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Proceedings of the Twenty-Eighth Annual Rutgers Turfgrass Symposium

James F. White, Jr. and Barbara Fitzgerald, Editors
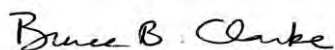
Director's Opening Remarks:

Welcome to the Twenty-Eighth Annual Rutgers Turfgrass Symposium. The Symposium was established in 1991 to provide Rutgers faculty, students, and staff with an annual forum for the exchange of ideas on a wide range of topics in turfgrass science. Over the years, this format has expanded to include presentations by colleagues at other institutions. I would like to thank Dr. Ambika Chandra (Department of Soil and Crop Sciences at Texas A&M University) who will present a keynote address entitled "Warm-season Turfgrass Breeding at Texas A&M," as well Dr. Geunhwa Jung (University of Massachusetts) and Dr. Lisa Beirn (Syngenta), and all of the Turf Center faculty and students who have agreed to present their research at this year's symposium. I would also like to thank Drs. Tom Gianfagna, Matthew Elmore, Donald Kobayashi, and Stacy Bonos for serving as session moderators and the Symposium Planning Committee comprised of Drs. Matt Elmore (Symposium Chair), Rong Di and Bruce Clarke, as well as Dr. Jim White and Ms. Barbara Fitzgerald (co-editors of the Symposium Proceedings) for their hard work in the preparation of this year's program. Without their efforts, this year's Symposium would not have been possible.

The faculty, students and staff in the Rutgers Center for Turfgrass Science continue to be recognized for excellence in research, teaching and outreach. In 2018, our graduate students received several major awards for their research accomplishments. Dr. Pat Burgess received the Musser International Turfgrass Foundation's Research Scholarship Award and James Hempfling was awarded the Watson Fellowship; these are two of the most prestigious graduate student awards in the country. Moreover, five of our students were recognized during the Graduate Student Poster and Oral Paper Competition at the Crop Science Society of America's Annual Meeting in Baltimore, Maryland. Trent Tate and Cathryn Chapman took First and Third Place honors, respectively, in the Turfgrass Management Oral Session, Phillip Vines and William Errickson were awarded First Place and Second Place, respectively, in the Plant Breeding Oral Session, and Phillip Vines and Stephanie Rossi each received Second Place honors in the Turf Genetics and Turf Physiology Poster Sessions, respectively.

Turf Center faculty continue to conduct outstanding research, undergraduate and graduate teaching, and continuing professional education and service programs in support of the Turfgrass Industry. In return, the Turfgrass Industry have donated their time and over $5 million in research grants, student scholarships, fellowships, buildings (the Ralph Geiger Education Complex and the Steven Hart Memorial Pavilion), equipment, and gifts to the Rutgers Turfgrass Program. We are indeed fortunate to have such an outstanding partnership with our Turfgrass Industry colleagues in the state, region and nation.

I welcome you to this year's Turf Research Symposium and hope that you will find it an enjoyable and a worthwhile experience.

Sincerely,

Bruce B. Clarke, Director
Rutgers Center for Turfgrass Science

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TWENTY-EIGHTH ANNUAL RUTGERS TURFGRASS SYMPOSIUM

School of Environmental and Biological Sciences, Rutgers University

January 11, 2019

Foran Hall, Room 138A

Friday, January 11, 2019

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

9:00 - 10:00 AM   **SESSION I: Germplasm Enhancement**
(Moderator: Tom Gianfagna)

9:00 – 9:20   **Rong Di** (*Department of Plant Biology, Rutgers University*) CRISPR-Gene Editing of 'Crenshaw' Creeping Bentgrass to Improve Stress Tolerance

9:20 – 9:40   **Phillip Vines** (*Department of Plant Biology, Rutgers University*) A High-Density Genetic Linkage Map and Quantitative Trait Locus Analysis for Salinity Tolerance and Dollar Spot Resistance in Perennial Ryegrass (*Lolium perenne* L.)

9:40 – 10:00   **William Meyer** (*Department of Plant Biology, Rutgers University*) Enhancing Cool-Season Turfgrass Germplasm by Collecting in the Centers of Origin

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

10:30 – 11:10 AM   **SESSION II: Mechanisms of Pesticide Resistance**
(Moderator: Matthew Elmore)

10:30 – 10:50   **Geunhwa Jung** (*Stockbridge School of Agriculture, University of Massachusetts – Amherst*) Multidrug Resistance Conferred by a Xenobiotic Detoxification Through Transcriptional Regulation in Filamentous Fungi

10:50 – 11:10   **Albrecht Koppenhöfer** (*Department of Entomology, Rutgers University*) Diagnosis and Management of Insecticide Resistance in Annual Bluegrass Weevil

11:10 – 11:20 AM   **Discussion session**

11:20 – 12:00 PM   **KEYNOTE: Ambika Chandra** (*Department of Soil and Crop Sciences, Texas A&M*) Warm-season Turfgrass Breeding at Texas A&M

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

1:00 – 2:00 PM   **SESSION III: Turfgrass Management**
(Moderator: Donald Kobayashi)

