

SECTION 5 PATHOLOGY AND NEMATOTOLOGY

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Fine Population Structure of *Discula destructiva*

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Nature of work: Dogwoods are prized ornamental trees native to North American forests. Two species, flowering (*Cornus florida* L.), and Pacific (*C. nuttalli* Audubon) dogwoods, have been recently the major subject of a devastating disease outbreak that has destroyed native dogwood stands throughout the eastern and western forests (6). Dogwood anthracnose, initially described as dogwood decline or lower limb dieback, causes dark leaf spots, blight of leaves and branches, and limb and trunk cankers. Lesions ultimately result in the death of the tree. Since original reports of the disease in Pacific (Washington, 1976) and flowering (New York, 1978) dogwoods, dogwood anthracnose has spread in the western coastal areas and Appalachian range, respectively. The sudden appearance and rapid dissemination of the disease has led to the hypothesis that the causal organism is an introduced pathogen (8,10).

Though infection was originally attributed to *Colletotrichum gloeosporoides* Penz. & Sacc. and *Gloeosporium corni* Green, the anthracnose-causing pathogen was later described as *Discula destructiva* Redlin (9), and in few cases as an undescribed *Discula* species. While both species of these filamentous *Discula* fungi were isolated from diseased flowering, Pacific, and Chinese (*C. kousa* Hance) dogwoods, a perfect stage has yet to be identified. The two species could be distinguished by growth habit on agar media and the ability to oxidize gallic acid (11), and at the genetic level by DNA amplification fingerprinting (DAF) analysis (10). DAF showed that *D. destructiva* was restricted in its genetic diversity, suggesting that the highly homogeneous nature of the pathogen results from either founder effects or genetic bottlenecks maintained by clonal reproduction. In contrast, *D. umbrinella* (Berk. & Br.) Morelet, a related European species considered an endophyte of beech (*Fagus sylvatica* L.), oak (*Quercus robur* L.) and chestnut (*Castanea sativa* Mill.), showed considerable genetic variation (7).

In this study, asexually reproducing fungal populations of the two *Discula* species were fingerprinted using a powerful nucleic acid scanning strategy capable of distinguishing closely related organisms. The fingerprinting approach is based on the generation of arbitrary signatures from amplification profiles (ASAP) (4), a dual-step amplification procedure that generates "fingerprints" from DAF amplification profiles using either conventional (5) or mini-hairpin oligonucleotide (3) primers. The objective was to analyze the population genetic structure of *D. destructiva* in North America and evaluate the origin and spread of the disease on west and east coasts.

A total 51 fungal isolates were collected from diseased symptomatic tissue from trees sampled in a wide variety of geographical locales in the United States and Canada. Mycelial DNA was extracted (10) and profiled by DAF analysis (3,5). In some cases, equal amounts of mycelia from a group of different isolates were bulked and DNA subsequently extracted. DAF reactions were assembled in a total volume of 20 μl , and amplified for 35 cycles of 10 s at 96°C and 10 s at 30°C in a oven thermocycler (3,5). ASAP reactions were assembled as DAF cocktails with mini-hairpin decamer primers (sequence: GCGAAGC-NNN; where N corresponds to any of the four DNA bases) at concentrations of 9 μM and template levels of about 0.1 $\text{ng}\mu\text{l}^{-1}$. Amplification products were separated in 10%T:2%C polyacrylamide-7M urea slab minigels (2) and stained with silver (1), and gels preserved by drying at room temperature. Bands (50-700 bp in length) were scored as present [1] or absent [0], and genetic relationships evaluated by phylogenetic analysis using parsimony (PAUP), and principal coordinate analysis (PCO) and unweighted pair group cluster analysis using arithmetic means (UPGMA), using Jaccard similarity coefficients and the NTSYS-pc program.

Results and Discussion: A novel scanning strategy for nucleic acid analysis capable of detecting increased levels of polymorphic DNA was used to examine the structure of the highly homogeneous dogwood anthracnose-causing *Discula* population. ASAP directed by primers containing a stable mini-hairpin structure at their 5' termini detected polymorphic DNA at levels 14-fold higher than DAF. Most mini-hairpin primers produced highly polymorphic patterns from monomorphic DAF sequences, enabling a study of fine population structure of the dogwood pathogen in the North American continent.

ASAP analysis was coupled with bulking of isolates representative of geographically-defined fungal subpopulations. The experimental strategy simplifies study of any large group of individuals, especially if they represent a highly uniform population at the genetic level (10), but provides more conservative estimates of diversity. ASAP differentiated a northwestern fungal subpopulation from northeastern and southeastern counterparts (Fig. 1). Furthermore, individual analysis of eastern isolates showed a reduction of diversity following the migration of the disease from coastal northeastern regions to the Appalachian mountains. This trend matched the historical dissemination of the disease through native flowering dogwood populations. Results showed that *D. destructiva* appears to have had separate centers of origin in west and east coasts of the North American continent, as subpopulations differed in genetic constitution and diversity. Introduction was probably independent from each other, and was influenced by regional biotic and abiotic factors and by a differential adaptation of the pathogen to Pacific and flowering dogwoods.

The study of a group of isolates from an orchard in Massachussets and from several locations in the Great Smoky Mtn. Natl. Park, showed that two Tennessee isolates were distinct from the very homogeneous Appalachian isolates examined. These observations suggest that natural selection and subsequent genetic drift within (disjunct) geographical pockets could result in the establishment of isolates with variant genetic constitutions, perhaps explaining the existence of a distinct and disjunct subpopulation selected to represent a southeastern boundary of the disease in 1994 (Fig. 1).

Significance to Industry: Initial DAF analysis established at the genetic level that *D. destructiva* is highly uniform throughout the range of the disease, and that the pathogen probably represents part of a single fungal population (10). This fungal homogeneity is consistent with the hypothesis that *D. destructiva* is an imported pathogen. However, ASAP analysis managed to dissect the homogeneous pathogen population in at least two clear subpopulations, suggesting the pathogen was separately introduced in both coasts of North America, perhaps on nursery stock. Obviously, dogwood anthracnose resulted from either: 1) migration of diverse fungal populations that were then severely reduced in genetic diversity by founder effects and genetic drift, or 2) introduction of few founder individuals that adapted and maintained their "clonal" characteristics. Our observations also suggest an insignificant role of nursery exchanges in fungal dissemination between coasts, at least during the initial establishment and progress of the disease. However, the nursery industry can still have great impact on the composition of the present and future fungal subpopulations. Despite phytosanitary inspection, the disease is difficult to detect and plant materials are presently being exchanged freely between coasts. Overall, our studies will help elect suitable control strategies for dogwood anthracnose, such as the management of environmental and cultural practices by the industry.

