The 2009 USGA Turfgrass and Environmental Research Summary is an annual compilation of projects currently funded by USGA's Turfgrass and Environmental Research Program. The summary contains research summaries of 65 projects funded in 2009.
The purpose of *USGA Turfgrass and Environmental Research Online* is to effectively communicate the results of research projects funded under USGA's Turfgrass and Environmental Research Program to all who can benefit from such knowledge. Since 1983, the USGA has funded more than 400 projects at a cost of $30 million. The private, non-profit research program provides funding opportunities to university faculty interested in working on environmental and turf management problems affecting golf courses. The outstanding playing conditions of today's golf courses are a direct result of *using science to benefit golf*.
2009 USGA Turfgrass and Environmental Research Summary

Jeff L. Nus

Since 1983, the United States Golf Association has funded more than 400 university research projects at 39 universities at cost of over $34 million. The Turfgrass and Environmental Research Program provides direction to these institutions and employs science as the foundation to benefit golf in the areas of turfgrass and resource management, sustainable development, and environmental protection. At the end of each year, the USGA provides a summary of the research conducted under this important national program and this report summarizes the results from 2009.

There are two primary goals of the research program. The first is to develop turfgrasses and cultural systems with better stress tolerance and reduced water requirements and pesticide use. To address the USGA's first research goal, 33 research projects were funded in integrated turfgrass management, physiology, breeding, genetics, and course construction practices. The second goal is to investigate environmental issues and sustainable resource management for golf courses. Four research projects that investigate the environmental impact of golf courses are reported.

The research program actively coordinates and supports research, associated educational programs, and other partnerships to benefit golf, the environment, and people. For example, the USGA, GCSAA, and National Turfgrass Evaluation Program (NTEP) together have developed turfgrass variety testing programs conducted on golf courses throughout the United States. In addition, the USGA works with state research foundations and superintendent chapters to fund applied research through the Grant-in-Aid Research Program. Thirteen research projects are included in this report under the Grant-in-Aid Research Program. USGA’s Turfgrass and Environmental Research Program also provides funding to the National Fish and Wildlife Foundation to support the Wildlife Links Research Program. Eight projects are currently being funded and summaries of their progress are included in this report.
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The United States Golf Association
Turfgrass and Environmental Research Program

Vision
Use science as the foundation to benefit golf in the areas of turfgrass and resource management, sustainable development and environmental protection.

Mission
Coordinate and support research, associated educational programs, and partnerships to benefit golf, the environment, and people.

Goals
Develop turfgrasses and cultural systems with enhanced stress tolerance and reduced supplemental water requirements, pesticide use and costs.

- Course Construction Practices
- Integrated Turfgrass Management
- Breeding, Genetics, and Physiology

Investigate environmental issues and sustainable resource management for golf courses.

- Environmental Impact of Golf Courses
- Wildlife and Habitat Management
### USGA Green Section Turfgrass and Environmental Research Project Grants in 2009

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<th>Project Area</th>
<th>Number</th>
<th>Grant $</th>
<th>% of Total</th>
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<tr>
<td>Course Construction Practices</td>
<td>1</td>
<td>30,000</td>
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<tr>
<td>Integrated Turfgrass Management</td>
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<td>240,456</td>
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<tr>
<td>Sustainable Management</td>
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<td>70,174</td>
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<tr>
<td>Pathology</td>
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<td>83,862</td>
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<tr>
<td>Entomology</td>
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<td>86,420</td>
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<tr>
<td>Breeding, Genetics, and Physiology</td>
<td>18</td>
<td>349,594</td>
<td>34.1 %</td>
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<tr>
<td>Cool-season Grasses</td>
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<td>Warm-season Grasses</td>
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<td>39,889</td>
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<td>New or Native Grasses</td>
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<td>49,846</td>
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<tr>
<td>Environmental Impact</td>
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<td>325,495</td>
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<tr>
<td>Fate and Transport</td>
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<tr>
<td>Wildlife Links Program</td>
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<td>Audubon International</td>
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<td>Outreach Programs</td>
<td>20</td>
<td>78,500</td>
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<td>Grant-in-Aid Research Program</td>
<td>14</td>
<td>46,000</td>
<td>4.5 %</td>
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<tr>
<td>Product Testing</td>
<td>5</td>
<td>31,000</td>
<td>3.0 %</td>
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<tr>
<td>Cooperative Research-Allied Assoc.s</td>
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</tr>
<tr>
<td>Total</td>
<td>67</td>
<td>1,024,045</td>
<td>100.0%</td>
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</tbody>
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2009 USGA Turfgrass and Environmental Research Committee

Chairman

Steve Smyers
Steve Smyers Golf Course Architects, Inc.
2622 W. Memorial Blvd.
Lakeland, FL 33815
v: 863-683-6100  f: 863-683-5888
ssgca@aol.com

Julie Dionne, Ph.D.
Royal Canadian Golf Association
272 Ile de France
Saint Basile le Grand
Quebec, CANADA
v: 514-705-3889  f: 905-845-7040
dionne@rcga.org

Ronald Dodson
Audubon International, Inc.
46 Rarick Road
Selkirk, NY 12158
v: 518-767-9051  f: 518-767-9076
rdodson@auduboninternational.org

Kimberly Erusha, Ph.D.
USGA Green Section
P.O. Box 708
Far Hills, NJ 07931-0708
v: 908-234-2300  f: 908-781-1736
kerusha@usga.org

Pete Grass, CGCS
1247 Ponderosa Drive
Billings, MT 59102
v: 406-855-6440
pgrass@gcsaa.org

Ali Harivandi, Ph.D.
University of California
Cooperative Extension
1131 Harbor Bay Parkway, #131
Alameda, CA 94502
v: 510-639-1271  f: 510-748-9644
maharivandi@ucdavis.edu

Michael P. Kenna, Ph.D.
USGA Green Section Research
P.O. Box 2227
Stillwater, OK 74076
v: 405-743-3900  f: 405-743-3910
mkenna@usga.org

Jeff Krans, Ph.D.
1258 LaFave Road
Manitowish Waters, WI 54545
v: 715-543-2114  f: 715-543-2114
jkrans@centurytel.net

Gene McClure
52 Golf Circle
Atlanta, GA 30309
v: 404-688-5000
lawmcclure@aol.com

James Moore
USGA Green Section Construction Educ.
770 Sam Bass Road
McGregor, TX 76657
v: 254-848-2202  f: 254-848-2606
jmoore@usga.org

Jeff Nus, Ph.D.
USGA Green Section Research
1032 Rogers Place
Lawrence, KS 66049
v: 785-832-2300  f: 785-832-9265
jnus@usga.org

Paul Rieke, Ph.D.
2086 Belding Court
Okemos, MI 48824
v: 517-349-2784  f: 517-355-0270
rieke@msu.edu

James T. Snow
USGA Green Section
Liberty Corner Road
P.O. Box 708
Far Hills, NJ 07931-0708
v: 908-234-2300  f: 908-781-1736
jsnow@usga.org

Clark Throssell, Ph.D.
Golf Course Supts. Assoc. of America
3280 Banff Avenue
Billings, MT 59102
chtrossell@gcsaa.org

Ned Tisserat, Ph.D.
Biological Sciences and Pest Mgmt.
Colorado State University
Fort Collins, Colorado 80523-1177
v: (970) 491-6527
Ned.Tisserat@ColoState.edu

Scott Warnke, Ph.D.
USDA-ARS
Room 110, Bldg 010A BARC-West
10300 Baltimore Avenue
Beltville, MD 20705-2350
v: 301-504-8260  f: 301-504-5096
warnkes@ars.usda.gov

James Watson, Ph.D.
3 Larkdale Drive
Littleton, CO 80123
v: 303-794-5346  f: 303-794-5346
jrwatson3@aol.com

Chris Williamson, Ph.D.
University of Wisconsin
246 Russell Labs
1630 Linden Drive
Madison, WI 53706
v: 608-262-4608  f: 608-262-3322
rcwillie@entomology.wisc.edu
Course Construction Practices

To take advantage of new ideas and technologies and to address the environmental and economic challenges of the coming decades, USGA sponsors research studies on the construction and establishment of golf courses. Preference is given to research studies that address issues related to:

1. Materials testing procedures for putting green rootzones and bunker sands
2. Alternative construction methods and materials
3. Effects of irrigation water quality on the selection of construction materials
4. Moisture-related problems in putting green rootzones
5. Calcareous sands
6. Various organic matter sources
7. Greens/surrounds interface construction and management
8. Alternative irrigation methods
9. How to amend various-textured native soils for use as tees (and possibly low-cost greens).

Interdisciplinary approaches are expected including the disciplines of soil physics, soil chemistry, soil microbiology, physiology, management, and pathology. Studies should give due consideration to environmental issues and to the use of alternative water sources for golf course irrigation.

Locations of the following projects funded in 2009 by the USGA Turfgrass and Environmental Research Program under the category of Course Construction Practices
This research investigates the dynamics of water movement through, and storage within, the rootzone of putting greens constructed using a geotextile atop a plastic support acting as the drainage structure (Airfield Systems design) compared to the same dynamics and storage in a green constructed with a gavel-based drainage structure (standard USGA design).

Constructed with a 100-mm layer of gravel to meet USGA recommendations, sand-based rootzone mixtures placed over gravel can hold water at the rootzone-gravel interface at tensions between 0 and 100 mm when watered to such a degree that drainage occurs. The Airfield Systems design replaces the gravel with a geotextile atop a 25-mm deep porous plastic support for drainage. The range of tensions at the rootzone-drainage structure in the Airfield Systems design is limited to between 0 and 25 mm water after drainage.

Sixty-three test columns were constructed from 332-mm (13-inch) inner diameter PVC pipe attached to a PVC sheet-lined plywood base. Twenty-seven of the columns were 420-mm tall and contained 100 mm of gravel the bottoms. The remaining 36 were 335-mm tall and had Airfield Systems’ AirDrain covered by a geotextile in the bottoms. Two tensiometers to measure the tension at the bottom of the rootzone were placed on top of the gravel or geotextile in each column. The columns then were filled with 300-mm deep rootzone mixture.

The treatments in the columns consisted of three rootzone mixtures, three gravels, and four geotextiles. The rootzone mixtures and the gravels ranged from the coarser to the finer sides of the USGA recommendations. The geotextiles consisted of one woven, two spunbond, and one needle-punch fabric. All geotextiles had an apparent opening size of about 0.2 mm. The three rootzone mixtures were placed over each of the three gravels and four geotextiles. There were three replicates of each treatment.

TDR probes from 21 columns could be monitored simultaneously, so we monitored each rootzone mixture across three gravels, four geotextiles, and three replicates. To test the water holding capacities, the test greens were watered in quantity sufficient to produce drainage, then the profile-averaged water contents were monitored at half-hour intervals for three days using a Campbell Scientific CR1000 datalogger. Tensions at the bottoms of the rootzones were monitored visually several times each day. Rootzone mixtures in the columns were sprigged with ‘MiniVerde’ dwarf bermudagrass and watered and fertilized as recommended. 300-mm long TDR probes were inserted vertically into the rootzones to monitor the effects of the treatments on the profile-averaged water contents.

In test runs conducted to date, greater tensions were always observed at the bottoms of the rootzones in the test greens containing the gravel treatments compared to those containing the geotextile-AirDrain treatments. Little differences in tensions were observed at the bottom of the rootzones of the three individual gravel treatments. Insignificant differences were observed in tensions at the bottom of the rootzones of the four geotextile-AirDrain treatments.

We observed greater water holding capacity of the AirField Systems designed green compared to the standard USGA test green. The differences in water holding capacity increased slightly with the coarseness of the sand in the rootzone mixture.

**Summary Points**

- Considerably greater water tensions developed at the bottom of rootzones in standard USGA gravel-based greens compared to Airfield Systems greens.
- Considerably lower water contents were observed in standard USGA greens compared to Airfield Systems greens.
- Airfield Systems greens held about 12 mm more water in the rootzone than the standard USGA greens—about three days more water for the grass to utilize.
Integrated Turfgrass Management

Improved turfgrasses developed for use on golf courses require management practices that provide quality playing surfaces while conserving natural resources and protecting the environment. A series of research projects are being funded with the aim of conserving natural resources by reducing the use of water, pesticides, and fertilizers. The objectives of these studies will focus on the following:

1. Develop cultural practices that allow efficient turfgrass management under unique conditions, such as poor quality soils, shade, and marginal quality water.

2. Determine the range of adaptability and stress tolerance of turfgrasses.

3. Evaluate direct and interacting effects of two or three cultural practices like mowing, irrigation, fertilization, cultivation, compost utilization, and develop programs to control pests and organic matter accumulation (thatch).

4. Investigate pest management practices such as biological, cultural, and mechanical controls, application of turf management practices utilizing IPM and reduced inputs, and pest modeling and forecasting.

The results of these studies should lead to the development of turfgrass management programs that conserve our natural resources and reduce costs, with minimal impairment of playing quality conditions or aesthetic appeal. We encourage regional cooperation among researchers where similar climatic and soil conditions exist.

Location of the following projects funded in 2009 by the USGA Turfgrass and Environmental Research Program under the category of Integrated Turfgrass Management
Developing Best Management Practices for Anthracnose Disease on Annual Bluegrass Putting Green Turf

James A. Murphy, Bruce B. Clarke, Joseph A. Roberts, Charles J. Schmid, and James W. Hempfling
Rutgers University

Objectives:
1. The multiple objectives of this research were organized into four field studies on annual bluegrass putting green turf that were designed to evaluate the main effect and interactions of: 1) irrigation quantity, 2) lightweight rollers and mowing equipment, 3) topdressing and foot traffic, and 4) nitrogen fertilization on anthracnose disease.

Start Date: 2008
Project Duration: three years
Total Funding: $90,000

Anthracnose, caused by Colletotrichum cereale, is a destructive disease of annual bluegrass putting green turf throughout the United States. The disease begins as small areas of yellowed turf (1 to 2 inches in diameter) with individual leaf blades eventually senescing resulting in an overall thinning of the turf canopy.

The frequency and severity of anthracnose outbreaks on putting greens has increased over the past decade and management practices employed to improve playability and increase ball roll distance on putting greens have been observed to be partly responsible.

Research completed in 2008 generated the following conclusions: 1) deficit irrigation causing wilt stress (e.g., 40% daily ET₀ replacement) increased the severity of anthracnose compared to greater irrigation quantities; 2) both sidewinder and triplex mounted vibratory rolling reduced disease severity compared to non-rolled turf under moderate disease pressure; and 3) sand topdressing reduced disease severity under both foot trafficked and non-trafficked conditions; unexpectedly, foot traffic decreased anthracnose regardless of sand topdressing level.

Previous research indicated that adequate N fertility to sustain moderate growth can reduce anthracnose severity. A study was initiated in 2007 to determine the effect of rate and frequency of summer soluble-N fertilization on anthracnose severity. Disease severity had a negative linear response to increasing amounts of N fertilization within the range of treatments studied. Nitrogen applied at the highest rate (0.1 lb per 1000 ft² every 14 days) or 14 days (0.2 lb per 1000 ft²) during the growing season provided the greatest reduction in anthracnose severity. The lowest anthracnose severity during the 2009 season was observed on plots treated with N at 0.2 lb per 1000 ft² every week.

A second soluble-N study was initiated in the summer of 2009 to determine the effect of greater soluble-N fertilization rates on anthracnose severity. Early in the season, 0.1 lb per 1000 ft² every 7 days had the greatest anthracnose severity within the range of treatments studied. As the season progressed, the N rates of 0.4 and 0.5 lb per 1000 ft² every week resulted in the greatest anthracnose severity. The lowest anthracnose severity during the 2009 season was observed on plots treated with N at 0.2 lb per 1000 ft² every week.

Spring granular-N fertilization reduced disease severity compared to autumn granular-N fertilization on all but the last sampling date (August 26, 2009). The interaction between season and granular-N rate (4 of 7 rating dates) indicated that greater granular-N rates reduced disease severity but only within the spring N-fertilization timing. Summer soluble-N fertilization influenced disease severity throughout most of the summer; N applied at 0.094 lb per 1000 ft² every week had the greatest reduction in anthracnose severity compared to N fertilizations every 2, 4, and 0 weeks. Plots that received no summer soluble N fertilization had the greatest anthracnose severity.

Summary Points
- Deficit irrigation (40% ET₀) induced wilt stress and intensified anthracnose severity. Irrigation at 80% ET₀ often resulted in the lowest anthracnose severity and best turf quality.
- Lighteweight rolling every other day with either roller type (i.e., sidewinder or triplex-mounted vibratory) effectively increased ball-roll distance and decreased anthracnose severity under moderate disease pressure.
- Sand topdressing initially increased anthracnose in 2007; however, continued weekly applications of sand reduced anthracnose severity by August 2007 and throughout 2008 under both foot trafficked and non-trafficked conditions. Moreover, daily foot traffic decreased anthracnose severity regardless of sand topdressing level. The combination of daily foot traffic with weekly sand topdressing resulted in the lowest disease severity and best turf quality in both 2007 and 2008.
- Nitrogen applied every 7 (0.1 lb per 1000 ft²) or 14 days (0.2 lb per 1000 ft²) during the growing season provided the greatest reduction in anthracnose severity; N applied every 14 days at 0.1 lb per 1000 ft² also reduced disease, albeit, to a lesser extent. Nitrogen applied at rates of 0.3 to 0.5 lb per 1000 ft² every 7 days increased anthracnose severity.
- Spring granular-N fertilization decreased anthracnose severity compared to autumn fertilization. Increasing granular-N rate within the spring fertilization timing reduced disease severity but had no effect during the autumn timing. Increased monthly soluble-N rate during the summer also decreased anthracnose severity.
Natural Enemies and Site Characteristics Affecting Distribution and Abundance of Native and Invasive White Grubs on Golf Courses

Carl T. Redmond and Daniel A. Potter
University of Kentucky

Objectives:
1. Determine identity and incidence of pathogens and parasitoids of Japanese beetle (JB) and masked chafer (MC) grubs on golf courses across Kentucky, the first such study in the transitional turfgrass zone.
2. Quantify site characteristics associated with particular grub species and natural enemies and prospect for potential new bio-control agents.
3. Evaluate how grass species and mowing height affect the susceptibility of white grubs to natural enemies in the field.
4. Test if a lag in buildup of natural enemies explains why Japanese beetle grub populations tend to reach outbreak densities on golf courses as the pest expands into new geographical regions and then stabilize over time.

Start Date: 2007
Project Duration: three years
Total Funding: $60,000

Biological insecticides and natural enemy conservation can reduce the need for chemical inputs on golf courses. We seek new pathogens having promise as bio-insecticides, and to clarify how site characteristics might be altered to enhance natural suppression of grub populations.

Grub survey kits were sent to 34 golf superintendents throughout Kentucky in late summer asking them to collect 30 grubs and a soil sample from their worst non-treated grub site. Six additional golf courses were intensively sampled in late August, mid-September, and early October to track natural enemy incidence over time. Grubs were identified, incubated for 30 days, and dissected to assess mortality from bacterial, fungal, or protozoan pathogens.

Masked chafers and Japanese beetles accounted for 64 and 30%, respectively, of grubs sent in by superintendents. Masked chafers also predominated on Lexington courses. Grub populations declined from about 18 m\(^{-2}\) in late August to about 5 m\(^{-2}\) in October and 2 m\(^{-2}\) the following spring owing to natural mortality agents.

Tiphia wasps, Metarthizium fumigus, Serratia (amber disease) and Puenibacillus (milky disease) bacteria, and entomopathogenic nematodes infected 2, 5, 8, 20, and 18% of the masked chafers grubs, and 0, 3, 5, 12, and 19% of the Japanese beetle grubs sent in by superintendents, but much higher mortality from particular agents was seen on some courses. Ovavesicula, a protozoan that reduces egg production by adult Japanese beetle, was uncommon in KY, but gregarines (Stichospora) infected 26% of Japanese beetle grubs in the spring. The latter two pathogens were absent or uncommon in masked chafer grubs.

Replicated stands of irrigated turfgrasses were sampled for grub species preference and incidence of parasitoids and pathogens. Of the fairway-height grasses, zoysiagrass and bermudagrass had the highest incidence of masked chafer grubs. Masked chafers predominated in creeping bentgrass, whereas Japanese beetles favored perennial ryegrass. Japanese beetle populations were highest in rough-height grasses, outnumbering masked chafers 2-4 fold.

Two years of rating skunk damage showed greatest foraging in creeping bentgrass and perennial ryegrass at fairway height. There was little to no skunk activity in fairway-height zoysiagrass or bermudagrass, or in rough-height grasses. Milky disease was the most common pathogen of masked chafers, present in both fairway and rough cut grass (rates to 24% and 20% for fairway zoysiagrass and rough-cut Kentucky bluegrass, respectively).

**Summary Points**
- Masked chafers and Japanese beetles accounted for about 66 and 30% of the grub infestations, respectively, on surveyed Kentucky golf courses. Tiphia wasps, milky disease, and other pathogens accounted for moderate to high natural mortality at some sites. Nematodes isolated from masked chafer and Japanese beetle grubs, and a protozoan pathogen (Stichospora sp.), are being evaluated as potential bio-insecticides.
- Turfgrass species and mowing height affected the species makeup of grubs and natural enemies. Skunk foraging damage was greatest in cool-season grasses at fairway height and least in warm-season grasses despite the presence of grubs.
- Japanese beetle grubs surveyed across the eastern and central United States showed a trend for higher pathogen loads in eastern states with longer history of infestation than in central or Midwestern states.
Seasonal Life History and Suitability of Horticultural Plants as Nectar Sources for *Larra bicolor*, a Parasitoid of Mole Crickets in the Northern Gulf Coast

David W. Held
Mississippi State University

**Objectives:**

1. Determine the seasonal life history of *Larra bicolor* in the northern Gulf Coast.
2. Determine the suitability of flowering plants as nectar sources for *Larra bicolor*.
3. Determine if incorporating wildflowers on golf courses will facilitate the establishment of *Larra* wasps on that site.

**Start Date:** 2006  
**Project Duration:** three years  
**Total Funding:** $29,232  

*Larra bicolor*, an introduced ectoparasitoid of mole crickets, has spread into the northern Gulf region (Mississippi and Alabama) from where it was originally introduced (Florida). This creates an opportunity for golf course superintendents in this area to utilize this biological control agent. This project is the first empirical research on *Larra bicolor* conducted outside of Florida. Therefore, it is necessary to determine the seasonal flight period in the extended range of this wasp.

From 2006 to 2008, wasps were monitored on four sites in south Mississippi. Across all sites, wasps were generally active June through October with an additional month of activity in fall 2007. This seasonal activity of *L. bicolor* is similar to that reported from the Florida panhandle (Gadsden Co.). Wasp activity was observed on flowers from about 13 hours (0700 to 1900 h CST) in August and 9 hours (0900 to 1700 h CST) in October. During this study, sunrise and sunset were 0520 h and 1845 h (August), and 0600 and 1730 h (October), respectively. Sunrise, and not temperature, likely trigger activity.

Air temperatures in August when wasps became active were 27-28° C, whereas wasps were active when air temperatures were 18.3° C in October. In August, a maximum of 14 females and 31 males were observed at once on flowers compared to 3 and 25, respectively, in October. Males were almost always present on flowers during the hourly observations. Females, however, were observed on flowers for distinct periods (3 to 9 hours) each day. Females spend their time mainly hunting for hosts only returning to flowers for nectar as needed.

In 2007 and 2008, we established a replicated garden of 16 plant taxa of flowering plants in 0.5 × 0.5 m plots arranged in a randomized complete block design. Only two taxa, Pentas and Spermacoce, were visited by *L. bicolor*. White pentas was readily foraged upon by *L. bicolor* and numbers of wasps were significantly different from either pink or red-flowered pentas. White and yellow are common colors attractive to day active insects such as these wasps. However, there were several taxa in the garden with yellow and lighter colored flowers that went unvisited by *L. bicolor*. Volatiles likely determine wasp visitation to these flowers.

Attempts to trap or passively monitor this species have been previously unsuccessful. In fall 2008, an experiment was conducted using pan traps of various colors as attractants. Despite activity on adjacent flowers, wasps were not recorded in the traps. In 2009, trapping studies were conducted using phenylacetaldehyde (PAA) and floral extracts of Spermacoce and Pentas as lures. The objective was to verify the results of a previous study that demonstrated that *L. bicolor* could be trapped using PAA and floral volatiles as a way to collect specimens for research or to passively monitor the continued spread of *L. bicolor*.

Field plots containing white-flowered pentas have been established on four holes at Grand National Golf Club, Opelika, AL. These plants will serve as release points for *L. bicolor* on that course. Wasps will be collected from golf course sites in Mobile and Baldwin Counties in Alabama and released on the flowers at dusk.

Due to heavy summer rains, wasps were unable to be collected in mass during August or September. Manual collection is slow and inefficient, but we anticipate release of four females (and males) per planting during October and early November.

**Summary Points**

- *Larra bicolor* have a 4-5 month activity in the northern Gulf region to parasitize mole crickets. Wasp activity is mostly during daylight hours. Although the impact of insecticides on *L. bicolor* have not been evaluated, turf managers wanting to conserve these biological controls should consider making insecticide applications later in the day as numbers of foraging wasps wane.
- In two trials, pentas, particularly white pentas, recruit equivalent numbers of *L. bicolor* as flowering *Spermacoce verticillata*. Wasps don't appear to be visually attracted to flowers and experiments using volatile attractants are being conducted.
Spring dead spot is the most devastating and important disease of bermudagrass where it undergoes winter dormancy. The disease is caused by one or more of three fungal species in the genus *Ophiosphaerella* (*O. herpotricha*, *O. korrae*, or *O. narmari*). The disease results in unsightly dead patches on fairways, tees, and bermudagrass greens, giving way to the encroachment of weeds and costly management efforts to eliminate weeds and re-establish grass in the affected area.

The overall goal of this study is to enhance our understanding of the interaction between *O. herpotricha* and its bermudagrass host, and how environmental and host factors influence this interaction.

We have inserted two different fluorescent reporter genes (red and green) into the fungus and examined root and stolon infection of various bermudagrass cultivars. We were able to document differences in infection and colonization for roots and stolons of grasses that vary in their response to the disease. Colonization and necrosis of epidermal and cortical cells was observed for the cultivars ‘Midlawn’ (resistant) or ‘Tifway’ (susceptible), while the stele remained uninfected and intact.

For a *Cynodon transvaalensis* accession, epidermal and cortical cells and the stele were colonized, but became much less necrotic than the other cultivars. A confocal scanning laser microscope was also used to produce 3-dimensional images of the fungus in and on bermudagrass roots further supporting observations that the fungus grows intercellularly (between cells) in roots before eventually penetrating and infecting root cells.

These studies have permitted the formation of hypotheses as to the mechanism of plant death when colonized by the fungus and exposed to cool temperatures that induce dormancy. Conventional wisdom would predict that for a resistant plant the fungus would either not be able to gain entry or that fungal growth would be restricted due to the host mounting an aggressive defense response visible as hypersensitive defense necroses.

However, from our data, we hypothesize that a resistance response to spring dead spot could include the fungus living endophytically in root and stolon tissues without causing severe necrosis due to the inability of a grass to recognize and respond to the invading fungus.

Conversely, susceptible plants' recognition of the fungus would elicit an extreme host defense response including a hypersensitive reaction (HR), whereby infected host cells undergo programmed cell death in an attempt to limit or stop pathogen growth. This has been shown to be the case with a few plant fungal disease interactions.

To test this hypothesis, we have inoculated a series of *C. dactylon*, *C. dactylon* x *C. transvaalensis* interspecific hybrids, and *C. transvaalensis* cultivars and collected samples to test for hallmark tags of the hypersensitive response.

**Summary Points**

- Colonization and necrosis of roots were extensive in ‘Tifway’ and less in ‘Midlawn’.
- The *C. transvaalensis* accession was extensively colonized, but with only limited necrosis.
- Disease resistance could be due to the inability of the plant to respond necrotically to the fungus.
- This information will be used to enhance bermudagrass disease resistance through traditional breeding efforts at Oklahoma State University.
Biological Control of Black Cutworms on Golf Courses
Using a Baculovirus and Natural Enemies

Andrea Bixby and Daniel A. Potter
University of Kentucky

Objectives:

1. Evaluate AgipMNPV, a naturally occurring baculovirus, as a bio-insecticide for season-long and multi-year preventive control of black cutworms (BCW) on golf courses.
2. Compare infectivity and persistence of AgipMNPV to BCW in sand-based and soil-based putting green and fairway height creeping bentgrass habitats.
3. Investigate compatibility and possible synergism of AgipMNPV with soil insecticides used for grub control on golf courses.
4. Investigate compatibility of endophytic and other insect-resistant turfgrasses with biological control of black cutworms by AgipMNPV.

Start Date: 2007
Project Duration: three years
Total Funding: $60,000

Biological controls, once established, can provide prolonged suppression of insect pests on golf courses. In 2003, a cutworm-specific virus was found decimating populations of black cutworms (BCW) on Kentucky golf courses. Virus-infected BCW rupture and spread millions of infective virus particles onto foliage and thatch.

The virus, identified as Agrotis ipsilon multiple nucleopolyhedrovirus (AgipMNPV), was amplified by feeding it to BCW, harvesting their cadavers, and mixing the concentrated virus particles with water. This crude biological insecticide was applied to turf to see if it would infect resident larvae.

Testing showed that it quickly killed young larvae, but that larger ones required higher dosages and fed for several days before being killed. When applied to 12 whole tees of two golf courses, the virus gave 78% and 33-41% control of newly hatched larvae after one week and one month, respectively, but residual control of larger larvae (45-56%) lasted only about a week.

During summer 2009, virus was applied to paired plots on creeping bentgrass greens and mixed-grass surrounds to compare its residual activity in those habitats. Residual efficacy was determined 3 days, and 2 and 5 weeks after application by implanting mid-sized BCW and determining how many were killed by virus infection. Three-day-old virus residues provided 50-60% control in both greens and surrounds, but infection dropped to only 10-20% in both settings after two weeks, and was almost nil by five weeks after application. A virus-based insecticide could provide short-term control of BCW on golf courses, but it probably would need to be re-applied at regular intervals.

Parasitoids are insects that parasitize other insects. When BCW were implanted on golf course tees, 30% of larvae recovered after 10 days in the field had been parasitized. A survey of BCW parasitoids was initiated in 2009 to provide better understanding of how to conserve and recruit these beneficial insects for biological control. For instance, naturalized areas on golf courses could promote beneficial insects by providing pollen, nectar, alternative hosts, or shelter that can attract and sustain natural enemies of pest insects.

Studies were initiated in 2009 to determine the species, natural history, and impact of parasitoids of BCW in golf course habitats. Sentinel eggs and newly hatched larvae were placed in the field monthly in four locations: tees, fairways, and roughs near or far away from naturalized areas on three Kentucky golf courses.

We hoped to determine how grass height and proximity to naturalized areas affect parasitism, but the study was inconclusive because ants consumed most of the eggs and larvae.

