Symposium Organizing Committee

James A. Murphy, Chair
Bruce B. Clarke
Barbara Fitzgerald
James F. White, Jr.
Ning Zhang

Proceedings of the Nineteenth Annual Rutgers Turfgrass Symposium

Ning Zhang and Barbara Fitzgerald, Editors

Rutgers Cooperative Extension educational programs are offered to all without regard to race, religion, color, age, national origin, gender, sexual orientation or disability.
Associate Director’s Opening Remarks

Welcome to the nineteenth Annual Rutgers Turfgrass Symposium at the School of Environmental and Biological Sciences/NJAES, Rutgers University. This symposium was started 19 years ago as an annual meeting to update Center for Turfgrass Science members and our stakeholders on current issues in turfgrass science. We are especially pleased to have Dr. John Stier from the University of Wisconsin and Dr. Yaling Qian from Colorado State University as invited lecturers. It is important to recognize our organizing committee: Drs. Ning Zhang, Jim Murphy, Jim White, Bruce Clarke and Ms. Barbara Fitzgerald for putting together an outstanding program. We are especially thankful to the proceedings editors, Dr. Ning Zhang and Barbara Fitzgerald.

The faculty of the Turf Center continue to be recognized for excellence in research, teaching and extension. In 2009, Dr. Stacy Bonos received the Early Career Award for Excellence in Plant Breeding from the multi-state Plant Breeding Coordinating Committee, the Environmental Stewardship Award from the New Jersey Turfgrass Association, the Early Career Achievement Award from the Northeast Division of the American Phytopathological Society (NED-APS) and the Merle V. Adams Award for Outstanding Junior Faculty from Rutgers Cooperative Extension. I was very fortunate to be inducted into the New Jersey Turfgrass Association’s Hall of Fame in 2009. Moreover, our graduate students were recognized for their research accomplishments at several national meetings: Joseph Roberts won the best graduate student oral presentation award at the NED-APS annual meeting in Quebec City, Canada as well as 1st place in the graduate student competition for both his oral and poster presentations in the Turfgrass Management Section (C-5 Division) at the Crop Science Society of America (CSSA) annual meeting. Ms. Lisa Beirn also was awarded 1st place honors by the Turfgrass Breeding Committee for her oral presentation (C-5 Division) at the same CSSA meeting in Pittsburg, PA.

The faculty and staff of the Center for Turfgrass Science continue to have a great impact nationally and internationally. We will be hosting the International Turfgrass Research Conference in New Jersey in 2017, with Dr. Bruce Clarke as the President of International Turfgrass Society (ITS). Dr. Jim Murphy was recently chosen to represent the United States on the ITS board for 2009-2013 and was elected to be the C-5 Chair of the CSSA in 2010. Finally, a record number of scholarships (over $100,000) were awarded to our students in 2009. We continue to have a very rewarding relationship with the turfgrass industry in the tri-state region.

Thanks for participating in this year’s symposium.

Sincerely,

William A. Meyer
Associate Director, CTS
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Monday, January 11, 2010

8:30 - 9:00 AM Registration, Coffee and Donuts

9:00 - 10:00 AM SESSION I: GERmplASM ENHANCEMENT AND BIOCOnTROL
(Moderator: Dr. Faith Belanger)

9:00 – 9:20 Dr. Stacy Bonos (Department of Plant Biology and Pathology, Rutgers University) Breeding Bentgrass Species for Improved Disease Resistance and Stress Tolerance

9:20 – 9:40 Dr. William Meyer (Department of Plant Biology and Pathology, Rutgers University) Breeding Improvements in Tall Fescue and Kentucky Bluegrass

9:40 – 10:00 Dr. Donald Kobayashi (Department of Plant Biology and Pathology, Rutgers University) The Atkins Approach to Biological Control: Does the High Protein/Low Carbohydrate Diet of Lysobacter Enzymogenes Specify Host Preference?

10:00 - 10:30 AM Discussion and Coffee Break

10:30 – 12:00 PM SESSION II: TURFGRASS MANAGEMENT AND THE ENVIRONMENT
(Moderator: Dr. Thomas Molnar)

10:30 – 10:50 Dr. James Murphy (Department of Plant Biology and Pathology, Rutgers University) Nitrogen Fertilization Effects on Anthracnose of Annual Bluegrass

10:50 – 11:10 Dr. Yaling Qian (Department of Horticulture and Landscape Architecture, Colorado State University) Carbon Sequestration Potential of Turfgrass Ecosystems

11:10 – 11:20 AM Discussion session
11:20 – 12:00 **Keynote: Dr. John Stier** (Department of Horticulture, University of Wisconsin-Madison) *Rain Gardens, Urban Runoff, and Groundwater Recharge*

12:00 - 1:00 PM **Lunch and Poster Session**

1:00 – 2:00 PM **SESSION III: TURFGRASS PHYSIOLOGY**
(Moderator: Dr. Thomas Gianfagna)

1:00 – 1:20 **Yan Xu** (Department of Plant Biology and Pathology, Rutgers University) *Proteomic and Metabolic Analysis of ipt-Transgenic Creeping Bentgrass with Improved Heat Tolerance*

1:20 – 1:40 **Dr. Eric Lam** (Department of Plant Biology and Pathology, Rutgers University) *Chemical Chaperones as Potential Tools to Suppress Cell Death in Plants and Induce Stress Tolerance in Crops and Turfgrass*

1:40 – 2:00 **Dr. James F. White, Jr.** (Department of Plant Biology and Pathology, Rutgers University) *Is Plant Endophyte-Mediated Defensive Mutualism the Result of Oxidative Stress Protection?*

2:00 – 2:30 PM **Discussion and Coffee Break**

2:30 – 3:30 PM **SESSION IV: TURFGRASS PESTS – BIOLOGY AND CONTROL**
(Moderator: Dr. Barbara Zilinskas)

2:30 – 2:50 **Dr. Ning Zhang** (Department of Plant Biology and Pathology, Rutgers University) *Molecular Detection of Fungal Turfgrass Pathogens*

2:50 – 3:10 **Dr. Stephen Hart** (Department of Plant Biology and Pathology, Rutgers University) *Metamifop: A New Postemergence Herbicide for Crabgrass and Goosegrass Control in Cool-Season Turf*

3:10 – 3:30 **Dr. Lemma Ebssa** (Department of Entomology, Rutgers University) *Biological Control of Black Cutworm in Golf Course Turf*

3:30 – 4:00 PM **Discussion/Closing Remarks**
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PLENARY PRESENTATIONS
Breeding Bentgrass Species for Improved Disease Resistance and Stress Tolerance

Stacy A. Bonos, Eric Weibel, Mathew Koch, Robert Shortell, and Laura Cortese

Department of Plant Biology and Pathology, Rutgers University

Creeping bentgrass (*Agrostis stolonifera*) is currently the most widely used bentgrass for golf courses. Its prostrate growth habit and ability to produce vigorous spreading stolons, allow it to tolerate low cutting heights and quickly recover from damage. This makes creeping bentgrass a good choice for golf course putting greens in the cool- temperate and in some cases the warm-humid climates. However, creeping bentgrass, as a species, is very susceptible to dollar spot disease (caused by *Sclerotinia homoeocarpa* F.T. Bennet) and typically requires regular fungicide applications. Since 1996, we have been breeding creeping bentgrass for improved dollar spot resistance and have made some dramatic improvements. Other diseases including brown patch disease (caused by *Rhizoctonia solani* Kühn) and more recently copper spot (*Glomocercospora sorghi*), and anthracnose (*Colletotrichum cereale* Manns sensu lato Crouch, Clarke and Hillman) can also occur with regular frequency when a reduced fungicide program is used. Unfortunately, cultivars with improved dollar spot resistance don’t necessarily also show improvements for other diseases. We are in the process of combining dollar spot resistance with germplasm that contains resistance to these other important diseases in order to develop new cultivars with multiple disease resistance.

Colonial bentgrass (*Agrostis capilaris* L.) also known as brown top, has traditionally been used as a lawn and golf course grass in areas of Northern Europe and New Zealand that have mild (cool humid) summers. Compared to creeping bentgrass, colonial bentgrass has a more upright growth habit and spreads by short rhizomes instead of stolons. Colonial bentgrasses typically do not require aggressive cultural management inputs. Additionally, DaCosta and Huang documented that colonial bentgrass exhibits faster recovery from drought stress compared to creeping bentgrass. The drought recovery, reduced maintenance requirements and increased dollar spot resistance of colonial bentgrass make it an interesting choice for golf course fairways during a time where environmental conservation, using lower inputs and energy conservation are on the minds of many people.

However, colonial bentgrass does have a major weakness affecting its use in temperate areas of the US. It is susceptible to brown patch disease and loss of turf density (damage) can be strongly evident during the months of June through September. It has been the goal of the breeding program at Rutgers for the past 10 years to improve brown patch resistance in colonial bentgrass so that it may be more useful as a potential grass for fairways and tees. We have made dramatic improvements in brown patch resistance in colonial bentgrass in the past several years and we hopeful that this could improve the use of colonial bentgrass on low maintenance fairways throughout the northern area of the country.
Velvet bentgrass (*Agrostis canina* L.) forms the finest textured and most dense turf of the bentgrasses and can nearly resemble green velvet when managed properly. It produces erect tillers and spreads with short stolons. Velvet bentgrass spreads more aggressively than colonial bentgrass but not as vigorously as creeping bentgrass. It is more tolerant of dollar spot and brown patch than creeping and colonial bentgrass respectively, however it tends to be more susceptible to copper spot and a root Pythium disease (caused by *Pythium* spp.). We have made some improvements in these diseases but more breeding is needed for better resistance to these diseases.

In addition to disease resistance, we have been evaluating bentgrasses for traffic tolerance. Velvet bentgrass and the higher density creeping bentgrasses tend to have better traffic tolerance. Colonial bentgrasses tended to have good to fair traffic tolerance. We are currently breeding bentgrasses for improved traffic tolerance and incorporating it into our disease resistant germplasm.

Breeding for salinity tolerance is also an important focus of the Rutgers bentgrass breeding program. Salinity tolerance will be important for the future of turfgrass management as regulations on the use of potable water on turf areas increases. We evaluated creeping, colonial and velvet bentgrass for salinity tolerance under field conditions. Creeping bentgrasses were the most tolerant followed by velvet and colonial bentgrass. The bentgrasses that have been developed for improved turf quality and disease resistance also tend to have improved salinity tolerance over older cultivars.