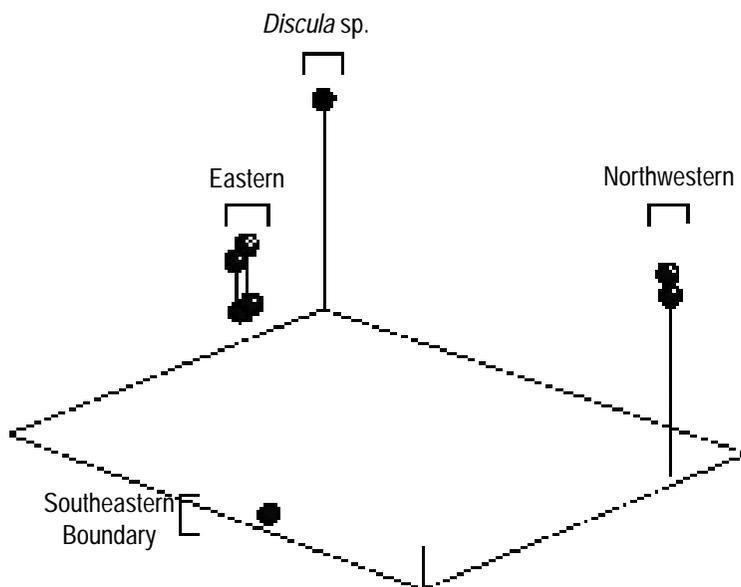
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Figure 1: Principal Coordinate Analysis (PCO) of ASAP fingerprints from bulked fungal samples representing eastern and western subpopulations of *D. destructiva*. DAF profiles originally generated with the octamer GTAACGCC were fingerprinted using 9 mini-hairpin decamers.



Canker and Shoot Blight Diseases of Leyland Cypress

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Nature of Work: Leyland cypress (*X Cupressocyparis leylandii*) is becoming a popular landscape plant in Tennessee. It is most often used in groupings or as screens as it is fast growing. While Leyland cypress has been relatively disease free in landscape plantings in Tennessee to date, canker diseases have been very damaging in other parts of the United States and in other countries (1,4,5,6).

Results and Discussion: During the spring and early summer of 1996, the University of Tennessee's Plant and Pest Diagnostic Center received sixteen samples of Leyland cypress exhibiting shoot dieback and/or resinous cankers. The diseases identified include: Seiridium canker (six samples), Botryosphaeria canker (three samples), Kabatina blight (six samples) and Sphaeropsis tip blight (one sample).

Seiridium canker was characterized by twig and branch dieback and sunken cankers on main stems. Although *S. cardinale* has been the most common species associated with cankers on cypress (4,5,6) only *S. unicolorne* was consistently observed in cankered tissue. This represents the first report of *S. unicolorne* on Leyland cypress in Tennessee. Environmental stress may predispose trees to infection. This disease has been associated with prolonged subfreezing temperatures (5). Also, Monterey cypress in Southern California was more likely to be infected with Seiridium canker when exposed to periods of high temperature with little rainfall (4). Fungicides such as chlorothalonil and thiophanate-methyl have been used successfully to protect artificially inoculated trees from Seiridium canker (2). However, this does not appear to be a practical management practice for landscape plantings as fungicide sprays would have to be applied indefinitely throughout the growing season. Infected shoots and branches should be pruned out and severely infected plants removed. Transmission is most likely through splash dispersal, air-borne spores and possibly infected nursery stock.

Kabatina blight has been reported to cause a tip dieback of one-year-old growth on juniper, arborvitae and cypress (3). *Kabatina juniperi* and *K. thujae* are the most common pathogens associated with this disease. During early spring infected shoots turn yellow-brown and then reddish brown. Small cankers may be observed at the base of the infected shoots. Fruiting bodies of the fungus may be observed in the canker with a 10X hand lens. *Kabatina* spp. apparently are wound pathogens unable to infect healthy plant tissue (3). Wounds made by insects, freeze injury and/or ice damage may serve as entry points for this fungus. Infected shoots should be pruned out. This disease is probably confused with Phomopsis blight of juniper which occurs later in the growing season on new flushes of growth in late spring and early summer.

Botryosphaeria canker is a disease found on a wide variety of plants around the world (3). This disease has a reputation of attacking plants predisposed to infection by drought, freeze injury and wounds. Botryosphaeria canker, caused by *Botryosphaeria dothidea* may be found on Leyland cypress in landscape plantings and nurseries. Symptoms on Leyland cypress include branch dieback and bleeding, resinous cankers. Cankers are often centered on twig or branch stubs. Infected trees may be so disfigured that they need to be removed. Infection most likely takes place during wet, humid weather during late spring and early summer when conidiospores of *Fusicoccum* sp. are produced. Diseased branches should be pruned out. In landscape plantings trees should be irrigated and mulched to prevent drought stress which may predispose trees to infection.

Significance to Industry: This is the first report of Seiridium canker in Tennessee caused by *Seiridium unicorne*. Canker diseases on Leyland cypress appear to be related to cultural and site problems in landscapes and winter injury. Trees that receive little or no irrigation appear to be most at risk.

