4 = some leaf spotting and $\leq$ 10% defoliation, 5 = lesions noticeable and $\leq$ 25% defoliation, 6 = lesions numerous and $\leq$ 50% defoliation, 7 = lesions very numerous and $\leq$ 75% defoliation, 8 = numerous lesions on few remaining leaves and $\leq$90% defoliation, 9 = very few remaining leaves covered with lesions and $\leq$ 95% defoliation, and 10 = plants defoliated. Significance of treatment effects were tested by analysis of variance and Fisher's protected least significant difference (LDS) test (P=0.05).

Results and Discussion: While average temperatures in 2006 were above the seasonal norm for June and July, rainfall totals for April, May, and June were well below the 30-year average for the study location. The pattern of above average temperatures and below average rainfall in the spring and early summer was repeated in 2007.

Dry late winter and spring weather patterns may have slowed disease development on photinia. As indicated by a disease rating of 5.0, the non-treated photinia suffered moderate leaf spotting in the lower and mid-canopy as well as approximately 25% premature defoliation (Table 1). When compared with the non-treated control on the 17 May 2006 rating date, all fungicides except for the All-In-One monthly drench treatment greatly reduced the severity of Entomosporium leaf spot. In addition, the All-In-One drench failed to give the same level of disease control as was provided by foliar applications of Daconil Ultrex, Bayer Disease Control, Immunox, and Ortho Rose Pride,

which gave similar control of Entomosporium leaf spot on photinia. Daconil Utrex and Bayer Disease Control-treated photinia were free of leaf spot symptoms.

While late winter and early weather patterns did not greatly differ, symptom severity on the non-treated controls, which suffered heavy leaf spotting and considerable premature defoliation, was higher in 2007 than in the previous year. As was noted in the previous year, disease ratings for the All-In-One-treated photinia were identical to those recorded for the non-treated control (Table 1). In contrast, all of the foliar-applied fungicides proved equally effective in controlling Entomosporium leaf spot on photinia. Again, no symptoms were seen on the Daconil Ultrex and Bayer Disease Control-treated photinia. Only a few spotted leaves were found on the photinia treated with Immunox and Ortho Rose Pride.

While the All-In-One drench product is offered to residential consumers as a simpler but more expensive alternative for controlling leaf spot and blight diseases on annuals, perennials, shrubs and trees in the home landscape, the more time consuming foliar fungicide applications gave proved to be the more effective method of controlling Entomosporium leaf spot on photinia. In two previous trials on field-grown roses (4,5), All-In-One failed to provide any protection from black spot. The fungicide tebuconazole found in All-In-One, when applied as a drench at a high enough rate, gave good black spot control on a hybrid tea rose in one (4) but not a second (5) study. Foliar applications of the same tebuconazole active ingredient as Bayer Disease Control gave superb Entomosporium leaf spot control in this study. Apparently, the tebuconazole concentration or application rate of the All-In-One drench product is insufficient for effective control of a leaf spot disease on photinia. In contrast, fungicides such as Bayer Disease Control, Immunox, and Ortho Rose Disease Control that are marketed for use in residential landscapes proved equally effective in controlling Entomosporium leaf spot on photinia as the commercial fungicide standard Dacnoil Ultrex.

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Table 1. Comparison of soil- and foliar-applied retail and commercial fungicides for the control of Entomosporium leaf spot on red-tip photinia in 2006 and 2007.

	Application		Disease Rating*	
Treatment and Rate/gal	Placement	Interval	2006	2007
Daconil Ultrex 0.2 oz	Foliar Spray	2 wk	1.0**	1.0
Bayer All-In-One 3.6 fl oz	Drench	4 wk	3.7	7.0
Bayer Disease Control 0.75 fl oz	Foliar Spray	2 wk	1.2	1.0
Immunox 1 fl oz	Foliar Spray	2 wk	1.3	1.2
Ortho Rose Pride 0.5 fl oz	Foliar Spray	2 wk	1.8	2.3
Untreated Control	---	---	5.0	7.0
LSD (P=0.05)	--	--	1.5	1.6

*Entomosporium leaf spot severity was rated on 17 May 2006 and 16 May 2007 using a modified 1 to 10 Florida peanut leaf spot scoring system.
**Means separation was according to Fisher's protected least significant difference (LSD) test (P=0.05).