Due to the cross-pollinated nature of bentgrasses, multiple traits can be incorporated together into one cultivar. As mentioned above we have made dramatic achievements in dollar spot resistance in creeping bentgrass and brown patch resistance in colonial bentgrass. Our goal is to incorporate resistance to other diseases, traffic and salinity tolerance into the germplasm we have already selected in an effort to develop cultivars that require less pesticides and utilize fewer water resources.
Breeding Improvements in Tall Fescue and Kentucky Bluegrass


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The Rutgers Turfgrass breeding program has been involved with the improvement of Kentucky bluegrass (Poa pratensis) and tall fescue (Festuca arundinacea) for over 40 years. This is a report on some recent epidemics of stem rust (Puccinia graminis), crown rust (P. coronata) and stripe rust (P. striiformis) on Kentucky bluegrass cultivars and experimental lines in New Jersey. Recently, Dr. Jon Bokmeyer completed his Ph.D. dissertation studying brown patch (Rhizoctonia solani Kühn) resistance in tall fescue. The results of his thesis indicate that some changes can be made in breeding tall fescue that will enhance the levels of resistance in this species.

Kentucky Bluegrass Breeding

Stem rust can be a severe disease during the late summer or early fall on Kentucky bluegrass especially when reduced nitrogen fertility is maintained. During the past 40 years, differences in cultivar susceptibility have been noted in our turf trials. In the past 9 years, varieties such as ‘Midnight’ that were reported as resistant for over 20 years in New Jersey were found to be susceptible in 2002 during seedling establishment. Since that time mature turf of the ‘Midnight’ and ‘Midnight’ types were found to be moderately resistant to stem rust while the cultivar ‘Ulysees’ was found to be extremely susceptible as a mature turf.

Crown rust is usually a common disease of low maintenance perennial ryegrass (Lolium perenne) and tall fescue turf in the cool fall periods. Stripe rust is a severe disease in seed production fields in the Pacific Northwest especially during periods of cool nights. This past fall, crown rust caused a severe epidemic on seedling turf at Adelphia Research Center (Freehold, NJ) on the ‘Midnight’ and ‘Midnight’ types and much less severe disease level on the mature ‘Midnight’ types in the 2008 trial at Adelphia. At Hort Farm II (North Brunswick, NJ) stripe and crown rust were both present in a severe epidemic in a fall seeding. The results of these trials will be reported.

Tall Fescue Breeding

Since the release of ‘Rebel’ tall fescue in 1980, there has been a gradual improvement each year in tall fescue cultivars that have evolved from conducting cycles of recurrent phenotypic selection for brown patch resistance. This has included mowed single-plant progeny and clonal spaced-plants that were inoculated with brown patch inoculum. These new cultivars also have improvements in density and persistence.
In recent studies on broad and narrow-sense heritability of brown patch resistance in tall fescue we found similar results to earlier studies indicating that this disease is greatly influenced by environmental conditions and that resistance is controlled by a combination of multiple genes. Data suggests that effective breeding for brown patch in the future should include more extensive progeny evaluation (replicated if possible) and tests of combining ability before parents are chosen for future cultivars.
The Atkins Approach to Biological Control: Does the High Protein/Low Carbohydrate Diet of *Lysobacter enzymogenes* Specify Host Preference?

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*Lysobacter enzymogenes* has been described as a bacterial antagonist of other microbes for over four decades. Recently, however, the species has emerged as a model microorganism in which to study biological control of plant diseases, including turfgrass diseases such as summer patch and Bipolaris leaf spot. Studies have indicated that the bacterium is capable of establishing pathogenic interactions, typified by intracellular infection, with many microbial eukaryotic hosts. The pathogenic host range of *L. enzymogenes* is restricted to lower eukaryotic hosts lacking evolved immune systems, further contributing to its attractiveness as a potential biocontrol agent for turfgrass diseases.

To gain a better understanding of how *L. enzymogenes* interacts with other microbes and improve its potential as a biological control agent, we initiated a project in 2007 to sequence the genome of the bacterium. Information gained from the nearly completed genome sequence, combined with experimental evidence, has provided valuable insight into the metabolic, ecological and pathogenic behavior of the bacterium. For example, the genome sequence has revealed that growth of *L. enzymogenes* does not appear to be auxotrophic for amino acids. However, *in vitro* studies indicate *L. enzymogenes* growth is optimal in a minimal nutrient salts medium when supplemented with combinations of amino acids or protein extracts. Encoded within the genome is the presence of nearly 100 proteolytic enzymes, many of which are secreted outside the cell through one of several protein translocation systems. This large number of enzymes is supportive for their important roles in nutrient acquisition, as well as for host-specified microbial antagonism. Gaining a better understanding of these and other processes within *L. enzymogenes* may lead to novel biocontrol and biorational approaches for controlling turfgrass and other plant diseases.
Nitrogen Fertilization Effects on Anthracnose of Annual Bluegrass

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Anthracnose, caused by Colletotrichum cereale Manns, is a devastating disease that is particularly severe on annual bluegrass (ABG) [Poa annua L. f. reptans (Hausskn) T. Koyama], especially during the summer months. Anthracnose can cause severe thinning of putting green turf and has become more prevalent in the United States as well as Ireland and the United Kingdom (Mann and Newell, 2005). Low N fertility management (as little as 4.9 kg ha\(^{-1}\) per month) is often used to limit vertical leaf growth and increase ball roll distance (green speed); however, reduced plant vigor and increased susceptibility to environmental stresses and diseases are potential consequences of this strategy.

Our initial study of N fertilization indicated that 4.9 kg ha\(^{-1}\) of N applied every 7-days reduced anthracnose disease severity compared to 28-day applications during the middle of the growing season (Inguagiato et al., 2008). Uddin et al. (2009) reported that increased N-fertility up to 24.5 kg ha\(^{-1}\) applied every 14-days can significantly reduce anthracnose severity. However, the specific nature of the disease response to low rates of soluble-N during the growing season has not been determined. Moreover, previous research has not clearly defined the possible role of late- or early-season higher N rate granular fertilization on anthracnose severity of annual bluegrass putting green turf (Danneberger et al., 1983; Inguagiato et al., 2008). Nor has the influence of the timing of granular-N fertilization on the frequency of low rate soluble-N fertilization during the growing season been defined.

Superintendents have frequently asked about the potential role, if any, of late- and early-season granular-N fertilization in suppressing anthracnose of annual bluegrass turf and are seeking guidance on the importance of this practice. Note that recent marketing of foliar (liquid) fertilization has been encouraging superintendents to reduce and possibly eliminate higher rate granular-N fertilization. Research is needed to provide insight into the feasibility of this approach to N fertilizer programming with respect to disease management.

Thus, our current research objectives are to:
1. Evaluate the effect of the cumulative rate of low rate soluble-N fertilization for suppressing anthracnose disease during the growing season (summer).
2. Evaluate the N rate effect of late- or early-season granular fertilization on anthracnose severity.
3. Determine whether late- or early-season granular-N fertilization alters (interacts with) the effect of frequent low rate soluble-N fertilization during mid-season on anthracnose severity.
Objective 1.

A 3-yr field trial was initiated in 2007 on ABG turf grown on a sandy loam with a pH of 6.4 in North Brunswick, NJ. The trial was mowed daily using a walk behind mower set at a bench height setting of 3.2 mm. Silica sand was applied as topdressing at 89 cm$^3$ m$^{-2}$ every 14-days and incorporated with a cocoa mat drag. Turf was irrigated to prevent wilt stress. Dollar spot (*Sclerotinia homoeocarpa* F.T. Bennett) and brown patch (*Rhizoctonia solani* Kühn) were prevented with fungicides shown to be ineffective at suppressing anthracnose on ABG in NJ. Annual bluegrass weevils (*Listronotus maculicollis* Dietz) were controlled with insecticide applications each year.

This study used a randomized complete block design. Fertilization treatments varying in N rate and frequency (4.9 kg ha$^{-1}$ every 1, 2, 4 and 8 wk and 9.8 kg ha$^{-1}$ every 2 and 4 wk) and initiation date (mid-May vs mid-June) resulted in six total N quantities (4.9, 9.8, 14.7, 19.6, 29.4, and 58.8 kg ha$^{-1}$) from initiation to August in each year of the study. Treatments were applied as an ammonium nitrate solution.

The field area used for this study in 2007 and 2008 had previously been inoculated with *C. cereale* isolate HFIIA (Inguagiato et al., 2008); disease outbreaks occurred naturally both years. In 2009, the field area was reinoculated with *C. cereale* isolate HFIIIB obtained from an adjacent field to ensure uniform infection. Inoculum was produced and applied according Inguagiato et al. (2008) using a concentration of 40,000 conidia mL$^{-1}$ between 1800 and 2000 h on 3, 4 and 11 July when the minimum air temperature was ≥21°C and relative humidity ≥90%. *C. cereale* was re-isolated from symptomatic tissue each year. Anthracnose severity was rated periodically from June through August using a line intersect-grid count method to calculate percent turf area infested (Inguagiato et al., 2008). Sequential disease assessments throughout each growing season were used to determine the area under the disease progress curve (AUDPC), a quantitative assessment of disease over time calculated by the trapezoidal method (Madden et al., 2007). Pearson product moment correlation between total AUDPC and total N rate was performed using means as the experimental units.

Turf receiving the greatest total N quantity (58.8 kg ha$^{-1}$ over 12 wks) had the least disease (AUDPC) during all years. These results agree with a previous report that anthracnose severity was reduced by N applied at 4.9 kg ha$^{-1}$ every 7-days (58.8 kg ha$^{-1}$ for 12 wks) compared to every 28-days (14.7 kg ha$^{-1}$ for 12 wks) during the growing season (Inguagiato et al., 2008). The response of anthracnose to the cumulative N in our study was linear in all years. Treatments initiated prior to disease development (mid-May) resulted in greater total N accumulation and reduced anthracnose severity compared to N treatments started at disease inception (mid-June).