1:00 – 1:20   **James Murphy** *(Department of Plant Biology, Rutgers University)*
Effects of Finer-Textured Topdressing Sand on Creeping Bentgrass
Putting Green Turf

1:20 – 1:40   **Qiang Chen** (*Department of Plant Biology, Rutgers University*)
Metagenomic Analysis Reveals Diversity and Effects of Bacterial
Endophyte Communities Associated with Turf Seeds

1:40 – 2:00   **Matthew Elmore** (Department of Plant Biology, Rutgers University)
Goosegrass Emergence and Control of Suspected Herbicide Resistant
Populations

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

2:30 – 3:30 PM   **SESSION IV: Turf Pathology**
(Moderator: Stacy Bonos)

2:30 – 2:50   **Lisa Beirn** (*Syngenta*) Not Your Grandfather's Dollar Spot: Resolving the
Taxonomy of this Important Turfgrass Disease

2:50 – 3:10   **Ning Zhang** (*Department of Plant Biology, Rutgers University*)
Understanding the Microbiome Associated with Turfgrasses

3:10 – 3:30   **Faith Belanger** (*Department of Plant Biology, Rutgers University*) What
We Know So Far About the *Epichloë festucae* Antifungal Protein

3:30 - 3:45 PM   **Discussion and Closing Remarks**

3:45 PM   **Social Hour and Poster Session**

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PLENARY PRESENTATIONS

CRISPR-Gene Editing of "Crenshaw" Creeping Bentgrass to Improve Stress Tolerance

Rong Di, Stacy Bonos and William Meyer

Department of Plant Biology, Rutgers University

Creeping bentgrass (*Agrostis stolonifera* L., *As*) is one of the most widely used cool-season grass species on golf courses. However, there is not one cultivar of creeping bentgrass that is considered completely resistant to dollar spot disease caused by *Sclerotinia homoeocarpa* F.T. Bennet. Creeping bentgrass is also stressed by heat and drought during summer months. The new and versatile CRISPR/Cas (clustered regularly interspaced short palindromic repeats-associated endonuclease) technology has provided an easy and precise gene editing platform to engineer all organisms. It has been used to knock-out disease susceptible genes in plants to improve disease resistance and stress negative regulators to enhance plant stress tolerance. We have developed the CRISPR/Cas gene editing platform for grass genomic modification. CRISPR-gene editing vector has been constructed to have the wheat U6 promoter driving the expression of guide RNA (gRNA) targeting to any chosen gene and the monocot codon-optimized Cas9 nuclease gene under the control of maize ubiquitin promoter. We have developed an efficient plant transformation protocol for "Crenshaw" creeping bentgrass by both gene gun bombardment and *Agrobacterium*-mediated gene delivery. The partial genomic DNA (gDNA) sequence of *DREB* (dehydration responsive element binding 2)-like gene of "Crenshaw" creeping bentgrass was identified by bioinformatics analysis and cloned by PCR (polymerase chain reaction). The 20-nucleotide *AsDREB* target sequence was chosen and the CRISPR-gene editing vector was constructed to transform the embryogenic calli initiated from "Crenshaw" creeping bentgrass seeds. Many transgenic "Crenshaw" plants have been produced by both gene gun and *Agrobacterium* transformation methods. A 277-bp gDNA fragment from each transgenic plant was PCR-amplified and analyzed by RFLP (restriction fragment length polymorphism) and genotyped by the Genetic Analyzer. Mutant plants were eventually identified by sequencing the edited *AsDREB* gene. Selected mutant and non-edited "Crenshaw" plants were tested for their drought tolerance under the controlled growth chamber condition. The plants were visually rated and plant health data including weight, leaf color and the NDVI (normalized difference vegetation index) were collected over a 5-day drought period. Some *AsDREB*-gene edited mutant "Crenshaw" plants were shown to be less stressed compared to the non-gene edited plants. Our developed CRISPR-gene editing platform can be applied to other turfgrass species and other agronomically important traits.

A High-Density Genetic Linkage Map and Quantitative Trait Locus Analysis for Salinity Tolerance and Dollar Spot Resistance in Perennial Ryegrass (*Lolium perenne* L.)

Phillip L. Vines, Josh A. Honig, Jennifer Vaiciunas, Christine Kubik, B. Shaun Bushman, Eric N. Weibel, Bruce B. Clarke, William A. Meyer, and Stacy A. Bonos

Department of Plant Biology, Rutgers University

Improved stress tolerance in widely used turfgrass species is an essential component for a sustainable turfgrass industry. The aim of this study was to develop molecular breeding tools to help improve salinity tolerance and dollar spot resistance in perennial ryegrass (*Lolium perenne* L.), a turfgrass species that is grown on every continent except Antarctica. In mild climates, perennial ryegrass is managed as a turfgrass in parks, golf course fairways and roughs, athletic fields, racetracks, and general landscaping areas. In lower latitudes, the species is used to overseed warm-season turf during periods of winter dormancy.