In another experiment, parasitoids accounted for similar (12-19%) mortality of BCW in field plots of creeping bentgrass and perennial ryegrass. Two parasitoid species that attack BCW eggs and three others that attack the larvae were identified. The most abundant parasitoid, a tachinid fly (Bonnetia comta), was reared to study its biology. The female flies, stimulated by BCW frass (feces), to give birth to maggots which are deposited near the entrance to a BCW burrow. The fly maggot crawls onto the victim's back when it emerges to feed. The maggot burrows in, feeds, and kills the BCW in a few days, then emerges from the shriveled victim and forms a pupal case from which a new fly emerges.

Experiments planned for 2009-10 explore chemical cues that attract the parasitic flies to golf turf, as well as compatibility of parasitoids with turf insecticides and endophytic turfgrasses.

Summary Points

- A virus-based biological insecticide has potential for short-term control of black cutworms (BCW) on golf courses.
- Five parasitoid species were found suppressing BCW populations on Kentucky golf courses. A study to determine if naturalized areas, which provide resources for beneficial insects, can increase parasitism of BCW was thwarted by high ant predation.
- Biology of a fly that parasitizes BCW was clarified and studies were initiated to evaluate it for augmentative or conservation bio-control.
Accurate Identification and Gene Expression in Relation to Virulence of Rhizoctonia Isolates Infecting Turfgrasses

Dilip K. Lakshman
USDA-ARS

Objectives:

1. Molecular identification of *Rhizoctonia solani* isolates pathogenic to turfgrasses using Universally Primed-Polymerase Chain Reaction (UP-PCR) and nucleic acid hybridization analysis.
2. Expressed Sequence Tag (EST) analysis for surveying genes and creating a gene database for *R. solani* with emphasis on genes affecting virulence and pathogenicity.

*Rhizoctonia* blights, variously named as brown patch, large patch, yellow patch, leaf and sheath spots, and brown ring patch, are caused by *R. solani sensu lato*. Symptoms are mostly determined by the isolates of the pathogen, species of turfgrass, and climatic conditions. For example, brown patch is a disease of cool-season grasses in the summer, caused by multinucleate *R. solani* (Tel: *Thanatephorus cucumeris*) isolates. Anastomosis groups (AG) 1, 2, 3, 4, 5 and 6 have been isolated from blighted grasses. Leaf and sheath spots in both cool- and warm-season grasses are caused by multinucleate *R. oryzae* and *R. zeae* (Tel: *Waitea circinata var oryzae/zeae*) in the summer. Yellow Patch of both cool- and warm-season grasses are caused by binucleate *R. cerealis* (Tel: *Ceratobasidium cereale*) AG-D isolates. Recently a new disease, Brown ring patch, has been reported to be caused by *W. cercinata var circinata*.

*Rhizoctonia* species and AGs are genetically heterogeneous groups and reported to differ in sensitivity to common fungicides and host susceptibility. The prevalence and severity of *Rhizoctonia* diseases on turfgrasses depend, among other factors, on infection by a particular species or AG (anastomosis group) of *R. solani*. Thus, minimization of chemical use as well as consistent and reliable management of *Rhizoctonia* diseases with genetic and biological methods will largely depend on identification of *Rhizoctonia* isolates to species and subspecies levels and knowledge of its virulence-regulating genes.

In a team effort with Dr. Brandon Horvath and Sajeewa Amaradasa, we collected a total of 448 *Rhizoctonia* samples from 5 locations in Northern Virginia and Maryland during the summer of 2008 and 2009. We selected a random sample of approximately 10% of the collected isolates for further studies. Isolates were grouped into species level using conventional approaches. AG group of each isolate was determined by anastomosis behavior with tester strains.

Genomic DNAs isolated from *Rhizoctonia* isolates are being analyzed employing molecular techniques like sequence analysis of the ribosomal DNA internal transcribed spacers (rDNA-ITS) regions and Universally Primed Polymerase Chain Reaction (UP-PCR) to group the *Rhizoctonia* isolates and compare them with conventional grouping methods. rDNA-ITS PCR products were sequenced and homology searched with rDNA-ITS sequences from the NCBI public database.

Analyzing *rDNA-ITS* sequence variabilities, we found that of the 54 isolates tested, 33 (61%) isolates belong to *R. solani*, 9 isolates (17%) belong to binucleate *Rhizoctonia*-like fungi, and 12 isolates (22%) belong to *R. zeae*/*W. cercinata var circinata*. Of the 33 *R. solani* isolates, 20, 12 and 1 belonged to AG2-2IIIB, AG 1-1B and AG-5, respectively.

UP-PCR products will be cross-hybridized to evaluate the possibility of developing a dot-spot hybridization detection method. Also, we will look for Sequence Characterized Amplified Region (SCAR) markers in the UP-PCR.

First time for any *R. solani* AG-4 isolate, we are investigating the expressed gene profiles through Expressed Sequenced Tag (EST) analyses of two virulence-differentiated and rDNA-normalized EST libraries (Objective 2). Approximately, 1,110 clones from each of the two libraries (total 2,220 cDNA clones) have been sequenced, generating 742 and 842 high quality EST sequences and 532 and 495 unigenes, respectively. Among the important genes identified from the two libraries are those involved in plant cell wall degradation, fungal cell wall protection, disease elicitation, avoidance of host and drug resistance, cellular metabolism, transport, division, and mating.

Summary Points

- We have collected a total of 448 *Rhizoctonia* isolates from golf courses and lawns of Virginia and Maryland. Analyzing the *rDNA-ITS* sequence variations of 54 randomly sampled isolates, we found that majority (i.e., 61%) of the isolates belong to *R. solani*, while 22% belong to *R. zeae*/*W. cercinata var circinata*, and 17% belong to *R. cerealis/Rhizoctonia*-like fungi. Of the identified *R. solani* isolates in this study, majority belong to AG2-2IIIB and AG 1-1B.
- Genome fingerprinting of *Rhizoctonia* isolates using UP-PCR and AFLP techniques are in progress.
- Analyzing the two virulence-differentiated EST libraries, we have identified 532 and 495 unigenes of *R. solani* AG-4. Among several identified genes are those with possible roles as elicitors of disease, in cell wall degradation, avoidance of host and drug resistance and mating.
Mole Cricket Sensory Perception of Insecticides

Eileen A. Buss and Olga Kostromytska
University of Florida

Objectives:
1. Determine the type, location, and abundance of different sensilla on the antennae and mouthparts of S. vicinus and S. borellii.
2. Demonstrate the physiological effect of insecticides on the mole cricket nervous system and/or ability of mole crickets to detect chemical stimuli.
3. Demonstrate the behavioral response of mole crickets to sub-lethal insecticide doses.

Start Date: 2007
Project Duration: three years
Total Funding: $84,208

The more that mole crickets (Scapteriscus spp.) move in the soil, the more damage they cause to turfgrass. Ideally, the insecticides used against them should stop their tunneling movements and kill them quickly. However, insecticides like acephate (Orthene), bifenthrin (Talstar), fipronil (TopChoice), imidacloprid (Merit), bifenthrin plus imidacloprid (Allectus), and indoxacarb (Advion) are neurotoxins, and their effect on mole cricket mobility is not well defined. They could either excite or inhibit the insect’s nervous system, which could lead to more or less tunneling before an affected mole cricket dies. In addition, the amount of time it takes to kill a mole cricket is also unknown because they tend to die in the soil. So, if spot treatments of neuroexcitatory insecticides are made, rather than “wall to wall” applications, more tunneling could occur at the edge of treated areas as the insects try to escape.

This first test was done to determine if the previously mentioned insecticides would excite or inhibit mole cricket nervous systems. We recorded the spontaneous nerve cord activity of tawny mole crickets using a suction recording electrode. Adults were dissected and their intact nerve cords were exposed within the abdominal cavity. For each specimen, the recording was conducted in saline solution for the first 5 minutes to establish a baseline. Then 10 µL of an insecticide solution was added to the abdominal cavity and recording continued for another 15 minutes. Technical grade insecticides were used (in 0.04% solution of DMSO), and saline alone and saline mixed with solvent (0.04% DMSO) were the controls.

Acephate, bifenthrin, fipronil, imidacloprid, and bifenthrin plus imidacloprid all had a significant neuroexcitatory effect, but indoxacarb and its metabolite were neuroinhibitory. Bifenthrin, fipronil, and bifenthrin plus imidacloprid caused the strongest neuroexcitatory effects on spontaneous neural activity.

We determined and compared the toxic effect of the insecticides by estimating the time needed for a product to kill 50% of the tested insects (LT50) for acephate, bifenthrin, fipronil, imidacloprid, bifenthrin plus imidacloprid, indoxacarb, and the indoxacarb metabolite (DCJW). Technical grade (95% and higher of active ingredient) insecticides were injected into the thorax of intact tawny mole cricket adults and nymphs, and the insects were held in petri dishes with moist sand and a food source. Mole crickets injected with saline solution of solvent served as controls. Their behavior was observed and mortality was recorded every hour for the first 12 hours and every 4 hours for the following 7 days.

Bifenthrin, fipronil, and bifenthrin plus imidacloprid provided the fastest mortality (38.3, 35.5 and 10.3 hours for adults, and 9.5, 10.4 and 6.5 hours for nymphs, respectively). Bifenthrin, fipronil, indoxacarb and its metabolite kill nymphs significantly faster than they kill adults.

Behavioral changes were noticed after treatment with most of the insecticides in the injection assay. Tawny mole crickets became immobile within 30 seconds after being injected with imidacloprid or bifenthrin plus imidacloprid, within 2-3 minutes after treatment with bifenthrin alone, and 1-2 hours after fipronil injection. However, mole crickets partially recovered after imidacloprid treatment and could walk but not tunnel.

Acephate increased the spatial movement and tunneling activity of mole crickets compared to the mole crickets injected with solvent solution only. Indoxacarb caused trembling, erratic leg and wing movements, and the insects kicked or jumped if disturbed. These data and observations correspond with the data in earlier behavioral assays. Our future research will focus on the ability of mole cricket antennae to detect insecticides using an electroantennogram.

Summary Points
- All tested insecticides (except indoxacarb and its metabolite) caused neural excitation, so their use could lead to increased tunneling, especially at sublethal doses.
- Fipronil, bifenthrin, and bifenthrin plus imidacloprid killed mole cricket adults and nymphs faster than imidacloprid, indoxacarb, the indoxacarb metabolite (DCWJ), and acephate.
- Fipronil, bifenthrin, and bifenthrin plus imidacloprid immobilized mole crickets within an hour, completely disrupting their tunneling activity at the tested dose.
- Insects injected with acephate and then held in petri dishes had increased tunneling and crawling.
- Imidacloprid caused immediate knockdown (within 30 seconds), but although the insects partially recovered, they could still walk but not tunnel.
The Efficacy of Spring Fungicide Applications Plus Organic Fertilizer for Controlling Spring Dead Spot of Bermudagrass

Maria Tomaso-Peterson
Mississippi State University

Objectives:
1. Determine the efficacy of spring and fall fungicide applications for reduction of spring dead spot incidence and severity.
2. Determine the effect of organic fertilizer for the reduction of spring dead spot incidence and severity and overall improvement of turf quality.

Start Date: 2007
Project Duration: three years
Total Funding: $30,000

Spring dead spot (SDS) is a serious root-rot disease of bermudagrass and is the most important disease of hybrid bermudagrasses managed as putting green and fairway turf. Aesthetically undesirable necrotic patches ranging from a few inches to several feet in diameter are evident in the spring and early summer in bermudagrass swards that experience a dormant period. Three fungal species in the genus Ophiosphaerella (O. korrae, O. herpotricha, and O. narmari) have been identified as the causal organisms throughout the United States and Australia. In Mississippi, O. korrae is the most common species; however, O. herpotricha has also been identified.

Results of a previous study conducted at Mississippi State University suggest the frequency of O. korrae in bermudagrass roots was greatest during winter dormancy and spring transition compared to summer and fall transition periods. As a result of O. korrae colonization in bermudagrass roots during the winter and spring, this study was initiated in the spring of 2007 in a 'Tifway' bermudagrass fairway with a history of spring dead spot. Symptoms of SDS were observed throughout the current study area in the spring of 2007.

The treatment plots (15 ft x 10 ft) are arranged in a split-plot randomized complete block design replicated four times. Fungicide treatments are the whole plot factor and nitrogen (N) source is the sub-plot factor (7.5 x 10 ft sub-plots). Fungicide treatments are applied during the spring and fall. The N sources include 12-2-12 organic fertilizer and 12-2-12 blend of inorganic fertilizer including ammonium sulfate (21-0-0), triple super phosphate (0-46-0), and muriate of potash (0-0-60) applied at 1.0 lb N per 1000 ft2 per month (May-October).

Spring dead spot severity (1 to 9; 9 = no disease) and incidence (% plot area) were determined in the spring of 2009. Spring green-up was quantified using an NDVI turf color meter, turfgrass quality (1 to 9; 9 = best) was rated each month, and SDS patch recovery was monitored.

Spring dead spot severity was moderate in 2009 across Mississippi and at the site of this fairway study despite persistent cold temperatures in late winter and early spring. Necrotic patches ranged in size from 4 to 15 inches in diameter. Three of the Rubigan treatments along with Eagle and Banner Maxx fungicides, applied in 2008, significantly reduced SDS severity and incidence in the spring of 2009. Rubigan treatments applied in the spring, fall, or spring and fall had less than 1.3 % SDS, while the control plots averaged 12% SDS. SDS severity ratings were also improved in those treatments compared to the control. Turfgrass quality in the Rubigan and Eagle treatments in May was improved (5.4 - 5.6) but not in the Banner Maxx plots (5.1) where turfgrass quality was similar to the control (4.6). Turfgrass quality was similar for all treatments the remaining months beginning in June 2009.

Neither the N source (organic vs inorganic) nor the interaction of fungicide and N source had an effect on SDS severity and incidence, turfgrass quality, or spring green-up. Soil pH in plots receiving organic N (pH=6.2) was significantly higher than the pH of plots receiving inorganic N (pH=6.0); however a pH difference of 0.2 is not likely to be biologically significant in this patho-system.

These results reflect the second year of this three-year study. Spring and fall fungicide treatments have been applied in 2009. The final SDS evaluations will be conducted in the spring of 2010. To date, Rubigan, applied in the spring, fall, or spring and fall and Banner Maxx applied in the fall have reduced SDS in the bermudagrass fairway study. Observations from the past two growing seasons indicate the N source applied at 1 lb N/1000 sq ft/month does not influence SDS the following spring.

Summary Points
- Spring, fall or spring and fall applications (2008) of Rubigan and single fall applications of Eagle or Banner Maxx significantly reduced SDS severity and incidence in the spring 2009.
- N source did not affect SDS severity or incidence in 2009.
- Spring green-up and turfgrass quality were similar for all treatments except in May 2009.
- Turf recovery from spring dead spot symptoms was evident in mid-June 2009.

Spring dead spot symptoms in a ‘Tifway’ bermudagrass fairway.
Developing Best Management Practices for Bermudagrass Control in Zoysiagrass Fairways

Scott McElroy and Mark Doroh
Auburn University

Objectives:
1. Evaluate the best-integrated practices for fairway conversion of bermudagrass (Cynodon dactylon) to 'Zorro' zoysiagrass (Zostera marina) turf through various cultural and chemical methods.
2. Evaluate the influence of various cultural practices on zoysiagrass competitiveness with bermudagrass.
3. Evaluate the competitive effects of various weed species on seeded 'Zenith' zoysiagrass (Z. japonica).
4. Evaluate new aryloxyphenoxypropionate (AOPP) herbicides for control of bermudagrass in zoysiagrass turf.

Turf renovation is increasingly popular where golf course superintendents wish to convert from older bermudagrass fairways to more desirable zoysiagrass. A successful renovation requires that the existing grass species be effectively controlled to permit the establishment of a monoculture of the preferred species. The efficacy of nonselective herbicide applications for bermudagrass control prior to establishment has been reported to vary. Regrowth of undesirable bermudagrass is a major problem with turf renovations, especially when converting to a different grass species. Severe contamination often leads to a poor turfgrass stand due to differences in color, texture, and growth-habit. Research is being conducted to evaluate integrated practices using herbicides and soil sterilants to convert a 'Tifway' bermudagrass fairway to 'Zorro' zoysiagrass.

The fairway conversion study was initiated in May 2009. Plots measured 5' X 10' and were arranged in a randomized complete block design with four replications. 'Zorro' zoysiagrass was sprigged at a rate of 10 bushels/1000 ft². At 15 weeks after establishment, EPTC and dazomet treatments applied in combination with glyphosate or siduron controlled bermudagrass well, yielding less than 15% bermudagrass cover. Zoysiagrass cover was greater than 80% at this rating using these treatments. Conversely, siduron and glyphosate controlled bermudagrass poorly (70% cover) and resulted in less than 30% zoysiagrass cover.

To evaluate the influence of cultural practices on zoysiagrass competitiveness with bermudagrass, a standard cup cutter was used to transplant common and 'Tifway' bermudagrass plugs into 'Zorro' zoysiagrass plots. Treatments included increasing rates of nitrogen with and without trinexapac-ethyl. Plugs were rated monthly for maximum diameter spread. At one month after the initial treatment, there were no significant differences among treatments. At two months after the initial treatment, trinexapac-ethyl resulted in the smallest diameter spread of 'Tifway' bermudagrass. Trinexapac-ethyl + 1.0 lb/1000 ft² nitrogen applied to common bermudagrass significantly increased diameter spread compared to other treatments.

Greenhouse studies were initiated in October 2009 to evaluate the competitive effects of crabgrass (Digitaria spp.) and goosegrass (Eleusine indica) on seeded 'Zenith' zoysiagrass (Z. japonica) establishment. The experiments are an additive design where 'Zenith' zoysiagrass seeding rate is held constant while weeds per unit area were increased. Plant counts and developmental stage will be observed weekly until 8 weeks after seeding. A final harvest and analysis of zoysiagrass dry weights among treatments will also be taken.

The evaluation of new AOPP herbicides for bermudagrass control study was initiated in June 2009. Three sequential applications of clodinafop, fenoxaprop, and metamifop applied alone and in combination with triclopyr were made at 3-week intervals. Plots were visually rated weekly for bermudagrass control and zoysiagrass injury using a 0-100% scale. Three weeks after the final application, clodinafop, fenoxaprop, and metamifop applied alone controlled bermudagrass poorly (less than 60%). Additionally, clodinafop and fenoxaprop caused unacceptable injury to zoysiagrass (greater than 30%). All treatments tank-mixed with triclopyr significantly reduced injury to zoysiagrass and increased bermudagrass control.

Summary Points

- EPTC and dazomet applied with glyphosate or siduron were the most effective practices for converting bermudagrass to zoysiagrass.
- Greenhouse studies have been initiated to assess the competitive effects of crabgrass and goosegrass on seeded zoysiagrass establishment.
- Fenoxaprop, and unlabeled clodinafop and metamifop, tank-mixed with triclopyr are safe to apply to zoysiagrass and effectively control bermudagrass.
Salinity Management in Effluent Water Irrigated Turfgrass Systems

Yaling Qian and David Skiles
Colorado State University

Objectives:

1. To determine spatial and temporal salinity accumulation patterns in soil profiles on golf course fairways with effluent water irrigation.
2. To evaluate different management practices for reducing sodium and salt accumulation in the soil.

Start Date: 2008
Project Duration: three years
Total Funding: $82,459

The constituents of recycled wastewater are complex and are dependent on the source. The main constituents include total dissolved salts, nutrient elements, and small quantities of organic compounds. Golf course superintendents often have concerns about salinity accumulation when effluent water is used for irrigation. Research studying salinity accumulation patterns at field scales has been limited. Real-time soil salinity and soil water content information would provide turf professionals with insight when trying to manage turf under effluent irrigation.

Currently, we are using two types of soil sensors to monitor soil water content, soil salinity, and soil temperature changes real-time and in ground. Twenty-four 5TE sensors (dielectric permittivity sensors) were installed on two fairways at the Heritage at Westminster Golf Course in Westminster, Colorado. At each fairway, 6 plots were established. On each plot, two 5TE sensors were installed at 15 cm and 30 cm below soil surface. A total of 12 sensors were installed on each fairway. Wire leads from each sensor were buried and connected to a data logger located at the edge of the fairway.

Sensor-measured soil electrical conductivity (EC) was compared to conventional saturated paste extracted soil EC to assess data accuracy. Significant linear correlation was observed between sensor-measured soil salinity vs. saturated paste extracted soil salinity (r = 0.77). This system has been collecting salinity, moisture, and temperature data from August to November 2008 and from March to November 2009.

Soil salinity at 15-cm depth ranged from 2 to 6 dS/m for Fairway 1 and from 1 to 4 dS/m for Fairway 10. The higher than average precipitation in 2009 reduced soil salinity from 3.0 dS/m to 1.5 dS/m measured at 30 cm below soil surface. The plots with higher soil salinity were at the edges of the fairways and likely experience increased compaction from golf cart traffic.

The plots with higher salinity also had greater percentages of clay content. The irrigation uniformity of study sites was 90%. Accumulation of salts appears to relate to precipitation patterns, soil texture, the degree of compaction, and drainage effectiveness.

At Heritage Golf Course, 9 putting greens and 9 fairways were selected for regular soil testing every 2-3 years for over a 10-year period since the initial conversion to effluent water irrigation. Ten years of soil testing data show a linear correlation for the increase of particular Mehlich III extractable elements. For example, soil sodium content increases overtime with R² of 0.84. In addition, we have also seen increases in Mg (R² = 0.81), Fe (R² = 0.81) extractable boron (R² = 0.27), and phosphates (R² = 0.80).

Increases in fairway soils ion content were observed for extractable boron (R² = 0.57), phosphates (R² = 0.57) and exchangeable Na (R² = 0.28).

To evaluate different management practices in reducing sodium and salts accumulations in the soil, calcium products (pelletized gypsum and liquid calcium chloride) and wetting agents (Dispatch and Primer Select) were applied to plots adjacent to the 5TE installations throughout the 2009 season. The calcium products were applied twice as a spring and fall application. Wetting agents were applied on 14-day rotation. Soil samples were taken before application and will be collected four weeks following the final fall application.

In 2009, we installed 6 Turf Guard sensors at the Common Ground Golf Course, a newly remodeled course that has transitioned to using effluent irrigation. Turf Guard sensor measurements can be accessed via a web-based interface. In lab testing, Turf Guard sensors showed strong correlation to a conventional saturated paste extracted soil EC measurement (r=0.73).

Summary Points

- Salinity and sodicity are among the concerns associated with effluent water irrigation.
- Fairways and putting greens irrigated with effluent water exhibited increased extractable sodium and phosphorus.
- Significant linear correlation was observed between 5TE salinity sensor-measured soil salinity, Turf Guard measured soil salinity vs. saturated paste extracted soil salinity.
- Accumulation of salts appears to relate to precipitation patterns, soil compaction level, soil texture, and drainage.
Cultural Practices, Environment, and Pathogen Biology: Studies for Improved Management of Large Patch of Zoysiagrass

Megan Kennelly, Jack Fry, Rodney St. John, and Dale Bremer
Kansas State University

Objectives:

1. Determine the effects of aeration, verticutting, and sand topdressing on large patch and investigate the biology of the interaction of cultural practices and disease.
2. Determine the effects of nitrogen source and time of application on disease development.
3. Study the environmental conditions associated with disease development in the field.
4. Compare large patch susceptibility of 34 new freeze-tolerant zoysiagrass genotypes.
5. Study the effects of different preventative fungicide application timing and correlate with weather conditions to develop better guidelines for fungicide deployment.

Start Date: 2008
Project Duration: three years
Total Funding: $46,806

Large patch, caused by *Rhizoctonia solani* AG 2-2, is the most common and severe disease of zoysiagrass in the transition zone. Knowledge is lacking about the interaction of cultural techniques, weather, and disease development. We are conducting field experiments at several sites to investigate these interactions. In preliminary experiments, spring aeration, verticutting, and sand topdressing surprisingly led to higher levels of large patch. As work progresses, we will better elucidate the influence of cultivation practices on large patch, and we will monitor the effects of weather on disease development. We will also investigate fungicide application timing and correlate it with environmental data to develop a model for optimal fungicide deployment if fungicides are used.

In 2009, we carried out the second year of cultural and fertility practices for Objectives 1 and 2. Plots were established at three sites (Manhattan, Olathe, and Haysville, KS). At all three sites, the experiments are set up as a split-plot with four replications. The main treatment plots are 12 x 20 feet. The main treatments are cultivation (aeration + verticutting + topdressing) vs non-cultivated. The subplot (12 x 10 feet) is fertility, either spring + fall or summer fertilization. For the spring + fall treatment, plots were treated with 1 pound N/1,000 ft² as urea (46-0-0), in both spring and fall.

The summer treatment was 2.0 pound N/1,000 ft² as polymer coated urea. To induce disease development, all plot areas were inoculated in September 2008 by taking out small turf cores, inserting *R. solani*-infested oats, and replacing the cores. Patch size was determined in spring 2009 by measuring two diameters of each patch (north-south and east-west) and taking an average.

There were no differences in thatch-level temperature between cultivated and non-cultivated plots. Water content was slightly lower in the cultivated plots. This supports our hypothesis that cultivation will reduce moisture and possibly reduce disease pressure.

We initially propagated 34 zoysiagrass lines in the greenhouse. Due to ongoing progress in another study by Dr. Fry, the lines of interest were narrowed down to 20. That is, in Dr. Fry's other study examining cold tolerance, quality, color, texture, etc, the "progeny of interest" have been reduced. The zoysiagrass is propagated vegetatively. Inoculations were performed once the turf was established for 5 months. The inoculations were conducted in a growth chamber and sheath blight symptoms were rated for disease severity.

Seven of the lines evaluated in the growth chamber had disease severity value less than that obtained for ‘Meyer’, the most commonly used zoysiagrass cultivar in our region. The growth chamber study will be repeated in early 2010. Analysis of field inoculations is in progress, and we will compare field and growth chamber studies.

Experiments were conducted as randomized complete block designs with four replicates for each treatment in an inoculated stand of zoysiagrass at the Rocky Ford Turf Center. Prostar (flu-tolanil) was applied on various dates and disease incidence was measured in spring 2009. For single applications, the best results were obtained with applications on September 16 and 23.

Summary Points

- Increase of patch size was highest in non-cultivated plots with spring+fall fertility.
- Water content was lower in cultivated plots, possibly reducing disease severity.
- Growth chamber work indicates some variation in susceptibility. Experiments will be repeated and compared with field studies.
- Differences in disease control were observed with different fungicide application timings.
The oriental beetle (OB) is the most important turfgrass insect pest in New Jersey, Connecticut, Rhode Island, and southeastern New York. Our overall objective is to investigate the dispersal biology of oriental beetle adults in order to improve the efficacy of mating disruption to control OB grubs in turfgrass.

Females should not be affected by their sex pheromone but may mate outside of pheromone-treated areas and migrate into treated areas to deposit eggs. However, female dispersal studies are hampered by the lack of female attractants and the fact that females are active around dusk. Females placed into the turfgrass between 5 and 7 pm in early July mostly dug into the soil or crawled short distances. Females that alighted flew 3-12 feet, after which they either did not alight again or could not be found. However, 1:1 sex ratios in black light traps captures suggested that female may fly significant distances.

OB male attraction to different pheromone sources was investigated in release and recapture field studies. Trécé Japanese beetle traps were placed in the ground with only the funnel part above ground in turf areas that had been treated with Merit in the previous year. The traps were lured with a virgin female placed in a metal mesh cage or with red rubber septa loaded with 0.3, 1, or 3 µg sex pheromone. Recapture rates of color-marked males released 6.25 to 100 ft downwind from the traps were determined after 24 hours. Recapture rates were higher for the 3 µg and 1 µg septa than for the 0.3 µg septa and the virgin females and declined logarithmically with distance for all pheromone sources.

Based on regression lines, 0% recovery for virgin female and 0.3, 1, and 3 µg septa was predicted at 92, 84, 140, and 842 ft, respectively. Capture of unmarked males from the background population was higher for 3 µg septa than 0.3 µg septa, with 1 µg septa and virgin females not significantly different from either.

We also compared male capture in Trécé Japanese beetle traps and cages each lured with a virgin female, or a red rubber septum loaded with 0.3, 1, or 3 µg pheromone. After 3 days exposure in a naturally infested turf area, OB male capture was more than 10-fold higher in traps than cages and was higher with the 3 µg and 1 µg lures than the 0.3 µg lures and virgin females. These data suggest that the use of virgin females can be substituted with the use of 0.3 µg lures.

Summary Points
- OB males are attracted to pheromone lures and formulation pellets from at least 200 feet distance, but attraction to females is weaker and usually under 100 ft.
- A 1:1 sex ratio in black light trap captures suggest that female OB may disperse similarly as males. Limited direct observations suggest limited dispersal of females over short periods.
- Traps used to evaluate the efficacy of mating disruption should use lower pheromone loads than previously used (0.3 µg vs. 30-300 µg) and red rubber septa with low pheromone loads (0.3 µg) could replace virgin females in cages used to evaluate mating disruption.
Inland saltgrass is a native warm-season grass which has excellent tolerance to water and soil salinity, drought, submer- sion, and sodic soils. As part of the domestica- tion process, it is necessary with any new species to determine a plant-pest profile.

Since all turfgrasses are suscepti- ble to weed invasion by annual and peren- nial plant competitors, herbicide tolerance is an important issue for proper turfgrass management. Typical responses of turfgrass to herbicides can include leaf tip burning, leaf bronzing, and general loss of color, yellowing (chlorosis), leaf necrosis, loss of stand density, or other plant growth regulator effects.

A six-year-old stand of ‘A49’ and ‘A50’ saltgrass was used for this test. The turf was mowed at 3.0 inches two to three times weekly, and irrigated to prevent drought stress. Twenty different compounds were selected representing different chemical families which are typically used for post-emergence grass, sedge, and broadleaf removal in warm-season turfgrasses.

These products were applied at their 1X maximum single application label rate, and also as a 2X spray solution application rate, which occurs in field spray overlaps. Methylated seed oil (MSO) or a 90% non-ionic surfactant were included with herbicides only when recommended by the product label.

Saltgrass plots were scored weekly for the percent of the total plot surface that produced necrotic leaves/shoots (percent plot straw) and the intensity of necrosis itself (degree of expression). All plots were also rated for overall turfgrass color, quality, and on one occasion, visual density using the NTEP rating visual assessment scoring system (1 = dead, 6 = acceptable, 9 = best possible).

Negative response to a particular herbicide usually occurred in the form of an enhancement of necrotic straw production, which usually reduced turf plot color, but always affected turfgrass quality. A notable exception was Banvel (dicamba), which slightly decreased the overall color of the turf (at 21 DAT), but otherwise did not affect overall quality and did not show an enhanced amount of canopy straw.