Beard et al. (1978) recommended 74 to 103 kg ha$^{-1}$ of N be applied over a 3 month period during the growing season to maintain ABG putting green turf. Our study
indicates that fertilization up to 59 kg ha\(^{-1}\) of N during this period will reduce anthracnose severity. The nature of the disease response above 59 kg ha\(^{-1}\) was not assessed, but based on the report of Uddin et al. (2009) further reductions in severity may be feasible. However, since high N fertilization (292 kg ha\(^{-1}\) yr\(^{-1}\)) has been shown to increase anthracnose severity on ABG fairway turf (Danneberger et al., 1983), additional research is needed to examine the impact of higher N fertility on anthracnose severity of ABG putting green turf. We currently have an ongoing trial that is assessing greater rates of soluble-N fertilization during the season.

Objectives 2 and 3.

Two trial areas were initiated September 2008 on ABG turf grown on a sandy loam in North Brunswick, NJ. Nitrogen fertilization treatments were arranged as a 3 x 4 x 4 factorial in a randomized complete block design with three replications. The first factor was the season for granular fertilization: spring, autumn and none. The second factor was the N rate of granular fertilization: 73, 147, and 220 kg ha\(^{-1}\) (1.5, 3.0, and 4.5 lb 1000-ft\(^{-2}\)). The third factor was the frequency of low rate soluble-N fertilization during the growing season: none and 4.6 kg ha\(^{-1}\) applied every one, two, or four weeks. This trial was repeated twice in space; anthracnose disease was allowed to develop in one trial area during 2009 while disease was suppressed with fungicides in the other trial area. The trial area where disease was suppressed will provide a second year (2010) of data from plots that have received two years of cumulative N fertilization, which may result in some treatments having a buildup of soil N that could impact findings related to Objective 3.

The trial areas were maintained similar to that described for objective 1 except for walk behind mowing, which was performed using a bench height setting of 3.4 mm. Anthracnose disease outbreak occurred naturally in 2009. Neighboring fields had previously been inoculated with \(C.\) cereale isolate HFIIA (Inguagiato et al., 2008) and were cut with the same mowers thus naturally infesting the trial areas. Anthracnose severity was rated periodically from June through August using a line intersect-grid count method to calculate percent turf area infested (Inguagiato et al., 2008).

Preliminary data analysis indicated that all three main effects had a significant impact on disease severity. Interestingly, disease severity was reduced when granular N fertilization was emphasized during the spring compared to autumn; however, the season of fertilization was not important at the lowest rate of granular N fertilization [73 kg ha\(^{-1}\) (1.5 lb 1000-ft\(^{-2}\))]. Generally, greater N rates applied by granular fertilization reduced disease severity compared to lower N rates. However, the N rate for autumn fertilization needed to be 220 kg ha\(^{-1}\) (4.5 lb 1000-ft\(^{-2}\)) for disease severity to be reduced; whereas, only 147 kg ha\(^{-1}\) (3.0 lb 1000-ft\(^{-2}\)) of N was needed for the spring granular program to reduce disease severity. Moreover, increasing N rate from 147 kg ha\(^{-1}\) to 220 kg ha\(^{-1}\) in the spring granular program did not improve disease suppression.
As expected, greater soluble-N rates during the season reduced disease severity; however, this factor interacted with granular N rate. Further data analysis is needed to interpret this interaction although it appears that disease severity can be suppressed with lower total amounts of N if a portion of the total N is applied as soluble-N during the season. A second year of data collection should clarify how much of the N should be applied as soluble-N during the season versus granular N fertilizer during the spring or autumn.

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Carbon Sequestration Potential of Turfgrass Ecosystems

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Land use changes to urban and suburban development are one of the global trends at unprecedented rates. Turfgrass is a major vegetation type in the urban and suburban environment. From lawns to parks, turfgrass is an integrated part of everyday life. Because of high productivity and lack of soil disturbance, turfgrass may be making substantial contributions to sequester atmospheric carbon. To determine soil organic carbon (SOC) changes in turf/soil ecosystems, several studies were conducted.

Study I. A total of about 690 historic soil testing datasets from parts of 16 golf courses in Colorado were collected. In addition, information on previous land use, soil texture, grass species and type, fertilization rate, irrigation, and other management practices were collected from these golf courses. The oldest golf course was 45 years old, and the newest golf course was 1.5 years old. Nonlinear regression analysis of compiled historic data indicated a strong pattern of soil organic matter (SOM) response to decades of turfgrass culture. Total carbon (C) sequestration continued for up to about 31 years in fairways and 45 years in putting greens. However, the most rapid increase occurred during the first 25–30 years after turfgrass establishment, at average rates approaching 0.9 and 1.0- ton ha\(^{-1}\) year\(^{-1}\) for fairways and putting greens, respectively. It was also found that past land use imparted a strong control of SOM baseline; fairways converted from agricultural lands exhibited 24% lower SOM than fairways converted from native grasslands.

Study II. The CENTURY ecosystem model was parameterized and adopted in turfgrass systems to evaluate C and N dynamics. To evaluate the CENTURY’s performance, two data sets were used: 1) long-term SOC data from golf courses, and 2) 3-year clipping yield data from a field study in Colorado. Model predictions of organic carbon accumulation compared reasonably well with observed SOC, with regression coefficients ranging from 0.67 to 0.83. That the CENTURY correctly simulates annual accumulative clipping yield. The CENTURY model was further used as a management supporting system to generate optimal N fertilization rates as a function of the age of turfgrass and with an aim to achieve adequate production under the constraint of minimal N out-fluxes. Simulated biomass and nitrate leaching data of various management scenarios (various fertilization rates with clippings removed or returned) were compared. The CENTURY model predicts that, as the age of turf ecosystem increases, N application rate needs to be reduced due to the greater amounts of N mineralized and recycled from SOM. At a loam soil site and under clipping returned scenario, the CENTURY model predicted that optimal productivity and minimum nitrate leaching (< 2 kg N ha\(^{-1}\) Yr\(^{-1}\)) could be obtained when annual N fertilization at 150, 100, 75, and 60 kg N ha\(^{-1}\) during 1-
10, 11-25, 26-50, and 51-100 years after turfgrass establishment, respectively. In contrast, under clipping removed scenario the CENTURY model predicted that, to achieve a comparable productivity and turf quality, N fertilization at 200, 150, and 140 kg N ha\(^{-1}\) per year would be required for the periods of 1-10, 11-50, 51-100 years after turfgrass establishment, respectively. Returning grass clippings back to turf/soil ecosystem can significantly reduce the fertilization requirements by 25% between 1-10 years after turf establishment, 33% between 11-25 years after establishment, 50% between 25-50 years after establishment, 60% thereafter.

**Study III:** Research was conducted to determine the rate of soil organic carbon (SOC) changes, soil carbon sequestration, and SOC decomposition of fine fescue (*Festuca spp.*), Kentucky bluegrass (*Poa pratensis* L.), and creeping bentgrass (*Agrostis palustris* Huds.) using carbon isotope techniques. Our results indicated that four years after establishment, about 17-24% of SOC at 0-10 cm and 1-13% from 10-20 cm was derived from turfgrass. Irrigated-fine fescue added the most SOC (3.35 ton C ha\(^{-1}\) yr\(^{-1}\)) to the 0-20 cm soil profile, but also had the highest rate of SOC decomposition (2.61 ton C ha\(^{-1}\) yr\(^{-1}\)). The corresponding additions and decomposition rates for non-irrigated fine fescue, Kentucky bluegrass, and creeping bentgrass in the top 20 cm soil profile were 1.39 and 0.87, 2.05 and 1.73, and 2.28 and 1.50 ton C ha\(^{-1}\) yr\(^{-1}\), respectively. Thus, the irrigated fine fescue added about 140% more SOC than did the non-irrigated fine fescue, and 55% more than irrigated-Kentucky bluegrass and creeping bentgrass. Irrigation increased both net organic carbon input to the soil profile and SOC decomposition. We found that all turfgrasses exhibited significant carbon sequestration (0.32 -0.78 ton ha\(^{-1}\) yr\(^{-1}\)) during the first 4 years after turf establishment. However, the net carbon sequestration rate was higher for irrigated fine fescue and creeping bentgrass than for Kentucky bluegrass.

In summary, C sequestration in turf ecosystems occurs at a significant rate that is comparable to the rate of C sequestration reported for US land that has been placed in the conservation reserve program. To evaluate total carbon balance, additional work is needed to evaluate the total carbon budget and fluxes of the other greenhouse gases in turfgrass systems.
Rain Gardens, Urban Runoff, and Groundwater Recharge

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Urban environments are increasingly being recognized for their potential to contribute to non-point source pollution of surface waters. Impervious surfaces such as rooftops and parking lots shed water during precipitation and snow melt, while urban areas have generally been designed to facilitate surface water flow toward storm sewers or directly into surface waters. Surface water pollution is enhanced by the rapid flow of stormwaters coming from impervious surfaces. During certain events, the sheer volume of water can overflow storm sewers or cause scouring along the banks of natural streams and rivers, exacerbating sediment and nutrient pollution. Phosphorus entering surface waters is often responsible for causing algal blooms, and correlates closely with sediment, with federal limits now provided for water entering lakes and rivers. Nitrate is another regulated nutrient as its supra-abundance (in excess of 10 mg L$^{-1}$) in water will negate that water source for drinking.

Rain gardens are intended to reduce stormwater runoff in urban areas and to enhance groundwater recharge. Rain gardens are flat-bottomed depressions, planted with “native vegetation”, which receive water flowing from impervious surfaces. The “native vegetation” is typically a mixture of wet and dry mesic prairie plants, which may or may not be native to the area. Ideally, the deep root system of the prairie plants facilitates drainage rates. The idea began in Prince County, Maryland, about 15 years ago. Since that time, various types of rain garden designs have been developed, with an equal number of models designed to estimate the runoff and percolate characteristics compared to the lawn areas they are intended to replace. Scientific research which actually measures runoff and leachate is essentially non-existent. The two primary components of rain gardens are berms and prairie plants. We hypothesized that the functional component of the rain garden are the berms, not the type of vegetation.

We constructed 16 rooftops to test our hypothesis. One set of objectives was to measure the amount of runoff and leachate water from rain gardens receiving water from the rooftops, and compare that to conventional lawn, a bermed area with lawn turf instead of prairie plants, and a prairie plant area without the berm. Each of the four treatments was replicated four times. All plots had a 5% slope. Rooftop runoff was funneled into the plots from a downspout connected to gutters on the downslope side of the roofs.