Salinity stress can be caused a number of ways including use of ice melting salts to clear roadways, saltwater intrusion in groundwater, applications of fertilizer and other soil amendments, among others. However, one of the biggest causes of salinity stress in turfgrass is the use of effluent irrigation water in semi-arid regions that do not receive adequate rainfall to flush the salt through the soil and/or in areas where potable water use is restricted. Thus, the development of salinity tolerant turfgrass cultivars will be paramount as water use limitations and restrictions continue to be issued for the management of turfgrass.

Dollar spot, caused by *Clarireedia jacksonii*, is a prevalent and persistent foliar patch disease of many warm-and cool-season turfgrass species. The disease occurs throughout the world and is of great economic importance; in fact, more money is spent by U.S. golf courses to manage dollar spot than any other turfgrass disease. Current recommendations for dollar spot prevention and control include various cultural management practices and strategic chemical applications. The use of a resistant turfgrass cultivar is among the most viable and promising disease control options.

In consideration of the aforementioned concerns, the objectives of this study were (i) to construct a high-density genetic linkage map for perennial ryegrass using a genotyping by sequencing approach, (ii) to conduct a quantitative trait locus analysis for salinity tolerance, and (iii) to conduct a quantitative trait locus analysis for dollar spot resistance in perennial ryegrass.

For the first objective, the goal was to use next-generation sequencing technology to identify single nucleotide polymorphism (SNP) DNA markers and construct a genetic linkage map with the 'I06 × A89' pseudo F$_2$ perennial ryegrass mapping population. Genomic DNA was extracted from young leaf tissue of both parents and all 118 progeny of the mapping population and used for preparation of DNA sequence libraries. Illumina MiSeq and HiSeq 2500 next-generation sequencing platforms were used to generate DNA sequences from the prepared libraries. A linkage mapping analysis framework, consisting of a series of software packages and programs, was used to identify SNP markers, conduct a linkage analysis, and visualize genetic linkage maps for our mapping population. In total, 1,506 SNP markers were mapped across 1,221.1 centiMorgan total map length. As expected, seven linkage groups were detected, which represents the base chromosome number of perennial ryegrass. The average length of the linkage groups was 215.1

centiMorgans, the average number of markers per linkage group was 174.4, and the average marker density was 1.2 SNPs per centiMorgan.

For the second objective, the goal was to collect phenotypic data for salinity tolerance and use that data, along with genotypic data, to perform a quantitative trait locus analysis to identify and map SNP markers associated with salinity tolerance. The mapping population was phenotyped for salinity tolerance in three field trials and one greenhouse trial. A series of software packages and programs were used for quantitative trait locus analysis using the combined phenotypic data for salinity tolerance and genotypic linkage analysis data. To date, a preliminary quantitative trait locus analysis for salinity tolerance has been completed using the 2017 field trial phenotypic data along with the unanchored genetic linkage map. There were twelve quantitative trait loci identified across four linkage groups in the salinity tolerant parent background and two quantitative trait loci identified across two linkage groups in the salinity susceptible parent background

For the third objective, the goal was to collect phenotypic data for dollar spot resistance and use that data, along with genotypic data, to perform a quantitative trait locus analysis to identify and map SNP markers associated with dollar spot resistance. The mapping population was established in a mowed spaced-plant trial and phenotyped for dollar spot resistance during 2017 and 2018. To date, a preliminary quantitative trait locus analysis for dollar spot resistance has been completed using the 2017 and 2018 phenotypic data along with the unanchored genetic linkage map. There were five quantitative trait loci identified across three linkage groups in the dollar spot susceptible background and six quantitative trait loci identified across three linkage groups in the dollar spot resistant parent background.

Currently, we are working to anchor the SNP-based genetic linkage map we have constructed with microsatellite DNA markers from previously published studies; this will allow for linkage group assignments. Additionally, phenotypic data for salinity tolerance and dollar spot resistance will be reanalyzed with the anchored genotypic linkage mapping data to complete the quantitative trait locus analyses. Findings from this study will have both basic and applied implications. Turfgrass breeders and various other researchers will benefit from the basic knowledge of the genetics associated with salinity tolerance and dollar spot resistance in perennial ryegrass, and these findings will likely provide a scientific foundation for subsequent research. Ultimately, however, this work will contribute to the development of improved turfgrass cultivars, which will improve sustainability of the turfgrass industry.

Enhancing Cool-Season Turfgrass Germplasm by Collecting in the Centers of Origin

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The true 'center of origin' for cool-season turfgrass species ranges from central Europe through south-central Mediterranean Europe to the mountainous region of northern Africa depending on the species (Beard, 2013). Since 1996, the Rutgers University Turfgrass Breeding Project and Mr. Pieter den Haan have collaborated to collect and increase cool-season grasses in Europe. All the collections were vegetatively propagated into 5 clonal replicates in the fall in Steenburgen, The Netherlands. Over the past 22 years, we have cooperated to establish over 45,000 genetic lines. These large plantings are evaluated each May and after selection over 17,000 germplasm collections have been received in August as processed seed since 1996. These collections are backcrossed into the adapted germplasm present in the Rutgers turfgrass breeding program.