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Control Strategies for *Phytophthora* Shoot Blight on Bedding Plants

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Nature of Work: *Phytophthora* shoot blight has emerged as a devastating disease of annual vinca or periwinkle (*Cathranthus roseus*). Although substantial plant death has occurred in production greenhouses, the destructive potential of this disease can readily be seen in commercial and residential plantings of annual vinca across Alabama and surrounding states (2). *Phytophthora* shoot blight probably is being introduced into landscape plantings on diseased annual vinca. Repeated disease outbreaks in annual vinca in the same landscape beds over a period of several years indicates that the causal fungus, *Phytophthora parasitica*, survives from year to year in the soil and plant debris. Strategies for controlling *Phytophthora* shoot blight, particularly in landscape plantings, are not well defined. Although Chase (1) reported some differences in the level of *Phytophthora* shoot blight among cultivars of annual vinca, nearly all of the cultivars screened suffered extensive and unsightly damage. The susceptibility of other annuals and perennials to attack by *P. parasitica* is largely unknown. Fungicides have been used successfully to control this disease in container-grown annual vinca (2,3). Best results have been obtained with recommended rates of Subdue 2E drenches or foliar applications of Subdue 2E and Aliette WDG + Fore (2). This study was initiated to identify bedding plants that could be successfully grown in *P. parasitica* infested beds and evaluate the efficacy of fungicides for the preventative control of *Phytophthora* shoot blight on annual vinca.

Prior to bed preparation in 1995, a 5-10-15 fertilizer at a rate of 400 lb/A was incorporated into a Benndale fine sandy loam soil at the Brewton Experiment Field in Escambia Co., AL. Several weeks prior to planting, the beds were fumigated with 250 lb/A of Terr-O-Gas 98 (98% methyl bromide + 2% chloropicrin). Flats of annual vinca cv. "Grape Cooler" were purchased from a garden center. Each plot consisted of four liners planted in two rows on 1 ft. centers. Treatments were randomized within four complete blocks. Plots were fertilized weekly with approximately 200 ppm of a 20-10-20 analysis fertilizer delivered through a drip irrigation system. Beginning on May 5, foliar applications of all fungicide treatments were using a CO₂ pressurized sprayer at the rates and schedules listed in Table 1. The final application date was August 25, 1995. The percentage of blighted shoots and plant survival was determined on August 30.

Beds for the 1996 bedding plant screening trial were prepared in April, 1995 using the procedures outlined above and then planted to annual vinca in May 1995. By late summer, the majority of plants had succumbed to *Phytophthora* shoot blight. The remains of those annual vinca from the 1995 trial were tilled into the soil the following March. Selected bedding plants were established on April 20, 1996. A complete list of the bedding plants included in this trial can be found in Table 2. Individual plots, which consisted of four liners of each bedding plant set in two rows on 1 ft. centers, were randomized in four complete blocks. The beds were mulched with aged pine bark. Plots were fertilized weekly with approximately 200 ppm of a 20-10-20 analysis fertilizer delivered through a drip irrigation system. Percent plant survival was determined on May 29, 1996.

Results and Discussion: From early May through the beginning of August of 1995, weather conditions at the Brewton Experiment Field were generally dry and unseasonably hot. By late July, a very light shoot dieback but no plant death was noted. In August, disease development followed heavy rains associated with Hurricane Erin. Although several treatments significantly improved plant survival as compared with the nonsprayed control, only Aliette 80W applied at a 2-week intervals effectively prevented the death of vinca due to *Phytophthora* shoot blight (Table 1). Also, significantly less shoot dieback was noted on annual vinca treated with Aliette 80W at 2 week intervals than on plants sprayed with the same fungicide at 4-week intervals. Aliette 80W applied every two weeks gave significantly better control of *Phytophthora* shoot blight than ASC66825 500F (fluazinam), Phytan 27, Subdue 2E, Pace 77W, and Daconil 2787 4.17F.

Although April, 1996 was unusually cool and wet, rainfall and temperature patterns in May were near normal. As expected, all the vinca cultivars quickly succumbed to *P. parasitica* (Table 2). By the end of May, few plants remained alive. The use of high rates of ammonium fertilizers may also have predisposed vinca to attack by *P. parasitica*. Also, numbers of surviving impatiens cv. Sun & Shade Red were significantly reduced. Although the survival of some of the other annuals and perennials did fall below 100%, no significant differences in the rate of survival of these plants were observed.

When used as a preventative treatment, Aliette WDG applied as a foliar spray at 2.5 lb of product per 100 gallons of spray volume at a 2-week interval controlled *Phytophthora* shoot blight on annual vinca. To obtain effective disease control, Aliette WDG was applied twice as often as the label specified. Applied at the recommended 4-week interval, Aliette WDG was ineffective. Simone (2) obtained better disease control on pot-grown annual vinca with a tank-mix combination of Aliette WDG plus Fore at 2.5 lb. and 5.0 lb. of product per 100 gallons of spray volume, respectively, than with the 2.5 lb. rate of Aliette WDG alone. Under weather conditions favorable for disease development, the other fungicides screened failed to control *Phytophthora* shoot blight. Subdue 2E also performed poorly against this disease in another trial (2).

Preliminary results indicate that a wide variety of summer annuals and perennials are possible replacements for annual vinca in blight devastated landscape beds. Other than several cultivars of the Sun & Shade impatiens and salvia, all the remaining selections until the end of May have thrived.

Significance to Industry: Aliette WDG gave effective, if rather costly control of *Phytophthora* shoot blight in a simulated landscape planting of annual vinca. Such a fungicide treatment program would, however, be impractical in the vast majority of residential and many commercial landscapes. Use of *Phytophthora*-resistant annuals and perennials offers an economical and effective alternative to an intensive fungicide treatment program. So far, selections of ageratum, begonia, blue daisy, celosia, coneflower, geranium, impatiens, marigold, nigella, petunia, salvia, scabosia, sweet basil, thyme, verbena, and zinnia generally performed well when grown in a soil infested with *P. parasitica*. Additional studies designed to examine the impact of plant selection as well as planting date and nitrogen source on the development and severity of *Phytophthora* shoot blight must be conducted.

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Table 1. Chemical Control of *Phytophthora* Shoot Blight on Annual Vinca.

Fungicide and rate/100 gal.	Spray Interval	Plant Survival	Blighted Shoots
	Wk	%	%
Aliette 80W 2.5 lb	2	100 a ¹	15 a
Aliette 80W 2.5 lb	4	56 abc	51 b
ASC66825 500F 8 fl oz	2	56 abc	60 bc
Phyton 27 40 fl oz	2	38 bc	76 bc
Subdue 2E 1.25 fl oz	4	38 bc	73 bc
Pace 77W 2 lb	4	31 bc	73 bc
Daconil 2787 4.17F	2	6 c	95 c
Untreated Control	2	6 c	96 c

¹Means within a column followed by the same letter are not significantly different.