Covered troughs made from PVC were placed down slope at the end of each plot to collect runoff water. Runoff water was measured after each precipitation or snowmelt event. Subsamples were collected for measuring sediment and total phosphorus. Low-tension wick lysimeters were installed 45 cm below the surface at the center of each plot. Lysimeters were installed by digging a lateral tunnel below the plot surface to avoid disturbing the soil above each lysimeter. Lysimeters were pumped once monthly to measure leachate volume and sample for nitrate.
Berms proved to be the functional component of the rain gardens, vegetation type having no effect on bermed plots. In the absence of berms, mowed turfgrass had significantly less runoff than unbermed rain garden plantings in about half the months of the study during the first year (Fig. 1). Unbermed rain garden plantings did not provide the dense ground cover of the lawn, which allowed surface water to continue to flow, often carrying sediment. The problem was particularly acute during the first autumn, winter and spring, with about 250 kg sediment ha\(^{-1}\) lost from the sites in runoff compared to about 10 kg ha\(^{-1}\) from the unbermed lawn and bermed lawn or rain garden planting treatments. Sediment loss in the unbermed rain garden treatments was substantially reduced by late spring of the first year, as annual weeds covered the soil between the prairie plants. Mulch would likely have accomplished the same reduction, but would have introduced another variable in the study. About one-third of the prairie plants in the rain gardens died during the winter and had to be replaced. In the second year, there were no statistically significant differences among treatments as precipitation and snowmelt largely occurred at rates, which could be absorbed by the soil regardless of berm or vegetation type.

Leachate, i.e. water collected below the root zone, was equivalent among the bermed treatments and the unbermed turf area in the first year, and significantly reduced in the unbermed rain garden treatments. In the second year there were no significant differences among treatments. Ancillary research has suggested that rain garden plantings have a higher evapotranspiration rate than cool-season lawn grasses (N. Balster, 2007, unpublished data), though our water balance data didn’t support that contention (Fig. 1). Leachate under all treatments averaged less than the U.S. EPA drinking water standard of 10 mg L\(^{-1}\) nitrate on an annual basis, though all treatments, including the unfertilized rain garden plots, had leachate during the late winter months which exceeded the U.S. EPA drinking water limit for nitrate.

Ultimately, our study showed that rain gardens per se don’t necessarily reduce runoff or improve groundwater recharge in the urban environment any better than a well-maintained lawn on a soil with a reasonable drainage rate (rain gardens aren’t recommended for clay soils). If large influxes of water need to be prevented from reaching storm sewers, swales or berms on the order of 15 cm height would be sufficient to slow water flow across receiving mowed turf areas for infiltration.
Fig. 1. Water balance of plots receiving rooftop runoff, Madison, WI. Prairie plantings = PP and are considered the quintessential vegetation for rain gardens.
Proteomic and Metabolic Analysis of *ipt*-Transgenic Creeping Bentgrass with Improved Heat Tolerance

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Cytokinins (CK) are known to regulate leaf senescence and affect heat tolerance, but mechanisms underlying CK-regulation of heat tolerance are not well understood. Comprehensive proteomic and metabolomic studies were conducted to identify proteins and metabolites altered by the expression of adenine isopentenyl transferase (*ipt*) gene controlling cytokinin synthesis and associated with heat tolerance in transgenic plants for a C₃ perennial grass species, *Agrostis stolonifera*. Transgenic plants with two different inducible promoters (*SAG12* and *HSP18*) and a null transformant (NT) containing the vector without *ipt* were exposed to 20°C (control) or 35°C (heat stress) in growth chambers. Two-dimensional electrophoresis and mass spectrometry analysis were performed to identify protein changes in leaves and roots in response to *ipt* expression under heat stress. Heat-induced changes in polar metabolites in the leaves and roots of the three lines were analyzed using GC/MS. Our results exhibited transformation with *ipt* resulted in protein changes in leaves and roots involved in multiple functions, particularly in energy metabolism, protein destination and storage, and stress defense. The abundance levels of six leaf proteins and nine root proteins were maintained or increased in at least one *ipt*-transgenic line under heat stress. Differential metabolic responses between NT and transgenic lines to heat stress mainly exhibited in the metabolism of certain amino acids, organic acids and carbohydrates.
Chemical Chaperones as Potential Tools to Suppress Cell Death in Plants and Induce Stress Tolerance in Crops and Turfgrass

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As sessile organisms, plants have evolved various mechanisms to sense and defend against stresses in their environment. Biotic stresses from phytopathogens can be manifested in increased programmed cell death (PCD), both in the form of a hypersensitive response in the case of incompatible interactions or as part of the pathological symptoms in the case of biotrophic or necrotrophic pathogens. Abiotic stresses such as heat and drought have also been shown to activate PCD that contribute to loss of crop yield. Genetic study of PCD control mechanisms in the model plant Arabidopsis has led us to the identification of the conserved Bax Inhibitor-1 (BI-1) protein as a rheostat for cell death activation that can modulate responses to various stresses, most likely integrated via the Endoplasmic Reticulum (ER)-stress response pathway. Pharmacological approaches with two chemical chaperones that can alleviate ER-stress in animal and plant cells showed that suppression of PCD by these agents can lead to increased tolerance of Arabidopsis and moss to certain stresses. Recent studies with wheat and preliminary studies with turfgrass showed that application of these chemicals can provide clear protection against the biotrophic fungal pathogen *Fusarium graminearum* in wheat and some level of protection for drought with the creeping bentgrass ‘Penncross’ (*Agrostis stolonifera* L.). Continued optimization of this approach to regulate PCD in plants by modulating the ER-stress pathway may provide novel chemical control of biotrophic and necrotrophic fungal pathogens as well as improved abiotic stress tolerance in crops and turf.

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Is Plant Endophyte-Mediated Defensive Mutualism the Result of Oxidative Stress Protection?

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In this presentation we will discuss the biology and beneficial effects of plant endophytes on host plants. The current explanation of endophyte protection (defensive mutualism) of host plants is based on the secondary metabolites (alkaloids) with antiherbivore properties produced by the symbiotic association between host plant and endophytes. We propose an alternative explanation of the mechanism of host protection through enhanced stress tolerance to oxidative stress. Several studies have demonstrated production of different compounds (phenolics) with antioxidant capacity in endophyte-infected plants. Endophytes may also produce mannitol, other carbohydrates and small molecules (proline) with antioxidant capacity. We suggest that enhanced antioxidant production by symbiotic plants may be the result of production of reactive oxygen species (ROS) by endophytes. In turn, symbiotic plants are protected from oxidative stress produced by plant diseases, droughts, heavy metals, and other oxidative stressors by the production of antioxidants. We report our results on chewing’s fescue grass (Festuca rubra var. fallax) infected with a clavicipitaceous endophyte (Epichloë festucae) on enhanced tolerance to ROS in the diquat dibromide test, production of antioxidants (phenolics), proline concentrations. Studies of the endophyte demonstrate that it produces reactive oxygen species peroxides and superoxides in culture and that ROS genes noxA and noxB are expressed in tissues of the plant. Future experiments are needed to evaluate the hypothesis that antioxidants are responsible for enhanced stress tolerance in endophyte-infected plants.
Molecular Detection of Fungal Turfgrass Pathogens

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The true fungi and oomycetes constitute the majority of pathogens that infect and damage turfgrasses. Early and accurate detection and identification of these organisms is critical for turf disease management. Traditionally, diagnosticians use direct observation or culturing of specimens to identify turfgrass pathogens. DNA macroarray is a new molecular tool, which offers a fast, culture-independent alternative for the detection of microbes from field samples. The advantage of the array technique is its remarkably high throughput compared to other detection methods. Hundreds of different pathogens can be detected in one reaction with one chip.

In this study, we designed a membrane-based DNA macroarray for two important turfgrass pathogens, *Rhizoctonia solani* and *Pythium aphanidermatum*, based on the internal transcribed spacer sequences of the rRNA genes (ITS). *R. solani* causes brown patch disease in cool-season grasses and large patch in warm-season grasses. *P. aphanidermatum* causes Pythium blight, crown and root rot, and cottony blight in most turfgrass species. Our DNA array included nine probes specific to each pathogen species. Six positive controls and nine internal controls were also spotted on the array. Array sensitivity was optimized by hybridizing labeled ITS PCR products of the two target species with three sets of test probes: 1) monomer oligonucleotide probes (20-25 nt), 2) dimers: two tandem repeats of the monomers (40-50 nt) and 3) dimers with an intervening poly-A between the two repeats (50-60 nt). The use of repeat sequence probes greatly increased the sensitivity of the macroarray. However, specificity was compromised when an intervening poly-A sequence was included. Therefore, dimers, without the poly-A, performed best in terms of both sensitivity and specificity. These findings will be used to develop a multiplex detection/identification system for major fungal and oomycete pathogens of turfgrasses that will facilitate early diagnosis and improved disease management.
Metamifop: A New Postemergence Herbicide for Crabgrass and Goosegrass Control in Cool-Season Turf