Over 3500 Kentucky bluegrasses (*Poa pratensis L.)* were received in New Jersey and seeded into turf trials. Approximately 7% of these Kentucky bluegrasses were selected for further increase in our nurseries. All of these collections were classified by their DNA using SSR markers as Eurasian types. We have also found collections from old US turf areas in the same group including Eagleton and Kenblue Kentucky bluegrass. Some of the bluegrass collections from Europe have had resistances to powdery mildew *Blumeria graminis* and rust (*Puccinia* spp.) and have been used in crosses with Rutgers developed varieties.

In the 2016 National Turfgrass Evaluation Program (NTEP) test of perennial ryegrasses (*Lolium perenne* L.), 29 of these entries entered by collaborators of Rutgers were sourced from 28-80 percent in their germplasm from European collections. Fifty percent of the useful sources of gray leaf spot (caused by *Pyricularia grisea*) resistance in perennial ryegrsses were from European selections.

Ten entries sourced from Rutgers in the 2018 NTEP tall fescue (*Schedonorus arundinaceus* (Schreb) Dumort) test were derived from European collections and contained anywhere from 10-38 percent European germplasm.

A large number of fine fescues including Chewings *(Festuca rubra L.* Supsp *fallax* (Thuill.) Nyman)*,* hard fescue (*F. brevilipa* R. Tracey) strong creeping red (*F. rubra* L. *rubra),* and slender creeping fescue (*F rubra* L. var. *littoralis* Vasey ex Beal) have been received and evaluated. Many of these fine fescues were associated with fungal endophytes along with many of the perennial ryegrasses and tall fescues. About 7% of the collected fine fescues have been used in the crosses with Rutgers advanced varieties.

In the large European collections of creeping bentgrass (*A gratis stolani* Fera L.), some of the lines from islands off of Spain have shown improved turf performance and disease resistance. Some of the colonial bentgrasses (*A. tenuis* L.) have shown improved lateral growth. The velvet bentgrasses from Europe are inferior to the collections made from US putting greens and fairways.

We feel confident that there has been great value to the turfgrass breeding program from the 22 years of collecting in Europe. It is obvious that the new collections have broadened the germplasm base at Rutgers.

References

Beard, J. 2013. Origins of north American turfgrasses. *In*: J.C. Stier, B.P Horgan, S.A. Bonos (eds) Turfgrass: biology, use and management. Agron. Monogr. 56. ASA, CSSA, and SSSA, Madison WI, pp 1-35.

Multidrug Resistance Conferred by a Xenobiotic Detoxification Through Transcriptional Regulation in Filamentous Fungi

Geunhwa Jung

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Fungi are known to utilize transcriptional regulation of genes that encode efflux transporters to detoxify xenobiotics leading to multidrug resistance, however to date it is unknown how fungi transcriptionally regulate and coordinate different phases (Phase I modification, Phase II conjugation, and Phase III secretion) of this detoxification system. In my talk I discuss evidence of an evolutionarily convergence between fungal and mammalian lineages, whereby all of xenobiotic detoxification genes, Phase I cytochrome P450 enzymes (CYP450s) and Phase III efflux transporters, are regulated by structurally unrelated transcription factors. Following RNA-seq analyses of a filamentous fungus, *Sclerotinia homoeocarpa*, the causal agent of dollar spot on turfgrasses, multidrug resistant (MDR) field strains were found to overexpress two sets of genes, CYP450s and ATP-binding cassette (ABC) efflux transporters for xenobiotic detoxification leading to multidrug resistance. Furthermore, a gain-of-function mutation of fungal-specific transcription factor (*S. homoeocarpa* xenobiotic drug regulator, ShXDR1), which was confirmed by a genetic transformation, is responsible for constitutive and induced overexpression of the Phase I and III genes. This is a first reported transcription factor involved in the xenobiotic detoxification. This fungal pathogen detoxifies xenobiotics through coordinated transcriptional control of CYP450s, biotransforming xenobiotics with different substrate specificity and ABC-transporters, excreting a broad spectrum of xenobiotics or biotransformed metabolites. A *Botrytis cinerea* strain, harboring the mutated ShXDR1, increased expression of Phase I and III genes, resulting in multidrug resistance. Furthermore, sequences of ShXDR1 are highly conserved among filamentous fungi which indicate the regulatory system is conserved in filamentous fungi. Better understanding of molecular mechanisms for xenobiotic detoxification in fungi holds potential for facilitating discovery of new antifungal drugs and development of resistance management strategies, and further studies of convergent and divergent evolution of xenobiotic detoxification in eukaryote lineages.

Diagnosis and Management of Insecticide Resistance in Annual Bluegrass Weevil

Albrecht M. Koppenhöfer[1], Olga S. Kostromytska[1,2], and Shaohui Wu[1,3],

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The annual bluegrass weevil (ABW), *Listronotus maculicollis*, is a serious and expanding pest of golf course turf in the Northeast and Mid-Atlantic regions of the United States and southeastern Canada. Its larvae can cause severe damage to tees, fairways, collars, and greens. This pest is one of the most difficult-to-manage turfgrass insect pests in North America due to the presence of several generations per year and concurrent presence of multiple life stages with increasing asynchrony during the growing season.