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Table 2. Survival of Selected Summer Annuals and Perennials in Beds Infested with *Phytophthora parasitica*.

Common Name	Cultivar	Survival ¹ %
Ageratum	Hawaii White	100 a ²
Begonia	Prelude Pink	94 a
Blue Daisy	Spring Merchen	81 ab
Celosia	Prestige Scarlet	100 a
Coneflower	Magnus	100 a
Coneflower	White Swan	94 a
Geranium	New Wave Scarlet	100 a
Geranium	Pinto Red Scarlet	100 a
Geranium	Ringo 2000 Light Salmon	100 a
Geranium	Saturn Formula Mix	100 a
Impatiens	Sun & Shade Coral	88 a
Impatiens	Sun & Shade Lavender	88 a
Impatiens	Sun & Shade Neon Rose	75 ab
Impatiens	Sun & Shade Red	63 b
Impatiens	Sun & Shade Violet	100 a
Marigold	American Indian Orange	100 a
Marigold	Girl Orange	94 a
Nigella	Mulberry Rose	100 a
Petunia	Deep Blue	100 a
Salvia	Firecracker Lilac	100 a
Salvia	Firecracker White	94 a
Salvia	Lady in Red	100 a
Salvia	Firecracker Salmon	100 a
Scabosia	Imperial Mix	100 a
Sweet Basil	Siam Queen	94 a
Thyme	—	94 a
Verbena	Formula Mix	100 a
Vinca	Bikini Formula Mix	6 cd
Vinca	Blush Cooler	19 cd
Vinca	Grape Cooler	25 c
Vinca	Peppermint Cooler	0 d
Zinnia	Crystal White	88 a

¹Stand count taken on May 29, 1996. ²Means within a column followed by the same letter are not significantly different.

Evaluation of Selected Indian Hawthorne Cultivars for Resistance to Entomosporium Leaf Spot

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Nature of Work: Indian hawthorn (*Raphiolepis umbellata*) is an attractive, usually dwarf-type flowering evergreen shrub with a dense, mounded or rounded canopy. The release of new cultivars has sparked renewed interest in indian hawthorn. The production and maintenance of many cultivars of indian hawthorn, however, has often been complicated by the disease Entomosporium leaf spot. This disease, which is caused by the fungus *Entomosporium mespili*, is especially common and destructive in states with humid, wet climates (3,4). An intensive and costly fungicide spray program has often been required to produce and maintain an attractive and vigorous indian hawthorn. Disease resistance offers the best long-term defense against Entomosporium leaf spot in both production nurseries and landscape plantings. Several cultivars of indian hawthorn have previously been identified with good disease resistance while others have proven highly susceptible to Entomosporium leaf spot (1). The objective of this study was to determine the resistance of newly released cultivars of indian hawthorn to Entomosporium leaf spot and compare their reaction to this disease with that of cultivars already in the nursery trade.

In March 1994, twenty one cultivars of indian hawthorn were established in a Benndale fine sandy loam at the Brewton Experiment Field (Zone 8a) on 5 ft. centers with 10 ft. between beds. The cultivars Rosalinda and Snow White were added in March of 1995 and Sea Breeze replaced *R. x delacourii* in April 1996. The experimental design was a randomized complete block with six (6) three-plant replications. Soil fertility and pH were adjusted according to the results of a soil fertility analysis. Twice during the spring, approximately one half cup of Osmocote 17-7-12 fertilizer was uniformly distributed around each plant. Plants were watered as needed using a trickle irrigation system. Hand weeding and directed applications of recommended rates of Roundup herbicide were used to control weeds. The beds were mulched with aged pine bark. Disease was visually assessed on April 5 and May 29, 1996 on a scale of 1 to 5 where 1 = no disease, 2 = 1% to 25%, 3 = 26%-50%, 4 = 51%-75%, and 5 = 76%-100% of the leaves damaged or prematurely lost due to Entomosporium leaf spot. A preliminary report has been published (2).

Results and Discussion: Weather patterns during the winter and early spring of 1996 at the Brewton Experiment Field were unusually cold and wet. Despite the often inclement weather, extensive disease development occurred sometime from late fall through the winter months. A similar pattern of disease development was also noted during the mild, wet winter of the previous year (2).

In early April, typical leaf spot symptoms of *Entomosporium* leaf spot were seen on all cultivars of indian hawthorn (Table 1). On the cultivars Olivia, F1, and Dwarf Yedda, symptoms were limited to a very light leaf spot and very little defoliation. Light to moderate spotting of the leaves and in some cases, a low level of disease-incited defoliation was noted on the cultivars Eleanor Tabor, Majestic Beauty, Snow White, Janice, Jack Evans, Indian Princess, F2, F3 and Clara. Heavy to near total defoliation along with spotting of most of the remaining leaves was observed on the cultivars Rosalinda, Spring Rapture, White Enchantress, Pinkie, Enchantress, Harbinger of Spring, Heather, F6, and Springtime.

In late May, little if any fresh leaf spot symptoms were found on the leaves of the cultivars F1, Dwarf Yedda, Olivia, and Indian Princess. Light to moderate leaf spot development, as indicated by disease ratings of 1.7 to 2.9, occurred on an additional ten cultivars of indian hawthorn. Consistently high disease ratings demonstrate that the cultivars Springtime, F6, Heather, Harbinger of Spring, Enchantress, Pinkie, White Enchantress, and Spring Rapture are very susceptible to *Entomosporium* leaf spot. The disease rating for the newly established cultivar, Sea Breeze, will be reported at a later date.

In the fall of 1995, a rapid decline in the health and vigor in all six blocks of the leaf spot resistant cultivar *R. delacourii*, was seen shortly after very heavy rains associated with hurricane Opal. Although the roots of symptomatic plants were rotted, no plant pathogenic fungi were detected with ELISA or were cultured from plant tissues. Apparently, *R. delacourii* is highly sensitive to- waterlogged or flooded soils and should be established only on raised beds in a well-drained soil. No decline symptoms were observed on any of the remaining cultivars of indian hawthorn.