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Field studies were conducted in 2008 and 2009 to evaluate postemergence grassy weed control and cool-season turfgrass tolerance to metamifop. Grassy weed control studies were conducted on high infestations of crabgrass and goosegrass, while turf tolerance trials were conducted on highly maintained, weed free stands of Kentucky bluegrass and creeping bentgrass. Metamifop was applied at rates ranging from 100 to 400 g ai/ha as single or sequential applications (at 3 week intervals) in crabgrass and goosegrass control studies. Fenoxyprop was included in all studies as a comparison at 100 g/ha. In 2008, single applications of metamifop at 200 or 400 were equally as effective fenoxyprop at 100 g/ha in controlling 3-4 leaf and 2-3 tiller crabgrass. In 2009, metamifop applied at 200-400 g/ha provided 81 to 94% crabgrass control 3 weeks after initial treatment (WAIT) and was equivalent to fenoxyprop applied at 100 g/ha. At 6 WAIT crabgrass control with single applications of metamifop at 200-300 g/ha was equivalent (ranging from 75-88% control) to fenoxyprop at 100 g/ha. However, increasing the rate of metamifop to 400 g/ha increased control to 96% which was superior to single applications of fenoxyprop. Sequential applications of both herbicides provided 95-99% crabgrass control 6 WAIT. In a crabgrass timing study, metamifop applied at 200 or 400 g/ha or fenoxyprop at 100 or 200 g/ha provided nearly complete control of 3-4 leaf crabgrass 4 WAT. Metamifop at 200 g/ha and fenoxyprop at 100 and 200 g/ha provided equivalent control of 1-2 tiller crabgrass at 4 WAT (79-88%). Metamifop at 400 g/ha provided 96% control which was superior to metamifop at 200 g/ha (79%) and fenoxyprop at 100 g/ha (85%). Control of 3-4 tiller crabgrass was equivalent (78-83%) when metamifop was applied at 200 g/ha and fenoxyprop applied at 100 or 200 g/ha. However, metamifop applied at 400 g/ha provided superior crabgrass control (95%) relative to the other treatments. Control of 6+ tiller crabgrass was equivalent (73 to 80%) with metamifop at 200 or 400 g/ha and fenoxyprop at 100 g/ha. However, control was greatest at 89% with fenoxyprop at 200 g/ha. Goosegrass control studies conducted in 2008 demonstrated that control of 3-4 leaf goosegrass was equivalent with metamifop applied at 200 g/ha and fenoxyprop at 100 g/ha. However goosegrass control was only 74% with metamifop applied at 100 g/ha. Control of 2-3 tiller goosegrass was equivalent with metamifop at 200 or 400 g/ha and fenoxyprop at 100 g/ha ranging from 83 to 94% at 4 WAT. Metamifop applied at 100 g/ha was ineffective for 2-3 tiller goosegrass control. The results of these studies suggest that metamifop shows excellent potential (that is similar to fenoxyprop) for multi-tillered crabgrass and goosegrass control at application rates ranging from 200-400 g/ha. In 2008 Kentucky bluegrass tolerance studies, single applications of metamifop applied at 200-800 g/ha and sequential applications applied at 200-400 g/ha caused less than 3% injury to Kentucky bluegrass. Fenoxyprop applied at 200 g/ha caused 18 and 11% injury at 10 and 22 days after treatment (DAT), respectively. In 2009, Kentucky bluegrass injury was not evident at metamifop rates of 800 g/ha or lower at 1 WAT. Metamifop applied at 1600 and 3200 g/ha caused 10 and
25% injury, respectively. At 3 WAT, visual injury was only evident at 3200 g/Ha of metamifop. In contrast, Kentucky bluegrass injury from fenoxyprop applied at 100 to 400 g/ha ranged from 14 to 40% at 1 WAT and increased to 10 to 66% at 2 WAT. Noticeable injury was still evident with fenoxyprop treatments at 3 WAT. In 2008 creeping bentgrass tolerance studies (maintained at 0.9 cm) injury ranged from 5 to 13% from initial applications of metamifop 1 WAT. However, injury was only evident from 800 g/ha metamifop 2 WAT. Sequential applications of these same rates of metamifop caused 10% or less injury to creeping bentgrass at any evaluation timing. In 2009, (creeping bentgrass maintained at 0.4 cm) initial applications of metamifop at 800 g/ha, fenoxyprop at 35 g/ha and quinclorac at 170 g/ha caused 9, 14 and 30% injury, respectively, 1 WAT. Injury from these treatments increased to 29, 23, and 35%, respectively, at 3 WAT. However, creeping bentgrass rapidly recovered and no injury was observed at 5 WAT. Metamifop applied at 200 and 400 g/ha and fenoxyprop applied at 18 g/ha did not cause significant injury to creeping bentgrass. Turfgrass tolerance studies suggest that metamifop may be used at much higher rates than required for weed control in Kentucky bluegrass leading to a much wider margin of safety relative to fenoxyprop. This may allow for more aggressive use of metamifop to control larger crabgrass and goosegrass as well as bermudagrass. The relative tolerance of creeping bentgrass to metamifop and fenoxyprop warrant further investigation. While far from conclusive, creeping bentgrass maintained at fairway and greens height appears to be tolerant to 200-400 g/ha of metamifop allowing for potential control of larger crabgrass plants relative to fenoxyprop at 18 g/ha.
Biological Control of Black Cutworm on Golf Course Turf

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The black cutworm (BCW), *Agrotis ipsilon* (Hufnagel) (Lepidoptera: Noctuidae), is one of the most important pests of golf courses because of the fondness of its larvae for bentgrass (*Agrostis* spp.). At the latitude of New Jersey, the BCW has three generations per year with damage occurring in mid-May to early June, late June through July, and late summer, respectively. Third instar and older larvae feed on stems and foliage which causes small dead patches, sunken areas, or pockmarks, and disrupts the uniformity and smoothness of the putting surface on greens. Foraging birds attracted to infested areas may further disrupt the surface.

Due to the extremely low tolerance for damage on tees boxes and putting greens, these surfaces are generally treated with insecticides. However, application of the commonly used broad-spectrum insecticides (pyrethroids, trichlorfon, carbaryl) adversely affects the natural enemies of BCW and other pests in addition to the adverse effect the chemicals have on the environment.

Entomopathogenic nematodes (EPN), may offer an alternative for BCW management without significant negative effects on arthropod natural enemies and environment. EPN can reproduce in the cadaver and recycle in the environment for continuous protection of the turfgrass areas from any further pest infestation.

In the present study we evaluated the virulence of several commercial EPN products against different BCW instars, determined the suitability of BCW instar for EPN reproduction, and tested efficacy and persistence of selected EPN products under field conditions.

**EPN screening**

To search for an effective EPN strain against BCW, 30-mL plastic cups (3 cm diameter) were filled with soil and one larval instar of BCW with diet was placed on the surface of the soil. Then, one of the following EPN commercial strains [*Steinernema carpocapsae* (Product name: Millenium), *S. riobrave* (BioVector), *S. feltiae* (Nemasys), *S. kraussei* (Nemasys L), *Heterorhabditis bacteriophora* (Nemasys G), and *H. megidis* (Nemasys H)] was applied directly on to the surface of the soil after the larvae had burrowed into the soil. The results indicated that up to 100% control of fourth and fifth instars can be attained using *S. carpocapsae*, *S. riobrave*, *H. bacteriophora*, or *H. megidis*. Superiority of these products were further confirmed with a potential of up to 100% fifth instar BCW control under a higher pest density in a bigger experimental arena (11 cm diameter plastic pot) in which creeping bentgrass was grown for the larvae to feed on. Furthermore, fifth and sixth instars were suitable for the reproduction of these
nematode species producing up to 65,000 nematodes per larva under laboratory conditions.

Field efficacy

Field plots were set up in a fairway creeping bentgrass at Horticultural Farm II, Rutgers University using a “plastic garden edging material” folded into a square and hammered 2.5 cm into the ground forming an experimental plot of 33 cm × 33 cm. Ten fourth instar BCW larvae were released into a square. *Steinernema carpocapsae, S. riobrave, H. bacteriophora,* or *S. feltiae* was applied to a plot. The experiment was conducted in May (night/day temperatures: 14.7/19.7 °C) and repeated in June (15.6/19.4 °C) and August (18.0/22.3 °C) 2009. Ninety-percent BCW control was achieved without any visible grass damage by using *S. carpocapsae,* or *S. feltiae* at a rate of $2.5 \times 10^9$ nematodes/ha.

Field persistence of the nematodes

*Steinernema carpocapsae, S. riobrave, H. bacteriophora,* or *S. feltiae* were applied at a rate of $2.5 \times 10^9$ nematodes/ha to 0.6m × 0.6m plots set up at different fields (sand based green, sandy-loam based green, fairway, and rough) at Horticultural Farm II, Rutgers University. The fields were established with creeping bentgrass except for the rough site which was Kentucky bluegrass. The experiment was conducted in June (average temperature, average humidity, and total precipitation during trial: 21.5 °C, 68.5%, 0.51 mm) and repeated in August (20.9 °C, 73.5%, 5.1mm) 2009. Soil samples from the treated plots were collected immediately after treatment, 4, 7, and 14 days after treatment (DAT). To quantify nematode persistence, soil samples collected on different dates were separately baited in the laboratory using an EPN susceptible insect species (*Galleria mellonella* larvae). As soil-dwelling organisms, EPNs are sensitive to high temperatures, desiccation, and UV light. After field applications, these factors may negatively affect EPN survival and hence persistence of the populations. High mortality particularly occurs in the first hours after applications until the EPN have reached the relative safety of the thatch and soil. In the current experiment, nematodes suspensions were applied in high water volume (5 mm) so that nematodes would be washed down to the ground but without any run-off. Thus, the nematodes well survived until four DAT with the highest survival for *S. riobrave* and *S. feltiae* (ca 45 – 60%). At 7 DAT, 55% of *S. riobrave* and *S. feltiae* survived at sandy-loam based green and fairway, respectively. Nematode persistence was the highest on fairway/sandy-loam based green followed by sand based green. Nematode persistence on the rough was the lowest. This may have been due to higher cut (3.8 cm) and greater thatch layer (6 years old) which may have reduce nematode penetration into the soil. On such greater thatch layer, high volume irrigation after nematode application may improve nematode penetration. In summary, with improved nematode application practices and under conducive weather conditions even in summer, inundative application of effective EPN products like *S. carpocapsae,* or *S. feltiae* have a residual activity of about a week. Prolonged golf course protection from BCW can be anticipated under suitable environmental conditions, in which the second generation nematodes from recycling in the cadaver may reproduce well and overtake.
POSTER PRESENTATIONS
Deep Transcriptome Comparison of Endophyte-Free and Endophyte-Infected Fine Fescue

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It is well established that the symbiotic fungal endophytes of grasses confer numerous benefits to their hosts. However, the details of the interaction are largely unknown. One of the outstanding questions regarding the plant-endophyte relationship is what are the factors contributing to maintenance of a compatible interaction. Understanding this may ultimately lead to the ability to infect important turfgrasses, such as Kentucky bluegrass, which do not naturally form associations with the fungal endophytes.

We are studying the host-endophyte interaction in fine fescue (Festuca rubra). We previously developed a set of endophyte-free and endophyte-infected fine fescues in which obvious phenotypic variations were documented among the plants (Johnson-Cicalese et al. 2000). Our goal is to obtain quantitative transcriptome comparisons between endophyte-free and endophyte-infected plants. Our hypothesis is that the host plant senses, in some as yet unknown way, the presence of the fungal endophyte and its transcriptome is altered in ways that are important to maintenance of the symbiotic interaction. Our approach is to use SOLiD-SAGE to compare the transcriptomes of endophyte-free and endophyte-infected fine fescue. With the SOLiD-SAGE method we can obtain a quantitative assessment of the plant and fungal genes that are expressed. We will be comparing the results from endophyte-infected and endophyte-free plants of the identical genotype. Because of the massive sequencing capability of the SOLiD system, we expect 10-15 million sequences from each sample, which will give an unprecedented amount of information on the genes expressed by the host and the endophyte. Differences between the samples in the plant genes will reveal which genes are affected by the presence of the fungal endophyte. We will also obtain quantitative data on the fungal genes that are expressed in the interaction. This information will be critical to understanding the basis of compatible interactions.