Synthetic insecticides have been the most commonly used tool in the management of ABW. Pyrethroids started to be widely used in the 1980s, because they can very effectively control ABW adults, thereby preventing oviposition and subsequent damage. Due to excessive pyrethroid use, by the 2000s many courses started to observe a decrease in the efficacy of this strategy. Pyrethroid resistance was first reported from southern New England. In a recently conducted survey throughout the area affected by the pest, 19% of participating golf courses suspected to have insecticide-resistant ABW populations, in the areas around the epicenter of the pest even higher rates (e.g., 28% New Jersey, 48% Connecticut, 55% Long Island, NY).

With growing awareness of pyrethroid-resistance issues, many superintendents have recently preferred to use the organophosphate chlorpyrifos for adult control. However, this organophosphate insecticide is less effective and troublesome from a toxicological and environmental standpoint. All other alternatives are larvicides including the diamides chlorantraniliprole (Acelepryn) and cyantraniliprole (Ference), the spinosyn spinosad (Conserve, MatchPoint), the oxadiazine indoxacarb (Provaunt), and the organophosphate trichlorfon (Dylox). However, according to field studies the efficacy of most insecticides against pyrethroid-resistant ABW populations seems to be reduced. This is not surprising, as pyrethroid-resistance in ABW seems to be at least in part due to enhanced enzymatic detoxification, a rather non-specific mechanism that breaks down active ingredients before they can reach their target sites in the organism.

We continue to work on the development of more sustainable ABW management practices such as plant resistance, biological control, and improved ABW monitoring. Meanwhile, careful measures need to be taken to prevent the development of resistance to synthetic insecticides in new populations and to new chemistries and to effectively manage already resistant populations.

Cross-resistance patterns to insecticides of several chemical classes among ABW populations with different levels of resistance to pyrethroids

We evaluated the degree and scope of ABW resistance, determined existing cross-resistance patterns, and confirmed laboratory findings under greenhouse conditions. The susceptibility of 10 ABW populations to insecticides of different chemical classes was assessed in topical, feeding and greenhouse assays. The level of susceptibility to pyrethroids varied significantly among

populations (LD_{50}s ranging 2.4 – 819.1 ng per insect for bifenthrin and 1.1 – 362.7 ng for λ-cyhalothrin in the topical assay). Three populations were relatively susceptible to pyrethroids, and seven populations had moderate to high resistance levels (resistance ratio RR_{50} for bifenthrin ranging 30.5 – 343.1). The toxicity of chlorpyrifos (RR_{50}s ranging 3.3 – 15.3), spinosad (RR_{50}s 2.4 – 7.7), the neonicotinoid clothianidin (RR_{50}s 4.2 – 9.7) and indoxacarb (RR_{50}s 2.8 – 9.7) was decreased for the pyrethroid-resistant populations. Toxicity data for bifenthrin and chlorpyrifos obtained under more realistic greenhouse conditions confirmed laboratory observations, indicating that the topical assay is an accurate method of detection and measurement of resistance level. Our observations expanded the previously known geographic range of ABW pyrethroid resistance to include the NY metropolitan area, New Jersey, and eastern Pennsylvania and provided clear evidence of cross-resistance not only within the pyrethroid class, but also to several other chemical classes.

Diagnostic dose assays for the detection and monitoring of resistance in ABW adults

Optimal diagnostic tools for resistance detection are crucial for efficient resistance monitoring and mitigation. To determine resistance levels to pyrethroids and chlorpyrifos in adult ABW, we developed a glass vial test with technical grade (> 95% active ingredient [AI]) in acetone spread on the inner surfaces of the vial and a Petri dish assays with formulated product applied on filter paper. Both tests separated different resistance levels among weevil populations. With the pyrethroid bifenthrin, susceptible, moderately resistant (RR_{50}s 12.2 – 95.7) and highly resistant (RR_{50}s 258.2 – 1760.9) populations were distinguished. With the organophosphate chlorpyrifos, susceptible, tolerant (RR_{50}s 2.4 – 6.7) and resistant (RR_{50}s 8.8 – 120.7) populations were distinguished. In validation assays, several bifenthrin and chlorpyrifos concentrations were needed to separate resistance levels in Petri dish (bifenthrin: 112.2 and 336.3 or 3362.5 mg AI m-2; chlorpyrifos: 3.4 and 33.6 mg AI m-2) and vial (bifenthrin: 112.1 or 1120.8 mg AI/m^2; chlorpyrifos: 2.2 and 11.2 mg AI/m^2) assays. The Petri dish assay with formulated bifenthrin and chlorpyrifos was the best option for ABW adult resistance detection and monitoring. It demonstrated sufficient discriminating power, accurately reflected resistance levels and was easier to conduct than the vial test. A single diagnostic concentration sufficed to separate susceptible and resistant populations. To determine different resistance/tolerance levels, two to three concentrations were necessary.