Of the twenty-two cultivars of indian hawthorn screened in a simulated landscape planting, Dwarf Yedda, Olivia, and Indian Princess have remained relatively disease free over a two year period at a location with a typically humid and often wet Gulf Coast climate. Although some light spotting of a few leaves was seen, disease-related defoliation was minimal and the foliage looked good. These cultivars could be maintained in a landscape and possibly grown in a nursery without protective fungicides. An additional eight cultivars demonstrated low to moderate levels of leaf spot resistance. Of these, the cultivars Clara and Eleanor Tabor over the two-year test period generally suffered the least amount of leaf spot damage. Corley (1) reported levels of resistance for the cultivars Majestic Beauty, Snow White, and *R. delacourii* similar to those obtained in this study. The cultivars Pinkie, Harbinger of Spring, Enchantress, Heather, White Enchantress, Springtime, Spring Rapture, and F6 are highly susceptible to *Entomosporium* leaf spot. The susceptibility of Springtime and Enchantress to *Entomosporium* leaf spot has been previously noted (1). Disease-related defoliation by early spring was severe and most of the remaining leaves on these cultivars were badly spotted. To produce salable container plants and maintain foliage quality in landscape plantings, an intensive and costly program consisting of multiple fungicide applications would be required to protect these cultivars from *Entomosporium* leaf spot. As a result, use of the leaf spot-susceptible cultivars in landscape plantings should be avoided.

Of the named cultivars evaluated, Indian Princess, Clara, Olivia, and Eleanor Tabor had growth habits which were most horticulturally desirable (thick, full plants with glossy green leaves).

Significance to Industry: The market for indian hawthorn has been limited, in part, by the susceptibility of many cultivars to *Entomosporium* leaf spot. The availability of disease resistant cultivars transforms indian hawthorn into a much more desirable, low maintenance woody shrub.

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Table 1. Reaction of Indian Hawthorn to Entomosporium Leaf Spot, 1996.

Cultivar	Disease Rating ¹		Cultivar	Disease Rating ¹	
	Apr 96 ²	May 96		Apr 96 ²	May 96
Springtime	4.9	4.7	F2	2.8	2.6
F6	4.6	3.6	Indian Princess	2.7	1.5
Heather	4.6	4.3	Jack Evans	2.7	2.6
Harbinger of Spring	4.6	4.0	Janice	2.6	2.6
Enchantress	4.5	4.7	Snow White	2.6	2.9
Pinkie	4.4	4.3	Majestic Beauty	2.5	2.7
White Enchantress	4.2	3.9	Eleanor Tabor	2.4	2.4
Spring Rapture	3.8	4.7	F1	2.1	1.7
Rosalinda	3.5	3.4	Olivia	2.1	1.9
Clara	2.9	2.3	Dwarf Yedda	2.0	2.0
F3	2.9	3.4	Sea Breeze ³	N.R.	N.R.
LSD	0.5	0.5		0.5	0.5

¹Disease was visually assessed on a scale of 1 to 5 where 1 = no disease, and 5 = 76% to 100% of the leaves diseased.

²Mean separation within columns according to Fisher's protected least significance (LSD) test (P = 0.05).

³Added to study in April 1996.

Evaluating Disease Resistance and Alternative Control Treatments for Varieties of Rose in the South

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Alabama

Nature of Work: Blackspot, caused by *Diplocarpon rosae* F.A. Wolf, is a fungus that causes black spots on foliage, defoliation, and premature death of roses, especially in the humid south where weather conditions are favorable to the development and spread of the disease (1). Many cultivated rose varieties that fall into the “old garden rose” class have more natural resistance to the blackspot fungus than some of the newer hybrid varieties that are popular today. Previous studies have shown that susceptibility of the blackspot fungus varies with rose species (2). The purpose of this study is to evaluate the performance of specific varieties of rose in the landscape, focusing on the control of blackspot through the use of resistant varieties and alternative chemical treatments and application times.

Plants were established at E.V. Smith Research Station, near Shorter, Alabama, in a randomized complete block design receiving full sun and supplemental water and nutrients through drip irrigation. Eleven different rose cultivars were visually rated weekly for percent disease, percent defoliation, flower production, and overall performance. Three fungal control treatments, plus a control treatment with no spray, were compared. The study included three replications of each treatment.

The treatments in this study included: 1) chlorothalonil applied on a 14-day interval, 2) an application of chlorothalonil around weather events that are conducive to disease development, 3) 1% Sunspray Ultra-Fine Horticultural Oil applied every 14-days, and 4) a no treatment control. Foliar sprays were applied until runoff.

We evaluated the performance of nine different “old garden” roses with two susceptible modern varieties included for comparison purposes. The eleven roses evaluated in this study were: 1) ‘Belinda’s Dream’, 2) ‘Carefree Delight’, 3) ‘The Fairy’, 4) ‘Floral Carpet’, 5) ‘F.J. Grootendorst’, 6) ‘Hansa’, 7) ‘Le Vesuve’, 8) ‘Red Mediland’, 9) ‘Seafoam’, 10) ‘Cary Grant’, and 11) ‘Love Potion’.

Results and Discussion: Preliminary data from May and June of 1996 were analyzed. Differences among treatments were determined using Duncan’s Multiple Range Test. Several significant differences in disease severity were observed among cultivars. After June ratings, ‘Hansa’, an old garden rose, had a significantly lower rating for disease severity than any other cultivar. ‘Carefree Delight’ and ‘Red Mediland’ had the highest disease severity ratings. ‘Red Mediland’ had significantly greater defoliation than other cultivars.

'The Fairy' and 'Hansa', both old garden roses, had the highest overall performance rating with 'Le Vesuve' and 'Cary Grant', a modern hybrid tea, consistently rated low. 'Le Vesuve', a china rose, is apparently more cold sensitive. A late cold snap after budbreak resulted in the loss of several of these roses and severe dieback of others. Since plants have recovered from the cold, ratings have improved.