Literature Cited

A New Threat to Switchgrass: Anthracnose Leaf and Stem Blight

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Panicum virgatum (switchgrass) is a warm-season perennial grass, which has recently gained fame as a renewable, plant-based bioenergy source. Breeding for high-yielding switchgrass cultivars has resulted in the establishment of extensive monocultures that may fall victim to disease epidemics if potential disease threats are not considered during the breeding process. Because switchgrass is a native plant, disease threats have been predicted to be minimal for this crop; however, switchgrass stands in Pennsylvania, New York and New Jersey have recently exhibited symptoms of anthracnose disease, contradicting this theory. In this study, we set out to determine the identity of the fungus responsible for anthracnose disease symptoms in switchgrass. Morphological data, multilocus DNA sequencing and phylogenetic analysis identified this pathogen as a novel species within the genus Colletotrichum. Growth chamber experiments fulfilled Koch’s postulates and provided the first experimental evidence that Colletotrichum is responsible for anthracnose disease of switchgrass. Our data show that this anthracnose disease is caused by a novel species of Colletotrichum, which we formally describe as C. navitas (navitas = Latin for energy). These findings suggest that pathogens may pose a significant threat to cultivated switchgrass despite its indigenous past, and that ongoing research is needed to determine the effects of C. navitas on commercially grown switchgrass cultivars in the field. The data generated from these experiments will be utilized to determine the importance of assessing anthracnose disease resistance during the breeding process.
Identification and Characterization of Rusts Infesting Cultivated Turfgrass Using Molecular Methods

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Rust is a common disease of cool-season turfgrasses that can decrease the aesthetic and economic value of many cultivated species, particularly Kentucky bluegrass (Poa pratensis L.). Chemical control of rust is costly and sometimes ineffective; therefore the use of resistant cultivars is important for the effective management of this disease. Over the past ten years, increased susceptibility to rust (Puccinia spp.) has been observed for several Kentucky bluegrass cultivars in the United States, most notably the once highly resistant ‘Midnight’ types. It has been theorized that new races or even new species of the pathogen may be responsible for this shift in cultivar susceptibility, but the data needed to test this hypothesis is lacking. In the current study, we are developing and utilizing molecular markers to evaluate turfgrass rust populations. To date, 66 rust infested leaf samples have been collected from graminicolous hosts in North America, the United Kingdom, Australia, and Chile. A reliable DNA extraction protocol was developed and the complete internal transcribed spacer (ITS) region and 5.8S ribosomal DNA portion of the samples were PCR amplified and sequenced. Assembled ITS sequences ranged from 682 to 701 base-pairs in length, including the partial sequences of the flanking 18S and 28S rDNA. Bayesian phylogenetic analysis of the aligned sequence data identified P. coronata (crown rust), P. graminis (stem rust), and P. striiformis (stripe rust) from rust infested grass samples. P. coronata was the most prevalent species (68% of samples), followed by P. graminis (27%), and P. striiformis (5%).

The species frequencies identified in our study contradict what has been frequently observed among turfgrass breeders in the field, where spore phenotype was the most common method used to identify rust species, thus suggesting that stem and stripe rust would be most prevalent. In addition to morphology, host-plant identity has played a role in field identification of rust species, where stem rust was traditionally associated with Kentucky bluegrass and crown rust with perennial ryegrass (Lolium perenne L.). However, in the current study, not only was crown rust shown to be the predominante species, but it was frequently found in association with Kentucky bluegrass hosts, indicating that the most common method for identifying these pathogens in the field -- spore pigmentation and host plant association -- are inadequate for accurate rust species identification.

To provide turfgrass breeders, pathologists and diagnosticians with an accurate, reproducible and rapid method for turfgrass rust species identification, we have developed a species-specific real-time PCR protocol using the ITS sequence data generated from this study. Probes were designed within ITS1, and in combination with rust specific primers, are capable of detecting ~ 3 x 10⁻³ ng of rust DNA. Although the ITS region was sufficient for rust species identification, these data provided low
intraspecific resolution, indicating the need for finer-scale markers to identify races within rust species. Exceptions to this generalization were observed, as in the case of the phylogenetic clustering of four isolates of stem rust and three isolates of crown rust. These sub-specific clusters were strongly supported by posterior probability values as unique groups and support the hypothesis of race structure within the turfgrass rusts. To better understand these relationships in the turfgrass rust population, amplified fragment length polymorphism (AFLP) analysis will be used to evaluate our rust collection in 2010.
Switchgrass (Panicum virgatum) is a perennial warm season grass (C4) native to most of the US with the exception of some Northwestern states. Switchgrass has been used in restoration, buffer strips, as a forage crop, and as an ornamental, but is now emerging as a source of alternative energy due to its native status, perennial life cycle, ability to grow on marginal land, and high yields with little inputs. However, little information is available on switchgrass yields in the northeastern US. The objectives of this study were to determine and compare yields of four switchgrass cultivars and corn (Zea mays) at two locations in New Jersey. Four switchgrass cultivars (Alamo, Carthage, Cave-in-Rock, and Timber) and corn were planted in a randomized complete block design at two locations: Pittstown, NJ and Upper Deerfield, NJ in spring of 2007. Due to establishment failure, the Pittstown location was replanted in spring of 2008. Switchgrass plots were established at a rate of 10 lbs PLS/acre. Locally grown Round-Up ready corn varieties were planted at each location at a rate of 28,000 seeds/acre. During establishment, irrigation was applied to promote germination and 2,4-D and dicamba were used to control broadleaf weeds. No herbicides or supplemental irrigation were applied in years 2 and 3. Both trials were managed for switchgrass production and received 50 lbs N/acre applied in mid-May of each year. A single fall harvest was made at each location and biomass yields were determined for Upper Deerfield in 2008 and 2009 and for Pittstown in 2009. Corn grain and stover was also harvested and separated to determine plant biomass and grain yield. Biomass yields for switchgrass ranged from 3.9 to 5.9 tons/acre. Corn grain yield under switchgrass management ranged from 46.6 to 53.1 bushels/acre. This was 19 to 32% lower than corn managed for corn production (adequate irrigation and 100 lbs N/acre). Cultivars Timber, Alamo, and Carthage had the highest yields at both locations. Conversion of biomass yields to ethanol yields resulted in 18 to 66% higher yields in switchgrass than corn grain and stover combined at both locations. Results found suggest that once an infrastructure for cellulosic ethanol is established, Timber and Alamo could be promising cultivars for biomass production in NJ as an alternative to corn.
Rain Garden Training for Professional Landscapers

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Rutgers Cooperative Extension offers a rain garden installation program for professional landscapers. The program was developed as part of a USDA CSREES National Water Program Grant and will continue to be offered in 2010. The rain garden training for professional landscapers provides the skills needed to install and maintain a rain garden and assists with marketing the new service offering. The rain garden training includes class lectures and a hands-on demonstration, which results in the installation of a community demonstration rain gardens. The professional landscapers receive a certificate of completion for the rain garden training and educational materials. The training program is offered in northern and southern New Jersey in Union and Gloucester counties. A brochure listing the professionals who completed the training has been published and distributed at public education programs. A website, http://water.rutgers.edu/Rain_Gardens/RGWebsite/landscaper.html provides stakeholders with an overview of the training program and the directory of professionals who completed the training. Program evaluations indicate the landscape professionals have improved their knowledge and will use the information from the training to promote the use of rain gardens to their customers, install rain gardens on client’s property and work with local municipalities to install rain gardens.
Recycling Home Construction Waste Benefits Turfgrass Establishment

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A case study examined the potential to utilize home construction waste materials as soil amendments for turfgrass establishment. In 2007, during construction of a 1500 sq. ft. addition to my home, I collected all waste materials and made an effort to find beneficial uses. With the exception of old asphalt roofing and scrap pressure-treated lumber, a recycling outlet or beneficial use was found for nearly every waste material.

The collected non-recyclable material filled about one-half of a dumpster and was sent to a landfill. The contractor had originally estimated that the accumulated construction waste would require three dumpsters. In the final analyses, a volume of two dumpsters of material was recycled. For the savings associated with this effort the contractor agreed to deduct $1000 from the final construction project invoice. This was a welcome compensation for the laborious recycling effort.

Waste materials that were diverted from the dumpster by hauling them to a recycling center included bottles, cans, scrap metal (such as used saw blades, nails, etc), and cardboard. Compostables (such as food waste, paper napkins, and sawdust from untreated lumber) were collected and composted. Fiberglass insulation scraps were collected and saved for augmenting the layer of insulation in the home attic. Gypsum board accounted for about one-third of the total amount of waste. This material (mostly calcium sulfate and paper) was applied to land area intended for lawn establishment. Once this material became moist after a rain, it was tilled into the soil. Soil compaction, an often serious and persistent problem following construction, was alleviated with use of the contractor’s backhoe by digging down one bucket depth and lifting and loosening the soil.

Homemade compost, supplemented with imported mushroom compost, was and spread in a one-inch layer over the intended lawn area and mixed with the surface, six inches of soil with tillage. No NPK fertilizer was necessary. Limestone was applied, as needed, based on soil tests. The lawn was seeded to Kentucky bluegrass in the fall of 2008. For my efforts, tons of waste was diverted to beneficial use and a beautiful dense green lawn was established.
Anthracnose Severity on Annual Bluegrass as Affected by Topdressing Programs and Cultivation Techniques

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Anthracnose is a destructive disease of cool-season turf caused by the fungus *Colletotrichum cereale* Manns. Disease frequency and severity have increased on putting greens over the last fifteen years, especially on annual bluegrass (*Poa annua*) turf. Such outbreaks have been attributed, in part, to the adoption of rigorous, stress-inducing turf management practices by golf course superintendents to address golfer expectations for fast putting greens. Previous research has shown that sand topdressing of annual bluegrass turf during the summer can reduce anthracnose severity, but the effect of spring topdressing on this disease has not been examined. Moreover, the effect of mid-season cultivation techniques, which injure leaves and other plant parts, on disease severity has not been studied. Three field studies were initiated in 2009 and will be conducted through 2010 to evaluate the impact of sand topdressing and cultivation on anthracnose severity of annual bluegrass. All studies were conducted on annual bluegrass turf grown on a Nixon sandy loam and maintained at a 3.2 mm cutting height in North Brunswick, NJ.