Resistance mechanism

To determine involvement of enzymatic detoxification in ABW resistance to pyrethroids, combinations of synergists (oxidase inhibitor PBO, glutathione transferase inhibitor DEM, esterase inhibitor DEF) and bifenthrin or chlorpyrifos were tested in laboratory bioassays against adults from seven ABW populations. Bifenthrin toxicity was significantly increased in presence of PBO (8-20 fold) and DEF (9-39 fold) which indicates involvement of oxidase and esterase systems as possible resistance mechanisms. DEM had a weak effect on bifenthrin toxicity for most populations. Synergists did not significantly affect chlorpyrifos toxicity in our study. While our study suggests that enhanced enzymatic detoxification plays a major role in the resistance of ABW to pyrethroids and some other insecticide modes-of-action, the involvement of other mechanisms like target site insensitivity cannot be excluded.

Pyrethroid-resistance level affects performance of larvicides and adulticides from different insecticide classes in populations of ABW

We examined the effect of pyrethroid-resistance level on the efficacy of adulticides and larvicides from different insecticide classes commonly used for ABW management through greenhouse and field studies. The tested populations had previously been determined to be susceptible, moderately resistant, resistant, and highly resistant to the pyrethroid bifenthrin. Field experiments were conducted in fairway areas with a history of ABW infestation over two years. Targeting adults, efficacy was significantly reduced for bifenthrin against the highly resistant population and for the spinosyn spinosad and the oxadiazine indoxacarb against the resistant and highly resistant populations. Efficacy of the organophosphate chlorpyrifos was not significantly reduced. No adulticide provided significant control of resistant and highly resistant populations. Targeting larvae, the efficacy of spinosad and the anthranilic diamide cyantraniliprole were marginally reduced against the highly resistant population. Significant reductions in efficacy and no significant control were observed for indoxacarb against the highly resistant population and for the neonicotinoid clothianidin, the anthranilic diamide chlorantraniliprole, and the organophosphate trichlorfon against the resistant and highly resistant populations.

Management recommendations

The pattern of insecticide efficacy against ABW populations with different resistance levels may vary somewhat between different golf courses based on their specific history of insecticide use. Nonetheless, the findings in this study combined with previous observations show a robust pattern that can serve as the base for recommendations for ABW control at different pyrethroid-resistance levels.

In view of the serious resistance issues, the first recommendation for ABW management is to minimize synthetic insecticide applications both in space and time as much as possible, whether managing susceptible or resistant populations. This goal can be achieved more easily by following the second recommendation, which is to shift control measures more from management of adults to management of larvae. This allows for more informed decisions on the need for applications because early larvicides allow monitoring adults past their peak densities in spring and late larvicides allow monitoring of actual larval densities.

For ABW populations with bifenthrin RR_{50}s above 50, management efforts should concentrate on larvae using the remaining effective larvicides cyantraniliprole, spinosad, indoxacarb, and, if still effective, trichlorfon and chlorantraniliprole. Against populations with bifenthrin RR_{50}s above 100, biological, biorational, and cultural control alternatives in rotation with the remaining effective synthetic insecticides cyantraniliprole, spinosad, and, if still effective, indoxacarb should be used to delay further resistance development.

To help with the implementation of these management recommendations, we have developed a simple Petri dish test using formulated bifenthrin and chlorpyrifos (see above) that could be used by consultants and diagnostic laboratories to help golf courses determine the resistance level of their ABW populations and thereupon base their management activities.

Warm-season Turfgrass Breeding at Texas A&M

Ambika Chandra

Texas A&M AgriLife Research

As an agricultural commodity, turfgrass impacts the lives of millions of people in many different ways including their physical and mental health, and social well-being. However, recent recurrent drought and increased demand on fresh water resources for human consumption means less potable water will be available for landscape irrigation while extreme weather patterns and temperature fluctuations are resulting in turfgrass winter-kill or heat-stress, especially in the transition zone. The turfgrass breeding program at the Texas AgriLife Research - Dallas focuses on developing resource efficient and stress tolerant cultivars of turfgrass species requiring lower inputs of water, pesticides, and fertilizer. We use conventional plant breeding approaches along with molecular biology tools for the genetic improvement of major warm-season turfgrass species for sustainable turfgrass production. The history of breeding, performance, and use of St. Augustinegrass and zoysiagrass will be presented with emphasis on research initiatives to improve drought tolerance, cold-hardiness and disease tolerance. Attendees will receive a comprehensive view of a warm-season grass breeding program working towards solving long-term problems challenging the economic and environmental viability of the southern turfgrass industry.

Effects of Finer-Textured Topdressing Sand on Creeping Bentgrass Putting Green Turf

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Sand topdressing of putting greens during the season is often avoided due to the potential of coarse sand particles interfering with play and dulling mower blades. This field trial determined the effects of eliminating coarse particles from topdressing sand (subsequently increasing the quantities of medium and fine particles) on the performance of creeping bentgrass maintained as putting green turf. The trial also assessed the ability of core cultivation and backfilling holes with medium-coarse sand to ameliorate the potential negative effects of finer-textured topdressing sands on turf performance and the physical properties at the surface of a putting green root zone.