Preliminary data also indicated a significant difference between treatments for disease severity, as well as overall performance. Treatments with 1% Sunspray Ultra Fine Horticultural Oil were rated higher for foliar damage than chlorothalonil. Overall performance ratings of plants were lower with the Sunspray treatment than any other treatment. These low ratings may be due to phytotoxicity on new growth. Phytotoxicity may result when horticultural oils are applied during prolonged periods of high temperature (3).

A disease other than Blackspot was observed on several of the old rose varieties. The disease was identified by the Auburn University Plant Diagnostics Lab as *Cercospora rosicola*, a pathogen that typically is not considered important on rose. In our study, this pathogen apparently has caused significant foliar damage on several of the cultivars being evaluated. Further observations may prove interesting because some roses that show resistance to *D. rosae* may not be resistant to *C. rosicola*. Treatments also appear to differentially affect each of these pathogens. Further research on the significance of *C. rosicola* is in progress.

Significance to Industry: Ecological problems associated with frequent fungicide applications as well as consumer demand for landscape maintenance with minimal low input, are a growing concern. Blackspot of rose is an unavoidable problem in the south. With Integrated Pest Management (IPM) strategies becoming more common in the horticulture industry, alternative treatments for disease control and identification of resistant roses for the landscape can help reduce the use of chemical fungicides, as well as convince the consumer that roses can be grown and enjoyed with less maintenance and chemical input.

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Weather-Based Scheduling of Fungicide for Blackspot Disease Control on Hybrid Tea Roses in Alabama

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Alabama

Nature of Work: Blackspot disease (caused by the fungus *Diplocarpon rosae*) can cause severe defoliation of rose plants in the Southern landscape detracting from their aesthetic beauty. In Alabama and the southeastern U.S., prevailing environmental conditions promote blackspot fungal pathogen development from March through November. Optimal disease control with the fungicide chlorothalonil requires frequent applications (every 7- to 10-days) to protect newly developing leaves and replace fungicide that is washed off with rain. Recent concerns focusing on the safety and environmental impact of frequent fungicide use have caused rose growers to consider control alternatives. Baking soda in solution with horticultural oil, for example, has been shown to reduce diseases on roses in New York state (2). However, in Alabama, baking soda and oil solutions have been observed to damage rose foliage probably due to the higher temperatures that prevail in the region (1). The purpose of our study was to evaluate the effectiveness of oil solution applications alternated with fungicide applications for minimizing foliar damage and blackspot disease development on hybrid tea roses.

In 1992, hybrid tea roses were planted in exterior beds in full sun with drip irrigation. The study was arranged in a randomized complete block design with three replications. Plots consisted of one each of three varieties (Cary Grant, Dolly Parton, and Princess Monaco). A 10-cm layer of pine straw was placed around the base of plants in March, 1995. Foliar sprays were applied 1 May through September and consisted of 1) weekly applications of chlorothalonil (1.3 g a.i./L); 2) weekly applications of horticultural oil solution (1% SunSpray Ultra-Fine Horticultural Oil) alternated with chlorothalonil when less than 0.63 cm rain occurred during the preceding week; 3) weekly applications of a sodium bicarbonate and horticultural oil solution (5.3 g/L NaHCO_3 [baking soda] and 1% horticultural oil) alternated with chlorothalonil, depending on the occurrence of 0.63 cm rain; and 4) no spray treatment. Applications of the horticultural oil solutions (oil alone and in suspension with baking soda) were alternated with chlorothalonil fungicide applications to avoid the phytotoxic effects previously observed (1). Oil solutions were applied weekly but substituted with the fungicide when no significant (≥ 0.63 cm) rain fell between spray dates since rain would remove some of the oil and reduce phytotoxic effects. Spray solutions were applied until run-off. Blackspot disease severity, plant vigor, and flower production were rated weekly and averaged over the season. Differences among treatments were determined based on Fisher's protected least significant difference ($P < 0.05$).

Results and Discussion: In 1995, disease ratings averaged 1.22 out of 5 (where 0 is a disease-free plant and 5 indicates a plant that is completely diseased). Blackspot disease was low throughout the season due to high ambient temperatures throughout the summer. Chlorothalonil was applied 22 times to plants under the weekly fungicide regime; eight fungicide sprays with 14 sprays of either oil solution were applied to plants under the alternating regimes.

Significantly less disease was observed on plants treated with any of the foliar sprays compared to the untreated control plants. Significantly more disease occurred on plants treated with either oil solution alternated with the fungicide compared to plants treated weekly with chlorothalonil. Average vigor of plants treated weekly with chlorothalonil was significantly higher than plants receiving applications of oil alternating with chlorothalonil or no foliar treatment. Average numbers of flowers on plants treated weekly with chlorothalonil or with oil alternating with chlorothalonil did not differ. No differences among treatments were calculated for plant defoliation.

Significance to Industry: Data indicate that chemical fungicide use can be reduced when weather conditions are appropriate by prolonging intervals between fungicide applications. Less frequent use of chlorothalonil, especially when combined with horticultural oil, may allow more disease than weekly applications of the fungicide. However, alternating applications of the horticultural oil with chlorothalonil can minimize defoliation due to blackspot disease, avoid phytotoxicity due to foliar treatments, and allow similar flower production as a weekly fungicide application.

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The Use of Film Forming Antitranspirants to Control Blackspot Disease on Roses

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Alabama

Nature of Work: In the southeastern U. S., the fungal disease blackspot, caused by *Diplocarpon rosae* F. A. Wolfe, is a common and highly destructive disease of cultivated roses. When the disease is severe, plants are defoliated resulting in a weakened plant with a shortened life span (Dobbs, 1984). Control of the disease is achieved through a combination of cultural practices and fungicidal sprays. Sanitation measures include the removal of fallen leaves, yearly replacement of mulch, avoidance of overhead watering, and the application of lime-sulfur spray to dormant canes (Bowen, *et al.*, 1995). Fungicide applications are applied preventively at 7- to 10-day intervals throughout the growing season. Twenty-five to 30 applications of fungicide are usually required to prevent and control blackspot in this region. The objectives of this study are to evaluate film-forming antitranspirants (1) as an alternative to fungicides for control of blackspot by limiting the dispersal of conidia, and (2) as a fungicidal extender capable of reducing the number and frequency of chemical or fungicide applications necessary for control of the disease.