The objective of the first study was to evaluate the effect of spring topdressing rate (0, 1.2 and 2.4 L m\(^{-2}\)) on anthracnose as well as the potential for this factor to interact with the effects of summer topdressing (0, 0.075, 0.15, 0.30 and 0.6 L m\(^{-2}\) every 14-d) on anthracnose severity. The trial used a 3 x 5 factorial arranged as a randomized complete block with four replications. Disease symptoms were slow to develop during the first year of this study; no treatment reached disease severity level greater than 10% until late July. Preliminary data analysis indicated that summer topdressing had a greater and more consistent effect on disease severity than spring topdressing. Both spring and summer topdressing at times reduced disease severity compared to no topdressing. Of the spring topdressing treatments, 2.4 L m\(^{-2}\) appeared to be the most effective rate at reducing disease severity. Sand topdressing at 0.30 L m\(^{-2}\) every 14-d during the summer appeared to be the lowest rate required to consistently reduce disease severity although further data analysis is needed to verify this finding.

The objective of the second study was to evaluate the impact of summer topdressing (0, 0.075, 0.15, 0.3 and 0.6 L per m\(^2\)) applied only after the initiation of anthracnose symptoms on disease severity. This trial used a 2 x 4 factorial arranged in a randomized complete block design with four replications; a non-topdressed control was also included. The four summer topdressing rates were applied one time or biweekly once disease severity had reached approximately 10% of the entire plot area (28 July). Data analysis is ongoing; however, a preliminary assessment suggested that initially lower topdressing rates increased disease while greater rates either had no effect or reduced disease severity. Towards the end of the evaluation period, biweekly topdressing was more likely to reduce disease severity than the single application of topdressing.
The objective of the third study was to assess the effect of mechanical injury on disease severity when cultivation was performed at the onset of disease symptoms. The cultivation techniques used included scarifying (6 mm depth), verticutting (3 mm depth), grooming (1.3 mm depth), solid-tine cultivation (5.7 cm depth with 6 mm wide tines on a 3.8 by 3.8 cm spacing) and a non-cultivated control. All four cultivation techniques were applied once on 24 July when disease symptoms had exceeded 10% of the plot area; weekly grooming (1.3 mm depth) was performed as a fifth cultivation treatment. The trial used a randomized complete block with four replications. Verticutting and scarifying treatments produced the greatest mechanical damage and were the only cultivation treatments that increased disease severity in August and early September. All three studies will be repeated in the same locations in 2010.
Genetic Variation and Physiological Traits in Drought Tolerance for Bentgrass

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Drought is a major abiotic stress, which often leads to decline in turf health and quality in various turf grass species. Creeping bentgrass (*Agrostis stolonifera*), one of the widely used species on golf courses, is sensitive to drought stress. Improving drought tolerance in creeping bentgrass is critically important for water conservation on golf courses. The genetic variation is an invaluable source for selecting drought-tolerant germplasm that can persistent drought. Knowledge of physiological traits associated with drought tolerance is essential for selecting drought-tolerant germplasm through conventional breeding and biotechnology. The objectives of the study were to compare genetic variation in drought tolerance among commercial available cultivars, transgenic line, and wild species, and to determine major physiological traits linked to drought tolerance in bentgrass. Eight cultivars of creeping bentgrass, a thermal bentgrass (*A. scabra*) collected from Yellowstone National Park, and a transgenic bentgrass line with a gene (*ipt*) for increasing cytokinin synthesis were subjected to drought by withholding irrigation for 7-10 days, and assessed with several measurements which included: turf quality, normalized difference vegetation index (NDVI), leaf relative water content (RWC), leaf electrolyte leakage (LEL), leaf photochemical efficiency (Fv/Fm), root electrolyte leakage (REL) and root respiration activity. These measurements, which were then tested for correlation with each other, encompass overall turf performance, water usage, photosynthetic health, and membrane stability during drought. Using these parameters cultivars were grouped into three groups of varying drought tolerance of high, medium or low tolerance: ‘Kingpin’, ‘L93’ and ‘Penncross’ consistently performed poorly under drought stress; the thermal grass ‘Ntas’ and transgenic ‘H31’, and ‘Declaration’ and ‘Pro’ were most tolerant; ‘007’, ‘Tyee’ and ‘Shark’ were intermediate. Correlation analysis showed that RWC and Fv/Fm were positively correlated to turf quality while LEL and REL were negatively correlated to turf quality under drought stress. Fv/Fm had the highest correlation coefficient with turf quality and NDVI among all parameters. While this study shows the relative performance of several cultivars the more important results are the correlation of the physiological parameters with drought tolerance. This information can be applied to improving selection techniques for breeding as well as understanding physiology mechanisms and genetic sources of drought tolerance.
Identification of Quantitative Trait Loci for Drought Tolerance Characteristics of Creeping Bentgrass

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Loss of turf quality due to limited water availability is a major concern in turfgrass management. Therefore, there is an urgent need to develop turfgrasses that are able to provide high quality turf with limited irrigation. Several different drought tolerance mechanisms have been identified that are utilized by turfgrasses but have yet to be fully genetically characterized. It is known that most are known to be controlled by multiple genes or quantitative traits; thus, identification of quantitative trait loci (QTLs) associated with drought tolerance would improve the selection efficiency of drought-tolerant germplasm and varieties. The objectives of this study were 1) to evaluate genetic variations in drought tolerance of a mapping population of bentgrass; and 2) to identify QTL markers associated with drought tolerance in creeping bentgrass. The mapping population with both parents (‘L-93’ and 7418-3) and 102 F2 progenies exhibited significant variation in drought symptom expression when exposed to drought stress under greenhouse and field conditions. QTL analysis was conducted on the phenotypic traits associated with drought tolerance (turf quality, electrolyte leakage, relative water content, water use efficiency, leaf area index, green leaf biomass, and osmotic adjustment). QTLs consistent in both years of the study will be discussed. The results indicate that some QTL markers associated with drought tolerance could be useful in marker-assisted selection of drought-tolerant turfgrass.
Hybrid Hazelnut Consortium: A Collaborative National Effort to Expand Hazelnut Production

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The Rutgers University Underutilized Perennial Crops Genetic Improvement Program recently joined forces with Oregon State University, the University of Nebraska, Lincoln, and the National Arbor Day Foundation to form the Hybrid Hazelnut Consortium. The goal of the consortium is to leverage previous investments, research developments, and genetic resources to greatly expand the production region of hazelnuts in the United States. At Rutgers, hazelnut (Corylus spp.) research began nearly fifteen years ago by Dr. Reed Funk as part of a temperate nut tree evaluation and improvement program. Hazelnuts quickly showed their merits and since 2000 we have had an active hazelnut breeding program that has expanded yearly and is demonstrating significant progress. Our major focus has been identifying and developing improved plants resistant to the disease eastern filbert blight, caused by the fungus Anisogramma anomala, which is the primary limiting factor of production in eastern North America. Much of the Rutgers’ work has been done in close cooperation with Oregon State University where hazelnut research and breeding has been ongoing for over forty years and the world’s largest collection of Corylus genetic resources are held. More recently, the demand for growing sustainable, low-input crops has inspired hazelnut research at the University of Nebraska and at the National Arbor Day Foundation, in Lincoln City, NE. At these locations efforts are focused on developing hybrid hazelnuts (Corylus avellana x C. americana) adapted to the harsh environment of the upper Midwest. By developing the Hybrid Hazelnut Consortium, we are better able to share and utilize resources, including land, and can more effectively evaluate and develop germplasm, breeding progeny, and potential new releases adapted to multiple locations and environments. In 2009, this partnership was awarded a USDA Specially Crop Research grant to build on efforts in areas of germplasm enhancement and cultivar development, genomics of host plant and pathogen (A. anomala), and national extension and outreach efforts. Based on our collaborations, the future of significantly expanding the regions of sustainable hazelnut production looks very bright.
Seasonal Wear Tolerance and Recovery in the 2005 NTEP Kentucky Bluegrass Test