Note that saltgrass has the tendency on its own, to retain lower leaves on its stems as part of the "natural" senescence process. This is noted even on the non-treated controls, which themselves always had some "straw" present. The overall effect on straw was reflected in the overall visual quality scores of the turf, which was used as the deciding factor in herbicide safety.

Both the non-ionic surfactant (applied alone at 0.5% v/v and at 1.0%) and MSO (applied at 1% and 2% applications) were safe. These treatments will be applied again in 2010 in order to determine consistency of response.

<table>
<thead>
<tr>
<th>Summary Points</th>
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<tr>
<td>🟦 Commonly used turfgrass herbicides were screened on saltgrass (Distichlis spicata) for tolerance at 1X and 2X label rates.</td>
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<td>🟦 Herbicide product treatments which were generally regarded as safe for saltgrass at rates/volumes applied here included:</td>
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<tr>
<td>Corsair (1X and 2X)</td>
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<td>Manor (1X and 2X)</td>
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<td>Drive XLR8 (1X)</td>
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<td>Certainty (1X and 2X)</td>
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<td>Velocity (1X and 2X)</td>
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<td>Quicksilver (1X and 2X)</td>
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<td>Dismiss (1X and 2X)</td>
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<td>2,4-D amine (1X)</td>
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<td>Banvel (1X)</td>
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<td>Image (1X)</td>
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<td>Illoxon (1X)</td>
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<td>Buctrill (1X and 2X)</td>
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<td>MSMA (1X and 2X)</td>
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<td>NIS (1X and 2X)</td>
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<td>MSO (1X and 2X)</td>
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<td>Revolver (1X)</td>
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<tr>
<td>🟦 Treatments which caused a slight decrease in quality for either a brief period of time or had marginal performance for at least two of the five rating events included:</td>
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<tr>
<td>Drive XLR8 (2X)</td>
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<td>Revolver (2X)</td>
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<td>Banvel (2X)</td>
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<td>Illoxon (2X)</td>
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<td>Spotlight (2X)</td>
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<tr>
<td>2,4-D (2X)</td>
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<tr>
<td>🟦 Treatments which caused an extreme loss of color, an enhanced straw condition, and subsequent loss of quality to below acceptable levels for at least two rating periods (over a 14-day period) included:</td>
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<tr>
<td>Monument (1X and 2X)</td>
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<td>Katana (1X and 2X)</td>
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<td>Tranxit (1X and 2X)</td>
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<td>Plateau (1X and 2X)</td>
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Screening Herbicides for Phytoxicity on Saltgrass (Distichlis spicata) Turf

Tony Koski
Colorado State University

Objectives:
To evaluate commonly used postemergence herbicides for tolerance and phytotoxicity on saltgrass maintained as a turf.

Start Date: 2008
Project Duration: two years
Total Funding: $6,000

Inland saltgrass, a native warm-season grass, has demonstrated excellent potential for turf use where adverse growing conditions make the use of traditional turf species difficult or impossible. This species possesses excellent salinity, heat, submersion, and traffic tolerance. Selections from more northern latitudes and higher elevations also display excellence cold tolerance.

Before cultivars of a new turf species are released for public use, it is essential that the cultural and management requirements of the new grass be studied. This includes an understanding of its response to applications of the most commonly used turf management herbicides. This study screened 20 herbicides commonly used for weed control in warm-season turf. The response of saltgrass to the herbicides (1X and 2X rates) was evaluated by rating for typical phytotoxic responses to herbicides, including discoloration (bronzing, yellowing, browning, burning) and/or loss of density.

A 4-year-old saltgrass stand (accession A138, established from plugs) was used for this test. The turf was mowed at 2.5 inches once or twice weekly, and irrigated to prevent drought stress. Twenty different compounds were selected representing different chemical families which are typically used for postemergence grass, sedge, and broadleaf control in warm-season turfgrasses. These products were applied at their 1X maximum single application label rate. They were also applied at a 2X rate, to simulate what occurs in overlaps during field application. Methylated seed oil (MSO) or a non-ionic surfactant (NIS) were used when recommended by the product label.

Plots were rated weekly for color, quality, density using the NTEP rating visual assessment scoring system (1 = dead, 6 = acceptable, 9 = best possible). Treatments were applied on August 22, with turf ratings conducted weekly from August 29 until September 25. Very cold temperatures during the week of September 20, including a light frost on September 21, caused the saltgrass in this study to become partially dormant.

With the exception of Monument, Katana, and TranXit, which caused a substantial and prolonged decrease in turf quality and color for more than half of the rating period at their respective 1X application rates, all other herbicides applied at the 1X rate in this trial in Fort Collins CO appeared to be moderately or totally safe for use on saltgrass. The 2X rates of Plateau, Drive XLR8, 2,4-D, Spotlight, Banvel, Revolver, and Illoxon did cause unacceptable injury lasting two weeks or more in this study, suggesting that these products be used with caution (and perhaps at lower than maximum label rates) to avoid the potential for turf injury in overlap areas.

The saltgrass plots used in this study had considerable natural browning evident prior to the application of the herbicides. Saltgrass shoots tend to retain lower leaves that are in the process of senescing, often giving the turf a somewhat brownish cast. This characteristic is less evident when turf is mowed at lower heights, perhaps because the lower (and more frequent) mowing increases turf density which essentially hides the dead leaves.

Summary Points

- Twenty commonly used postemergence herbicides were applied to established saltgrass turf in Fort Collins CO in late August, at 1X and 2X maximum allowable label rates for warm-season turf (bermudagrass or zoysiagrass). Their relative safety, as determined by visual rating of turf color, quality and density are:
  - Safe (no injury or minor phytotoxicity lasting less than one week) at 1X or 2X label rates:
    - Corsair 75 WDG
    - Manor 60 WDG
    - Image 70 DG
    - Sedgehammer 75 WDG
    - Buctril
    - Certainty 75 WDG
    - Velocity 17.6 SG
    - Quicksilver T&O
    - Dismiss
  - Safe at 1X maximum label rate; injury at 2X maximum label rate lasting more than two weeks:
    - 2,4-D amine
    - Plateau
    - Illoxan
    - Drive XLR8
    - Revolver
    - Banvel
    - Spotlight
  - Not Safe (significant injury lasting more than two weeks) at either 1X and 2X maximum label rates:
    - Katana 75 WDG
    - TranXit 25 DF
    - Monument 75WG

Saltgrass plots at Colorado State University, Fort Collins, CO.
Zoysiagrass (Z. japonica or Z. matrella) is increasing in popularity and availability with over 30 cultivars now commercially available. Zoysiagrass has historically been more widely utilized on golf courses in the upper transition zone. However, there has been a recent trend to plant zoysiagrass on golf courses in the lower transition zone and farther south. While use has increased, zoysiagrass is typically considered an alternative turfgrass for golf courses. Knowledge regarding the management of these new cultivars is critical as they are marketed and recommended for use.

Previous research in Texas found that turfgrass quality during summer was improved with higher nitrogen (N) rates, especially at the lower mowing height. Additional research has focused on mowing heights or fertility, but not a combination of the two. In South Carolina, Z. matrella had excessive thatch and scalping at high N rates (greater than 3 lbs N/1000 ft²), but thatch was not problematic in Z. japonica. Others concluded that 2 lb N/1000 ft² or less during the growing season was adequate in Missouri.

Experimental areas were sprigged in 2001 at the Arkansas Agricultural Research and Extension Center, Fayetteville, AR with ‘El Toro’, ‘Meyer’, and ‘Cavalier’ zoysiagrass. Plots were maintained from 2002-2007 using 1-2 lb N/1000 ft²/year. Fertilization treatments were initiated in May 2008 using sulfur-coated urea at 0, 2, 4, and 6 lbs N/1000 ft²/year applied on May 1, June 1, July 1, August 1, and September 1. Response was evaluated as turf quality, density, green-up, and scalping.

Separate areas of established ‘Meyer’ and ‘Cavalier’ zoysiagrass were used at the same location. Three nitrogen sources, urea, ammonium nitrate, and calcium nitrate, were applied as 2.0 and 4.0 lbs N/1000 ft²/year with each source. Application timings were the same as Study 1. Response was evaluated as turf color, quality, density, green-up, and scalping.

Results indicate that turf density is improved through cultivar selection and N fertility. ‘Cavalier’ consistently had greater turf density than ‘Meyer’ and ‘El Toro’. Increasing annual nitrogen applications to at least 2 lbs N/1000 ft²/year also improved turf density. In the spring of 2009, N rates to at least 4 lbs N/1000 ft²/year were observed to cause a delay in spring green-up and a decline in turf quality at the 1.5-inch mowing height. Turf quality was generally highest for ‘Meyer’ and ‘Cavalier’. Turf quality was never unacceptable (less than a 6 rating) for the unfertilized check plots in either year.

There was no advantage to fertilizing more than 2 lbs N/1000 ft²/year. Turf quality was never unacceptable for the unfertilized check plots.

Nitrogen source did not affect turf quality in the field.

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Experimental areas were sprigged in 2001 at the Arkansas Agricultural Research and Extension Center, Fayetteville, AR with ‘El Toro’, ‘Meyer’, and ‘Cavalier’ zoysiagrass. Plots were maintained from 2002-2007 using 1-2 lb N/1000 ft²/year. Fertilization treatments were initiated in May 2008 using sulfur-coated urea at 0, 2, 4, and 6 lbs N/1000 ft²/year applied on May 1, June 1, July 1, August 1, and September 1. Response was evaluated as turf quality, density, green-up, and scalping.

Separate areas of established ‘Meyer’ and ‘Cavalier’ zoysiagrass were used at the same location. Three nitrogen sources, urea, ammonium nitrate, and calcium nitrate, were applied as 2.0 and 4.0 lbs N/1000 ft²/year with each source. Application timings were the same as Study 1. Response was evaluated as turf color, quality, density, green-up, and scalping.

Results indicate that turf density is improved through cultivar selection and N fertility. ‘Cavalier’ consistently had greater turf density than ‘Meyer’ and ‘El Toro’. Increasing annual nitrogen applications to at least 2 lbs N/1000 ft²/year also improved turf density. In the spring of 2009, N rates to at least 4 lbs N/1000 ft²/year were observed to cause a delay in spring green-up and a decline in turf quality at the 1.5-inch mowing height. Turf quality was generally highest for ‘Meyer’ and ‘Cavalier’. Turf quality was never unacceptable (less than a 6 rating) for the unfertilized check plots in either year.

There was no advantage to fertilizing more than 2 lbs N/1000 ft²/year. Turf quality was never unacceptable for the unfertilized check plots.

Nitrogen source did not affect turf quality in the field.

University of Arkansas scientists apply fertilizer treatments to field plots of zoysiagrass in Fayetteville, AR. Results of these studies indicate that there is no advantage to using more than 2 lbs N/1000 ft²/year.
Breeding, Genetics, and Physiology

The quality and stress tolerance of turf is a product of the environment, management practices, and genetic potential of the grass plant. In many cases, major limitations to turf quality are stress effects, many of which can be modified or controlled through plant improvement. Projects are directed toward the development of turf cultivars that conserve natural resources by requiring less water, pesticides and fertilizers. Research projects that apply new biotechnological methods toward turfgrass improvement are considered. Among the characteristics most desirable in the new turfgrasses are:

1. Reduced need for pesticides by increasing resistance to disease, insects, nematodes, and weed encroachment
2. Increased shade tolerance
3. Reduced requirements for mowing, irrigation, and fertilization
4. Tolerance of non-potable water
5. Ability to survive high- and low-temperature extremes
6. Increased drought tolerance
7. Tolerance of intensive traffic
8. Tolerance of poor quality soils.

Research in the fields of biotechnology, genetics, cytogenetics, cytology, entomology, genetics, microbiology, nematology, pathology, physiology, and other sciences that support the project objectives and provide improved techniques for improving golf turf species will be considered.

Locations of the following projects funded in 2009 by the USGA Turfgrass and Environmental Research Program under the category of Breeding, Genetics, and Physiology
A Bentgrass Breeding Consortium to Support the Golf Industry

Michael Casler  Geungwha Jung  Scott Warnke  Stacy Bonos and Faith Belanger  Suleiman Bugharra
University of Wisconsin  University of Massachusetts  USDA-ARS  Rutgers University  Michigan State University

Objectives:

1. To develop elite clones of creeping bentgrass with multiple pest resistances and stress tolerances that can be delivered to the seed industry for use in synthesizing new creeping bentgrass cultivars broadly adapted to a range of ecological and environmental conditions including reduced pesticide application.

Activities for 2009 focused on field evaluations of dollar spot and snow mold on progeny from elite crosses, extraction of DNA from leaves, and the initial assay of DNA markers. Replicated field evaluations were established in Massachusetts, New Jersey, and Maryland in 2007 and 2008 and evaluated in 2009.

In 2010, data will be collected on disease resistances of field-grown plants and DNA markers. Selected plants will be used to create new hybrid plants, progeny populations, and releases of individual clones or genotypes to bentgrass breeders who are developing new varieties.

Summary Points

- Creeping bentgrass plants with improved resistance to both dollar spot and snow mold fungi have been identified and propagated.
- These plants will be released to private turf breeders for use in developing new and more disease-resistant creeping bentgrass varieties.
- This research has contributed to the identification of specific genes for resistance to snow mold and dollar spot diseases, which can be used to design more efficient and effective breeding methods based on using DNA marker technologies. These methods will be tested in the current phase of this project.
Conserving water in the landscape is critical to inhabiting the arid portions of the western United States. Native accessions of the inland form of saltgrass (*Distichlis spicata* var *stricta* (Torr.) Beetle) remained green, while turfgrass lines of blue grama, buffalograss, crested wheatgrass, and bermudagrass went dormant from lack of rainfall during the drought of 2000 and 2001.

Since saltgrass is non-domesticated, this research improves native germplasm by selecting for specific traits in order to make saltgrass more turf-like. We continued evaluation of the C2 population, with measurements of height, rust rating, general verdure, leaf shredding, and seed yield. In addition, this year we screened turf types for salinity tolerance.

For salinity screening, the germplasm was approximately 70 lines (from 3,000 lines) in the C2 nursery that had been chosen for general turf quality in 2008 (high shoot density, short height, and desirable appearance under drought). Line COAZ-01 was used as a check since Qian had found it, compared to other accessions, to have high turf quality, low leaf firing, high root growth, and high root viability across a range of salinity levels (12, 24, 36, 48 mmhos/cm).

Four plugs (16 cm in diameter and 16-cm deep) were dug for each line, potted, and acclimated for four weeks in tubs maintained at 2.5-cm depth of tapwater (0.8 mmho/cm, mountain stream source). Since saltgrass is a phreatophyte, sitting in water is assumed to have little effect on growth. At the end of four weeks, two plugs of each line were placed in salinity (12 mmhos/cm derived from commercial aquarium sea salt) tubs, with the solution at a 2.5 cm depth. Salinity was monitored and maintained daily. Every three days, a pump was used to manually water the surface of the pots to equilibrate the soil salinity with the saline solution.

After two weeks, the saline solution was raised to 36 mmhos/cm. Several days afterwards, many plants took on a darker green appearance than their counterparts in the tapwater tubes. We increased the salinity solution over two weeks to 48 mmhos/cm. Two weeks after the 48 mmho/cm threshold, plants in the saline treatments were compared to their counterparts in the tapwater treatment and assigned a number based on:

1. green = control color
2. darker green
3. darker green with leaf tips burned
4. attributes in 3 and dieback of male or female head shoots
5. half or more of all shoots dieback.

Eight lines showed no change in color, and were significantly different than the bottom ranked 23 lines. A total of 26 lines ranked higher than COAZ-01, which is considered to have excellent salinity tolerance. These results suggest a correlated response in salinity tolerance by selecting for turf traits (short height, rust resistance, shoot density, and seed yield) in saltgrass. Plants ranked high in salinity tolerance will be considered for parents.

We use a selection index in order to rank plants for selection as parents. In the first cycle, this took the form of the selection value of an individual line = (standardized spike numbers) + (standardized shoot density) - (standardized height) - (standardized rust rating). Standardization equalizes the scale between traits. The top 30-40 lines are then selected as parents.

Parents will be brought into the greenhouse this winter and induced to flower, and all crosses will be made between parents. Seed will be harvested, germinated in April in growth chambers, and theses will form the C3 nursery. Seed of elite crosses will be germinated in growth chambers over winter, grown out in the field, and observed for segregation of turf traits.

**Summary Points**

- Salinity tolerance screening resulted in 26 lines ranked higher than the previously preferred check.
- Parents are selected for winter crossing to produce a progeny population with improved turf traits.
- The breeding population with a genome of 38 chromosomes is being maintained.

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**Development of Seeded Turf-type Saltgrass Varieties**

**Dana Christensen and Yaling Qian**

**Colorado State University**

**Objectives:**

1. Evaluate C2 population. Select parents from these and intercross for the Cycle 3 population.
2. Evaluate segregation of elite crosses.
3. Evaluate inbreeding of closely related crosses.
4. Screen parents for ploidy level.
5. Screen for salinity tolerance.

**Start Date:** 2006  
**Project Duration:** three years  
**Total Funding:** $78,822

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**Plants on the left were at 48 mmhos/cm for two weeks and assigned a number 4 class. Plants on the right were grown with tapwater. All pots are the same genotype.**

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**2009 USGA Turfgrass and Environmental Research Summary**
As of October 30, 2009, over 1,800 promising turfgrasses and associated endophytes were collected in Italy, Atlas Mountains in Morocco, France, and Spain. These have had seed produced in The Netherlands and will be evaluated in New Jersey. Over 9,435 new turf evaluation plots, 120,000 spaced-plant nurseries, and 4,000 mowed single-clone selections were established in 2009.

Over 340,000 seedlings from intra- and inter-specific crosses of Kentucky bluegrass were screened for promising hybrids under winter greenhouse conditions and the superior plants were put into spaced-plant nurseries in the spring. Over 14,000 tall fescues, 6,000 Chewings fescues, 7,600 hard fescues, 60,000 perennial ryegrasses, and 12,000 bentgrasses were also screened during the winter in greenhouses, and superior plants were put in spaced-plant nurseries. Over 472 new inter- and intra-specific Kentucky bluegrasses were harvested in 2009.

The following crossing blocks were moved in the spring of 2009: 4 hard fescues (188 plants), 2 Chewings fescues (69 plants), 21 perennial ryegrasses (899 plants), 2 strong creeping red fescues (102 plants), 16 tall fescues (626 plants), 4 velvet bentgrasses (66 plants), 5 creeping bentgrasses (93 plants), and 6 colonial bentgrasses (125 plants).

To enhance our breeding for resistance to gray leaf spot, over 900 large pots of perennial ryegrass (selected the previous summer) were moved to a greenhouse in November. Increased daylengths were used in the spring to provide an early harvest of these pots. Harvest was one month early, and a new perennial ryegrass trial was seeded on July 9, 2009. Within 30 days, a severe gray leaf spot epidemic occurred and recombinations of the most resistant clonal sources will be made in 2010. Three new gray leaf spot-resistant ryegrasses released in 2009 were 'Revenge GLX', 'Palmer V' and 'Linedrive GLS'.

The breeding program in bentgrasses made good progress in finding new sources of turfgrass germplasm. The soon to be released 'Capri' colonial is an improved variety for this disease. Emphasis was also put into breeding for anthracnose and copper spot resistance in creeping, colonial, and velvet bentgrasses. 'Pinup' is a new improved creeping bentgrass increased in 2009.

'Capri' colonial bentgrass is an improved variety with better brown patch resistance, and 'Pinup' creeping bentgrass has improved dollar spot resistance.

Summary Points

- Continued progress was made in obtaining new sources of turfgrass germplasm. These sources are being used to enhance the Rutgers breeding program.
- Modified population backcrossing and continued cycles of phenotypic and genotypic selection combined with increasing sources of genetic diversity in turfgrass germplasm has resulted in the continued development and release of top performing varieties in the NTEP.
- Fifteen new tall fescues, 3 fine fescues, 5 Kentucky bluegrasses, 2 bentgrasses, and 3 perennial ryegrasses were released in 2009.
- 'Capri' colonial bentgrass is an improved variety with better brown patch resistance, and 'Pinup' creeping bentgrass has improved dollar spot resistance.
- Published or have in press over 10 referred journal articles in 2009.
Magnaporthe grisea. Gray leaf spot has been infected with gray leaf spot caused by a valuable cool-season turfgrass that can stand within a few days. Breeding rye-grass (GLS) can completely destroy ryegrass home lawns in the US. Gray leaf spot is a problem on perennial ryegrass in golf course fairways and on becomes a serious problem on perennial ryegrass cultivars. The immediate attention of host resistance in perennial ryegrass needs to the interaction between pathogen variability and perennial ryegrass resistance.

Our objective is to test whether there is a significant interaction between GLS pathogen variability and perennial ryegrass resistance. The two ryegrass parent clones (MFA, MFB) and five commercial resistant cultivars (‘Gray Star’, ‘Gray Fox’, ‘Grey Goose’, ‘Manhattan-5’, ‘Paragon GLR’) were included in the study. Two GLS resistant plants, Paragon GLR-1R, Paragon GLR-2R and two susceptible plants, Paragon-3S and Paragon-4S were selected from individual plants of ‘Paragon GLR’ based on a previous inoculation experiment.

Ryegrass plants germinated from seeds of the commercial cultivars and lines and the selected clonal plants were grown in French Hall greenhouse at the University of Massachusetts, Amherst. The plants were inoculated with GLS isolates under growth chamber conditions. Based on two inoculations, MFA, MFB, Paragon-1R, and Paragon-2R were resistant to all GLS isolates and Paragon-3S and Paragon-4S were very susceptible as expected. All commercial cultivars and experimental lines were moderately resistant to GLS isolates, but 2NKM-1 and ‘Gray Star’ were susceptible.

Additional inoculation experiments are needed for further confirmation. In addition, individual plants (5-10 per cultivar or line) will be randomly selected from each population of the resistant cultivars and experimental lines in order to further test pathogen and host interactions on an individual plant basis.

Due to the outcrossing nature of ryegrass, all ryegrass cultivars are synthetically different. Further inoculations using clonally replicated ryegrass plants and 13 geographically diverse GLS isolates will be performed to check whether the resistance in commercial cultivars differ from MFA and MFB. DNA markers significantly associated with QTLS for GLS resistance in the various sources will be developed for marker-assisted selection. Multiple disease resistance genes including gray leaf spot, crown rust, and leaf spot can be incorporated into an elite perennial ryegrass cultivar to strengthen host resistance as part of an integrated pest management strategy for turfgrass.

Summary Points

- Significant differences in pathogenicity among 13 gray leaf spot (GG9, GG11, GG12, BL00, LP97, Lin00, 05T-04, 02V-23.1, 04S-01, 06T-02, 11W-03, and 11W-07) and one rice (6082) isolates under growth chamber conditions.
- Seven recently improved perennial ryegrass cultivars and experimental lines with GLS resistance showed only moderate resistance to the 13 geographically diverse isolates. This result may indicate non-race specific resistance in perennial ryegrass.
- Preliminary results indicated a marginally significant interaction between gray leaf spot isolates and ryegrass germplasm under growth chamber conditions.
- GLS-resistant plants from the MFA x MFB mapping population will be crossed with Paragon GLR-1R and -2R plants. Those parent plants were transplanted to the field.
Breeding and Evaluation of Turf Bermudagrass Varieties
Yanqi Wu, Dennis L. Martin, and Charles M. Taliaferro
Oklahoma State University

**Objectives:**

1. Assemble, evaluate, and maintain *Cynodon* germplasm with potential for contributing to the breeding of improved turf cultivars.
2. Improve bermudagrass germplasm for seed production potential, cold tolerance, leaf firing resistance, and other traits that influence turf performance.
3. Develop, evaluate, and release seed- and vegetatively-propagated turf bermudagrass varieties.

**Start Date:** 2006  
**Project Duration:** three years  
**Total Funding:** $90,000  

Turf bermudagrass [*Cynodon dactylon* (L.) Pers., *C. transvaalensis* Burtt-Davy, and their hybrids *C. dactylon* x *C. transvaalensis*] is the most widely used turfgrass in the southern USA and throughout tropical and warmer temperate regions of the world. The OSU turf bermudagrass genetic improvement program made progress in the enhancement of turf bermudagrass germplasm and the development of experimental cultivars in 2009. Screening of 1,080 putative *F*1 progeny plants (*C. dactylon* x *C. transvaalensis*), field established in 2006, was completed in 2009 by selecting 14 superior progeny plants after evaluating winter color retention, spring green-up, winterkill, foliage color, texture, sod density, seedhead abundance, and overall turf quality for three years. The respective selected plants are being maintained in a greenhouse at the OSU Agronomy Research Station for use in establishing a replicated performance trial in 2010.

A field trial was continued to comprehensively evaluate eight OSU experimental synthetics for turf performance traits against clonal and seeded standard cultivars at the Turfgrass Research Center in 2009. Standard field performance parameters for fairway-type bermudagrass were assessed in this trial. In addition to the trial, two 2007-2012 NTEP ancillary bermudagrass trials inoculated with *Ophiosphaerella herpotricha* and *O. korrae* were continued in 2009. Tolerance to the disease will be assessed over the next three years in these trials.

A clonal turf bermudagrass selection nursery encompassing 1,500 putative *F*1 progeny plants of *C. dactylon* x *C. transvaalensis* field transplanted last year was screened for spring green-up, foliage color, texture, sod density, overall turf performance, and winter color retention in 2009. Large variations were observed in the progeny populations for various morphological, turf performance, and adaptation traits.

A broad-based breeding population formed by polycrossing tetraploid and desirable Chinese *Cynodon dactylon* germplasm accessions was evaluated for seed yield component and morphological traits to select approximately 10% superior individual plants as parents for the development of cycle one (C1) population and to select elite parents to make new synthetic experimental cultivars in 2010. More recently, a new experiment was initiated to identify clonal turf bermudagrass cultivars using Simple Sequence Repeat (SSR) markers. Currently, more than 90 SSR markers were developed for bermudagrass and more than 200 SSR markers developed from mining and testing the expressed sequence tags (EST) of *Cynodon* in the National Center for Biotechnology Information. Three experimental lines, OKC 1119, OKC 1134, and OKS 2004-2 are in the 2007-2012 NTEP National Bermudagrass Test and exhibited outstanding performance.

**Summary Points**

- A set of 14 superior clonal bermudagrass putative hybrids were selected in 2009 from a screening nursery for next step in-house comprehensive evaluation.
- A field trial to comprehensively evaluate eight new experimental synthetics was continued.
- Spring dead spot disease tolerance evaluation was continued in 2009.
- A clonal bermudagrass nursery of 1,500 putative hybrids was evaluated for various traits in 2009.
- A new experiment was initiated to molecular identify clonal turf bermudagrass cultivars using SSR markers.
- OKC 1119, OKC 1134, and OKS 2004-2, experimental bermudagrass varieties had outstanding performance in the 2007 NTEP National Test.
In 2009, several experiments were conducted to address these objectives. Crossing block consisting of 21 single crosses were tested for crossing compatibility and seed yield potential. Significant differences were observed among the single crosses tested for yield and yield components. As the result, high yielding compatible parents were identified. Some progenies of these parents were established to assess their turfgrass performance.

Advanced lines IV consisting of 104 entries including standard checks were evaluated for spring green-up, stand density, turfgrass color, and turfgrass quality in 2009. Significant differences were observed for these traits among the genotypes tested. Evaluations will continue for a several more years to identify the best genotypes for potential future release.

Similar data were collected on selections consisting of over 1,500 entries that were obtained from collections and hybridizations. Visual differences were observed though they were not replicated and analyzed. Evaluation will continue to identify and promote genotypes with superior turfgrass performance for a future advanced line replicated trial. In 2009, an additional 129 genotypes were added to the selections evaluations from collections made from diverse environments and management conditions. Some of these genotypes have potential shade tolerance and have been placed in a shade tolerance evaluation.

Buffalograss best performance is achieved when optimum management practices such as fertilizer rates, mowing heights, and irrigation rates are identified and met for the grass. Significant differences in turfgrass color and quality were observed among buffalograss genotypes in a mowing height by nitrogen nutrition evaluation. Data from this evaluation are being summarized for publication.

Attempts to establish buffalograss turfs from sprigs have been limited and successful stand establishment has been inconsistent. A study was conducted to determine whether accumulated growing degree-days (GDD) of harvested sprigs and cultivar have an effect on buffalograss sprig establishment. Two field studies were conducted on a Tomsk silty-clay loam (fine smectitic mesic Pachic Agriudolls). 'Legacy', a hexaploid, and 'Prestige', a tetraploid, were used in this investigation.

Sprigs harvested at 1,050 GDDs resulted in the best establishment for both vegetative cultivars. Results from this study will be useful in vegetative establishment of larger turfgrass areas, like fairways and roughs, using buffalograss sprigs.

Summary Points
- Buffalograss germplasm was enriched through collection and hybridization.
- Among the elite buffalograss genotypes evaluated at diverse environments, several had wide adaptation potential.
- Best compatible high yielding buffalograss parents were identified for future hybridization and cultivar development.
- Mowing height and N nutrition rates influenced buffalograss turfgrass performance and adaptation though interactions were not significant.
- Study results support the recommendation to establish buffalograss from sprigs harvested prior to 1,050 growing-degree-days.
Genetic Improvement of Prairie Junegrass
Eric Watkins
University of Minnesota

Objectives:
1. Determine the genetic potential of native prairie junegrass (Koeleria macrantha) germplasm for use as low-input turfgrass.

Start Date: 2007
Project Duration: three years
Total Funding: $30,000

Grass species that are native to North America should be better able to cope with our environment and could lead to overall reductions in inputs such as fertilizers, pesticides, and water. Prairie junegrass (Koeleria macrantha), which is native to the Great Plains of the United States, has shown the potential to be successfully used as a turfgrass in lower-input environments.

The species is widely distributed throughout much of the western United States, and it can also be found throughout much of Europe and Asia. Based on data that has been collected in recent years, this species appears to perform well in Minnesota under low-input conditions (no irrigation, limited nitrogen application, and no fungicide or insecticide applications).

Prairie junegrass has several attributes that would make it a useful low-input turfgrass in Minnesota including tolerance of doughty and alkaline soils, tolerance of sandy areas, survival of low and high temperature extremes, and reduced growth rate. 'Barkoe' was the first cultivar of this species specifically developed for use as a turfgrass. However, this cultivar was developed with ecotypes from Europe. We are proposing the development of a cultivar primarily derived from germplasm native to North America.

Developing a high quality turfgrass is not, by itself, adequate. In order to be used by consumers, an economically viable turfgrass cultivar must be able to produce sufficient quantities of seed. Non-selected populations of the species can produce seed for 4-5 years. Collections of natural ecotypes made in 2005 suggest that individual genotypes may possess the ability to be highly productive; however, it is unknown if it can produce economically adequate amounts of seed.

In order for a cultivar of this species to be developed. Accessions showed high levels of variability for this trait at both locations. An accession from Ireland (PI 430287) out-performed accessions from other regions for both mowing quality and overall turfgrass quality. This collection is characterized by dark green color, excellent mowing quality, and superior turfgrass quality. In a related study, we found that this accession had poor seed yields when grown in Minnesota.