The trial was initiated in May 2016 on a 19-month-old 'Shark' creeping bentgrass (*Agrostis stolonifera* L.) maintained at 2.8-mm on a sand-based root zone. A 3 x 2 x 2 factorially arranged randomized complete block design with four replications included the factors of sand size (medium-coarse, medium-fine, fine-medium), quantity of mid-season topdressing (244 and 488 g/m^2 every 10 to 14 days from June through October), and cultivation (non-cultivated or core cultivated plus backfill in May and October). Controls (no mid-season topdressing) at each level of cultivation were also included for comparisons resulting in 14 total treatments (Table 1). The medium-coarse sand met USGA recommendations for construction; whereas the fine sand content of the medium-fine and fine-medium sands exceeded 20% (by weight) and contained little to no coarse particles (Table 2).

Data collection during 2018 included: clippings to determine the quantity and particle size distribution of sand collected during mowing; falling-head infiltration rate; surface firmness (penetration depth); surface hardness (Clegg Soil Impact Tester, 0.5-kg and 2.25-kg missiles); volumetric water content (VWC) of the surface 0- to 38-mm and 0- to 76-mm depth zones; mat layer thickness, organic matter content (loss on ignition) and sand particle size distribution; visual rating of turf color, density and quality; and normalized difference vegetation index (NDVI).

Topdressing with medium-coarse sand increased the percentage of applied-sand collected during mowing compared to medium-fine and fine-medium sands. Additionally, the percentage of applied-sand collected in mower clippings was greater at 488 g/m^2 topdressing than the 244 g/m^2.

Water infiltration into the bentgrass surface was slowed by topdressing with sand that contained a greater amount of fine sand; however, core cultivation was capable of offsetting this effect and increased water infiltration.

Topdressing produced a firmer surface compared to the non-topdressed control at both levels of cultivation. Firmer surfaces were observed on plots that received the greatest rate of mid-season topdressing and were core cultivated. The Clegg Soil Impact Tester indicated that sand sizes only affected in surface hardness under non-cultivated conditions, where the softest surfaces were topdressed with the fine-medium sand.

Medium-fine and fine-medium sands increased the fineness of sand within the mat layer; however, surface wetness (VWC) frequently was greatest for only the fine-medium sand. Surface wetness

of the medium-fine plots was very similar to medium-coarse plots. Core cultivation was very effective at decreasing surface wetness of non-topdressed plots as well as plots topdressed with fine-medium sand. Plots that were core cultivated – and backfilled with medium-coarse sand – reduced the tendency to increase the fineness of sand in the mat layer of plots topdressed with sand containing more than 20% fine sand; however, the fine sand content was well above (9-10% more than) that of plots topdressed with medium-coarse sand.

Sand topdressing increased the mass-content of organic matter (kg/m^2) in the mat layer while the organic matter concentration (% by weight) was decreased (diluted) by topdressing sand. The 100 lbs. topdressing rate increased the mass-content and decreased the concentration of organic matter more than the 50 lbs. topdressing rate. Core cultivation reduced both the mass-content and concentration of organic matter in the mat layer.

Core cultivation reduced the normalized difference vegetation index (NDVI) values of the creeping bentgrass turf compared to non-cored plots throughout 2018. Similarly, plots topdressed at 488 g/m^2 had lower NDVI values than plots topdressed at 244 g/m^2. Thus, treatment combination that were more aggressive in managing thatch accumulation also lowered vegetative cover.

Data will be collected from this trial for a fourth growing season during 2019.

Table 1. Individual treatment combinations of sand size, topdressing rate, and cultivation as well as two controls (no mid-season topdressing) being evaluated on a 'Shark' creeping bentgrass turf grown on a sand-based root zone.

		Factors in the Experiment		
Treatment No.	Sand Size[†]	Topdressing Rate during Mid- Season[‡]	Cultivation[¶]	Annual Quantity of Sand Applied
		g/m^2		g/m^2
1	Medium-coarse	244	Non-cored	6,350
2	Medium-coarse	244	Core + Backfill	8,300
3	Medium-coarse	488	Non-cored	8,790
4	Medium-coarse	488	Core + Backfill	10,740
5	Medium-fine	244	Non-cored	6,350
6	Medium-fine	244	Core + Backfill	8,300
7	Medium-fine	488	Non-cored	8,790
8	Medium-fine	488	Core + Backfill	10,740
9	Fine-medium	244	Non-cored	6,350
10	Fine-medium	244	Core + Backfill	8,300
11	Fine-medium	488	Non-cored	8,790
12	Fine-medium	488	Core + Backfill	10,740
13	None	0	Non-cored	0
14	None	0	Core + Backfill	5,860

† First-mentioned size class represent the predominant size fraction in the sand.

‡ Topdressing applied every two weeks from 10 June through 12 October (10 applications). Topdressing at 244 g/m^2 represented a 'dusting' quantity (O'Brien and Hartwiger, 2003); whereas, topdressing at 488 g/m^2 filled the surface thatch and lower verdure layers.

¶ Core cultivation to the 40-mm depth was performed twice a year (10 May and 2 November) using 13-mm diameter hollow tines spaced to remove 10% of the surface area annually. Coring holes were backfilled with 2,930 g/m^2 of medium-coarse sand. At the time of coring, non-cultivated plots were topdressed with the respective sand size at 1,950 g/m^2 to fill the surface thatch and verdure layers to the same extent as plots that were cored and backfilled.