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Increased use of sports fields and other recreational sites presents a difficult challenge for turfgrass managers responsible for maintaining uniform and safe natural playing surfaces. Kentucky bluegrass (*Poa pratensis* L.) is one of the most commonly used turfgrass species for sports fields grown in cool-season climates and cultivar recommendations for sports fields based on the specific season of play would be useful. The objective of this study was to assess the wear tolerance and recovery of Kentucky bluegrass in spring, summer, and fall. One-hundred twenty-one entries, including the 2005 National Turfgrass Evaluation Program (NTEP) Kentucky Bluegrass Test, were established in September 2005 on a well-drained Nixon loam at Horticultural Research Farm II in North Brunswick, NJ. The test was mowed 2 to 3 times per week with a reel mower at a height of 3.8-cm and was irrigated as necessary to limit drought stress. Eighteen passes of simulated wear were applied in October 2006 (fall), July 2007 (summer), and April 2008 (spring). The section of each plot that received wear was visually assessed for fullness of turfgrass canopy (0-100% scale; 0=absence of turfgrass canopy and 100%=full canopy) before wear (C_{BW}), immediately after wear (C_{W}) and 20 days after wear (C_{20DAW}). Canopy retention immediately after wear was calculated as: C_{W}-C_{BW}, where a less negative value indicated better canopy retention. The redevelopment of the turfgrass canopy during recovery was assessed as: C_{20DAW}-C_{W}. Analysis of variance was performed on data arranged in a randomized complete block design. Entries were replicated three times in each season; replications were nested within season-year (spring, summer, and fall). Means were separated using Fisher’s protected least significant difference (LSD) test at *P* < 0.05. Kentucky bluegrass had the least C_{W} after spring wear (summer=fall>spring); however, C_{W}-C_{BW} ranked spring>fall>summer. Thus, low C_{W} during spring was due to incomplete green-up (low C_{BW}). Season-years did not differ for C_{20DAW}; however, season-years did effect recovery as measured by C_{20DAW}-C_{W} and ranked spring>summer=fall. Greater C_{20DAW}-C_{W} during spring was due to rapid growth of Kentucky bluegrass during spring. Kentucky bluegrass entry responses were dependent on season-year. ‘Julia’, CPP 822, and ‘Sombrero’ (CP 76-9068) had the greatest C_{W} and C_{20DAW} after spring, summer, and fall and were ranked the best for canopy retention (C_{W}-C_{BW}) during two or more season-years. The lowest C_{W} and poorest canopy retention across all three season-years was exhibited by DLF 76-9075 and ‘Mermaid’ (DP 76-9081), respectively. The following had the poorest C_{W}-C_{BW} in two or more season-years: A95-410, PSG 366, ‘Volt’ (A98-999), A00-1400, SPTR 2959, ‘Glenmont’, ‘Starburst’ (STR 2703), DLF 76-9075, ‘Empire’ (A01-299), Mermaid, PST1A1-899, A99-2377, A97-1560, ‘Barduke’ (BAR VV 8536), A00-247, ‘Belissimo’, ‘Kenblue’, ‘Futurity’ (A99-3119), RAD-762, BAR VK 0710, and A00-99.
Sand topdressing on putting green turf enhances surface aesthetics and playability. Anthracnose, caused by *Colletotrichum cereale* Manns, is a disease of annual bluegrass [*Poa annua* L. f. reptans (Hausskn) T. Koyama], which is more severe on stressed turf and was thought to be enhanced by sand topdressing and foot traffic. A two year field study was initiated in 2007 to evaluate the effect of foot traffic and sand topdressing on anthracnose severity in North Brunswick, NJ. A split-plot design with foot traffic [none and 392 footsteps m$^{-2}$ d$^{-1}$ (200 rounds d$^{-1}$)] as the main factor and sand topdressing (none and 0.3 L m$^{-2}$ wk$^{-1}$) as the subplot factor was used on an annual bluegrass putting green established on a Nixon sandy loam and maintained at a 3.2 mm cutting height. Foot traffic was initiated 14 June 2007 and 6 June 2008 and continued through 4 September 2007 and 7 September 2008, respectively. Sand topdressing was applied from 14 May through 28 August 2007 and 15 May through 29 August 2008. Nitrogen was applied bi-weekly at 4.9 kg ha$^{-1}$ resulting in a total of 29.3 kg ha$^{-1}$ during the time treatments were applied. Foot traffic was applied after morning mowing as straight line passes (5 d wk$^{-1}$) by students wearing golf shoes equipped with soft spikes. Anthracnose was rated using a line-intersect grid counting method. Surprisingly, foot traffic reduced anthracnose severity as much as 27% regardless of the level of sand topdressing during both years. Sand topdressing resulted in a subtle increase in disease severity during June and July 2007. However, disease severity decreased by August as topdressing continued in 2007. Disease severity was either not affected or reduced by topdressing during 2008. Interactions between treatments occurred three times during the two year study. The interaction in 2007 (two dates) indicated that the reduction in disease severity caused by topdressing began sooner in plots that also received foot traffic. In 2008 (one date), the reduction in disease severity resulting from topdressing was only apparent in plots that received regular foot traffic. Although visual symptoms of wear stress were evident in foot traffic plots, the treatment combination of foot traffic (5 d wk$^{-1}$) and weekly sand topdressing resulted in the best turf quality by the end of both seasons. Results indicate that the practice of sand topdressing should not be terminated, even under conditions of intense foot traffic, when anthracnose develops on annual bluegrass putting greens.
Effect of Photoperiod and Temperature on the Production Rate of Tillers and Rhizomes in Tall Fescue

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Temperature and photoperiod are key factors regulating plant growth and development. The objective of this study was to investigate effects of temperature and photoperiod on tillers and rhizomes formation in tall fescue. Different genotypes were selected based on tillering ability and/or rhizome production. Plants were grown in growth chambers with a combination of different photoperiods and day/night temperatures; short photoperiod (9 hours) and low temperature (day/night at 15/10 °C) (SL), long photoperiod (18 hours) and low temperature (LL), short photoperiod and high temperature (day/night at 25/15 °C) (SH), and long photoperiod and high temperature (LH). Each treatment was replicated four times in four growth chambers. The SL plants showed significant increases in the number of tillers in two clonal lines. The LL plants also showed increase in the number of tillers. The number of rhizomes was increased under LL and LH conditions in three clonal lines. Our results suggested that the responses of tiller and rhizome development to temperature and photoperiod varied with genotypes; in addition, longer photoperiod appeared to promote rhizome formation, regardless of temperature while lower temperature stimulated tiller production in some genotypes of tall fescue. This study would be helpful in determining the performance of tall fescue under the longer/shorter photoperiod and variable temperature conditions in different parts of United States.
Use of Mesotrione for Annual Bluegrass (\textit{Poa annua} L.) Control at Kentucky Bluegrass Establishment

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Field studies were conducted in the fall of 2007 to the spring of 2009 to evaluate the response of newly seeded Kentucky bluegrass cultivars to mesotrione applied at planting (PRE), and PRE followed by (fb) sequential treatments four weeks after turfgrass emergence (WAE) at rates ranging from 0.28 to 2.24 kg ai/ha. In separate studies annual bluegrass control in newly seeded ‘Midnight II’ Kentucky bluegrass was evaluated with mesotrione applied PRE fb sequential treatments 4 and 8 WAE at 0.14 to 0.56 kg/ha. All applications were made with a single 9504E nozzle \textit{CO}_2 pressured sprayer calibrated to deliver a total 375 L/ha at 220 kPa. Experimental designs were a strip-plot with four replications for the Kentucky bluegrass cultivar study and a randomized complete block with four replications for the annual bluegrass control study. Kentucky bluegrass cultivars ‘America’, ‘P-105’, ‘Midnight II’, ‘Avalanche’, Kingfisher’, ‘Washington’, ‘Bedazzled’, ‘Thermal’, and ‘Award’ were seeded on 8-28-07, and 9-16-08 at 1.7 kg/ha in 1.8 m rows with a drop spreader. Annual bluegrass control studies were initiated on 9-13-07 and 09-22-08. Kentucky bluegrass cover and annual bluegrass control were visually evaluated in December and the following spring on a scale of 0 (no cover or control) to 100 (complete cover or control). In the Kentucky bluegrass cultivar study significant cover reductions were not evident across all Kentucky bluegrass cultivars at rates of 0.28 and 0.56 kg/ha. Cover reductions were evident on some cultivars at rates of 1.12 and 2.24 kg/ha and sequential applications further reduced cover. A high degree of intraspecific variability was evident with cultivars ‘Thermal’ and ‘Washington’ showing the most tolerance while ‘Kingfisher’ and ‘Avalanche’ were the most sensitive. In the annual bluegrass control studies, nearly complete control of winter annual broadleaf weeds such as chickweed, henbit, oxalis and veronica were observed at all application rates. ‘Midnight II’ cover was not significantly reduced by mesotrione. Annual bluegrass control ranged from 61 to 94% and increased with increasing mesotrione rate. Annual bluegrass control was 83% at 0.28 kg/ha averaged across the three application regimes. Applying a sequential application of mesotrione at 4 WAE increase annual bluegrass control to 80% from 74% compared with a single PRE application. Applying a third application of mesotrione at 8 WAE did not further increase annual bluegrass control compared with two applications. The results of these studies suggest that the overall tolerance of Kentucky bluegrass is excellent and mesotrione can be safely used at establishment for high levels but not complete annual bluegrass control.
Identification and Functional Analysis of Drought-Responsive Proteins in Kentucky Bluegrass Cultivars Differing in Drought Tolerance

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Drought is one of the major limiting factors of plant production worldwide. The objective of this study was to investigate protein changes associated with drought tolerance in two Kentucky bluegrass (Poa pratensis L.) cultivars. Plants of ‘Brilliant’ and ‘Midnight’ were subjected to drought stress by withholding water for 15 days in growth chambers. The leaves were harvested at 10 and 15 days after drought treatment. ‘Midnight’ maintained higher relative water content and photochemical efficiency, and lower membrane leakage than ‘Brilliant’ at 15 d of drought stress. Proteins were extracted and separated by difference gel electrophoresis. Eighty-eight protein spots were differentially accumulated in response to drought stress in at least one cultivar. The sequences of these protein spots were analyzed using mass spectrometry and 64 spots were identified. Many proteins involved in amino acid metabolism or energy metabolism were down-regulated in both cultivars, and most of these proteins had higher level in ‘Midnight’ than in ‘Brilliant’ cultivar. The abundance of two proteins related to protein stability (60 kDa chaperonin and 70 kDa heat shock cognate) was decreased under drought only in ‘Brilliant’, suggesting that heat shock proteins may contribute to superior drought tolerance in ‘Midnight’.
Leaf Proteomic Responses to Drought Stress in Two Bermudagrass Cultivars Contrasting in Drought Tolerance

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Drought is one of the major abiotic stresses that affect plant growth. To investigate drought tolerance mechanisms in warm-season turfgrass species, a physiological and comparative proteomic analysis was carried out in two bermudagrass (Cynodon spp.) cultivars differing in drought tolerance. Two cultivars (‘Tifway’ and ‘C299’) of bermudagrass developed gradual water stress over 15 days without watering, while the control treatments were maintained well-watered. Drought stress significantly increased leaf relative electrolyte leakage and decreased leaf chlorophyll fluorescence (Fv/Fm), as well as relative water content. ‘Tifway’ exhibited better drought tolerance than ‘C299’.

Total proteins of leaves were extracted from well-watered and drought-stressed plants at 10 days after treatment, separated by two-dimensional gel electrophoresis (2-DE) and stained with colloidal coomassie brilliant blue (CBB). Out of about 750 protein spots reproducibly detected, 32 proteins had increases in abundance and 22 proteins exhibited decreases in abundance, and no changes were detected in the remaining number of proteins. All the drought-responsive proteins were excised from gels and subjected to mass spectrometry analysis, leading to identification of 51 proteins involved in metabolism, energy, cell growth/division, protein synthesis, and disease/defence. The results suggest that the superior drought tolerance in ‘Tifway’ could be mainly attributed to higher photosynthetic capacity, as indicated by more stable photosynthetic proteins and less degradative energetic proteins and greater levels of antioxidant enzymes related to the control of reactive oxygen species (ROS).