This study confirms that the USDA NPGS accessions can provide valuable turf quality traits; however, our previous research has indicated that few of these accessions have adequate levels of seed production when grown for seed in Minnesota. Seed production evaluations in Minnesota have also shown that material collected in the northern half of the United States has superior seed production to most non-native accessions. Combining the positive characteristics of the native and non-native populations in our breeding program may provide the best possibility for the development of a useful low-input cultivar.

Summary Points
- Mowing quality of some non-native collections is superior than native germplasm
- Many accessions have adequate turfgrass quality under low-input conditions.
- Integration of traits from diverse germplasm should be effective in the development of a low-input cultivar.
The identification of turfgrass cultivars that can tolerate irrigation with alternative water sources while maintaining safe, acceptable quality would result in a community and industry more accepting of voluntarily utilization of alternative water sources. Cultivars of perennial ryegrass, creeping bentgrass, and Kentucky bluegrass are being tested under both greenhouse and field conditions. This research will provide recommendations to golf course superintendents regarding salt tolerant cultivars, advance turfgrass breeding and genetics, as well as help conserve our natural resources.

Two greenhouse screening runs were conducted on clones from five perennial ryegrass cultivars at four salinity levels (1, 5, 10, and 15 dS/m). Significant differences were observed between salinity treatments with the highest salinity treatments causing the most injury to perennial ryegrass plants. Clones of ‘Palmer III’ exhibited the highest percent green ratings compared to other cultivars.

Two greenhouse screening runs were conducted on 21 Kentucky bluegrass cultivars at four salinity levels (1, 6, 9, and 12 dS/m). The cultivars exhibiting the highest percent green ratings were ‘Eagleton’, ‘Liberator’, and ‘Cabernet’ and cultivars and selections with the lowest percent green were a Texas x Kentucky bluegrass selection, A03TB-246, ‘Baron’, and the Kentucky bluegrass selection A03-84.

Two greenhouse screening runs were conducted on eight bentgrass cultivars at four salinity levels (1, 4, 8, and 12 dS/m). Unlike perennial ryegrass and Kentucky bluegrass, individual clones of bentgrass cultivars responded variably to salinity stress. In all greenhouse runs for all cool-season turfgrass species, there were significant differences between salinity treatments with higher salinity treatments causing more injury to turfgrass plants. Additionally, percent green ratings were highly correlated to clipping yields, root weights, and shoot weights for all species.

Twenty-one Kentucky bluegrass cultivars, 22 perennial ryegrass cultivars, and 15 bentgrass cultivars were established in the fall of 2006 and spring of 2008. They were evaluated for salt tolerance in the summer of 2007, 2008, and 2009 by treating with a salt solution (EC = 10 ds/m) three times per week throughout the growing season of each year. Significant differences were observed among cultivars and selections for all species under field conditions. Field results are significantly correlated to greenhouse salt chamber results.

Clones of perennial ryegrass were established in the fall of 2007 and treated with a salt solution with an EC of 10 ds/m. Initial broad-sense heritability was estimated to be 0.78 and indicates that a large proportion of the variation could be contributed to genetic effects.

**Summary Points**

- Significant differences were observed in clones of five perennial ryegrass cultivars treated with 4 different salinity levels (1, 5, 10, and 15 dS/m) under greenhouse conditions.
- Four clones of 'Palmer III' and one clone of 'Applaud' exhibited the highest percent green ratings compared to clones of ‘Paragon GLR’, ‘Brightstar SLT’, and ‘Nui’.
- Significant differences were observed in 21 Kentucky bluegrass cultivars treated with 4 different salinity levels (1, 3, 6, and 9 dS/m) under greenhouse conditions. The cultivars exhibiting the highest percent green ratings were ‘Eagleton’, ‘Liberator’, and ‘Cabernet’. The cultivars and selections with the lowest percent green were a Texas x Kentucky bluegrass selection, A03TB-246, ‘Baron’, and the Kentucky bluegrass selection A03-84.
- In all greenhouse runs for all cool-season turfgrass species, there were significant differences between salinity treatments with higher salinity treatments causing more injury to turfgrass plants. Percent green ratings were highly correlated to clipping yields, root weights, and shoot weights for all species.
- Significant differences were observed among cultivars and selections for all species under field conditions.
- Results were significantly correlated to greenhouse salt chamber results and indicate that we may use the greenhouse screening technique to select cultivars with improved salinity tolerance that will also exhibit similar field responses.
- Initial broad-sense heritability of salinity tolerance in perennial ryegrass was estimated to be 0.78.
Turfgass improvement for drought tolerance requires a comprehensive, integrated understanding of the physiological traits and genetic basis underlying trait variations in diverse natural populations. In recent years, association mapping has been developed as a novel and more powerful mapping technique. This technique uses a natural plant population to correlate genes with a phenotypic trait of interest and serves as an alternative method for mapping quantitative trait loci (QTL). Compared to linkage mapping in traditional bioparental populations, association mapping offers three main advantages: increased mapping resolution, reduced research time, and greater allele numbers.

Phenotypic traits were evaluated in the natural population of perennial ryegrass (Lolium perenne L.) established in three locations varying in soil texture in Indiana for the first year. Across all population, leaf wilting ranged from 1 (no wilting) to 9 (severely wilted), leaf water content ranged from 60 % to 82 %, canopy temperature ranged from 29°C to 38°C, and chlorophyll fluorescence ranged from 0.69 to 0.85 under drought stress, respectively. Other traits associated with plant metabolisms are being characterized. Large variations in drought tolerance across the mapping population provide a good basis for future gene and trait association analysis.

A total of 109 simple sequence repeat (SSR) markers were used to screen 192 diploid perennial ryegrasses for assessing genetic diversity, population structure, and molecular variance among and within populations. The overall genetic diversity and polymorphic information content of mapping population was 0.80 and 0.79, respectively.

The STRUCTURE identified five subpopulations (G1 to G5). The genetic diversity was 0.76, 0.75, 0.74, 0.79, and 0.75 for G1, G2, G3, G4, and G5, respectively. Genetic distance was the largest between G2 and G3 (0.69) and the smallest between G1 and G3 (0.21), and the results were consistent with population pair-wise Fst test.

The Molecular Variance Analysis indicated that genetic variation among and within populations were 8.6 % and 91.4 %, respectively. As population structure (associated with local adaptation or diversifying selection) or familial relatedness (from recent co-ancestry) can result in spurious associations of genes with traits, it is necessary to examine population structure to enhance accuracy of association mapping.

**Summary Points**
- The natural mapping population showed large variations in drought tolerance.
- The overall genetic diversity of mapping population was 0.80.
- Five subpopulations were identified among the natural mapping population.
- Genetic variation among and within populations were 8.6 % and 91.4 %, respectively.
A research site was assigned and planted using greenhouse-grown bermudagrass plugs on June 22, 2007 at the Oklahoma State University Turfgrass Research Center, Stillwater, OK. The site was specifically constructed to host this and future shade-selection projects. The research site receives mid to late afternoon shade, depending on season, from a dense, mature evergreen canopy on the west side of the site. The site meets the most important parameters for effective shade research. Late afternoon vegetative shade is provided by conifers on the west side of the plots. These conifers also provide root competition and reduce the predominately westerly airflow. Maple trees have been planted along the south side of the site and redbud trees along the east side to increase the duration of vegetative shade. We attempted to increase the duration of shade at the site in 2008 by planting vines along a hoop structure, but we had limited success. Due to limitation and uneven density of natural shade, a 75% black woven shade cloth was installed above the plots to provide shade in the middle of the day.

The study consists of 45 bermudagrass selections and four standards, 'Celebration', 'Patriot', 'Tifton 4', and 'Tifton 10'. 'Celebration', 'Tifton 4', and 'Tifton 10' were chosen for their potential shade tolerance and 'Patriot' was chosen for its likely poor shade tolerance. The bermudagrass selections were collected primarily from China, Africa, Australia, and other nations. Each bermudagrass was replicated five times on the shade site that is in full sun for about 48% of each day and on an adjacent site that is in full sun for about 90% of each day. Visual turf quality (TQ) and NDVI (normalized difference vegetation index) were assessed every two weeks in 2009 for five rating dates from June 5 to August 13, 2009.

In 2008, shade stress occurred on the shade site for 12% longer each day than on the sun site. This short duration of shade stress caused an average 4.9% decline in TQ and a 3.4% decline in NDVI in 2008. On May 7, 2009, a black woven shade cloth with 75% light reduction (10 ft x 160 ft) was installed on a hoop structure to provide longer and even more uniform shade for the shade site. Consequently, the shade intensity duration increased from 12% in 2008 to 52% in 2009. Also, it increased decline in TQ from 4.9% in 2008 to 12% in 2009 and a decline in NDVI of 3.4% in 2008 to 7.4% in 2009.

The bermudagrass selections differed significantly in TQ and in NDVI both in full sun and in shade in both 2008 and 2009. Data indicate that there is significant variation among selections in both sun and shade. The best performing cultivar in both full sun and shade was 'Patriot'. In 2010, photosynthesis will be measured from the 10 best selections, 10 worst selections, and four standards in May, July, and September with a LI-6400 portable gas exchange system.

Summary Points

- Photosynthetically active radiation was measured continuously to determine the amount of shade stress present in the shade site.
- Turfgrass quality ratings were made every two weeks by collecting visual and NDVI ratings.
- In 2009, the shade site received 48% of the solar irradiance received on the sun site.
- Turfgrass visual quality ratings and NDVI indicated significant diversity among selections.
- The mean visual turf quality decline between like selections in full sun to shade was 12%, and the mean decline in NDVI quality was 7.4%.
Identification of Quantitative Trait Loci (QTL) Associated with Drought and Heat Tolerance in Creeping Bentgrass

Bingru Huang, Stacy Bonos, and Faith Belanger
Rutgers University

Objectives:
1. To evaluate variations in drought and heat tolerance for two mapping populations of bentgrass segregating for disease resistance.
2. To identify phenotypic traits associated with drought and heat tolerance.
3. To identify QTL markers associated with drought and heat tolerance utilizing the available linkage maps.

Start Date: 2008
Project Duration: three years
Total Funding: $89,912

The creeping bentgrass mapping population (L93-10 x 7418-3) segregating for dollar spot resistance was evaluated for their variations in drought tolerance in 2008. A second bentgrass mapping population was evaluated for drought tolerance in a greenhouse in 2009. This is a hybrid population that was originally developed by crossing creeping (Agrostis stolonifera L.) and colonial bentgrass (Agrostis capillaris L.) by interspecific hybridization for the introgression of colonial genes for dollar spot resistance into creeping bentgrass and for development of colonial linkage map.

The F2 population, progeny of a creeping/colonial (Hybrid 15) x creeping (9188) cross, was used for drought screening. The population was propagated and maintained in a greenhouse until establishment and exposed to drought treatment from 5/10-5/20/2009, during cool greenhouse temperatures to prevent additional heat stress. Soil volumetric water content (SWC) was monitored and was not significantly different among plants prior to drought treatment (approximately 25%). The duration of water being completely withheld for drought treatment was 10 days, when the SWC reached 8%. Measurements of drought tolerance characteristics included turf quality, relative water content, electrolyte leakage (EL), canopy temperature depression (CTD), relative water content (RWC), water use efficiency, and chlorophyll content.

The phenotypic traits associated with drought tolerance and the molecular marker data previously generated for the two mapping populations were subjected to QTL analysis. Molecular markers and chromosomal locations with significant LOD scores were identified as QTLs associated with the phenotypic traits of drought and heat tolerance.

Several possible QTLs associated with drought tolerance were identified in two bentgrass populations. Under drought stress of both the field and greenhouse studies, possible QTLs were identified on chromosomes 3, 5, 8, 11, and 13, of which two chromosomal locations on groups 3 and 11 overlapped for multiple traits. The QTL on group 3 was identified for both FvFm and for RWC whereas the QTL on group 11 was for TQ and carbon to nitrogen ratio.

QTL analysis of the data taken in the drought stress study in the greenhouse for the hybrid population resulted in highly significant QTLs on three different chromosomes, groups 4, 5 and 10. The location of the QTL on group 5 overlapped for the two traits EL and CTD. TQ mapped to 3 locations, 2 on group 10, and 1 on group 4. This data is the result of only a single greenhouse study which will be repeated in the winter of 2010.

For heat tolerance QTLs, potentially important regions of the genome include chromosome groups 3, 6, 11, 12, and 13 identified for the traits including TQ, green leaf biomass, leaf area index, and FvFm. Groups 11 and 13 overlapped for both heat and drought data, indicating that these could also be important chromosomal locations for the combined heat and drought tolerance.

Markers associated with multiple traits and with both heat and drought tolerance are particularly important which will be further analyzed for future use in marker-assisted selection.

Summary Points
- Phenotypic variations in drought and heat tolerance exist in the creeping bentgrass and hybrid bentgrass mapping population developed for disease resistance.
- Significant QTLs were identified for various traits that were good indicators for stress tolerance and turf performance under below optimal conditions such as TQ, EL, and RWC.
- Important chromosomal locations for heat and drought tolerance may exist on 5 different linkage groups for the creeping bentgrass population and 3 different linkage groups in the hybrid population.
- Regions of the chromosome with significant overlap for both heat and drought tolerance and for multiple stress tolerance parameters such as CTD and EL may be the most useful in future studies for development of marker-assisted selection procedures.
The New England velvet bentgrass germplasm collection is a collaborative effort by researchers at the University of Rhode Island and the University of Massachusetts. More than 250 accessions have been collected from old golf courses throughout New England. In 2009, the entire collection was evaluated for salt tolerance in the greenhouse at URI, and for genetic color, growth rate, and dollar spot in the field at URI. Copper spot resistant accessions identified in 2008 were re-screened to confirm resistance.

Velvet bentgrass (Agrostis canina) has two ploidy levels, diploid (2n=14) and tetraploid (2n=28) referred to subsp. canina and subsp. montana. A flow cytometry analysis was carried out in the velvet bentgrass collection to clarify the ploidy level of the accessions. After the analysis, 74% (159 individuals) of the accessions were found diploid supported with velvet phenotypes and 26% (56 individuals) were found tetraploid. Out of 83 EST-SSR primers previously screened, 10 pairs were selected because of their polymorphism and quality of amplification.

Velvet bentgrass has excellent tolerance to lower levels of sunlight, nitrogen, and water. Velvet bentgrass is considered native to New England and coastal regions as far south as Maryland. The stress tolerance genes found in velvet bentgrass need to be preserved as potentially irreplaceable genetic resources.

The entire collection was evaluated for salt tolerance in the greenhouse at URI from February until June 2009. Three clones of each accession were transferred to pots filled with sand and placed in an ebb-and-flow hydroponics system. The plants were irrigated with a nutrient solution amended with sodium chloride. The salt concentration was increased every two weeks from 1,000 ppm to 8,000 ppm.

At the end of each two-week period, all the plants were photographed using a digital camera and a portable light box. SigmaScan software was used to measure retention of green foliage by calculating the number of green pixels in each image. At 8,000 ppm salt, ‘SR7200’ retained 4% green cover, and ‘Greenwich’ retained 10%. The velvet bentgrass accessions ranged from 0% green cover to 80% cover. Thirty-nine accessions were significantly more salt tolerant than either cultivar, and an additional seven were more tolerant than ‘SR7200’. Thirty-one of these accessions retained more than 50% green cover.

Three replications of the germplasm collection were established in the field at URI as spaced-plants on 18-inch centers and mowed at fairway height. This field trial is being used to evaluate genetic color, turf growth, and disease resistance. Each accession was photographed in September 2008 and July and November 2009 using a digital camera and controlled lighting. The digital green color index has been calculated for each accession, producing quantitative data for color comparisons.

The collection was transplanted to the field in June 2008 as 5.5-cm plugs. Holes were cut into an established chewings fescue turf using a standard cup cutter, so each plug was surrounded by a 2.6-cm wide ring of bare soil. The diameter of each plant was measured in July 2009. Four accessions failed to survive. Diameters for the others ranged from 6 cm to 23 cm. Most of the accessions more than doubled in size; 42 accessions more than tripled in size despite competition from the chewings fescue.

The field trial was visually evaluated for dollar spot resistance in August 2009 following a severe natural disease outbreak. Accessions were rated from 1-9, with 9 indicating no disease. Eighty-seven out of 233 accessions (37%) showed no sign of disease.

**Summary Points**

- Collaborative studies continue with several researchers in New England.
- Significant progress has been made to identify germplasm that have enhanced resistance to biotic and abiotic stresses.
Evaluation and Development of Poa Germplasm for Salt Tolerance

Paul Johnson
Utah State University

Joseph Robins and B. Shaun Bushman
USDA-ARS

Objectives:
1. To identify salt-tolerant Poa germplasm that can be incorporated into breeding and genetics efforts.
2. To identify genes whose RNA transcript levels vary between control and high-salinity treatments.

Start Date: 2007
Project Duration: three years
Total Funding: $75,237

One of the greatest challenges confronting the turf industry is water. Irrigation water is in short supply due to the rapidly growing population, especially in the North American West. Golf course superintendents and other landscape managers are being asked to deal with a very difficult situation—to use less irrigation water, use lower quality water sources, and allow more use of the turfgrass areas. Therefore, turfgrass with high turfgrass quality and greater salt tolerance is essential to meet these expectations.

Extensive breeding efforts in Kentucky bluegrass (Poa pratensis L.) have been conducted by both universities and private companies to improve turfgrass quality traits and pest resistance and some stress tolerance traits. However many Poa species, and most of the available germplasm within Poa pratensis, has not been rigorously evaluated for salt tolerance and turf characteristics in the arid West climate. Similarly, there is not a good understanding of the genetic control of salt tolerance in Poa. Germplasm screening and detailed genetic studies of salinity tolerance are needed for breeding programs to effectively develop salt-tolerant bluegrasses.

We assembled a large and diverse collection of Poa germplasm, check varieties, and other species to evaluate in field conditions and controlled-environment salinity evaluations. Seedlings were subjected to increasing salt concentrations until the last plants had been killed by the treatment.

We also conducted suppressive-subtractive hybridization (SSH) to evaluate genes that are differentially expressed under salt stress. We selected two accessions identified in the screening procedure, one identified as salt tolerant and one salt intolerant. We then applied salt stress to one set of each accession with another set in control conditions-no salt stress.

RNA was extracted from shoot/crown and root tissues, purified, quantified, and then sequenced and analyzed to identify the differentially expressed genes in the tissues. The resulting sequences of the gene transcripts are being compared to a plant sequence database to assign identities to the transcripts based on sequence homology.

Based on two runs of the salinity screening experiment, several germplasm sources were identified with consistently high-salinity tolerance during both runs of the study. The salinity tolerance of some of the germplasm sources was similar to that of the tall fescue and perennial ryegrass check entries. The better performing germplasm has been moved into greenhouse and field hybridization experiments.

The SSH procedure was completed for eight samples making four comparisons of salt and non-salt stressed tissues in both shoot/crown tissues and root tissue. Comparisons were made in “both directions”, meaning that genes over-expressed in salt-treated, and over-expressed in control conditions, were investigated. The SSH libraries were cloned, and over 700 clones were selected and sequenced. After comparison to gene databases, the majority of sequences were identified. Most of these annotated genes were involved in some type of plant stress. Some have roles in stress response, some are unidentified, and some only have identities to other stress-induced gene libraries.

The many genes that can be further investigated provide important insights into how Kentucky bluegrass responds to, and tolerates, salt stress at a molecular level. In addition, over 100 sequences had no similar gene found in other grasses. Additional validation of a subset of genes is underway using quantitative real-time PCR (qPCR), wherein we will test differential expression of genes with known salt-response functions.

Summary Points
- We have observed significant variation in salt tolerance among a diverse set of Poa species and Poa pratensis germplasm.
- The most salt-tolerant accessions exceeded the tolerance of Kentucky bluegrass check varieties and approached or exceeded the tolerance of perennial ryegrass and tall fescue check varieties.
- Over 700 genes related to salt response were cloned and sequenced, some with known involvement in salt tolerance in other plant species.
Genetic Enhancement of Turfgrass
Germplasm for Reduced-input Sustainability

Kevin Morris
National Turfgrass Federation, Inc.

Scott Warnke
USDA-ARS

Objectives:
1. The objective of this research, conducted at the USDA, ARS’ Beltsville Agricultural Research Center in Beltsville, MD, is to use genetic and biotechnology approaches to identify and develop turfgrass germplasm with improved biotic and abiotic stress resistance. Efforts will be made to identify molecular markers associated with desirable traits and to combine useful traits into germplasm able to grow with reduced inputs.

Start Date: 2007
Project Duration: three years
Total Funding: $50,000

There is a tremendous need to improve the stress tolerance of turfgrass. To address this broad objective, we have undertaken the following five projects:

1. Development of Danthonia spicata as a Low Maintenance, Native Turfgrass Species

A key aspect to the development of Danthonia spicata (Poverty grass) as a low-maintenance turfgrass species is obtaining a better understanding of the biology of the species. Initial observations indicate that there is variation present in the species, and there may be more than one species present in natural stands.

A seeding rate trial was established at the University of Maryland turfgrass center using three different seeding rates. The results of the first year indicate no significant difference in turf quality between the three seeding rates.

Genetic diversity of selected Danthonia spicata germplasm was established using 347 AFLP markers. The results indicate that the species is primarily self-fertile, however, low levels of outcrossing are likely to occur. Genetic markers will be used to establish estimates of outcrossing rates in natural populations and controlled crosses will be attempted.

2. Field Screening of Bentgrass Germplasm for Resistance to Important Turfgrass Diseases

A field trial containing clonally-propagated plants from a bentgrass mapping population, developed by Dr. Geunhwa Jung at the University of Massachusetts, was established at the University of Maryland turfgrass center. The study involves approximately 300 entries replicated three times, plugged into a ryegrass turf.

The results indicate no significant difference in turf quality between the three seeding rates.

AFLP marker results indicate that Danthonia spicata (Poverty grass) is primarily a self-fertile species, however, a small percentage of outcrossing does occur.

A high level of dollar spot (Sclerotinia homoeocarpa) infection in 2009 caused differing responses among bentgrass germplasm planted in Maryland field trial.

Tall fescue plant introductions (PIs), inoculated with Rhizoctonia solani and R. zeae, showed varying degrees of tolerance to both diseases.

1,300 Miniature Inverted-repeat Transposable Elements (MITEs) display markers were used in a diversity analysis of Agrostis species. The markers established the relationship between ancestral diploid material and cultivated varieties, and showed reduction in genetic diversity in new cultivars.

Summary Points
- AFLP marker results indicate that Danthonia spicata (Poverty grass) is primarily a self-fertile species, however, a small percentage of outcrossing does occur.
- A high level of dollar spot (Sclerotinia homoeocarpa) infection in 2009 caused differing responses among bentgrass germplasm planted in Maryland field trial.
- Tall fescue plant introductions (PIs), inoculated with Rhizoctonia solani and R. zeae, showed varying degrees of tolerance to both diseases.
- 1,300 Miniature Inverted-repeat Transposable Elements (MITEs) display markers were used in a diversity analysis of Agrostis species. The markers established the relationship between ancestral diploid material and cultivated varieties, and showed reduction in genetic diversity in new cultivars.
There is a nationwide effort to use native grass species in turf systems, as most species used now are typically non-native.

From July to September 2007, almost 300 clones from seven species of perennial range grasses were collected in Arizona which included curly mesquite, false grama, sprucetop grama, wolftail, blue grama, black grama, and hairy grama. These clones were propagated and grown under near-optimal conditions in a greenhouse.

In early January 2008, and again six weeks after a "grazing" (severe defoliation) event, data were collected from plants actively growing in the greenhouse. These included (1) plant height (2) plant diameter in two directions, and visual scores for (3) innate plant density, and (4) overall turf quality (1-5, 5 = best for the latter traits).

Height/width ratios (lower values suggest a "wider" than "taller" growth habit) before and after defoliation were relatively consistent up to a value of 5, for pre- and post-defoliation measurements, respectively. Stem and box plot data show that there may be considerable useful variation for growth habit in curly mesquite, wolftail, and sprucetop grama.

These species had a low H/W ratio before defoliation. After a single defoliation event, most clones exhibited increased height, with some variation remaining for low H/W ratio growth response for curly mesquite, wolftail, and sprucetop grama. Other species showed very little variation for H/W ratios following defoliation. When plotted against the average of turfgrass quality and density scores after defoliation, clones with the lower height/width ratio values had the greatest numerical mean quality-density averages.

Based on the greenhouse results, 100 clones across all species were selected and planted as replicated plant propagules in a mowed spaced-plant nursery. Plants were mowed 3 times per weekly at a 3.0-inch height. Plant width data showed good growth in plant size from August 2008 to May 2009, with many bunch grass species having girths of 80-120 cm, or more, along with high shoot densities.

By June, the overwhelming majority of clones developed necrotic centers, which seriously detracted from turf quality. Among the bunchgrasses, sprucetop grama (B. chrondoisoides) had the least expression of this trait. Both blue and black grama selections did not produce satisfactory quality turf parameters under the high heat conditions realized, but sprucetop grama did.

Most wolftail clones (Wolfii tailii) exhibited extreme lateral growth, but produced too many necrotic stems for turf acceptance. Seed collection efforts of sprucetop grama have increased.

**Summary Points**

- 300 clones from seven species of perennial range grasses were collected from a 150-mile radius of Tucson. 100 bunchgrass and 100 stoloniferous grasses were selected for field evaluation under regular mowing.
- Most bunchgrass species either had poor shoot density or produced concentric necrotic "straw rings" within the first year's growth. After one summer of field screening, sprucetop grama has the largest percentage of its selected plants with good turf quality under mowed field conditions.
**Bermudagrass and Seashore Paspalum Cultivar Response to the Sting Nematode**

Wenjing Pang, William T. (Billy) Crow, and Kevin E. Kenworthy  
University of Florida

**Objectives:**

1. Determine the range of response (resistance or tolerance) of bermudagrass and seashore paspalum cultivars to the sting nematode and identify the best performing cultivars.
2. Investigate if a proposed alternative method for assessing sting nematode response is as effective, or more efficient, than traditional methods.

**Start Date:** 2008  
**Project Duration:** three years  
**Total Funding:** $31,407

Recent cancellation of fenamiphos (Nemacur, Bayer Cropscience) has resulted in the need of better nematode management tactics. Currently, Curfew Soil Fumigant (Dow Agrosciences) is the most effective management for sting nematodes. However, Curfew provides only short-term control, is expensive, and environmental restrictions highlight the need for alternative options. Utilization of resistant or tolerant cultivars is the most efficient, least costly practice for nematode management on turf.

Separate glasshouse and field experiments for bermudagrass and seashore paspalum are being conducted to assess the range of these species for response to sting nematode. Seventeen cultivars of bermudagrass and seven cultivars of seashore paspalum were tested in separate greenhouse experiments. Six weeks after turfgrass establishment, grasses were inoculated with *B. longicaudatus* at 50 nematodes per container. Experiments were harvested 90 days after inoculation. Percent reduction in the root length of inoculated plants compared with the uninoculated control was calculated.

For all cultivars, the inoculated treatments led to reductions in total root length compared with the uninoculated controls, which ranged from 5 to 36%. Maximum reduction of 36% was found for ‘Floradwarf’ while the minimum reduction of 5% was observed on ‘TifSport’. ‘TifSport’ and ‘Patriot’ suffered little root damage with the root length percent reduction less than 10% and exhibited resistance. ‘Patriot’ exhibits tolerance (little root damage with an increase in nematodes). On the other hand, maximum root damage (more than 30% root length reduction) occurred for ‘Floradwarf’, ‘Champion’, ‘TifEagle’, ‘MiniVerde’, and ‘Tifton 10’, identifying them as susceptible cultivars.

Sting nematodes reduced the root length of all seashore paspalum cultivars from 12 to 27% compared with the uninoculated treatments. Root length reduction was less than 15% on ‘SeaDwarf’ and ‘Sealsle Supreme’. However, sting nematode population was increased four-fold on these two cultivars. ‘SeaSpray’ and ‘Aloha’ were the most susceptible out of the seven cultivars tested.

In 2008, two field plot experiments were conducted. Nematode population in each plot was assayed before planting of grasses on the same day. Soil samples were collected every 90 days after planting. Data in March and June both indicated that cultivars ‘Champion’, ‘Floradwarf’, ‘Tifgreen’, ‘MiniVerde’, and ‘TifEagle’ were the most susceptible cultivars, and ‘TifSport’ was the most resistant cultivar.

For seashore paspalum, no difference in the sting nematode population was observed among the three cultivars ‘Sealsle 1’, ‘Aloha’, and ‘SeaDwarf’ in March; however, population on ‘Sealsle 1’ was higher than that on ‘SeaDwarf’ in June.

Both field and greenhouse studies indicated that ‘TifSport’ was the most resistant bermudagrass cultivar while ‘Champion’, ‘Floradwarf’, ‘MiniVerde’, and ‘TifEagle’ were susceptible cultivars. Also it showed a trend that greens-type bermudagrass cultivars support higher population of *B. longicaudatus* than those cultivars used on fairways.

In 2009, 13 cultivars of bermudagrass and seven cultivars of seashore paspalum were planted. Nematode populations in each plot were assayed before planting of grasses on the same day. Soil samples were collected every 90 days after planting. There was no difference in sting nematode population among these plots either in April or July.

**Summary Points**

- In the greenhouse, sting nematodes reduced the root length of all bermudagrass cultivars, and the reduction ranged from 5 to 36%. The most tolerant bermudagrass cultivars were ‘TifSport’ and ‘Patriot’.
- ‘TifSport’ was the most resistant bermudagrass cultivar while ‘Champion’, ‘Floradwarf’, ‘MiniVerde’, and ‘TifEagle’ were susceptible bermudagrass cultivars to sting nematodes both under greenhouse and field conditions.
- Both greenhouse and field studies indicated that greens-type bermudagrass cultivars supported higher populations of *B. longicaudatus* than those cultivars used on fairways.
- Sting nematodes reduced the root length of seashore paspalum cultivars from 12 to 27% in the greenhouse; however, the reduction was not different among cultivars.
- Sting nematode populations at harvest increased on all seashore paspalum cultivars under greenhouse conditions. However, ‘SeaDwarf’ and ‘Sealsle Supreme’ exhibited best levels of tolerance among cultivars tested.
Breeding Turf-type Annual Ryegrass for Salinity Tolerance

L. R. Nelson
Texas AgriLife Research

Objectives:
1. To identify improved salt-tolerant genotypes and introgress the tolerance into adapted turf-type genotypes of annual ryegrass and increase seed of these populations for experimental testing.
2. To select plants of turf-type annual ryegrass which are infected with a fungal endophyte and determine if presence of the endophyte provides this population with increased tolerance to high salt concentrations.
3. To test the experimental lines of annual ryegrass under high salinity growing conditions in hydroponic and field conditions and release the line as a salt-tolerant variety.