Materials and Methods: Eighty plants of each of three hybrid tea roses (Cary Grant, Princess Monaco, and Dolly Parton) were established at the E. V. Smith Research Center near Shorter, Alabama. Plots of plants consisted of each of the three cultivars, and 20 plots were arranged in each of four replications. Treatments were arranged in a randomized complete block design. Treatments included: (1) no treatment for high disease comparison; (2) 1% SunSpray oil applied weekly as long as there was a rain event between applications, when no rain occurred, chlorothalonil was applied at 1.3 g a.i./L (1.1 lbs. a.i./100 gal.); (3) weekly applications of chlorothalonil for low disease comparison; (4) Wilt Pruf at 5% applied every 2 weeks; (5) Nu Film 17 at 2% applied every 2 weeks; (6) Vapor Gard at 2% applied every 2 weeks; (7) Stressguard at 0.25% applied every 2 weeks; (8) Transfilm at 0.25% applied every 2 weeks; (9) Nu Film 17 at 1% alternated with chlorothalonil as in oil treatment; (10) Vapor Gard at 1% alternated with chlorothalonil as in oil treatment; and (11) Stressguard at 0.5% applied every two weeks. All treatments were applied with a CO₂ backpack sprayer, and sprayed on foliage until run-off. Treatments began May 3, 1996. All plants were evaluated weekly for disease, defoliation, vigor, and number of flowers.

Results and Discussion: Differences among treatments were determined using Duncan's Multiple Range Test comparisons. Preliminary results for May and June indicate that none of the treatments were as effective in suppressing disease and defoliation as weekly chlorothalonil applications. However, several treatments resulted in statistically similar disease or defoliation compared with weekly chlorothalonil treatment, including both the Nu Film 17 at 1% and the SunSpray oil alternated with chlorothalonil around weather events.

Significance to Industry: Since two of the treatments employed one half (SunSpray treatment) and one-third (Nu Film 17 treatment) the amount of fungicide that would normally have been applied, then results show that the use of antitranspirants and oils can reduce the amount of fungicide necessary to control blackspot disease. Further research should indicate the best programs for rates and timing of applications.

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Variation in Dogwood Anthracnose Resistance in Half-Sib Families of *Cornus kousa*.

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Tennessee

Nature of Work: In 1989, Polly Hill of Vineyard Haven, Massachusetts, wrote a letter to **American Nurseryman** magazine that some of her *Cornus kousa* selections were afflicted with dogwood anthracnose (*Discula destructiva*). This was confirmed by Richard Bir, NCSU extension nursery specialist. Previous to this, all *C. kousa* were presumed resistant. I asked Mrs. Hill for seed from some of her susceptible and resistant trees. She graciously provided us with fruit from 16 trees in her collection, half of which she considered "good" trees (not showing anthracnose symptoms) and half "bad" (showed anthracnose symptoms). It was my intention to germinate the seed, grow the seedlings, and challenge them for dogwood anthracnose resistance. We hoped this might give a clue as to heritability of resistance or susceptibility to the disease. We cleaned the seed, stratified it, germinated the seedlings in a greenhouse, potted them and grew them under ordinary nursery conditions to 2-gallon size. In late spring 1993, they were transported to a test plot in the Great Smoky Mountains National Park. They were placed by a stream where there was an overhead canopy of natural dogwoods (*Cornus florida*) infected with *Discula*. The containers were watered as needed and nature was allowed to take its course, raining spores down on the test plants from overhead. Dr. Mark Windham and staff scored the plants periodically for dogwood anthracnose infection on a modified Horsfall-Barrat scale that indicated the percent foliage diseased. This was a blind evaluation where they did not know the identity or disease resistance of the parent tree. Plants were transported back to the research nursery for further container culture. Those families showing good anthracnose resistance were planted at the UT Arboretum in Oak Ridge in 1996 for further observation and evaluation.

Results and Discussion: Table 1 gives the parentage and other details relative to each half-sib family we tested. Even the worst half-sib family (1E) scored better than an average *C. florida* tree would have, based on previous experience. Weather conditions were hotter and drier than normal, also depressing the expression of symptoms somewhat. Since this was open-pollinated seed, the male parent was unknown but assumed to be either a self or a cross with one of Polly Hill's other *C. kousa* trees. A small group of 7 plants labelled as 7W had the best score for *Discula* resistance (2.1). However, they were mis-labelled and probably are a sub-set of the 8E ('Big Apple') family which only scored 9.9 and was significantly worse. Regrettably, there did not seem to be any correlation or pattern of resistance or susceptibility linked to that of the pistillate parent. For example, we had duplicate collections from two separate 'Julian' trees. One of these families scored relatively high in *Discula* resistance (4.1) while the other scored only 10.2. In contrast the two 'Steeple' families (9W, 6W) scored about the same. Based on these results, neither susceptibility nor resistance seems to be a simple dominant gene. It may be that dogwood breeders will encounter a complicated inheritance system for *Discula* disease resistance. They may be forced to raise two or more successive generations, perhaps utilizing backcrossing, in order to combine anthracnose resistance with desirable horticultural characters. The other alternative is to rely on chance discovery of resistant trees, or screening large populations for resistance and good horticultural characters.

Significance to Industry: These studies represent the first efforts we know of to track the inheritance of *Discula* resistance in dogwood, utilizing half-sib families of *C. kousa*, a species which contains a range of genotypes from resistant to susceptible. Further work on breeding *Discula* resistance in *C. florida* utilizing putatively resistant clones and horticultural cultivars is an ongoing effort in Tennessee.

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Table 1. Mean scores for % diseased foliage, standard deviation, and range of scores for half-sib families of *C. kousa* trees challenged for *Discula destructiva* resistance in 1993.