A
nnual and perennial ryegrass are considered to be susceptible to high salinity. Genetic selection for tolerance to high salinity will be carried out by two methods. First, a salt tank greenhouse screening method will be conducted to select salt tolerant plants at Overton. Secondly, a screening procedure will be utilized under high-salt field conditions at Pecos, Texas.

Selection of genotypes with improved salt tolerance were planted in the field in a high-salinity soil and irrigating with high-salinity water at Pecos, Texas. Selection of seed heads of best plants took place in May of 2009.

In the greenhouse, seed were planted into an organic mix in flats. When plants were in the 2-3 leaf stage, entire flats were immersed in water with a salt concentration of 3,100 ppm (4.9 dS m⁻¹). Thereafter, flats were immersed every 3 to 4 days. Salt concentration was increased gradually over 60 days until a water salt concentration of 15,900 ppm (25.4 dS m⁻¹) was accomplished. At that time, 80% of plants were dead. Remaining plants (300 plants) cross-pollinated and produce seed in the spring of 2009. This seed, as well as the seed from Pecos, is being tested for salt tolerance in 2010.

In the greenhouse trials, the salt concentration was at 5,000 ppm at first immersion. Salt concentrations were gradually increased until near the end of the screening period, when salt concentrations were 9,800 ppm. The actual rating for each entry should increase from one date to the next date. This is useful to judge how severe the "salt damage" was. Late in the rating period of both trials, many ratings were at or near 8, meaning many plants were dead.

A sixth replication of all entries was grown in flats with identical conditions, except it was immersed in well water and not in saline water. Ratings were also made on entries from this treatment. We had a significant amount of senescence in the untreated (salt) plants. This could have been due to poor light quality, poor water quality, or some other factor. The entry TF-152 appears to be tolerant to high salt.

Summary Points
- Differences for salt tolerance between annual and perennial genotypes were obtained in both greenhouse and field experiments.
- Early generation salt-tolerant germplasm seed was increased at Overton in 2009.
- This salt-tolerant germplasm will be tested in 2010 to determine if it truly is improved for salt tolerance.
Evaluating Methods for Vegetative Propagation and Enhancement of Seed Production of Greens-type Poa annua Cultivars

David R. Huff
Pennsylvania State University

Objectives:
1. Establish efficient vegetative propagation methods of greens-type Poa annua for sod production and for establishing/renovating golf course putting greens.
2. To scale-up our results for greens-type cultivar production to a larger commercial level by collaborating with sod producers.
3. To release a genetically stable, vegetative greens-type cultivar exhibiting superior putting green quality and stress tolerance.

Start Date: 2008
Project Duration: two years
Total Funding: $20,000

Poa annua L. has been part of the game of golf for over 130 years, however despite repeated attempts to breed improved strains, currently there are no commercial sources available of high quality greens-type Poa annua. Developing such commercial products would allow superintendents and architects an opportunity to immediately begin utilizing the perennial greens-types of Poa annua putting surfaces rather than having to wait for the natural evolution of high quality greens-types from the wild and weedy invasive annual types.

The greens-type phenotype in Poa annua L. is essentially a dwarf annual bluegrass plant where a number of traits (including shortened culms, shortened tillers, short leaves, reduced number of spikes, and single-branched inflorescences) are linked together thus inherited as though they were a single trait. As a result, the greens-type phenotype is a reduction in plant stature as compared to the annual-type.

In previous studies, GA3 and 1-naphthaleneacetic acid (NAA) were used in plant hormone bioassays while auxin signaling and GA biosynthetic genes were PCR-cloned and used for expression analysis on annual-type and greens-type Poa annua. The results indicated that the greens-type biotype is responsive to GA3, resistant to NAA, and GA20 oxidase is up-regulated in comparison to the annual-type. Moreover, a 518 bp length fragment was cloned from Poa annua and shows high sequence homology to Auxin F-Box sequences from rice, maize, and Arabidopsis.

RT-PCR and Real-Time analysis suggest that the greens-type phenotype is correlated to a reduction in the expression of this Auxin F-Box like gene which are known to regulate GA expression. These results suggest that the greens-type Poa annua dwarfism is the result of hormonal cross-talk between the auxin and gibberelin biosynthetic pathways.

Previous genetic research also suggests that regulation of the Auxin F-Box-like gene expression is potentially altered from environmental stimuli. Perennial greens-type phenotypes result from the action of mowing which causes an alteration in Auxin F-Box-like gene expression resulting in repression of the plant growth hormone gibberellic acid (GA) signaling pathway. In the absence of the mowing stimulus, the GA pathway progressively becomes unsilenced resulting in reversion of the greens-type plants back to the annual type. The annual type is undesirable as a putting surface and requires years of mowing in order to develop a perennial greens-type form.

We believe that the genetic stability of perennial greens-type Poa annua is capable of being maintained through vegetative propagation in combination with mowing. We have established vegetative plots using different source plant materials including aeration cores, solid sod, and shredded sod/plugs.

We also evaluated vegetative establishment in the greenhouse in combination with the exogenous applications of GA in an attempt to further reduce the time required to achieve full coverage. Mowing was deemed to be the most critical factor to maintaining the greens-type phenotype. Using these methods, we successfully propagated enough vegetative material to establish a series of replicated trials of greens-type Poa annua cultivars at five different universities.

This research project will now begin to extend these vegetative propagation techniques towards the commercial sod production of greens-type Poa annua to a larger scale approaching that of commercial sod production. Seed and vegetative materials produced in 2009 will provide the necessary planting material for us to work directly with interested sod producers for an evaluation of scaled-up production.

Summary Points
- Our research continues to indicate that the dwarf nature of perennial greens-type Poa annua results from an epigenetic inhibition of the biosynthetic pathway for the plant growth hormone gibberellic acid (GA).
- We demonstrated our ability to vegetatively propagate greens-type Poa annua to an extent of satisfying requests from five universities participating in the Northeast 1025 Project titled: "Biology, Ecology, and Management of Emerging Pests of Annual Bluegrass on Golf Courses".
- We have begun collaborating with commercial sod producers in Pennsylvania and Illinois to begin scaling up our vegetative propagation techniques of greens-type Poa annua.
Comparative Irrigation Requirements of 30 Cultivars of Kentucky Bluegrass under a Large Rainout Facility in the Transition Zone

Dale Bremer, Steve Keeley, Jack Fry, and Jason Lewis
Kansas State University

Objectives:

1. Develop a novel method for concurrently comparing irrigation requirements among 30 cultivars of turfgrasses using a large rainout facility at Kansas State University.
2. Produce a database of relative irrigation requirements for 30 cultivars of Kentucky bluegrass.
3. Partition cultivars of Kentucky bluegrasses into irrigation-requirement categories of "high, medium, and low."
4. Conduct dry-down and genetic rooting potential experiments in a greenhouse to evaluate responses to drought and physiological characteristics among the same cultivars as those tested in the field.

Efficient use of irrigation water on turf is becoming more crucial in the U.S. Information is needed about relative irrigation rates among newly-released cultivars of turfgrasses. A large rainout shelter near Manhattan, Kansas offers an opportunity to compare the irrigation requirements of multiple turfgrass cultivars in the stressful climate of the transition zone. By shielding rainfall from the turfgrasses, plots can be irrigated individually on an as-needed basis over a period of several weeks or months to determine the total irrigation requirements among cultivars given similar field conditions. Turfgrasses with similar visual qualities but with lower irrigation requirements may offer significant water savings to turfgrass managers.

Twenty-eight cultivars of Kentucky bluegrasses and two Texas bluegrass hybrids were selected for this study from among and within 11 different phenotypic groups, based largely on performances in the 2004 NTEP tests. Plots, replicated three times per cultivar, were prepared and seeded under the rainout shelter in September 2006. Plot preparation included cultivation, fumigation, leveling, and insertion of 30-cm deep metal edging around individual plots to prevent lateral movement of water.

There were two summer dry-down periods. Plots were well-watered until early June 2007 and 2009, after which turfgrasses were allowed to dry down without irrigation or precipitation until the first sign of wilt. Individual cultivars were evaluated daily and irrigated with one inch of water when approximately 50% of the plot area showed visible symptoms of wilt. Each plot was manually irrigated, and irrigation quantity and date was recorded. General turf performance was also evaluated daily by visually rating turf quality.

The total amount of water applied averaged over the two summers varied significantly among cultivars and ranged from 8 to 20 inches during the four-month period. Visual quality also varied substantially among cultivars. In general, when considering both visual quality and water requirements, cultivars in the Compact America and Mid-Atlantic groups performed better (higher quality, lesser water requirements) and "Common" types poorer (lower quality, greater water requirements) among phenotypic groups, although there was significant variability even among cultivars within each group.

The same cultivars used in the field study were evaluated for rooting characteristics including maximum root length extension, surface area, mean root diameter, and root biomass in a greenhouse at Kansas State University using root tubes. Briefly, turfgrasses were planted into clear polyethylene root tubes that were filled with fritted clay; polyethylene tubes were then inserted into opaque PVC pipe (sleeves).

Root growth was monitored periodically along the side of the clear root tubes. There were broad ranges in rooting characteristics among cultivars at each depth. A number of cultivars had maximum rooting depths below 90 cm. Differences among phenotypic groups were less pronounced although root surface area was lower in Mid-Atlantic and Compact America groups than in "Common" types.

Summary Points

- Twenty-eight cultivars of Kentucky bluegrasses and two Texas bluegrass hybrids were established under a large rainout shelter located in the transition zone.
- Individual cultivars were evaluated in the field from June through September, 2007 and 2009 for their visual quality and total water requirements during repeated dry-downs.
- Cultivars in the Compact America and Mid-Atlantic groups generally performed better (higher quality, lesser water requirements) and "Common" types poorer (lower quality, greater water requirements) among phenotypic groups although there was significant variability even among cultivars within each group.
- There was a broad range in the rooting characteristics among cultivars.
- Greenhouse rooting characteristics measured were not correlated with water applied in the field study.
The public is concerned about the effects of golf courses on the environment. In response to this concern, the USGA has conducted research examining the fate of pesticides and fertilizers since 1991. The USGA continues to support scientifically based investigations on the environmental impact of golf courses. The focus remains on research to understand the effects of turfgrass pest management and fertilization on water quality and the environment.

Research on best management practices evaluates pesticide and fertilizer programs for golf courses in order to make turfgrass management recommendations that protect environmental quality. The research is conducted on university experiment stations and golf courses. The projects evaluate pesticides or nutrients that pose an environmental risk, and identify cultural practices that minimize volatilization, surface runoff, and groundwater contamination.

Pesticide and nutrient fate models are used to predict the environmental impact of turfgrass pesticides and fertilizers. From 1991 through 1997, research sponsored by the USGA demonstrated:

1. Measured nitrogen and pesticide leaching was minimal and that surface transport (runoff) posed a greater problem for golf courses, especially on heavy textured soils in high rainfall areas of the country.
2. The turf/soil ecosystem enhances pesticide adsorption and degradation that greatly reduces the amount of chemical that moves below the rootzone.
3. Current agricultural fate models need modification to predict the fate of pesticides and fertilizers applied to turfgrasses grown under golf course conditions.

The results of USGA-sponsored pesticide and fertilizer fate research is being used to calibrate and validate existing pesticide fate models for turfgrasses managed under golf course conditions.

Locations of the following projects funded in 2009 by the USGA Turfgrass and Environmental Research Program under the category of Environmental Impact
Utilizing Reduced-risk Pesticides and IPM Strategies to Mitigate Golfer Exposure and Hazard

J. Marshall Clark, Raymond Putnam, and Jeffery Doherty
University of Massachusetts

Objectives:

1. Determine the level of hazard of volatile and foliar dislodgeable residues of the reduced-risk pesticides carfentrazone, halofenozide, and azoxystrobin following total course and full-rate applications.
2. Determine the effect of partial course application strategies (e.g. tees and greens only) and post application irrigation on volatile and foliar dislodgeable pesticide residues following full-rate applications of carfentrazone, halofenozide, and azoxystrobin.
3. Model the relationship of volatile and dislodgeable foliar residues vs. actual golfer exposure using urinary biological monitoring techniques or, for pesticides that are not amenable to biomonitoring, dosimetry techniques.

This study seeks to determine actual levels of golfer exposure to “reduced-risk” pesticides following application to turfgrass. The fate of pesticides after application largely determines how much of it is available for potential human exposure.

We have analyzed pesticide residues in the air and on turfgrass (dislodgeable foliar residues, DFR) in 45 pesticide applications using either chlorpyrifos, carbaryl, cyfluthrin, chlorothalonil, 2,4-D, MCPP-p, dicamba, imidacloprid, carfentrazone, and azoxystrobin. This season (2009), two applications of the reduced-risk insecticide halofenozide were made. Analyses of these samples are in progress.

This study also evaluates best management practices for reducing golfer exposure to reduced-risk turfgrass pesticides. While many “standard” pesticides have been removed from use, new reduced-risk pesticides have been added to the IPM practitioner’s toolbox. To date, there is no dosimetry or biomonitoring data on these reduced-risk pesticides, which exhibit low mammalian and environmental toxicity, low potential for groundwater contamination, low pest resistance potential, and are compatible with IPM.

To determine precisely how much of the environmental residues is actually transferred to golfers during a round of golf, volunteers are used for dosimetry (measuring pesticide residues on full-body cotton suits and personal air samplers) and biomonitoring (measuring urinary metabolites). We will compare the biomonitoring and dosimetry results for these reduced-risk compounds with those previously determined for chlorpyrifos, carbaryl, cyfluthrin, 2,4-D, MCPP, dicamba, chlorothalonil, and imidacloprid. This season (2009), we determined exposure in 16 rounds of golf following application of halofenozide with post-application irrigation.

Hazard quotients (HQs) less than or equal to 1.0 indicate that the exposure resulted in a pesticide dose at which adverse effects are unlikely. To date, HQs determined for chlorpyrifos, carbaryl, cyfluthrin, 2,4-D, dicamba, MCPP, chlorothalonil, imidacloprid, carfentrazone, and azoxystrobin.

Summary Points

- Researchers have evaluated exposure in 16 rounds of golf following the application of halofenozide (2009). They will compare these results with those of previous experiments on chlorpyrifos, cyfluthrin, carbaryl, chlorothalonil, 2,4-D, MCPP-p, dicamba, imidacloprid, carfentrazone, and azoxystrobin.
- Determination of golfer exposure to reduced-risk pesticides will provide a novel dataset for these IPM-friendly compounds.
Optimization of Vegetative Filter Strips for Mitigation of Runoff from Golf Course Turf

Barbara DeFlorio, J. Marshall Clark, Jeffery J. Doherty, Guy R. Lanza, and Om Parkash
University of Massachusetts

Objectives:

1. Use selected plant species in a field study to evaluate the efficacy of vegetative filter strips (VFS) and their most effective arrangement.
2. Determine the fate of pesticides retained in VFS and the major mechanisms of degradation.

Start Date: 2008
Project Duration: three years
Total Funding: $90,000

The loss of pesticides and nutrients into surrounding bodies of water and the resulting decreases in water quality has led to the use of best management practices on golf courses. One such practice is the use of vegetative filter strips (VFS) to intercept runoff water and thus prevent its loss and the loss of any associated pesticides and nutrients to surrounding water bodies.

Joint greenhouse and field studies have been implemented to evaluate selected plants for their effectiveness in removing pesticides and nutrients from turfgrass runoff waters that enter vegetative filter strips (VFS). A greenhouse pot study determined five species (big blue stem, blue flag iris, eastern gama grass, prairie cordgrass, and woolgrass) most effectively removed the six selected pesticides (two fungicides, two herbicides, and two insecticides) from a silt loam soil.

In 2008, a run-on plot, consisting of 12 VFS planted in replicates of three (unvegetated, random mixture of plants, succession of plants, and turfgrass cut to three heights), was established. An overhead simulated rainfall system was constructed similar to those used in previous USGA-funded runoff studies in Minnesota. This growing season, we installed additional lysimeters 1’ under-ground and conducted two studies using an estimated runoff volume generated during a 1-year storm event of 25.4 gallons over the course of 24 hours.

The purpose of the first application was to determine the runoff generated from VFS that had been planted in 2007. Soil presaturation was achieved prior to the initiation of the storm event by applying irrigation for 6 hours (~2.4”). The 25.4 gal of run-on was applied to the top edge of each VFS as a water mixture with bromide (15.1 g/gal) via a solvent transfer pump. The run-on volume was applied over a 2-hour interval with the first hour in the presence of "rainfall" and the second hour without.

Runoff water was continuously collected at the bottom of the VFS. Three-minute grab samples were collected in 30 mL bottles for the first 30 minutes after the initiation of run-on and analyzed for bromide. There were little differences in runoff volumes from the VFS planted as turfgrass (0.5 gal), mixture of plants (0.2 gal), and succession of plants (0.3 gal) compared to the bare strips (7.1 gal). Bromide was detected in the runoff from the bare plots only (average time to bromide detection was 6.5 minutes).

The second run-on application occurred in the presence of overhead simulated rainfall. Soil presaturation was achieved prior to the initiation of the storm event by applying 0.8 inches/hr of rainfall for 9 hours, followed by 15 hours without rainfall. Three hours of overhead rainfall overlapped with the run-on for the last hour only.

The overhead rainfall produced a four-fold increase in average runoff volumes and a greater distinction between treatments (1.2, 1.2, 4.6, and 27.3 gal over the course of 2 hours for turfgrass, mixture of plants, succession of plants and bare plots, respectively). We collected 60 runoff samples, 84 subsurface water samples from 1’ lysimeters, 108 subsurface water samples from 5’ lysimeters, and 648 soil core samples that will be analyzed for pesticides.

We collected 132 30-mL samples to be analyzed for bromide. Bromide was detected after 3 minutes for the bare plots. In 2010, a 5-year storm event scenario will be tested.

Summary Points

- A one-year rain event has been simulated twice on the VFS, once using irrigation and once using an artificial rainfall system.
- Preliminary bromide data indicates that bromide (and presumably pesticides) are being intercepted by the vegetative plots.
- 900 samples are currently being analyzed for pesticides.
- The pesticide application will be repeated next year using a 5-year storm scenario.

<table>
<thead>
<tr>
<th>VFS type</th>
<th>Irrigation Average Total Volume (gal)</th>
<th>Average Time to Bromide (min)</th>
<th>Overhead Rainfall Average Total Volume (gal)</th>
<th>Average Time to Bromide (min)</th>
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<td>Turfgrass</td>
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<td>BDL</td>
<td>1.2</td>
<td>BDL</td>
</tr>
<tr>
<td>Mixed planting</td>
<td>0.2</td>
<td>BDL</td>
<td>1.2</td>
<td>TBA</td>
</tr>
<tr>
<td>Succession planting</td>
<td>0.3</td>
<td>BDL</td>
<td>4.6</td>
<td>TBA</td>
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<tr>
<td>Bare plots</td>
<td>7.1</td>
<td>6.5</td>
<td>27.3</td>
<td>3.0</td>
</tr>
</tbody>
</table>

BDL = below detection limit (0.1 ppm); TBA = to be analyzed; VFS = vegetative filter strip

Table 1. Average total runoff volumes (gal) and time (min) to bromide detection in runoff water from vegetative filter strips during a simulated 1-year rain event utilizing irrigation or overhead rainfall.

2008 USGA Turfgrass and Environmental Research Summary
The USGA initially funded research at Michigan State University to determine nitrogen fate and leaching from a Kentucky bluegrass turf in 1991. Similar to previous research, the initial research at MSU conducted from 1991 through 1993 indicated that there was minimal risk of nitrate-nitrogen leaching from Kentucky bluegrass (*Poa pratensis* L.) turfgrass. Since the summer of 1998, percolate samples have been collected from the same monolith lysimeters and analyzed for nitrate-nitrogen (NO₃-N). As of 2009, the turfgrass area has now been under continual fertilization practices for 19 years with percolate collection for the last 11 years consecutively.

From July 1998 through 2002, lysimeters were treated annually with urea at a N rate 98 kg N ha⁻¹ (24.5 kg N ha⁻¹ application⁻¹) and a high N rate of 245 kg N ha⁻¹ (49 kg N ha⁻¹ application⁻¹). From 1998-2002 for the high N rate, there was a dramatic increase in NO₃-N leaching from 5 mg L⁻¹ in 1998 to 25 mg L⁻¹ in 2002. During the same time frame, there was a modest increase in NO₃-N leaching from 3 mg L⁻¹ in 1998 to 5 mg L⁻¹ in 2002.

In 2003 the N rate was reduced to 196 kg N ha⁻¹ for the high N rate, while the low N rate remained at 98 kg N ha⁻¹. Since 2003, phosphorus from triple superphosphate (20% P) has been applied at two rates, 49 and 98 kg P ha⁻¹ split over two applications. The phosphorus application dates coincide with nitrogen application dates in the spring and autumn.

In 2003, the concentration of NO₃-N leaching from the high N rate treatment did not decline from the previous years. The average NO₃-N concentration leached from the low and high N rate treatments was 6.3 and 31.6 mg L⁻¹. In 2004, the concentration of NO₃-N leaching from the high N rate treatment declined drastically from previous years. The average concentration of NO₃-N in leachate for the high N rate was 8.5 mg L⁻¹. This was a decrease in NO₃-N concentration of 23.1 mg L⁻¹ from 2003.

For the low N rate, the average concentration of NO₃-N in leachate for the low N rate was 1.2 mg L⁻¹. From 2004 through 2009, the mean NO₃-N concentration for the low and high N rates was 2.6 and 7.9 mg L⁻¹.

The concentration of phosphorus detected in leachate remains very low regardless of treatment. The mean concentration of phosphorus detected in leachate since initiating phosphorus treatments in 2003 has been less than 0.02 mg L⁻¹.

### Summary Points
- The mean NO₃-N concentration from 2004 through 2009 is less than 8 mg L⁻¹.
- Results continue to indicate low amounts of phosphorus leaching.

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**Long-term Nutrient Fate Research**

**Kevin W. Frank**

Michigan State University

**Objectives:**

1. Determine nitrate-nitrogen and phosphorus leaching from a turfgrass stand that has been continually fertilized for 18 years.
2. Continue data collection from the Long Term Nutrient Fate Research Area at MSU; currently we have data collection for 11 years consecutively.

**Start Date:** 2003

**Project Duration:** 11 years

**Total Funding:**

- 2008-2010: $34,800
- 2000-2002: $64,612

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The concentration of phosphorus detected in leachate remains very low regardless of treatment. The mean concentration of phosphorus detected in leachate since initiating phosphorus treatments in 2003 has been less than 0.02 mg L⁻¹.
Golf courses represent excellent sites for examining the potential migration of pharmaceutical personal care products (PPCPs) and endocrine-disrupting compounds (EDCs) toward the water table. Recently, arid southwestern cities like Las Vegas, Phoenix, and Palm Springs have been installing distribution systems that allow golf courses to utilize reuse water. Additionally, reuse water is sometimes supplied to infiltration basins that augment natural recharge to phreatic aquifers.

Assessing and evaluating the vadose-related attenuation processes may help determine whether accumulation of these compounds occurs in shallow groundwater systems. Many studies over the last two decades show that turfgrass is extremely effective in retaining and biodegrading trace contaminants such as many pesticides. In particular, research shows that the thatch layer, which is the matted layer of grass debris and soil organic matter near the surface, is a highly active "biofilter." This project is investigating the fate and transport of PPCP/EDCs to determine the effectiveness of turf in mitigating their vertical movement (i.e., leaching).

An inter-laboratory comparison of sample analyses has been completed. The results from the two laboratories are fairly consistent and are considered acceptable. Additional samples will be sent to both labs for comparative analysis and evaluation. Aerobic and anaerobic soil conditions were evaluated for their effectiveness in degrading PPCPs. Most PPCPs were stable in anaerobic conditions versus being attenuated in aerobic soil conditions. Drainage has been observed in all 12 of the high leaching fraction lysimeters while less than half of the low leaching fraction lysimeters are draining. Three of the 17 compounds have been detected in the drainage samples that were selected based on meeting pore volume displacement goals. The three are bisphenol A, primidone, and sulfamethoxazole. It is possible that the bisphenol detections are from equipment contamination and the research team is evaluating that possibility.

The field plot study is almost complete. There are two soil types - sandy loam and loamy sand - and two irrigation rates, low and high, being studied. Early results are:

**Sandy loam soil**

The same four PPCPs were detected in the low and the high irrigation rates, but generally at higher concentrations under high irrigation. They are: primidone, sulfamethoxazole, trimethoprim, and carbamazepine. Removals through the soil were about 59, 99, 87 and 85%, respectively.

**Loamy sand soil**

Three of the four PPCPs detected in the sandy loam soil leachate were also detected in loamy sand leachate at the low irrigation rate. All four were detected at the high irrigation rate. Removals were about 77 - 88%.

Initial data from all field sites is being collected and analyzed. Equipment issues at Industry Hills have been an ongoing challenge for data and sample collection. Financial hardships at the course in Palm Springs have lead to a temporary delay in data and sample collection.

The research project is on schedule and is scheduled for completion in July 2010. The research team is looking to extend some of the sampling of the soil column experiment and golf course field sites past the July 2010 deadline. Achieving the soil/water balance equilibrium would be of interest in order to get a complete picture of the degradation and drainage flux parameters.

**Summary Points**

- Aerobic and anaerobic soil conditions were evaluated for their effectiveness in degrading PPCPs. Most PPCPs were stable in anaerobic conditions versus being attenuated in aerobic soil conditions.
- Three of the 17 compounds have been detected in the drainage samples that were selected based on meeting pore volume displacement goals.
- Primidone, sulfamethoxazole, trimethoprim, and carbamazepine were detected in the low and high irrigation rates, but generally at higher concentration under high irrigation. Removals through the sandy loam soil were about 59, 99, 87, and 67%, respectively.
- Three of the four PPCPs detected in the sandy loam soil leachate were also detected in loamy sand leachate at the low irrigation rate. All four were detected at the high irrigation rate. Removals were about 77 - 88% in loamy sand soil.
Audubon International strives to educate, assist, and inspire millions of people from all walks of life to protect and sustain the land, water, wildlife, and natural resources around them. In 1991, Audubon International launched the Audubon Cooperative Sanctuary Program for Golf Courses (ACSP), an environmental education program designed to help golf courses play a significant role in enhancing and protecting wildlife habitat and natural resources. The ACSP provides an advisory information service to help golf courses conduct environmental projects and achieve positive recognition for their efforts.

In addition, the Audubon Signature Program works closely with planners, architects, managers, and key stakeholders of new golf course developments to merge wildlife conservation, habitat enhancement, resource conservation, and environmental improvement with the economic agenda associated with the development. Involvement in the development stages of a project enables Audubon International to ensure that biodiversity conservation, environmental quality, and sustainable management are built into the project and continue after construction is completed.

On average, golf courses in the Audubon Cooperative Sanctuary Program will convert 22 acres of turfgrass into natural areas. "At Smock Golf Course in Indianapolis, we choose to naturalize many areas on our course; The key part of that decision was base largely on enhancing the player's experience and ongoing monitoring of the same experience, visually, pace of play, etc. An unexpected side benefit was our improved image/identity from our customers, and community's perspective" states Jan Tellstrom, PGA Professional.

Diamondback terrapins are small turtles that spend their lives in brackish estuaries and are listed as "Species of Special Concern" by the State of New Jersey. Excluder traps installed at Marriott's Seaview Resort and Spa protect diamondback terrapin eggs safe from golfers and predators such as fox and birds.

An interesting point about this particular eagle nest is that it is located between two Signature Program member properties—Raptor Bay Golf Club, FL, and West Bay Golf Club, FL, to the north. So, not only one, but two properties, made design decisions based on the proximity of the nest to land to be developed.

Interpretive signage at the entry to the Sequia Major wetland shows plant and animal species that may be encountered along the trail and boardwalk within the wetland. The golf course was integrated around the large, lush wetland that has been designated an Area of Natural Interest by the Generalitat of Catalonia. PortAventura Golf Course, Tarragona, Spain.

2009 USGA Turfgrass and Environmental Research Summary
The Audubon Cooperative Sanctuary Program for Golf Courses

Josh Conway
Audubon International

Objectives:

1. Enhance wildlife habitats on golf courses by working with the golf course superintendent and providing advice for ecologically sound course management.
2. Encourage active participation in conservation programs by golfers, golf course superintendents, golf officials, and the general public.
3. Recognize the people who are actively participating in environmentally responsible projects.
4. Educate the public and golfing community about the benefits of golf courses and the role they play relative to the environment and wildlife.

Start Date: 1991
Project Duration: ongoing

Audubon International is a non-profit environmental organization that envisions communities becoming more sustainable through good stewardship of the natural environment where people live, work, and recreate through responsible management of land, water, wildlife, and other natural resources. Since 1991, it has worked in partnership with the USGA to offer the Audubon Cooperative Sanctuary Program for Golf Courses (ACSP), an award-winning education and certification program that promotes ecologically-sound land management and the conservation of natural resources on golf courses. In addition, Audubon Signature Programs provide comprehensive environmental education and planning assistance to new developments.

Today, more than 2,300 golf courses in 36 countries participate in the ACSP for Golf Courses. More than half of those enrolled have developed an environmental plan to guide management of the golf course and 688 have achieved certification for their outstanding best practices. We also awarded 793 "Certificates of Achievement" to recognize golf courses for outstanding accomplishments to improve wildlife habitat, save water, conserve energy, and reduce waste.

Audubon International provided environmental planning services to 164 projects (137 golf-related) development projects in 37 U.S. states and in 11 countries, covering 75,000 acres of land, through the Audubon Signature Programs in 2008/09. Five projects achieved certification, bringing the total number of golf courses that have been designated as Certified Audubon Signature Sanctuaries to 72 in 26 US states and 5 countries, 32 of which were recertified.

An analysis of environmental improvements on certified golf courses compared with golf courses that are enrolled but not yet certified revealed significant differences. Certified courses are two-to-four times more likely to be involved in environmental projects, such as exotic plant removal; prairie, wetland, and stream restoration; and endangered/threatened species reintroduction.

Efforts to enhance wildlife habitats on golf courses are done on a regional and site-specific basis. In addition to participation in the program's environmental management and certification activities, golf courses are invited to participate in wildlife monitoring events and activities. Birdwatching teams from 47 golf courses each identified and average of 56 bird species during the 2009 North American Birdwatching Open, a 24-hour bird count conducted at the height of bird migration in March, April, and May. (Start date based on location.)

Audubon International encourages active participation in conservation programs by golfers, golf course superintendents, golf officials, and the general public through a variety of outreach and education activities. In 2005, Audubon International, the United States Golf Association, and The PGA of America launched the Golf and the Environment Initiative to foster environmental awareness, action, and positive results throughout the golf industry. A web site, www.golfandenvironment.com serves as a clearinghouse of golf and environment information. In addition, a Green Golfer pledge drive to engage golfers has been launched. Currently there are over 7,000 Green Golfers who have taken the pledge and over 100 Golf Courses involved in the Green Golf Challenge.

Over 2,000 people took part in seminars and field training conducted by Audubon International staff in 2008/09. More than 500 conservation organizations are directly involved with golf courses as a result of their participation in the ACSP.

Summary Points

- Number of golf courses enrolled in the Audubon Cooperative Sanctuary Program (ACSP): 2,209
- Number of golf courses enrolled in the Audubon Signature Program: 137
- Total number of acres registered in ACSP for Golf Courses: 564,185
- Number of conservation organizations directly involved with golf courses as a result of their participation in the ACSP: >500
- Average number of bird species sighted on 72 golf courses participating in the 2007 North American Birdwatching Open: 56
- Number of Certified Audubon Cooperative Sanctuary Golf Courses: 688
- Number of Certified Audubon Signature Golf Courses: 72
- Total number of golf courses certified in Environmental Planning: 1,336
- Number of golf course personnel and others educated by Audubon International in 2005/06 through seminars, conference presentations, and site visits. > 2000
The Turfgrass Information Center (TIC), a specialized unit at the Michigan State University Libraries (MSU), contains the most comprehensive publicly-available collection of turfgrass education materials in the world. TIC has over 150,000 records in its primary database, the Turfgrass Information File (TGIF), with over 40% linked to the full-text of the item.

The aim of this ongoing project is to provide exhaustive access to the turfgrass research and management literature, regardless of age or language or format of the source material. With much of that rich heritage previously inaccessible to the majority of even heavily-networked turf scientists, TGIF intends to aggressively continue to move towards providing “a turfgrass library on your desktop.” And, of course, there has to be a physical turfgrass library behind the virtual one, where the cumulative record of turfgrass science is collected, preserved, and made available.

Location of the Turfgrass Information File (TGIF) housed in the Michigan State University Libraries in East Lansing, Michigan.
Since construction started in 1984, the Turfgrass Information File (TGIF), a cooperative project of the USGA and the Michigan State University Libraries' Turfgrass Information Center, has offered access to the turfgrass literature in support of more effective turfgrass research and management. From initial print-only search results by mail or fax from the database, to dial-up access beginning in 1988, to internet access in 1993, to World Wide Web access beginning in 1997, TGIF has steadily grown along with turfgrass science through that time.

Today, turfgrass researchers and students at more than 60 subscribing academic institutions worldwide, corporate and individual subscribers, as well as professional members of the following organizations all have access to TGIF:

- American Society of Golf Course Architects
- Asociación Española de Greenkeepers
- Canadian Golf Superintendents Association
- Golf Course Superintendents Association of America
- Midwest Association of Golf Course Superintendents
- Sports Turf Association
- Sports Turf Managers Association
- Turfgrass Producers International
- Wisconsin Golf Course Superintendents Association

Online use of TGIF data continues a trend of steady, upward growth, reflecting TGIF’s fundamental role in:
- supporting more efficient and thorough support for ongoing turfgrass research,
- expanding golf course superintendents’ ability to verify product claims and evaluate documented research,
- permitting students to explore new concepts within the turf industry,
- enabling all users to discover international diversity in turf management practices as reflected in the literature published worldwide.

TGIF indexes academic journals, professional and trade magazines, organizational publications and special reports, annual research reports, government documents, theses and dissertations, book chapters and books, fact sheets, manuscripts, booklets, registration statements, CD-ROM discs, extension bulletins, etc., as well as the ever increasing galaxy of online materials, such as video clips, podcasts, online presentations, web documents, digitized blueprints, and webinars.

TGIF’s digitization efforts, beginning first with and featuring USGA content such as:

- USGA Green Section Record archive, 1921-present
- USGA Turfgrass and Environmental Research Online, 2002-present
- USGA Turfgrass and Environmental Research Summary, including USGA Green Section Research Progress Reports, 1983-Present

has now expanded to include materials from 17 additional organizational, agency, academic institutions, or individual partners from around the world. In addition, digitization of relevant turf-related theses and dissertations, and out-of-copyright or non-copyrighted ‘classic’ turfgrass works are undertaken in order to permit direct access to complete content.

The Michigan State University Libraries continue to actively seek support to build the TIC Endowment to underwrite ongoing operational costs. Contributions from organizations, industry, foundations, other academic institutions, and individual donors to the Endowment have currently surpassed $2.3 million.

Summary Points:
- Number of TGIF records: over 156,000
- Digitized or online periodical projects hosted in cooperation with partners: 8 public sites, with 3 under construction.
- 7 TGIF-accessible sites, with 3 under construction
- Different periodicals monitored routinely for TGIF inclusion (past or present): 647
- Percentage of TGIF records now linking to full-text content: over 40%
- Number of 2009 TGIF data displays: 1 million +
USGA’s Turfgrass and Environmental Research Program relies on science to attain answers that will help ensure the long-term success of the golf course management industry. Frequently projects may span several years. Many times, however, golf course superintendents need answers to very applied problems to help them meet the many challenges of properly managing golf courses. The Grant-in-Aid Research Program was created to address this need. This program allows directors of all eight USGA Green Section regions to identify applied problems and the appropriate researchers in their regions to solve those problems. Research projects funded under this program most often include cultural aspects of golf course management. Examples include what fungicides work best on a particular disease, or the management of new turfgrass cultivars, renovation techniques, safe and effective use of herbicides, insecticides, or fertilizers. These projects are usually short duration (one to three years), but can offer golf course superintendents answers to practical, management-oriented challenges that they can put into use quickly.
Aggressive organic matter dilution programs are intended to slow loss of aeration porosity and subsequent infiltration rates thereby allowing superintendents to more easily manage their putting greens and lessen the effects of summer bentgrass decline. Our research was done on 9-yr-old ‘Penn A4’ practice putting greens at the Independence Golf Club near Richmond. Prior to initiation of the study, analysis of four randomly-selected cup cutter cores revealed a thatch/mat layer (~0-2" deep) with 5.8% organic matter and an infiltra-
tion rate of 11 inches per hour.

Various combinations of small tines (0.25" inside diameter, id), big tines (0.50" id), and verticutting (3-mm blade) were imposed in late March and early September to provide a range of seasonal surface removal from 0% to 26.6% (Table 1). Verticutter blade spacing was 1", while depth was 0.75"; tine spacing was 1.33" X 1.5", with a coring depth of 2". Heavy sand topdressing of approximately 12 ft³ (1,200 lbs/1000 ft²) was applied on both days of cultivation, supplemented by four light topdressings of 0.15 ft³ every 4-6 weeks between cultivations, for a seasonal total of about 24.6 ft³. Cultivation treatment had no effect on soil temperature, soil moisture, or ball roll distance throughout the 2008 season and were not measured in 2009. At the end of 2008, only those coring treatments that removed 14.8 to 19.6% significantly reduced % OM relative to the untreated control (Table 1). Use of smaller tines-alone, verticutting-alone, or combinations of the two, failed to reduce % OM in 2008.

At the end of 2009, all treatments, except verticutting alone, significantly decreased % OM in the thatch/mat layer relative to the control. Coring spring and fall with 0.5" ID tines on a tight spacing (1.33" x 1.5") to remove approximately 9.8% surface area to a depth of 2" resulted in the least OM (3%) over the two years. These data imply that verticutting to a depth of 0.75" does not remove enough depth of material for adequate organic matter dilution, even though this procedure removes a large amount of surface area (11.8%) with each pass.

To track percent cover or recovery rate following cultivation treatments in 2009, digital images were taken every 7 to 14 days with a light box and analyzed with SigmaScan software. Linear regression was then used to predict the number of days required for each treated plot to return to 99% cover or a non-disrupted putting surface. Fastest spring recovery of 24 days was measured for treatments including small tine coring and verticutting. Large diameter coring or small diameter coring + verticutting on the same day required 31 to 36 days for spring recovery.

Late summer/early fall recovery data were very similar for cultivation treatments that remained the same as their spring counterpart. Verticutting treatments recovered in only 21 days, while large diameter coring-alone required only two extra days of recovery (38 versus 36 days), relative to the spring. Fastest early September recovery of 7 days was with 0.25" tine core aeration + 3-mm blade verticutting where only 2.5% surface removal occurred. Data indicate that as little as 10% surface area removal via spring and fall coring may be sufficient for Virginia conditions, while 15 to 20% annual removal should almost always keep you on the safe side. While verticutting alone provides fast healing, our data indicate that it needs to be combined with at least one annual coring for adequate results.

## Summary Points

- The least disruptive treatment in terms of percent removal (Trt 2, 10%) healed quickly (24 to 34 days) and reduced thatch/mat OM to an acceptable level of 3.4% after two years.
- Verticutting alone each spring and fall (Trt 3) resulted in the second fastest recovery of any treatment (21 to 24 days), but failed to significantly reduce OM to a level below the untreated.
- Treatment 4 resulted in the least amount of days of disruption over the season (38), while also reducing OM to an acceptable 3.7%. Total recovery time was 7 days less than verticutting-alone, with a slightly faster rate of OM reduction.
- Using large tines (0.5" ID) at a close spacing both spring and fall each year (19.6% surface removal, Trt 6) worked best in terms of final OM at 3.0%, but required approximately two extra weeks each season for recovery relative to two passes with small tines or verticutting alone.
- Being ultra-aggressive by removing 26.6% surface area (Trt 7) per year did not work in this trial. Recovery time was significantly delayed without achieving greater OM dilution relative to treatments that removed 10 to 20% surface area.

### Table 1: Cultivation treatments and percent surface area removed.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Surface Area Removed (%)</th>
<th>March 28</th>
<th>Sept 4</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Untreated check</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2. 0.25” tine core aeration X2</td>
<td>5.0</td>
<td>5</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>3. 3-mm blade verticutting</td>
<td>11.8</td>
<td>11.8</td>
<td>23.6</td>
<td>23.6</td>
</tr>
<tr>
<td>4. 0.25” tine core aeration + 3-mm blade verti</td>
<td>2.5</td>
<td>2.5</td>
<td>4.8</td>
<td>4.8</td>
</tr>
<tr>
<td>5. 0.5” tine core aeration + 0.25” tine core</td>
<td>9.8</td>
<td>9.8</td>
<td>19.6</td>
<td>19.6</td>
</tr>
<tr>
<td>6. 0.5” tine core aeration X2</td>
<td>9.8</td>
<td>9.8</td>
<td>19.6</td>
<td>19.6</td>
</tr>
<tr>
<td>7. 0.5” tine core aeration + 3-mm blade vertic</td>
<td>9.8</td>
<td>9.8</td>
<td>19.6</td>
<td>19.6</td>
</tr>
<tr>
<td>8. 0.25” tine core aeration X2</td>
<td>5.0</td>
<td>5</td>
<td>14.8</td>
<td>14.8</td>
</tr>
</tbody>
</table>

2009 USGA Turfgrass and Environmental Research Summary
Impact of Sand Type and Application Rate of Fairway Topdressing on Soil Physical Properties, Turfgrass Quality, Disease Severity, and Earthworm Castings

Jason J. Henderson and Nathaniel A. Miller
University of Connecticut

**Objectives:**
1. Determine whether particle size distribution and/or application rate will affect turfgrass quality, disease incidence, and earthworm activity.
2. Quantify the effects of particle size distribution and topdressing layer depth on soil physical properties.
3. Use the resultant data to make recommendations to improve the practice of fairway topdressing.

Although selecting a sand that meets USGA specifications for particle size distribution may not be necessary, it is often subjectively suggested that the sand not be too fine or too coarse.

The cost of a USGA sand does not improve the practicality of implementing this program for many golf courses. The impact of using sand that does not meet USGA specifications, however, has not been thoroughly investigated. Particle size distribution will likely affect infiltration and water retention at the playing surface. Topdressing materials that are too fine may retain excess moisture, whereas, a sand that is too coarse may predispose a large portion of the course to moisture stress. The short- and long-term impact of topdressing native soils is unknown.

This study was initiated on an ‘L-93’ creeping bentgrass (Agrostis stolonifera) stand managed as a golf course fairway at the University of Connecticut Plant Science Education and Research Facility in the summer of 2007.

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**Start Date:** 2007
**Project Duration:** two years
**Total Funding:** $6,000

Fairway topdressing is a relatively new cultural practice that is being adopted by several golf course superintendents throughout the United States to improve playing conditions. Some of the benefits reported have been improved drainage, less disease, and firmer fairways. The benefits to fairway topdressing seem unanimous, but the practice requires a significant budget, considerable labor, time, and commitment to implement properly. Additionally, many questions remain unanswered with regards to topdressing material selection, application rates, and the turfgrass management implications as the topdressing layer accumulates.

Sands used in USGA putting green construction and subsequent topdressing have been thoroughly researched to optimize macroporosity while maintaining sufficient water holding capacity. However, due to the strict specifications, these sands are prohibitively expensive when considered for use on larger fairway acreage. Therefore, recommendations for fairway sands are often very general.

This study was initiated on an ‘L-93’ creeping bentgrass (Agrostis stolonifera) stand managed as a golf course fairway at the University of Connecticut Plant Science Education and Research Facility in the summer of 2007.

- **Summary Points**
  - Topdressed plots showed a faster green-up response than the untreated control plots in mid-April regardless of sand type. Plots that received higher rates of application exhibited a greater greening response than plots receiving lighter rates of application.
  - Topdressed plots exhibited less dollar spot incidence than untreated plots. Plots that received higher rates of topdressing had less dollar spots than plots that received lower rates of topdressing, regardless of sand type.
  - Topdressed plots exhibited less earthworm castings than untreated plots. Plots that received higher rates of topdressing had less earthworm castings than plots that received lower rates of topdressing, regardless of sand type.
  - Topdressed treatments had higher resistance to penetration than the untreated control plots, demonstrating a firmer surface than the untreated controls. The fine sand had the greatest resistance to penetration, followed by the medium sand and the coarse sand, respectively. Plots receiving higher rates of topdressing exhibited greater firmness than plots receiving the lower rates.
  - Untreated controls had the highest volumetric soil moisture content in the top 2" of the playing surface compared to all topdressing treatments. The fine and medium sand treatments hold more water than the coarse sand treatments. Regardless of sand type, the higher the rates of application, the less water is held in the top 2" of the playing surface.
Growing concern over the sufficiency and variability of present water supplies in western Texas and other areas of the arid southwest led to the examination of several reduced-input turfgrass species for water conservation. Buffalo grass \( \text{Buchloe dactyloides} \) (Nutt) Engelm.] is a warm-season turfgrass that shows excellent drought, cold, and salinity tolerance. Increasing awareness and acceptance of buffalo grass as a viable turfgrass option in arid environments requires investigation into conversion techniques for its establishment.

Research was conducted at the Texas Tech Turfgrass Research Station during the summer of 2008 on an established bermudagrass rough. The parameters evaluated included four seedbed preparation treatments and two buffalo grass seeding rates. Bermudagrass was sprayed with glyphosate at 32 oz/A using small plot spray equipment five weeks prior to seedbed preparation. A second application (32 oz/A) was applied after bermudagrass began to regrow (one week prior to seeding).

All plots were scalped following herbicide application and subsequent desiccation of bermudagrass. Seedbed preparation treatments consisted of verticutting in two directions, verticutting plus topdressing (0.6 cm layer), verticutting plus topdressing plus aeration (hollow-tine), or no seedbed preparation. 'Texoka' buffalo grass was examined at seeding rates of 3 or 4 lbs/1000 ft\(^2\) and was planted on June 1, 2009. A starter fertilizer was applied at seeding, and all plots were lightly brushed to ensure good seed to soil contact.

Plots were irrigated daily by an automated irrigation system that applied approximately 2 inches of water per week. Plots were mowed once a week to a height of 2 inches with a rotary mower. Treatments were arranged in a randomized complete block design with four replications of treatments. Buffalo grass conversion was visually evaluated weekly for the first 2 months and monthly thereafter using a scale of 0 (no cover) to 100% (complete cover).

Chemical control of bermudagrass was more successful in year two when glyphosate was applied in split applications. Bermudagrass regrowth during the duration of the trial was minimal. Buffalo grass establishment increased when seed was applied at the higher rate (4 lbs/1000 ft\(^2\)) regardless of seedbed treatment.

Verticutting, aeration, and topdressing treatments increased buffalo grass cover by 34 to 56% and 57 to 77% when seeded at 3 and 4 lbs/1000 ft\(^2\), respectively, 2 MAP (months after planting) compared to no seedbed preparation. Percent buffalo grass cover 3 MAP was 71, 81, and 86% for topdressing, aerification plus topdressing, and verticutting plus topdressing treatments seeded at 4 lbs/1000 ft\(^2\), respectively.

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Comparison of Chlorothalonil, Propiconazole, and Iprodione Products for Control of Dollar Spot and Brown Patch Diseases

Peter Landschoot and Michael Fidanza
Pennsylvania State University

Objectives:

1. To determine if different fungicide products containing chlorothalonil, propiconazole, or iprodione provide differences in control of dollar spot and brown patch diseases of bentgrass maintained as a golf course fairway.

Start Date: 2008
Project Duration: two years
Total Funding: $6,000

Chlorothalonil, propiconazole, and iprodione are among the most popular and effective fungicides for controlling foliar diseases of turfgrasses. Patent protection for the original chlorothalonil, propiconazole, and iprodione products has expired, and several 'post-patent' products are now available to golf course superintendents for use in disease control programs. Questions remain about the handling, mixing, and efficacy of individual products.

Three fungicide trials were conducted on bentgrass maintained as a golf course fairway at two different locations in Pennsylvania (a golf course in the southeastern portion of the state and a research facility in central Pennsylvania). Treatments included three chlorothalonil products (Daconil Ultrex, Echo Ultimate, and Chlorothalonil DF), four different propiconazole products (Banner MAXX, Propiconazole 14.3, Propensity 1.3ME, and Spectator Ultra 1.3), and three iprodione products (Chipco 26GT, Ipro 2SE, and Raven). All products were applied at the same rate and timing for each active ingredient.

Results for two growing seasons showed differences in disease severity among active ingredients in two of the three trials, and a few differences among products with the same active ingredient in all three trials. At the southeastern Pennsylvania site, few differences in dollar spot incidence (number of spots per plot) were observed among chlorothalonil, propiconazole, and iprodione active ingredients. No differences in dollar spot incidence were detected among the chlorothalonil products or the propiconazole products in any rating date in both years; however, on the majority of rating dates, one product provided better dollar spot control than chlorothalonil and propiconazole.

Iprodione products generally showed better dollar spot control than chlorothalonil and propiconazole products. With one exception (one trial during 2009), chlorothalonil products included in this study performed similarly in controlling dollar spot. Generally, chlorothalonil and iprodione products provided better brown patch control than propiconazole products. No differences among propiconazole-containing products were observed with respect to dollar spot control in any of the dollar spot trials. Differences in brown patch control were detected among certain products on one rating date in both years; however, on the majority of rating dates, no differences were detected among propiconazole products.

No differences in dollar spot severity (visual ratings of disease severity on a scale of 0 to 10) were observed among the chlorothalonil products in 2008. However, in one of the tests during 2009, the Chlorothalonil DF treatment showed a higher level of dollar spot severity compared to the other chlorothalonil products. No differences in dollar spot severity were detected among the propiconazole products in either year of the study. Two of the three iprodione products (Chipco 26GT and Ipro 2SE) showed similar dollar spot incidence during both years of the study. One iprodione product, Raven, showed greater disease incidence compared to Chipco 26GT and Ipro 2SE on two of the seven rating dates in 2008. However, a new (2009) formulation of Raven performed similarly to the other iprodione products in 2009.

Two tests were conducted at the central Pennsylvania site: one on creeping bentgrass with a history of severe dollar spot infestations, and the other on colonial bentgrass with a history of moderate dollar spot and brown patch infestations. In both years of the study, differences in dollar spot severity were observed among chorothalonil, propiconazole, and iprodione active ingredients, with iprodione generally showing less dollar spot than chlorothalonil and propiconazole.

No differences in dollar spot severity (visual ratings of disease severity on a scale of 0 to 10) were observed among the chlorothalonil products in 2008. However, in one of the tests during 2009, the Chlorothalonil DF treatment showed a higher level of dollar spot severity compared to the other chlorothalonil products. No differences in dollar spot severity were detected among the propiconazole products in either year of the study. Two of the three iprodione products (Chipco 26GT and Ipro 2SE) performed similarly with respect to dollar spot severity in 2008. Raven showed higher severity ratings on three of the nine rating dates. In 2009, the new formulation of Raven was not significantly different from Chipco 26GT and Ipro 2SE with respect to dollar spot severity.

In the colonial bentgrass trial, all fungicide treatments were applied prior to brown patch symptom development. By mid-summer 2008 and 2009, differences in brown patch severity were observed among chorothalonil, propiconazole, and iprodione fungicides; with chlorothalonil and iprodione generally showing less disease than propiconazole. No differences in brown patch severity were observed among the chlorothalonil products or the iprodione products on any rating date.

Summary Points

- Iprodione products generally showed better dollar spot control than chlorothalonil and propiconazole products at the central Pennsylvania location. However, in the southeastern Pennsylvania trial, the three different active ingredients were very similar in controlling dollar spot.
- Generally, chlorothalonil and iprodione products provided better brown patch control than propiconazole products.
- With one exception (one trial during 2009), chlorothalonil products included in this study performed similarly in controlling dollar spot and brown patch.
- No differences among propiconazole-containing products were observed with respect to dollar spot control in any of the dollar spot trials. Differences in brown patch control were detected among certain products on one rating date in both years; however, on the majority of rating dates, no differences were detected among propiconazole products.
- Of the three iprodione products, Chipco 26GT and Ipro 2SE provided the most consistent dollar spot control. Raven showed less dollar spot control in 2008 when compared with Chipco 26GT and Ipro 2SE; but a new formulation of Raven performed similarly to the other iprodione products in 2009.
Leaf Cuticle Characteristics and Foliar Nutrient Uptake by a Cool-season and Warm-season Turfgrass

Michael D. Richardson, James C. Stiegler, Douglas E. Karcher, and Aaron J. Patton
University of Arkansas

Objectives:

1. Determine the seasonal dynamics of the turfgrass cuticle in a cool-season and warm-season turfgrass species managed under putting green conditions.
2. Compare the seasonal uptake of foliar nitrogen by a cool-season and warm-season turfgrass species managed under putting green conditions.

Start Date: 2008
Project Duration: two years
Total Funding: $6,000

Foliar fertilization refers to the process of nutrient uptake through the foliage or other aerial plant parts. Foliar fertilization has been observed to be an increasingly common practice in today's golf course management. Recent surveys of Arkansas golf course superintendents indicate that nearly all golf course superintendent use foliar fertilization on some area of their golf course, and this method of nutrient application often comprises a major portion of annual nitrogen (N) inputs to putting greens.

Although previous research has documented uptake of N by turfgrass leaves in controlled-environment studies, there have been no studies which demonstrate its potential effectiveness in a field environment. It is known from previous agricultural research that environmental factors and seasonal dynamics of leaf cuticle characteristics can influence the foliar absorption of N solutions. Therefore, the aim of this project is to increase scientific understanding of the turfgrass leaf cuticle, while assessing foliar uptake of N during a two-year putting green research trial.

Experimental areas of 'Penn A1' creeping bentgrass and 'Tif Eagle' ultradwarf bermudagrass were developed on an established sand-based putting green in Fayetteville, Arkansas. The greens were maintained according to typical management practices for the region. Foliar uptake of N was studied each month from May to September to determine if foliar uptake was consistent across the growing season.

An isotopic tracer technique that allows for positive identification and direct measurement of fertilizer N in the plant tissue was used in the study. Solutions of 15N-labeled urea (~2 atom %) were sprayed at two different rates to represent a low and high rate (0.10 or 0.25 lb. N / 1000 ft²) common to foliar fertilization rates of golf course superintendents. For a 24-hour period after treatment, plots received no irrigation or rainfall in order to track only foliar absorption of N. Plant tissues were sampled at 1, 4, 8, and 24 hours after application. In addition, an enclosed chamber with an internal acid trap was installed within the plots after treatment to estimate the volatilization of N as ammonia from the plots. Plant cores were sampled and analyzed for total wax load associated with the cuticle and for components of the cuticle wax.

Results from our two-year field trial indicate that both species are receptive to foliar uptake of urea-N and ammonia volatilization losses were minimal (averaged < 2% of applied N). Ranges of 24-57% and 32-67% of the fertilizer N applied were recovered in leaves/shoots at 1 hour after treatment across months in 2007 and 2008, respectively. Peak foliar absorption was normally observed around 4 hours after treatment and the vast majority of the N, which would enter the leaf tissue after 24 hours, had already been absorbed at an earlier time interval.

Foliar uptake, when measured as a percentage of N applied, was significantly reduced at higher application rates. While this effect was statistically significant and observed on both species, it was not likely agronomically significant as the difference was only 5% (low vs. high rate) when averaged over all treatment dates and sampling times.

Absorption of urea-N by bentgrass and ultradwarf bermudagrass leaves was affected by month of application and year. Depending on month and/or year of application, values for foliar uptake of urea-N at 24 hours ranged anywhere from 36-70% of that which was applied on both species. Overall, foliar absorption efficiency, after spraying urea-N to putting green turf, was variable and there were no consistent seasonal trends associated with these differences.

Chemical extraction of bentgrass leaf cuticle wax in the laboratory showed a highly significant relationship between total wax load and percentage of N recovered in leaf/shoot tissue; however, the variability associated with our foliar uptake values could not be fully explained by cuticle wax amounts alone. Clearly, the foliar absorption process is complex and continued research is needed to better understand the overriding influences and factors in order to maximize the efficiency of foliar applications on golf course putting green turf.

Summary Points

- Both creeping bentgrass and ultradwarf bermudagrass greens are receptive to foliar uptake of urea nitrogen.
- Most of the nitrogen applied to putting green turfgrass foliage is absorbed in the first 4 hours after application.
- Foliar uptake in creeping bentgrass was reduced during warmer months, suggesting a change in the composition of the leaf cuticle.
- Loss of foliar-applied nitrogen to ammonia volatilization appears to be minimal.
Zoysiagrasses offer increased density, improved shade tolerance, and reduced fertility requirements compared to other species historically used on golf course fairways in the transition zone. Consequently, the use of zoysiagrass in the transition zone is increasing.

One of the most troublesome weeds to control in zoysiagrass is common bermudagrass (*Cynodon dactylon*), as physiological similarities between these species often render them susceptible to the same herbicide chemistries. Although research has shown that applications of fluazifop (Fusilade II) mixed with triclopyr (Turflon Ester) can provide bermudagrass suppression with minimal zoysiagrass injury, superintendents still struggle to control bermudagrass infestations in zoysiagrass fairways.

Research was initiated in 2009 at the East Tennessee Research and Education Center in Knoxville, TN on a stand of ‘Zenith’ zoysiagrass infested with common bermudagrass. Plots measuring 5’ X 5’ were arranged in a 4 X 6 factorial design replicated three times. Herbicide treatment and application timing served as factors in this study.

The four herbicide treatments evaluated were: (1) fluazifop at 6 fl oz/A + triclopyr at 32 fl oz/A; (2) fluazifop at 12 fl oz/A + triclopyr at 32 fl oz/A; (3) fluazifop at 18 fl oz/A + triclopyr at 32 fl oz/A; and (4) untreated check. These treatments were applied at 6 timings: 200 growing-degree-days (GDD), 450 GDD, 825 GDD, 1,275 GDD, 1,775 GDD, and 2,250 GDD. Yearly accumulated GDD were calculated beginning on January 1.

Bermudagrass suppression and zoysiagrass injury were assessed weekly using a visual scale from 0 (no injury; vigorously growing, green turf) to 100% (severely injured, necrotic, brown turf) until suppression/injury subsided. Bermudagrass suppression and zoysiagrass injury were also assessed quantitatively by collecting relative chlorophyll index data with a hand-held chlorophyll meter. While this experiment is still ongoing, current results indicate that bermudagrass is most susceptible to applications of fluazifop and triclopyr surrounding periods of dormancy. The greatest degree of suppression was observed for applications made to bermudagrass transitioning out of winter dormancy (200 GDD) in spring and entering winter dormancy (1,775 GDD) in fall.

At 35 days after treatment, suppression for applications at 200 GDD and 1,775 GDD measured greater than 82% compared to less than 20% for applications made at other points during the growing season. This response was observed for all fluazifop + triclopyr treatments evaluated. Increasing the rate of fluazifop from 6 to 18 fl oz/A did not improve the efficacy of treatments applied to bermudagrass transitioning out of winter dormancy in spring or transitioning into winter dormancy in fall.

While the maximum-labeled use rate of fluazifop for bermudagrass suppression is 6 fl oz/A, ‘Zenith’ zoysiagrass exhibited tolerance to higher application rates. With the exception of treatments applied in spring (200 GDD, zoysiagrass injury measured less than 15% for all fluazifop rates evaluated. Regardless of fluazifop rate, spring (200 GDD) applications induced greater zoysiagrass injury at 7 days after treatment than any other timing.

Preliminary results suggest that superintendents should structure herbicide programs for bermudagrass control to deliver treatments when this species is transitioning into or out of winter dormancy. This study will be repeated in 2010 to confirm treatment responses observed in 2009.

**Summary Points**
- Preliminary results suggest that bermudagrass is most susceptible to applications of fluazifop and triclopyr when transitioning out of winter dormancy in spring or transitioning into winter dormancy in fall.
- ‘Zenith’ zoysiagrass exhibited tolerance to rates of fluazifop greater than 6 fl oz/A in 2009.
- In 2009, increasing the rate of fluazifop above 6 fl oz/A did not improve efficacy for treatments applied to bermudagrass transitioning out of winter dormancy in spring or transitioning into winter dormancy in fall.
Winter damage to perennial ryegrass (Lolium perenne) and annual bluegrass (Poa annua) is a significant concern for turfgrass managers in northern climatic regions. The problems associated with winter injury of turf are complex and can result from several interacting factors including low temperature kill, crown hydration, desiccation, and low temperature fungi. Among these factors, research suggests that these cool-season grasses are particularly sensitive to winter temperature fluctuations and direct low temperature kill.

The physiological factors that predispose perennial ryegrass and annual bluegrass to freezing injury require further investigation. Therefore, one of the primary objectives of this project is to assess underlying physiological and biochemical factors that contribute to variability in freezing tolerance among cool-season turfgrasses. The long-term goal is to utilize this information to aid in the selection of freeze-tolerant turfgrasses and to help identify management practices that will maximize winter survival of these species.

To date, we have completed two controlled-environment studies that examined major physiological and biochemical changes during cold acclimation of selected freeze-tolerant and freeze-susceptible perennial ryegrass accessions from the University of Minnesota. In summary, we found that freezing tolerance was associated with rapid accumulation of water soluble carbohydrates in crowns during cold acclimation, particularly sucrose.

In addition, tolerant accessions exhibited a higher capacity to alter crown membrane composition (individual lipid classes and saturation levels) that could help to maintain membrane integrity at low temperatures. Therefore, the accumulation of protective compounds, such as sucrose, along with changes in lipid composition during cold acclimation may represent critical mechanisms to help to lower cellular freezing point and improve cellular stability of perennial ryegrass at low temperatures.