<u>pistil parent</u>	<u>tree ID</u>	<u>P.Hill rating</u>	<u># plants tested</u>	<u># plants grown</u>	<u>mean* score</u>	<u>std. dev.</u>	<u>range</u>
***	8W	***	7	***	2.1	0.8	1.3 - 2.9
'Snowbird'	13W	bad	20	85	3.0	1.1	1.9 - 4.1
'Blue Shadow'	9E	best	3	11	4.0	0.7	3.3 - 4.7
'Julian'	13E	bad	17	71	4.1	2.3	1.8 - 6.4
'Steeple'	9W	good	82	210	4.3	2.3	2.0 - 6.6
unnamed	7W	good	11	33	4.6	2.8	1.8 - 6.4
unnamed	5E	bad	11	23	5.4	1.6	3.8 - 7.0
'Steeple'	6W	good	19	73	5.7	2.0	3.7 - 7.7
unnamed	12E	worst	17	24	6.6	3.0	3.6 - 9.6
'Square Dance'	2W	bad	44	61	6.8	3.9	2.9 - 10.7
unnamed	7E	bad	5	33	9.8	7.6	2.2 - 17.4
'Big Apple'	8E	good	36	207	9.9	6.3	3.6 - 16.2
'Julian'	13E	bad	35	dup.	10.2	7.0	3.2 - 17.2
unnamed	11W	good	51	66	11.5	5.9	5.6 - 17.4
unnamed	2E	so-so	46	89	12.0	6.7	5.3 - 18.7
unnamed	1E	bad	7	37	21.0	6.4	14.6 - 27.4

* represents % diseased foliage

Resistance to Powdery Mildew in Flowering Dogwood

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Tennessee

Nature of Work: Powdery mildew epidemics were common in fields of flowering dogwood (*Cornus florida*) in 1995. Economic losses due to powdery mildew were because the result of stunting of seedlings and lack of plant growth in older trees. In the fall of 1995, perithecia (sexual fruiting bodies of the fungus) were observed and the pathogen was identified as a *Microsphaera* species. The objectives of this project were 1) to screen popular cultivars of the flowering dogwood for resistance to powdery mildew and 2) to evaluate the frequency of natural resistance in dogwood seedlings.

Trees of 19 dogwood cultivars were evaluated for resistance to powdery mildew at a wholesale nursery in Tennessee. All trees were located in the same field and were the same age. One hundred trees of each cultivar were selected at random and evaluated for signs of powdery mildew using the following scale: 0 = healthy, 1 = powdery mildew on <2% of foliage, 2 = powdery mildew on > 2% but <10% of foliage, 3 = powdery mildew on >10% but <25% of foliage, 4 = powdery mildew on >25% but <50% of foliage, and 5 = powdery mildew on >50% but <100% of foliage, and 6 = 100% of foliage infected. Cultivars evaluated included 'Autumn Gold', 'Cherokee Brave', 'Cherokee Chief', 'Cherokee Daybreak', 'Cherokee Princess', 'Cherokee Sunset', 'First Lady', 'Holman's gold', 'Mystery', 'Pygmy', 'Plena', 'Purple Glory', 'Red Beauty', 'Rubra', 'Sisk Gold Variegated', 'Springtime', 'Spring Grove', 'Stokes Pink', and 'Welchi'.

In a separate study, leaves (upper and lower surfaces) of 13,280 flowering dogwood seedlings were examined for signs of powdery mildew infections. Eighty-eight seedlings were found to be free of mildew and were tagged. In late fall, 56 of the tagged seedlings were obtained from the nurseryman. Seedlings were potted in 1 gallon containers with pine bark media amended with nutrients and allowed to remain dormant for 10 weeks. Seedlings were then placed in a greenhouse to break dormancy. After leaf break, seedlings were arranged in a completely random design along with 12 control seedlings and 20 dogwood trees with severe powdery mildew infections. Plants were examined for 7 weeks for signs of powdery mildew and evaluated using the following scale: 0 = healthy, 1 = 1-2 leaves with mildew, 3 = >2 leaves but <25% of foliage with mildew, 3 = >25% but <50% of foliage with mildew, 4 = >50% but <1—% of foliage with mildew, 5 = 100% of foliage with mildew

Results and Discussion: Dogwood cultivars varied in their resistance to powdery mildew (Table 1). The cultivar 'Cherokee Brave' (deep pink or red bracts) was the only cultivar that demonstrated good resistance to this disease. Variegated cultivars usually displayed the most severe infections of powdery mildew. Of white flowering dogwood cultivars, 'Springtime' and 'Pygmy' displayed the best resistance, but neither cultivar appeared to be as resistant as 'Cherokee Brave'. Resistance to powdery mildew has been previously reported in cultivars of kousa and crosses between *C. florida* and *C. kousa* (1).

Of the 56 seedlings placed in the greenhouse trial, 12 seedlings were selected as having good resistance to powdery mildew (fig. 1). Many of the nonselected seedlings demonstrated partial resistance (significantly less incidence of powdery mildew than the controls) but were not as resistant as the seedlings designated as greenhouse selections in Fig. 1. Selected seedlings have been repotted into 5 gallon containers and are currently being grown for bud wood. In future studies, they will be evaluated for resistance to powdery mildew and for superior horticultural characteristics such as tree form and bract size.

Significance to Industry: Results of this study reveal considerable variation in resistance to powdery mildew in cultivars of flowering dogwood. The only cultivar identified in this study as having good resistance was 'Cherokee Brave'. Natural resistance to powdery mildew does occur in dogwood seedlings, but at a low frequency. Twelve seedlings were identified with good resistance to powdery mildew and will be evaluated for release as cultivars.

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Table 1. Evaluation of 19 dogwood cultivars for resistance to powdery mildew (0 = healthy, 6 = 100% of foliage infected).

Cultivar	Score
Cherokee Brave	1.0 a
Springtime	2.6 b
Pigmy	2.6 b
Cherokee Chief	3.4 c
Cherokee Princess	3.8 cd
Autumn Gold	4.0 cd
Plena	4.3 d
Rubra	4.6 de
Mystery	4.8 e
Stokes Pink	4.9 ef
Cherokee Daybreak	5.0 efg
Purple Glory	5.3 efg
Red Beauty	5.3 efg
Spring Grove	5.4 fg
First Lady	5.9 gh
Hohman's Gold	5.9 gh
Cherokee Sunset	6.0 h
Welchi	6.0 h
Sisk Gold Variegated	6.0 h

abcdefgh Means followed by the same letter do not differ according to Duncan's New

Multiple Range Test ($p \leq 0.05$).