Further work is currently being conducted to compare freezing tolerance characteristics of annual bluegrass and creeping bentgrass (Agrostis stolonifera), a freeze-tolerant turfgrass species. Controlled-environment experiments will focus on loss of freezing tolerance (deacclimation) in response to fluctuating soil temperatures and its implications on winter hardiness.

The specific objectives of this project are: (i) quantify the critical temperature thresholds required for deacclimation of annual bluegrass and creeping bentgrass; and (ii) examine early physiological and biochemical changes in response to deacclimating temperatures, including changes in lipid composition, protein expression, and plant carbon balance. This project will be part of a larger collaborative initiative among researchers and turfgrass managers in the Northeast and Canada on applying a comprehensive approach to study winter injury of annual bluegrass.

The results from controlled-environment experiments will be further tested in field trials at the Joseph Troll Turf Research Center. All research results will be posted on the University of Massachusetts Northeast Winter Injury Initiative website, which is available to turfgrass professionals.

Summary Points
- The accumulation of protective compounds, such as sucrose, along with changes in lipid composition during cold acclimation, represent critical mechanisms to help to lower cellular freezing point and improve cellular stability of perennial ryegrass at low temperatures.
- Research is underway to identify practices and conditions that can minimize annual bluegrass turf loss due to winter injury.
- Data obtained from university research will be updated on a website (www.umass turf.org/winterinjury), which is an essential component for disseminating important winter injury information to turfgrass professionals.
**Preliminary Investigation on the Epidemiology of Fairy Ring in Turfgrass**

**Michael Fidanza, Ph.D.**  
Pennsylvania State University

**Objectives:**

1. To develop a technique to facilitate the successful isolation of fairy ring-causing basidiomycete organisms from a turf/soil sample.  
2. To develop a technique to induce the vegetative mycelium from fairy ring-causing basidiomycetes to produce a basidiocarp (i.e., fruiting body or mushroom) that could be used to accurately identify the organism.  
3. To examine weather conditions associated with the appearance of fairy ring symptoms in turf.

**Start Date:** 2009  
**Project Duration:** one year  
**Total Funding:** $3,000

Visual symptoms of fairy ring in turfgrasses are categorized as type I, type II, type III, and all three types can occur alone or in various combinations at the same site. Type I is the most severe, characterized by necrotic or dead turf in circle or arc patterns and often accompanied by soil hydrophobicity and localized dry spot conditions. Type II symptoms are easily identified by the appearance of rings or arcs of stimulated, rapidly growing turf that is typically darker green in color than the surrounding turf. Type III symptoms are simply the appearance of the basidocarps or "fruiting body" (i.e., mushrooms or puffballs) of the fungal causal agent. Fairy ring symptoms in turf are attributed to a basidiomycete fungal organism that interacts with the soil/rootzone biological, chemical, and physical environment thus indirectly causing turf damage or reduction in turf stand quality and function.

Fairy ring is often confirmed in turf if the fairy ring-causing basidiomycete is successfully isolated from the affected area and examined under a microscope. Most mushroom or toadstool-producing fairy ring fungi have distinct clamp connections formed along their hyphal strands. The puffball-forming fungi, however, do not typically form clamp connections.

In the field, the basidiocarp is the most accurate way to identify the species. According to the literature, over 60 species of fairy ring-causing basidiomycetes exist, and their range of susceptibility to fungicides labeled for fairy ring or cultural practices to suppress fairy ring symptoms has not been determined.

Specific extraction and isolation of basidiomycetes from the soil, soil rootzone, or thatch is difficult, tedious, and time consuming, and contamination from the many other fungi and bacteria in the soil is often the result. Most basidiomycete organisms collected from fairy ring-affected turf sites in the Mid-Atlantic region grew well on either potato dextrose agar or malt extract agar, especially when either growth media was amended with an antibiotic. Future research will investigate additional media amendments that may facilitate rapid extraction and growth of these fairy ring-causing basidiomycete fungi from turf/soil samples.

Currently, molecular techniques involving "DNA fingerprinting" are being investigated at North Carolina State University, as well as additional methods of soil extraction and isolation. In this investigation, the basidiomycetes grown in pure culture were transferred into a potting soil mix containing wood chips since these fungi are classified as wood-decaying fungi and therefore prefer a diet of lignin, which is also a major component of thatch. The soil mix was subjected to a series of wet/dry cycles to induce fungal colonization of the soil mix and eventually to produce a basidiocarp or mushroom. Future research will examine methods to rapidly produce basidiocarps to make rapid and accurate identification of the fairy ring-causing species.

Fairy ring symptoms on greens at Riverwind Golf Course (West Deptford, NJ) were visually monitored from May through September 2009 for the appearance of fairy ring symptoms. Weather information was obtained from the Philadelphia International Airport, located directly across the Delaware River from the golf course. Therefore, airport weather data was chosen to represent environmental conditions within this Mid-Atlantic region. Weather data obtained was air temperature (minimum, average, and maximum), relative humidity (minimum, average, and maximum), and natural rainfall (inches).

Five fairy ring outbreaks of type I symptoms were observed in June through August. Although the weather conditions were not strongly correlated with fairy ring outbreaks, an observed trend revealed the onset of type I fairy ring symptoms at this location corresponded to a maximum air temperature > 80°F with no precipitation (i.e., hot and dry conditions) after rainfall occurred (i.e., warm and wet conditions). Therefore, these "wet/dry" cycles may be the key to understanding fairy ring epidemiology in turf.

### Summary Points

- Basidiomycete fungi attributed to fairy ring symptoms in turfgrasses are difficult to isolate from the soil and thatch of affected turf areas, although progress is being made in this area.
- Techniques are being tested to develop a reliable and rapid procedure to identify the species of the fairy ring-causing basidiomycete, and thus may lead to improved recommendations for control.
- With a wide and diverse number of basidiomycetes that cause fairy ring, symptoms can be observed any time of year. However, destructive or type I fairy ring symptoms are often seen during periods of hot/humid and dry weather.
I
n the Intermountain Pacific Northwest, cool-season turfgrasses are typically growing from April through September. With such a short growing season, timing of disruptive aeration practices, which reduce golf green playability, can be difficult.

Twelve aeration dates (every two weeks from April 15 to July 1 and August 15 to November 1, 2008) and two sand treatments: (black sand, BS; and tan sand, TS) were applied at each date in a randomized complete block split-plot design with 4 replications. One week prior to each aeration date, each corresponding sand treatment received 293 kg ha⁻¹ fertilizer (10-4-16).

Aeration was accomplished with a core cultivator (GreensAire 24) with 1.27-cm diameter., side-eject hollow tines at a tine depth of 7.62 cm on 5-cm centers. Cores were removed and topdressing sand was applied. Tan sand topdressing treatments were applied at 40,000 kg ha⁻¹. Black sand topdressing treatments received tan sand at 20,000 kg ha⁻¹ brushed in followed by black sand at 20,000 kg ha⁻¹ to duplicate how black sand is commonly used by golf course superintendents.

Soil temperature was recorded semi-weekly until full recovery. Soil temperature was measured at 7.5 cm depth with a digital thermometer. Full recovery was determined when no visible damage from cultivation was observed in 95% of the plot area.

The most days to recovery occurred in the spring. There was a decrease in the number of days to recovery from the May 1 aeration to the May 15 aeration. Sand types did not differ except at June and September aeration when black sand, had fewer days to recovery than tan sand. In 2008, days to recovery steadily decreased from the May 15 to August 15 aeration. Following the August 15 aeration, the days to recovery increase, but the black sand treatments increased at a slower rate than the tan sand treatments.

There was an increase in soil temperature between the May 1 and May 15 aeration. Soil temperatures were highest in the summer months, as expected, and decreased by the September 15 aeration. Sand type resulted in soil temperature differences, with black sand being higher than tan sand. This was especially noticeable September 15 when the black sand treatments recovered faster than the tan sand treatments.

Black sand had as much as a 25% increase in quality over tan sand. In addition, a decrease in turfgrass quality was noted during the summer. There was a 25% increase in turfgrass color ratings due to black sand at the April 15 aeration. This was the largest difference in color between the two sand types.

Within each rating date, black sand, had a higher color rating than tan sand. The improvement in turfgrass color from the use of black sand was the most dramatic effect seen from the use of black sand. Initially it was thought that there may be a fertility response from the black sand, but a nutrient analysis showed an insignificant amount of plant available nutrients in both sand types.

Summary Points
- The best time to aerate in the Intermountain Pacific Northwest would be about May 15 and August 15.
- Black sand was able to reduce the number of days to recovery in the late fall.
- Black sand resulted in increased quality and color in the spring and fall.
This project compares fertility, traffic, drought survival, and divot recovery of eight bermudagrass genotypes. Included genotypes are commercial cultivars 'Tifway', 'TifSport', 'Celebration', 'Floratex', 'Riviera', and three University of Georgia experimental lines (Hybrid #1, St-5, and T-11).

In Florida there is interest in using new cultivars of bermudagrass on golf course tees and fairways. As an example, 'Celebration' is increasingly being utilized for new courses and renovations. There is limited information comparing the performance of 'Celebration' to standards such as 'Tifway'. Therefore, information involving fertility, traffic, drought response, divot recovery, and their interaction would benefit golf course superintendents trying to make appropriate decisions regarding cultivar selection.

Plots are being evaluated for genetic color, density, turf quality, winter color, thatch accumulation, divot recovery, and drought survival (dry-down and recovery). Artificial traffic is applied to a portion of the plot using a modified Cady traffic simulator that was constructed using a Ryan GA-30 aerifier. Divots are removed from the plots using a divot machine constructed from a modified clay pigeon thrower.

This study is being conducted at the University of Florida Plant Science Research and Education Unit located in Citra, Florida. The plots are approximately three-years-old, having been established in 2006. Main plots (cultivar) are laid out in a randomized complete block (RCB) design with three replications with each main plot being approximately 14' x 14' in size. Each cultivar is split into traffic and non-traffic plots 14' x 7' in size. Each traffic treatment is split into three nitrogen rates (0.5, 0.75, and 1.0 lbs/1000 ft²) applied as a Harrell's 15-5-15 turf fertilizer blend with 50% slow-release nitrogen once every two weeks. Treatments were initiated on August 1, 2009 and continued through November 23, 2009 with visual quality and photographs for digital image analysis being collected once every two weeks. Initial drought response observations are expected to be conducted during the 2010 growing season.

Initial results indicated no difference in turf visual turf quality, color, or density as a result of nitrogen rate. This is supported by digital image analysis which also indicated no relationship between nitrogen rate and percent green cover or digital green color index values. This indicates that the current nitrogen rates are too low and/or too narrow in range to establish differences in growth response.

Traffic treatments reduced green cover by 10-20% in each cultivar at the end of the first season of data collection. Comparing overall turfgrass visual quality along with density and digital green color index as calculated through image analysis, we observed that Hybrid 1, ST5, and 'Celebration' consistently yielded the best visual quality ratings when subjected to traffic stress. 'Celebration' consistently produced the darkest green color, while 'Celebration', ST5, Hybrid 1, and T11 maintained the highest plant density under weekly traffic treatments.

When considering traffic tolerance and divot recovery, the cultivars consistently yielding the quickest divot recovery and best traffic tolerance were Hybrid 1, ST5, 'Celebration', and T11.

Summary Points

- Nitrogen rates of 0.5, 0.75, and 1.0 lb N/1000 ft² are too narrow in range to result in N-rate treatment differences
- 'Celebration' has the darkest genetic color as supported by visual ratings and DGCI analysis
- 'Celebration', ST5, Hybrid 1, and T11 maintained the highest plant density of the eight cultivars after 14 weeks of weekly traffic treatments.
- Cultivars that performed well under traffic stress also recovered most quickly from divot damage.
The chemical and physical properties of a putting green rootzone can be largely altered by the changes of organic matter over time. Meanwhile, spatial distribution and decomposition of organic matter, both quantity and quality, may be reflected from the soil fertilities. Also, the fate of chemicals are affected by the status of soil properties. Environment and cultural practices influence the dynamics of soil properties. However, little information is available for the soil organic matter with depth based on analysis of putting green rootzone mixtures.

The objective of this study was to characterize vertical distribution of soil organic matter as affected by putting green establishment and age. The study consists of two parts. The first part is aimed at investigating space variations of selected soil properties which are used as measures (indicators) of soil organic matter. The second part focused on testing the potential of using FTIR in predicting soil properties.

Soil samples in this study were collected in 2006 from four experimental putting greens that were constructed in sequential years from 1997 to 2000 at the University of Nebraska. Putting green age was 6, 7, 8, and 9 years-old. Treatments are consisted of two rootzone mixtures: a sand/sphagnum peat mixture at 80:20 (v/v) and a sand/sphagnum peat/soil mixture at 80:15:5 (v/v/v). Two nutritional programs during the year of establishment were also included.

Four-inch deep cores of soil samples were collected from each plot and subdivided evenly into 12 layers. Soil pH, cation exchange capacity (CEC), total C, total N, and mineralizable N were measured in each layer. CEC was estimated by ammonium acetate extraction method at pH 7. Total N, organic C of samples were analyzed by the dry combustion method. The 7-day short-term anaerobic incubation method was used to measure mineralizable N. Lignin and cellulose content of the top layer was quantified by modified gravimetric method.

Result to date suggested a significant putting green age effect on lignin and cellulose content in the top layer. For CEC value, establishment effect was not significant, however, age effect was significant. Two-way interactions between establishment and age were also significant. CEC was observed to increase with putting green age due to organic matter accumulation. No significant effect of treatment was observed for pH and mineralizable N.

All the subsamples had been subjected to FTIR analysis. Data analysis of this part is in progress. However, preliminary results showed a good potential to predict some selected properties of putting green rootzones.

Summary Points
- Generally, soil pH increases and soil CEC and mineralizable N decreases from the top to the bottom of the soil profile across years and treatments.
- The soil CEC and pH dynamics in different years after the establishment of putting greens are correlated to accumulation of organic matter.
- No significant difference was observed on mineralizable N among different putting green ages due probably to the balance of N immobilization and mineralization. Mineralizable N as an indicator of labile organic matter will be discussed later.
- Establishment treatments (accelerated and control) had little effect on CEC, pH, and mineralizable N.
- Significant age effect of putting greens was observed for lignin and cellulose content in the top layer. Lignin and cellulose increased in the upper rootzone as the age of putting greens increase.
Unraveling Billbug Seasonal Ecology to Improve Management: Developing a DNA-based Larval Identification Tool

Douglas Richmond, Brandon Schemerhorn and Mohamed Abdelwahab
Purdue University

Objectives:
1. Identify key regions in billbug rDNA sequences that can be used to identify and differentiate major billbug pest species.
2. Create species-specific DNA primers for these same rDNA regions that will allow for rapid and dependable identification of field-collected billbug larvae.

Billbugs are increasingly being recognized as a serious threat to golf course turf across the United States. The larvae of this diverse group of insect species (the billbug species complex) damage both warm- and cool-season turfgrass by feeding on or inside the stems, crowns, roots, stolons, and rhizomes. Recent expansions in the range of several billbug species, possibly driven by increasing interstate movement of turfgrass sod, have resulted in a nationwide collage of billbug species assemblages. The resulting variation in seasonal life histories, behavior, and ecology that often accompany such novel species interactions have made satisfactory management difficult to achieve in many regions.

Although adult monitoring can be used to estimate billbug species composition, insight into billbug larval population dynamics is needed to improve management strategies and identify which species are actually responsible for damage to turfgrass. Because the damaging larval stages of these insects cannot presently be identified to species, the seasonal population dynamics of many common billbug pests remains unresolved, leaving researchers and superintendents with little scientific foundation on which to base and refine management programs.

The long-term goal of this project is to clarify billbug seasonal ecology and elucidate new management opportunities. In working toward this goal, the first phase of this research is focused on developing a DNA-based billbug larval identification tool that will facilitate the basic, regional studies of billbug seasonal ecology that are needed to improve management.

Several hundred billbug adult and larval specimens were collected from various locations across the U.S. Specimens include all known billbug pest species; the bluegrass billbug (Sphenophorus parvulus), the lesser billbug (S. minimus), the unequal billbug (S. inaequalis), the Denver or Rocky Mountain billbug (S. cicatristriatus), the hunting billbug (S. venatus), and the Phoenix billbug (S. phoeniciensis). Adult specimens are identified to species using classic morphological characters. After the identity of adult specimens is confirmed, several regions of the rDNA are extracted, amplified, and sequenced to determine which of these regions are most useful for differentiating billbug species.

Portions of the rDNA coding sequences are highly conserved even between distantly related species, allowing the application of ‘universal’ primers for amplification from any species. Non-coding rDNA spacer sequences, however, can be highly variable in length and sequence between closely related species. Concerted evolution acting on rDNA arrays maintains sequence homogeneity within species as it drives differentiation between species, a pattern that explains the utility of rDNA for species-diagnostic assays.

The ITS2 region (internal transcribed spacer region 2) between the 5.8s and 18s ribosomal DNA sequences has been the first target of our investigation. Based on the size and sequence of ITS2, results to date indicate that this region will allow differentiation between S. parvulus (338 base pairs) and S. minimus (463 base pairs) based on the size (number of base pairs) of the region alone. Furthermore, ITS2 will allow us to dependably differentiate these two species from S. cicatristriatus, S. venatus, and S. inaequalis, all of which share an ITS2 region of identical size (348 base pairs) and sequence.

However, because ITS2 does not provide the differences in size or sequence necessary to dependably differentiate all of the billbug species of concern, efforts are being re-directed to examine several other promising regions within the ribosomal RNA multigene family (ITS1, CO1, and IGS).

Summary Points
- A DNA-based billbug larval identification tool could provide researchers with the means to gain a more complete understanding of the seasonal biology of this emerging pest complex on a region-by-region basis, leading to improved management programs.
- The ITS2 region of the rDNA multigene family will allow differentiation between S. parvulus (338 base pairs) and S. minimus (463 base pairs) based on the size (number of base pairs) of the region alone.
- ITS2 will also allow us to dependably differentiate these two species from S. cicatristriatus, S. venatus, and S. inaequalis, all of which share an ITS2 region of identical size (348 base pairs) and sequence.
- Because ITS2 does not provide the differences in size or sequence necessary to dependably differentiate all billbug species of concern, future efforts will focus on other regions within the ribosomal RNA multigene family (ITS1, CO1, and IGS).
**Comparison of Cultivation Techniques in Turfgrass**

**Kai Umeda**  
University of Arizona

**Brian Whittark**  
USGA Green Section

**Objectives:**

1. Compare deep solid-tine aerification with a soil-slicing implement to reduce soil compaction, salinity, and sodium in turfgrass.

**Start Date:** 2008  
**Project Duration:** one year  
**Total Funding:** $3,000

Soil compaction is a problem associated with turfgrass management due to continuous traffic contributed by equipment, golf carts, and players. Irrigation water quality is less than optimal with the use of reclaimed water and the high sodium content contributes to poor soil structure, reduced water infiltration, limited rooting depth, and less favorable nutrient availability for growing and maintaining high quality turfgrass.

Two aeration implements were compared in field experiments, a Vertidrain equipped with 0.625 inch by 10-inch solid tines and a Blec GB1500 soil-slicer with 10-inch slit spacing (6 slits with 0.75-inch width) set to a depth of 4 to 10 inches. On each of four golf courses in 2006, a fairway was cultivated one time with treatments replicated three times in a randomized complete block design and soil samples were collected and analyzed at intervals after cultivation.

Additional soil compaction measurements were observed with the Clegg impact hammer device and infiltrometers attempted to measure water penetration and did not provide consistent data. All aeration treatments exhibited beneficial effects that were observed for 2 weeks after treatment (WAT). Sodium (Na+) reduction was observed by all aeration treatments. Blec treatments tended to enhance Na+ reduction compared to the Vertidrain.

Blec treatments reduced Na+ 25% compared to 18% for the Vertidrain at one site, nearly 20% versus 8%, respectively, at two other sites, and no changes were observed at the fourth golf course. Na+ reduction decreased at 4 and 8 WAT when soil samples were analyzed.

In 2007 and 2008 at only two golf courses, gypsum at 100 lb/1000 ft² was applied in addition to aeration done 1, 2, or 3 times at monthly intervals in June, July, and August. In 2007, electrical conductivity (EC) tended to be reduced at one site for the untreated treatments with or without gypsum compared to the cultivated treatments. Only the Vertidrain treatment done three times exhibited a significantly lower EC at only one site.

Application of gypsum following cultivations contributed to enhanced reduction of Na+ levels. Soil analysis indicated that in 2008, Na+ levels were variable among treatments at most sampling dates. Similarly, the EM-38 salinity measurements were variable among treatments. The TruFirm data was not consistent within treatment replicates.

At the end of the season, gypsum added to both the Blec and Vertidrain cultivations showed only slightly better Na+ reduction relative to the untreated. At one site, only the Vertidrain showed improvement with slightly lowered Na+. Turf quality and density was visually worse for non-cultivated treatments.

In 2008, additional measurements were made for irrigation precipitation rate and distribution uniformity was calculated, salinity was measured using the EM-38, and the TruFirm instrument measured turf firmness. Soil analysis indicated that in 2008, Na+ levels were variable among treatments at most sampling dates. Similarly, the EM-38 salinity measurements were variable among treatments. The TruFirm data was not consistent within treatment replicates.

**Summary Points**

- Cultivation techniques exhibited short-term reduction of salinity and Na+ levels.
- Blec and Vertidrain cultivations were effective in reducing Na+ levels though not consistent at all sites or all years.
- Blec and Vertidrain cultivations were effective in reducing Na+ levels for up to 2 weeks.
- Application of gypsum following cultivations contributed to enhanced reduction of Na+ levels.
- Benefits of multiple cultivations were significant for only one implement in one year.
- Small size plots were not conducive for high variability encountered with irrigation distribution uniformity and soil variability at golf course sites.
- Measuring water infiltration, soil compaction, or turfgrass firmness was not consistent among cultivation treatments.
- EM-38 measurements were not effectively correlated to laboratory soil analysis for salinity measurements.
Wildlife Links Program

Golf courses offer excellent opportunities to provide important wildlife habitat in urban areas. With more than 17,000 golf course in the United States comprising in excess of two million acres, great potential exists for golf courses to become an important part of the conservation landscape.

Wildlife Links is a cooperative program with the National Fish and Wildlife Foundation that funds innovative research, management, and education projects that help golf courses become an important part of the conservation landscape. The United States Golf Association is providing through the Wildlife Links Program which began in 1996. The objectives of Wildlife Links are to:

1. Facilitate research on wildlife issues of importance to the golf industry.
2. Provide scientifically credible information on wildlife management to the golf industry.
3. Develop wildlife conservation education materials for the golf industry and golfers.
4. Implement wildlife monitoring programs that will improve management on golf courses.

Location of the following project funded in 2009 through the Wildlife Links Program administered by the National Fish and Wildlife Federation
Reproductive Success and Habitat Use of Painted Buntings on Golf Courses in Coastal South Carolina

Patrick G.R. Jodice, Mark Freeman, and Jessica Gorzo
Clemson University

Chris Marsh
Low Country Institute

Objectives:
1. Locate nests of Painted Buntings during two breeding seasons via nest searching on study courses.
2. Measure various metrics of reproductive success.
3. Measure various metrics of nesting habitat at the nest, patch, and territory scale (e.g. nest substrate and height, distance to edges, canopy cover, shrub density) and relate to reproductive success.
4. Conduct surveys of Painted Buntings and other songbirds on golf courses in coastal South Carolina and relate presence to landscape-scale and patch-scale habitat variables.

Summary Points
- Only 18 Painted Bunting nests were located in two breeding seasons; nests were difficult to locate and well-hidden.
- Nests were located in a variety of plant types/trees/habitat structure ranging from low, dense scrub to moss nests in upper reaches of mature oaks.
- Nest failures were common and occurred during both incubation and chick-rearing.
- Brown-headed Cowbirds, a nest parasite of songbirds, were common on golf courses and cowbird eggs and chicks were frequently identified in Painted Bunting nests.
- A new research focus for 2010 will use surveys from multiple golf courses to determine landscape- and patch-scale habitat variables associated with Painted Bunting occurrence.

The Painted Bunting (Passerina ciris) has been placed on the Audubon Watch List due to a steady population decline since 1966. Painted Buntings also are a high priority species in Wildlife Action plans in both North and South Carolina. Painted buntings have established two breeding populations in the US, one in the southcentral states and one in the coastal southeast. The southeastern distribution ranges from southern North Carolina to northern Florida.

Survey data show that many buntings inhabit urban and agricultural landscapes in the northern coastal and southern and central inland portions of their range. Within the coastal counties of South Carolina, golf courses and golf course communities are common and becoming more prevalent. This region was historically characterized by a mix of forest, agricultural land, and rural communities. The coastal counties are now characterized by a highly developed urban matrix with large parcels of isolated green space comprised of golf courses. This changing landscape presents management concerns for Painted Bunting populations.

Our research seeks to assess the potential conservation value of golf courses in the Beaufort County area of South Carolina for Painted Buntings. During the breeding season of 2008 and 2009, we searched the Spring Island and Chechessee Creek Clubs in Okatie, South Carolina for nests of Painted Buntings. Once found, nests were monitored until they succeeded or failed. We recorded signs of success or failure, nest height, nest plant species, and GPS location.

During these two seasons, we only located 18 nests. Of these, 13 were located in wax myrtles or shrub/scrub habitat and the remainder in trees. Three nests occurred in clumps of Spanish moss that were located high in mature oaks. This appears to be a unique nesting substrate for Painted Buntings and one that maybe restricted to habitats with mature, large oaks.

Most nests were located during incubation. The best cue for nest presence was behavior of adult birds around the nest, although capturing brief windows of adult activity near nests over the study area was difficult. Nest failure was common during both incubation and chick-rearing. Brown-headed Cowbirds (a nest parasite that lays its eggs in nests of songbirds) were common on golf courses and cowbird eggs and chicks were regularly identified in Painted Bunting nests. We also located and mapped the habitat use of 14 groups of Painted Bunting fledglings from summer 2008. Data from both the nest locations and fledgling groups are being incorporated into GIS data layers.

During 2010, we will incorporate a new analysis that will use GIS data to examine small- and large-scale landscape characteristics of Painted Bunting locations on golf courses in the region. We will assess vegetation characteristics with presence/absence of painted buntings, thus lending insight to land management techniques for southeastern coastal golf courses. Unlike our previous efforts of nest searching which focused on two courses, we will instead conduct surveys on a number of courses (10-20) in the region.

Habitat variables and landscape features to be considered that also are of interest to golf course managers include presence of salt marshes, presence or density of houses, connectedness to other golf courses or undeveloped habitats, presence of buntings in adjacent habitats, vegetation type and species composition of out-of-play areas, shape, size, and extent of out-of-play areas, and extent of developed land cover within the course.
The southern Appalachian Mountain region harbors an exceptional amphibian diversity that is dominated by salamanders. These salamanders are integral ecological components of the headwater ecosystems they inhabit where they often account for the majority of vertebrate biomass. They are found in close association with headwater stream habitats, forming communities that are comprised of five to nine species from the genera Desmognathus, Eurycea, Gyrinophilus, and Pseudotriton. These salamanders all have biphasic life cycles consisting of an aquatic larval stage that is followed by a terrestrial adult stage.

Stream salamanders are dependent upon both the stream and the surrounding riparian habitat for foraging, breeding, and dispersal. Though less conspicuous and least studied, the larval stage is essential to the persistence of or the reestablishment of adult salamanders in the surrounding riparian habitat. Managing landscapes with an eye for both human use and preservation of biodiversity can create a win-win situation for stakeholders and wildlife. Considering that the average golf course consist of more than 150 acres of green space (70% is rough, non-play areas) and there are more than 17,000 golf courses in the U.S. that total over 2.2 million acres, we suggest there is great potential for golf courses to serve as sanctuaries for many wildlife species if the habitat needs of species are present.

Our project measured stream salamander abundance and diversity in order to make biologically relevant management suggestions to improve the quality of golf courses for stream salamanders. We sampled 10 golf courses in the mountainous region of the southern Appalachians within a 25-mile radius of Highlands, NC to compare upstream, through-course, and downstream areas. We also conducted chemical analyses of leaf litter in streams to test for the presence of insecticides, herbicides, and fungicides that may affect the survival of aquatic larvae.

During the 2009 summer field season, Mark Mackey (PhD student) and two field technicians were able to catch over 2,300 salamanders of seven species on the 10 golf courses involved in this study. They sampled salamander larvae using leaf litter bags that were placed in streams to act as aquatic refugia. Each course contained 30 leaf bags, with a total of 300 leaf bags in the study. Each of the 300 leaf bags was checked three times throughout the field season producing a total of 1,015 salamanders (Figure 1).

Adult salamanders were sampled by visiting the golf course streams at night. Two researchers would systematically search each bank of the stream, catching or noting all salamanders that were encountered. There were six stream transects per course with a total of 60 in the study. Each of these transects was surveyed three times throughout the field season, producing a total of 1,200 adult salamanders.

A total of seven species of larvae and adults were captured on the 10 golf courses (Figure 2). Twelve different habitat measurements were recorded and quantified at the 60 stream transects at the beginning, middle, and end of the summer field season. Aquatic habitat characteristics measured include pH, dissolved oxygen, water temperature, percent sedimentation, percent of substrate cover (sand, pebbles, conglomerates), amount of coarse woody debris, amount of cover rocks, water surface velocity, stream width, and stream depth.

**Summary Points**

- A total of 2,215 salamanders of seven species were caught on the 10 golf courses during the 2009 field season.
- The 300 total leaf bags were checked three times throughout the field season, producing 1,015 larval salamanders.
- The 60 total stream transects in the study were surveyed on three different nights, resulting in 1,200 adult salamanders.
- Data on 12 important aquatic and terrestrial habitat characteristics, including chemical contaminants, were quantified in order to better understand the influence of golf course management techniques on the stream salamander communities.
Brown-headed Nuthatch Enhancement Study

Mark Stanback
Davidson College

Objectives:

1. Assess the importance of pine density and competition with eastern bluebirds on the spatial distribution of Brown-headed Nuthatch nests.
2. Assess the numerical response of brown-headed nuthatches to the experimental exclusion of eastern bluebirds.
3. Assess the ability of eastern bluebirds to usurp nest sites from brown-headed nuthatches.
4. Provide golf course managers in the Southeast with recommendations to increase the numbers of brown-headed nuthatches on their golf courses.

Start Date: 2007
Project Duration: two years
Total Funding: $7,400

The Brown-headed Nuthatch (Sitta pusilla) is a cooperatively breeding bird endemic to the southeastern United States. But for nearly half a century, its numbers have been in decline. At the same time, another cavity-nesting species has increased dramatically in number in the same region - the Eastern Bluebird (Sialia sialis). Bluebirds tend to fare well in a variety of human-altered habitats and have been the beneficiaries of nest box programs throughout their range.

Nest boxes on six golf courses near Davidson, North Carolina have been monitored since 2001. For each nest box, we measured the distance to the nearest three pine trees (Pinus echinata). The distance to the third was our measure of pine density. Boxes for which the third closest pine was less than 50 meters away were considered to be in "pine-rich" habitat. Boxes for which the third closest pine was more than 50 meters away were considered to be in "pine-poor" habitat.

We modified a subset of the nest boxes to exclude bluebirds. Standard 1.5" entrance holes accommodate both bluebirds and nuthatches; 1.25" holes accommodate nuthatches, but are too small for bluebirds. Prior to the 2005 breeding season, we randomly assigned boxes on three golf courses to the 1.25" treatment.

Results indicate that pine density had no significant effect on nesting by nuthatches. Hole size, however, had a highly significant effect on nuthatch settlement: nuthatches settle where competition with bluebirds is minimized, regardless of local pine density.

Inter-specific competition with bluebirds thus has a major influence on nuthatch distribution. We recorded the number of nuthatch nests on three courses with a subset of 1.25" entrance holes vs. the number on three other courses on which all boxes had standard "bluebird-friendly" 1.5" entrance holes. We monitored boxes in 2004 to ensure that all six courses were similar in their lack of nuthatch numbers. Boxes for which the third closest pine was more than 50 meters away were considered to be in "pine-poor" habitat.

For the 2005, 2006, and 2007 breeding season, we monitored the numbers of nuthatch nests on both experimental and control courses. Numbers of nuthatches increased in each year of the study, suggesting that bluebirds competitively exclude nuthatches from available habitat.

These results clearly demonstrate that nuthatches flourish only where bluebirds are excluded. Prior to the 2008 breeding season, we reversed the treatments on our six golf courses: our three "bluebird-friendly" courses became "nuthatch-friendly" and vice-versa. If bluebirds are truly the determinant of the distribution and abundance of Brown-headed Nuthatches on golf courses in the Southeast, we should expect in 2008 to find bluebirds nesting in boxes that contained nuthatches in 2007.

Of the 32 boxes containing nuthatch nests in 2007, 31 contained bluebird nests in 2008. The competitive superiority of bluebirds is unquestionable. In the absence of bluebirds, nuthatch numbers increase; in the presence of bluebirds, nuthatch numbers plummet.

Golf course managers throughout the US are encouraged to make a subset of course boxes unavailable to bluebirds (via a smaller entrance hole). Course managers can ensure that Brown-headed Nuthatches can thrive. For golf courses in the distributional range of Brown-headed Nuthatches, we recommend that at least one-third of boxes be provided with small holes.

Brown-headed Nuthatch Enhancement Study

Brown-headed Nuthatch numbers have declined throughout the Southeast. Their supposed dependence on old growth pine forests - and susceptibility to habitat alteration - is usually blamed for this decline. We offer an alternative hypothesis - that nest site competition with a burgeoning Eastern Bluebird population is also responsible.

Brown-headed Nuthatch numbers increased dramatically on three golf courses where bluebirds were excluded from one-third of nest boxes. Control courses had few if any breeding nuthatches over the same period.

When bluebird-friendly holes were returned to experimental boxes, bluebirds quickly evicted resident nuthatches.

To prevent monopolization of nest boxes by bluebirds, golf courses in the Southeast should provide smaller entrance holes on a subset of their nest boxes.
We have been working on three local golf courses: Twin Run Golf Course (Hamilton, OH), Hueston Woods Golf Course (College Corner, OH), and Oxford Country Club (Oxford, OH). We have reared cricket frogs in enclosures located in six golf course ponds—three with a 1-meter terrestrial buffer zone and three that were mown up to the edge. We found that the presence of the buffer zone did not affect larval development, as predicted. However, there were large differences in survival among golf courses, suggesting that local management strategies may make some golf courses more suitable for amphibians than others.

We marked and released cricket frogs at the sites where they were reared to determine if presence of the buffer zone influenced terrestrial survival. The number of animals released varied between sites (11–108 individuals) because of differences in the number of animals that reached metamorphosis at each pond. Students searched ponds in the spring and did not recover any marked cricket frogs. This result may have been due to relatively low numbers that were released or because the terrestrial habitat on the course was not suitable.

We released cricket frogs in mown or unmown grass at the Hueston Woods Golf Course. Cricket frogs were dipped in fluorescent powder and released. Hours later, students returned to examine the pattern of the path cricket frogs left to determine if they oriented toward the pond, and how their movement differed between mown and unmown habitat. Preliminary evidence suggested that frogs could use unmown grass for faster and straighter travel, but that frogs preferred taller unmown grass.

For choice studies, cricket frogs were released in eight enclosures that contained both mown and unmown habitat. After three days, a barrier was inserted and the habitats were thoroughly searched for the cricket frogs. Preliminary evidence indicates that cricket frogs were most often found in the unmown habitat, suggesting that this habitat was more favorable to growth and survival.

In another study, cricket frogs were reared with various potential predators found in golf course ponds (bluegill sunfish, grass carp, crayfish, or no predators) and presence or absence of sublethal level of the insecticide imidacloprid, a common insecticide used by one of the golf courses in our study area. We found that fish species essentially eliminated cricket frog tadpoles from ponds—even the grass carp, which is an herbivore frequently stocked in golf course ponds. Preliminary analyses of another study suggest that canopy cover can reduce cricket frog populations, suggested that open to intermediate canopy cover is best for cricket frog populations.

Our results are suggesting that maintaining unmown habitat in areas that are out-of-play and adjacent to ponds may allow cricket frog populations to use golf course ponds for their full life cycle.

**Summary Points**

- Golf course ponds are often suitable for aquatic development and survival because they are open-canopy ponds located in a green area. However, all fish species and some contaminants may put populations at risk, so their use should be minimized in and around wetlands.
- Providing unmown terrestrial habitat adjacent to a pond may be critical for terrestrial survival of amphibian populations, because cricket frogs selected unmown grass over mown grass.
- Field studies on golf courses show that cricket frog tadpoles can survive in ponds with or without buffer zones, which suggests maintaining unmown habitat around all of the pond is not necessary for aquatic life stages. However, terrestrial life stages need adequate unmown habitat. Therefore, leaving part of a pond unmown in out-of-play areas will provide essential terrestrial habitat for cricket frogs and should allow populations to persist at these sites.
Population and Community Responses of Reptiles to Golf Courses
Matt Goode
University of Arizona

Objectives:
1. Examine diversity, distribution and relative abundance of reptiles using the golf course.
2. Implant radio transmitters into Gila monsters, and use fluorescent powder tracking, to examine movement patterns, habitat use, and behavioral responses to man-made features of the environment.
3. Using radiotelemetry data, examine movement patterns and home range characteristics of tiger rattlesnakes in response to features of the golf course and surrounding residential development.
4. Inform residents, golfers and golf course personnel about local reptiles via interactions in the field and through formal presentations to club members and residents.
5. Develop recommendations for golf course designers and managers that can be used to retrofit existing courses and design new courses to maximize benefits to reptiles.

Start Date: 2008
Project Duration: two years
Total Funding: $59,994

The demand for golf course communities in Arizona has steadily increased, especially as aging baby boomers transition into retirement. The more information we have on how to design and manage golf course communities, the better we will be at maximizing the benefits that golf courses can provide for wildlife. We hope to strengthen these potential benefits by means of education and involvement of golf course professionals and the local community, and through development of recommendations for both retrofitting and design of new courses.

We used mark-recapture to determine relative abundance and distribution of reptiles using golf courses in Arizona. We have documented literally thousands of tortoises, snakes, and lizards using various features of golf courses. Irrigated vegetation along fairways and surrounding greens and tee boxes are used well out of proportion to their availability, while open fairways appear to be avoided, especially by snakes. Areas of the golf course where natural desert has been incorporated into the design of the course correspond to areas of greater relative abundance of most species.

We have been radio tracking Gila monsters to gain a better understanding of how they are responding to the golf course and its surroundings. Gila monsters tend to spend more time on the edges of fairways and greens, presumably searching for prey in the dense, irrigated vegetation. We have observed individual Gila monsters to change the location and use of their home ranges as newly constructed homes become more common. Interestingly, the Gila monsters have started to concentrate more of their activity in areas immediately adjacent to the golf course that are off limits to development.

In 2008, we began using a new technique that allows us to quantify the exact movement path taken by a Gila monster. We obtained data from 102 Gila monster powder tracks, greatly bolstering our data set. The technique involves the use of fluorescent powder that we apply to the Gila monster by gluing rabbit fur onto its belly and loading up the fur with the powder. With a UV light, we can follow its exact track. This technique allows us to determine if Gila monsters are avoiding certain features of their habitat, such as fairways and roads. Using radiotelemetry, we only get the straight-line distance between successive fixes, but fluorescent powder allows us to determine the exact distance moved. So far, we have found that Gila monsters typically move up to two times farther than their straight-line distances would indicate.

We added data from an ongoing, long-term study of tiger rattlesnakes to our objectives. This data set includes information from over 60 individual snakes, located over 8,000 times, which is an unprecedented sample size for snake radiotelemetry. We have observed changes in home range size, use, and configuration in response to both features of the golf course and the construction of new homes.

Summary Points
- Reptile species vary in their use of golf course features, but most species tend to avoid open fairways.
- We have observed a greater abundance of reptiles using thick, irrigated vegetation adjacent to fairways and greens.
- The placement and layout of the golf course helps to determine the distribution of reptiles, and certain species alter their distributions to take advantage of irrigated vegetation and natural areas that are incorporated into the golf course as hazards or rough.
- Using radiotelemetry and fluorescent powder tracking, we have determined that Gila monsters tend to avoid open fairways, and tend to utilize irrigated areas along the edges of the golf course out of proportion to their availability.
- Tiger rattlesnakes change the configuration of their home ranges to accommodate features of the golf course and newly constructed homes.
Freshwater turtles are experiencing declines worldwide with habitat loss cited as a major cause. Expanding urbanization contributes to this habitat loss and causes population isolation, road-kill, and habitat degradation by contaminants.

Urban wetlands, particularly golf course associated wetlands, may create "oases" from these threats due to controlled access, nutrient enrichment and hence biological productivity, and restrictions on road traffic. Golf courses could provide valuable habitat for freshwater turtles because they generally provide open-water wetland surrounded by a variety of natural habitat types in largely unroaded areas with controlled human access.

During the 2009 field season, we trapped turtles in a total of 75 wetlands ("ponds") from June 1 through August 15 along an urban-rural land use gradient in the vicinity of Syracuse, New York. We characterized the ponds based on an initial assessment of whether they were in primarily "urban", "golf course", or "protected area" contexts. Of these 75 ponds, 32 were on golf courses (42%), 20 were in urban areas (26%), and 25 were in protected areas (32%).

A total of 335 turtles were captured in the initial 3-month trapping period. Of these, 190 (57%) were snapping turtles (Chelydra serpentina), 144 (43%) were painted turtles (Chrysemys picta), and 1 was a musk turtle (Sternotherus odoratus).

For each turtle we made morphological measurements (carapace width and length, height, plastron width and length, and pre-cloacal length) and fitness measurements (weight, leech load, and algal cover). We also noted the sex, number of growth annuli, whether turtles had eggs, and abnormalities such as deformities or injuries.

Initial analyses indicate that the proportion of traps catching at least one snapping turtle varied with comparable relative abundances in wetlands on golf courses (36% traps catching one snapping turtle) and protected areas (40%) versus higher abundances in wetlands in heavily urbanized areas (67%). The proportion of traps catching at least one painted turtle also varied among wetlands on golf courses (10%), protected areas (28%) and urban areas (38%).

In terms of species composition, golf course wetlands may be more favorable to snapping turtles than to painted turtles (51% females) versus urban areas (38%) and protected areas (21%), whereas the proportion of females among painted turtles caught did not vary among wetlands on golf courses (53%), urban areas (48%), and protected areas (43%).

For the 2010 field season, we will expand the number of sampled wetlands and incorporate other wetland types. We will also measure habitat configurations in detail at each site: wetland depth, shoreline composition, vegetation within and surrounding the wetland, number of basking sites, substrate, and water quality analyses.

Summary Points

- 75 wetlands were sampled in golf course (42%), urban (26%), and protected area (32%) contexts resulting in 335 turtles captured.
- Golf course wetlands (1) supported turtle populations comparable in size to protected areas but lower than heavily urbanized areas, and (2) had a large percentage of snapping turtles compared to painted turtles (79% snapping, 21% painted).
- Golf course wetlands had sex ratios closest to the expected 1:1 ratio.
- Next season's research will expand the trapping effort to additional golf courses and new wetland types and landscape contexts, as well as examine relationships between turtle populations and landscape and habitat variables in greater depth.
Insects that inhabit dead-wood are an important but under-appreciated natural resource within North America. Within the dead-wood insects, there is an incredible diversity even just within the beetle species. The larvae of these beetles help to break down dead-wood and release nutrients back into the soil, while many of the adult beetles are often associated with flowers and are important pollinators. Furthermore, these insects serve as a major food source for birds, mammals, and other woodland animals. Unfortunately, the services that these insects provide to ecosystems are often overshadowed by the damage caused by the few invasive and native pest insects. Nevertheless the conservation of dead-wood insects is crucial for the long-term health of managed and natural landscapes.

Dead-wood habitat has become increasingly rare as agriculture and urban land use has spread. While most urban environments are poor choices for the conservation of dead-wood habitat, golf courses, by incorporating dead trees or logs into their landscapes, are uniquely suited to this purpose and with minimal effort could serve as nodes of biodiversity for surrounding landscapes. Management of golf courses to promote dead-wood habitat would not only increase the number of beneficial decomposers but would also serve to increase the number of beneficial predators of wood-boring insects. These predators, such as woodpeckers, are effective natural defenses against both pest and invasive species.

While the conservation of dead-wood habitat on golf courses should be relatively easy to achieve, it will require the evaluation of several criteria. We used saproxylic beetles to determine two of those criteria. Research was conducted at the Birck-Boilermaker Golf Complex at Purdue University and utilized both the established Ackerman Hills course and the newer Kampen course. Sentinel logs were used as a non-intrusive and replicable method of determining the beetle species in an area. This technique consists of leaving standardized lengths of logs in an area over the summer and then rearing out the adult beetles in the lab for identification.

To determine the effect of fairway management on dead-wood habitat quality, four sentinel log transects were placed at the edge of fairways, extending perpendicularly into adjacent wooded or grassy areas. These transects consisted of paired sets of logs, freshly cut oak and dried oak, at 1, 2, 4, 8, 16, and 32 meters from the turf edge.

To examine which environments present on these golf courses are the best habitat for dead-wood boring insects, we placed 10-meter grids of four sentinel logs in three different types of environments: grassy meadow, managed glade, and forest. These groups of sentinel logs consisted of one log each of freshly-cut maple, freshly-cut pine, dried maple, and dried pine. All sentinel logs were placed on the courses in April 2009 and will be removed at the end of November 2009.

To rear the adult insects out of individual logs, banks of rearing tubes were constructed in our lab using 12-inch ID black HDPE culvert pipe.

Finally we have begun formulating a list of at least 40 Midwestern golf courses and digitizing aerial photographs of those courses in GIS. We will use the digitized maps of Purdue's courses to develop a value of connectivity for each habitat patch using an area-weighted distance measure. This measure will then be compared to the diversity and abundance of beneficial species at the sampling locations. These findings will allow us to develop recommendations towards the management of dead-wood habitat on these courses and to project those recommendations onto other courses throughout the Midwest.

**Summary Points**

- Research is underway to determine the effect, if any, of fairway management practices on dead-wood insect species, both pest and beneficial, as well as the most appropriate habitat for conserving dead-wood habitat for beneficial species.
- Seventy-two individual rearing tubes have been constructed at Purdue University and will be loaded with sentinel logs at the end of November 2009.
- Work has begun to digitize approximately 40 Midwestern golf courses using aerial photographs and GIS. These digital maps will allow for spatial analysis of habitat patches within each course and improve conservation recommendations.

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**Balancing Pest Management and Biodiversity**

**Conservation on Golf Courses**

Jeffrey D. Holland and Douglas S. Richmond

Purdue University

**Objectives:**

1. Determine the best managed golf course habitat for dead-wood insects and the optimal distance from fairway management practices for that habitat.
2. Digitize approximately 40 Midwestern golf courses to determine the current amount and connectivity of potential dead-wood habitat.

**Start Date:** 2009  
**Project Duration:** two years  
**Total Funding:** $7,304
American Bullfrogs (*Lithobates* (=*Rana* catesbeiana)) are invasive in the western US, including Colorado, and present several threats to native amphibian diversity. Bullfrogs are aggressive predators and competitors of many native western amphibians, and they are thought to have the potential to act as a reservoir host for a harmful fungal amphibian pathogen (*Batrachochytrium dendrobatidis*, referred to as Bd).

Along the Front Range region of Colorado, east of the Rocky Mountains, bullfrog populations have expanded at the same time that the area has undergone rapid human development. We believe that bullfrogs are responding to this land use change by taking advantage of the artificially permanent water bodies that accompany suburbanization. Unlike our native amphibians, bullfrogs require permanent bodies of water to have successful development because their tadpole stage lasts more than a year.

In the western US, agricultural expansion followed by urbanization has led to the creation of many artificial but permanent ponds (e.g. cattle ponds and suburban ponds) where they had not previously existed. In this same region, native leopard frogs have undergone drastic declines in recent decades that coincide with land use change, invasive bullfrog expansion, and the appearance of the amphibian pathogen, Bd. Therefore, learning about the regionally specific biology of invasive bullfrogs is key to understanding how we may mitigate their negative effects on vulnerable native species.

We are examining the hypothesis that bullfrogs are a reservoir host for Bd, meaning that they carry and spread the pathogen without experiencing disease. And finally, we are exploring a method of reducing bullfrog breeding by conducting dry-downs of bullfrog breeding ponds during the off-season, thereby breaking their larval development cycle.

During the summer of 2009, we conducted our first field season on golf courses in the Colorado Front Range. In total, we surveyed 95 ponds from 21 different golf courses located between Fort Collins and South Denver. Of the 95 ponds surveyed, we found invasive bullfrogs present at 38 ponds (40%) with active breeding at 24 ponds (25%). This is a major success because we have confirmed our prediction that golf courses would provide a good opportunity to study the distribution of this invasive species.

Golf courses are located in suburban landscapes and often use municipal water sources (e.g., channelized ditch canals) to fill their ponds. We believe that bullfrogs are using these canal systems as a pathway for dispersal and the artificial ponds (such as those on golf courses) as a place for breeding. We found native amphibians at only a handful of ponds (4%), and these included a few chorus frog and western toad tadpoles.

Overall, native amphibians were very rare. Of the 38 ponds where we observed bullfrog populations, 35% of them were found to be positive for Bd. This result is a strong indicator that bullfrogs do serve as reservoir hosts for this harmful amphibian pathogen and lends further support to our argument that bullfrog mitigation is needed.

### Summary Points
- Invasive bullfrogs threaten native amphibian diversity in Colorado.
- Invasive bullfrogs are very common on golf courses in Colorado as a result of municipal water sources that support their dispersal and breeding.
- Of the 38 bullfrog populations surveyed on golf courses, 35% were positive for a fungal amphibian pathogen (Bd), indicating that bullfrogs are an increased threat due to their role as a pathogen reservoir.
- Mitigating bullfrog breeding sites by conducting temporary dry-downs of golf course ponds will be the focus of the upcoming year’s activities.
Every year golf course superintendents are introduced to new products in the marketplace. Without results from properly designed, objective research, superintendents are asked to make buying decisions based on word-of-mouth, previous experience from colleagues, or the representative of the product. Several surveys indicate that golf course superintendents desire side-by-side product evaluations to assist them in making product purchases. The result of this desire for this type of information is the Product Testing Program section of USGA’s Turfgrass and Environmental Research Program. Currently, USGA is funding three projects that fall into this category of USGA-supported research, including cooperators at several sites across the country (see map below).
Evaluation of Plant Growth Regulators and Biostimulants for Use in Managing Summer Bentgrass Decline

Bingru Huang
Rutgers University

Objectives:
1. To investigate whether the application of selected PGRs and biostimulants to a bentgrass putting green would enhance turf quality during summer months.
2. To determine whether these products would help alleviate summer bentgrass decline by delaying leaf senescence and promoting root growth.

Start Date: 2007
Project Duration: two years
Total Funding: $20,000

Summer bentgrass decline is a major concern of superintendents growing creeping bentgrass on putting greens across the country. It is characterized by thinning turf canopy, leaf senescence, and root dieback. Incorporation of management practices such as use of natural products or plant growth regulators (PGRs) that may promote shoot and root growth would favor creeping bentgrass survival in the summer.

Some organic materials such as seaweed extracts are common ingredients in many biostimulant products. They are rich in organic and mineral compounds and often exhibit activity of plant hormones such as cytokinins and auxin. Some biostimulant products also claim to increase soil microbial density and activity by incorporating microbial inoculums, which in turn enhances turfgrass quality through increased organic matter decomposition and nutrient availability.

We proposed to investigate whether foliar application of selected PGRs and biostimulants would alleviate decline in creeping bentgrass growth during summer months and to examine their effects on leaf senescence and root growth. The study was conducted in two consecutive summers (2007 and 2008) on a 4-year-old ‘Penncross’ bentgrass green built to USGA greens specifications at Hort Farm II, Rutgers University, North Brunswick, NJ. This report summarizes data of two years.

The following selected products that are extensively used in the golf course maintenance or pure chemicals with great potential to be marketed as stress reducers were tested: Primo Max (from Syngenta, 0.125 oz/1,000 sq ft): Inhibitor of gibberellic acid synthesis and vertical shoot growth. TurfVigor (from Novazymes, 15 oz/1,000 sq ft): Combines patented high impact microbial strains, and macro and micronutrients. CPR (from Emeraled Isle Solutions, 6 oz/1,000 sq ft): a blend of natural sea plant extracts, micronutrients, and a surfactant. Aminoplex (from Grigg Brothers, 2 oz/1000 sq ft): A proprietary mixture of 15 plant-based L amino and organic acids, complex polysaccharides, and natural hormones. Aminoethoxyvinylglycine (AVG, from Sigma, 25 uM): an ethylene synthesis inhibitor that suppressed leaf senescence and helped to maintain greener turf for an extended period in a recent growth chamber study when creeping bentgrass was exposed to 35°C. 6-benzylamine (BA, from Fisher, 25 uM): a synthetic cytokinin that demonstrated functions in delaying leaf senescence and improving heat tolerance in creeping bentgrass in controlled-environment conditions.

Two control treatments were included for comparison. Nutrient control (full strength Hoagland’s solution): complete nutrient solution including all kinds of macronutrients and micronutrients to sustain plant growth. Water control: plants were sprayed with water.

We evaluated turf quality, chlorophyll content, photosynthesis rate, leaf area index (indicator of canopy density), green leaf biomass (indicator of canopy color), and root growth. Root growth was examined by measuring total root surface area and root biomass.

Summary Points
- Turf quality of TurfVigor- and CPR-treated plots were consistently higher than that in the water and nutrient control plots during the whole treatment period (late June to mid-September) in both years. Nutrient control plots were not statistically different from the water control plots for most parameters. The promoting effects of Primo on turf quality started from mid-August in 2007 and early July in 2008. Aminoplex, AVG, and BA had positive effects only in August 2007, and July and August 2008.
- Shoot growth of creeping bentgrass was promoted and leaf senescence was mitigated when treated with several products. TurfVigor and Primo promoted all physiological parameters on most sampling dates in both years. CPR consistently reduced decline in turf density and increased leaf chlorophyll content but did not affect photosynthetic rate. BA had positive effects on chlorophyll content and photosynthetic rates in July of both years. AVG promoted turf density and Aminoplex promoted photosynthetic rate on a few sampling dates in 2008.
- Root growth of creeping bentgrass was not consistently affected by any of the product during the whole treatment period. Root surface area was significantly increased in TurfVigor-treated plots in mid-August 2007 and mid-September 2008. Root biomass was significantly increased by TurfVigor and Primo treatments on a few sampling dates in 2007; in 2008, TurfVigor, CPR, Aminoplex, and AVG treatments all had positive effects on a few sampling dates.
- Our results suggested that proper use of selected biostimulants and PGRs could promote turf growth and alleviate summer bentgrass decline in warm climates.

Summer bentgrass decline is a major concern of superintendents growing creeping bentgrass on putting greens across the country, especially in the southern states and the transitional zone.
Buffalograss experimental lines and cultivars established in 2007 and 2008 were evaluated for turfgrass performance during the 2009 season at nine locations. Trials were established in Tucson, AZ; Fort Collins, CO; Lawrence and Wichita, KS; Las Cruces, NM; Mead, NE; Logan, UT; Blacksburg, VA; and Yakima, WA. There were 17 entries at each location. 'Bison', 'Bowie', 'Cody', and 'Texoka' were used as seeded standards, and 'Legacy', 'Prestige', and 'NE 609' were used as vegetative standards.

There were five seeded and five vegetative experimental lines in addition to the industry standards in the trials. The experimental design was a randomized block with entries replicated three times. Turfs were mowed at 2.0 inches with clippings returned. Two lbs N/1000 ft² of was applied as one pound applications in June and July, and irrigation was applied at 50-60% of potential evapotranspiration. Annual grasses and broadleaves were controlled as needed.

Considerable amounts of data were collected for most locations. Seeded and vegetative genotypes were analyzed separately. The seeded experimental lines, NE-BFG-07-02, NE-BFG-07-03 and NE-BFG-07-04E had the best overall turfgrass quality ratings across most environments, indicating their wide adaptation potential. These were followed closely by NE-BFG-07-09 and NE-BFG-07-10, which both showed a wide range of potential adaptation. The other experimental lines showed more specific adaptation.

The vegetative experimental lines performed differently than the seeded types. The standard entry, 'Legacy', had the best overall performance, but it was followed closely by NE-BFG-07-09 and NE-BFG-07-10, which both showed a wide range of potential adaptation. The other experimental lines showed more specific adaptation.

Four one-acre blocks (i.e. one each of NE-BFG-07-01, NE-BFG-07-02, NE-BFG-07-03, and NE-BFG-07-08) were established in 2008 to evaluate their seed yield potential, using larger scale harvesting equipment. The first harvest was made in August 2009. NE-BFG-07-02 had best seed yield in the first year of harvest for these four experimental lines. In 2010, an additional acre block will be added to evaluate NE-BFG-07-04E.

The turfgrass evaluations and larger seed production trials will be continued through 2010. At the end of this time, decisions will be made on the potential release of these experimental lines. With this in mind, trials were initiated in multiple locations to obtain the necessary data to support applications for plant variety protection of the seeded types and patenting of the vegetative types. This process was initiated to enhance the potential release process.

Summary Points

- Significant differences were observed among genotypes tested for most traits.
- Seeded genotypes showed the widest range of adaptation and performance over years and locations.
- Vegetative genotypes were more location specific in their adaptation and performance.
- Large test plots were initiated in 2008 for evaluation of seed yield using large-scale harvesting equipment.
- Large test plots were harvested for the first time in August 2009 with NE-BFG-07-02 having the highest seed yield of the four lines evaluated.
- PVP trials were initiated to collect necessary data to facilitate the release of the seeded types determined to have the best turfgrass performance and adaptation.
Assessment of Commercially Marketed Filter Materials for Tile Drainage Outlets on Golf Courses

Kevin W. King
USDA-ARS

James F. Moore
USGA Green Section

Jim C. Balogh
Spectrum Research, Inc.

Objectives:
1. Assess the feasibility and effectiveness of commercially marketed filters designed to strip nutrients (nitrogen and phosphorus) and pesticides (chlorothalonil and metalaxyl) from drainage waters exiting managed turf areas.
2. Identify and demonstrate the efficacy of using both synthetic and natural products in a field scale application of the end-of-tile filter.

Start Date: 2005
Project Duration: four years
Total Funding: $42,200

Tile drainage and other subsurface drainage features are considered essential by turfgrass managers to maintain water tables at depths necessary for healthy plant growth, maintain sufficient water and air in soil void space, to stimulate essential microbial activity, avoid rutting and soil compaction by maintenance equipment, and to allow site use soon after heavy rains. Subsurface drainage increases the subsurface movement of excess water and facilitates infiltration.

Nutrient and pesticide transport through subsurface drainage systems may become a component of surface runoff if the drainage water discharges directly into surface water or onto the surface offsite or downslope. Subsurface drains conveying water directly into a stream or pond will bypass natural and managed filtering processes, including upland and riparian buffer zones. This research is designed to address the potential for end-of-tile filters to significantly reduce the transport of nutrients and pesticides from golf course tile drainage outlets to surface waters. In addition to the commercially available filters, other synthetic and natural by-products will be identified and used to demonstrate the effectiveness of end-of-tile filters in a managed turf setting.

The research is being conducted in two phases. The first component is a controlled laboratory experiment designed to evaluate the filter’s effectiveness while operating at flow rates comparable to those measured in the field. The second component is a before-after field assessment of the filters. The field study is being conducted at two different golf courses, one located in Texas and the other in Minnesota. The Texas site is located at Ridgewood Country Club in Waco, TX. The assessment is taking place on the drainage pipe that collects both surface and subsurface drainage waters from a significant portion of the course.

For the laboratory study, a hydrograph generator was created to simulate tile flow discharge. The hydrographs were generated from a supply reservoir containing a solution of nitrate nitrogen, dissolved phosphorus, chlorothalonil, and metalaxyl. The water was pumped through the filter assembly. Samples were collected prior to entering the filter and after flowing through the filter.

The filters were a blend of equal parts by weight of activated carbon, activated alumina, and zeolite. The filters substantially reduced the amount of dissolved phosphorus, chlorothalonil, and metalaxyl from the drainage water. Removal of nitrate was not as great. The removal was inversely proportional to the flow rate and varied depending on the location on the hydrograph (rising limb, peak flow, or receding limb). Preliminary findings from the field study in Minnesota indicate significant reductions in ammonium, nitrate, total nitrogen, and total phosphorus. No results are yet available for the pesticides. Filters containing newly identified materials have recently been installed at the Texas site.

Future research will investigate different activated carbons and different by-product mixes, inclusion of a denitrification barrier prior to/after the filter, long-term sorption capability, efficiency dependence on influent concentrations, optimizing contact time, and scaling for larger applications.

Summary Points

Laboratory Study
- Substantial loading reductions were measured for dissolved reactive phosphorus (51.6%) and chlorothalonil (58.2%), while intermediate reductions for metalaxyl (28.8%) and minor reductions for NO3-N (4.7%) were measured.
- For all contaminants, the discharge flow rate was inversely related to percent removal and was consistent across all tested hydrographs.
- The efficiency of the filters varied depending on the location within the hydrograph. A greater percent removal was observed during the rising limb compared to the peak and receding limbs.

Field Study
- Preliminary data from the Minnesota field site suggests that the filter is significantly reducing the amount of ammonium, nitrate, total nitrogen, and total phosphorus.
- Additional materials have recently been identified and implemented at the field site in Texas.