Table 2. Sand size distributions of the three topdressing sizes, and the mat layer and underlying root zone at the initiation of the trial; USGA construction specification provided for reference.

Topdressing Sand Size	Particle Diameter (mm)/Size Class				
	2.0-1.0	1.0-0.5	0.5-0.25	0.25-0.15	0.15-0.05
	Very Coarse	Coarse	Medium	Fine	Very Fine
	---------------------------- % retained (by weight) ----------------------------				
Medium-coarse	0	33.8	57.7	8.4	0.1
Medium-fine	0	0.1	76.7	22.7	0.5
Fine-medium	0	5.7	25.8	66.8	1.7
Profile Layers					
Mat Layer†	0.1	25.3	56.4	15.4	2.7
Rootzone	6.9	25.3	44.6	17.2	4.1
USGA construction specification	≤10	≥60		≤20	≤5

† Size distribution of sand in 45 core samples of the mat layer collected before the initiation of treatments in May 2016.

Metagenomic Analysis Reveals Diversity and Effects of Bacterial Endophyte Communities Associated with Turf Seeds

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Endophytes have been shown to enhance field performance of turf grasses by improving germination rates, promoting plant growth, and increasing resistance of grasses to environmental stresses. Similar benefits to grass hosts have been shown for both fungal and bacterial endophytes. In previous studies, it has been shown that optimal seed germination and seedling growth in grass seedlings occurs when seeds carry a small diverse community of endophytic bacteria (Verma et al., 2017; Verma and White, 2018). Some of the bacteria (alpha-Proteobacteria and Sphingobacteria) in seeds may enter into the cytoplasm of eukaryote cells (Ettema and Anderson, 2009; Thomas et al., 2010), and as a consequence are likely carried in the interior of seeds within the embryonic cells; other bacteria (e.g., gamma-Proteobacteria, beta-Proteobacteria, Actinobacteria and Firmicutes) have been found to be carried on seed surfaces in bract tissues that adhere to seeds (White et al., 2018). Optimal seed germination and seedling development only occurs when seeds carry the full compliment of bacteria vectored within the seed, and those carried on seed surfaces (Verma and White, 2018). Removal of seed surface vectored bacteria results in seedlings that germinate poorly and seedlings that show diminished development in terms of root growth and function (Verma et al., 2017; White et al., 2018). Very little is known about natural processes that affect the development of the seed bacterial endophyte community. It has been observed that seed germination tends to be better in grasses from climates where seed is exposed to moisture during the seed maturation process. We hypothesized that dry climate restricts the growth of the seed-surface bacterial community, and that absence of the surface bacterial community results in poor seed germination and diminished seedling development. We employed metagenomic analysis and experiments using seeds produced in areas with differing levels of moisture to test this hypothesis. Seed samples were classified as either high moisture or low moisture climates during the seed maturation period. Seed samples were then subjected to metagenomic analysis to examine the diversity of bacteria associated with seeds. We conducted the study by sequencing the V3-V4 region of 16S rDNA sequences with Illumina MiSeq. By comparing the bacterial community in several grass genera, we found that *Lolium* (including *Lolium arundinaceum*; average of 20851 reads/sample) possessed more than twice the reads of fine fescues (average of 9211 reads/sample). Also, *Lolium* had a higher proportion of alpha-Proteobacteria, Actinobacteria, but a lower proportion of Firmicutes*,* beta-Proteobacteria and gamma-Proteobacteria. The results showed that seed from moist climates tended to show a higher diversity of bacteria associated with seeds. In our seed germination tests, alpha-Proteobacteria and Sphingobacteria abundances (with lower abundance of other bacterial groups) were positively associated with germination times and negatively associated with germination rates. Gamma-Proteobacteria abundance showed a beneficial effect with a positive correlation with germination rate and a negative correlation with germination time. To further examine these correlations, we conducted analysis at genus level. Seeds bearing a high percentage of *Paenibacillus, Erwinia,* and *Pseudomonas* showed higher germination rates and shorter germination times. In this analysis, abundance of several genera in alpha-Proteobacteria and Sphingobacteria (and lower abundance of other bacterial groups) was again negatively associated with germination rates and positively

with germination times; these genera included *Agrobacterium, Methylobacterium,* and *Pedobacter*. The genus-level results matched with the class-level results. On average, alpha-Proteobacteria, gamma-Proteobacteria, Actinobacteria, and beta-Proteobacteria took 26.91%, 21.12%, 14.84% and 12.33% of the total bacteria in communities associated with the seeds. This overall correlation could explain why *Lolium* seeds tended to germinate better than fine fescues in our experimental assays. The results of the metagenomic study and experiments support the hypothesis that seeds produced in dry regions lack a well-developed community of bacteria on seed surfaces, and this results in poor germination and seeding growth. Further, this study suggests that germination and growth of grass seedlings produced in dry regions could be improved by seed treatments to increase presence of gamma-Proteobacteria, beta-Proteobacteria, Firmicutes, and Actinobacteria that vector on seed surfaces.

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Goosegrass Emergence and Control of Suspected Herbicide Resistant Populations

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Goosegrass (*Eleusine indica*) is a problematic warm-season annual grassy weed of cool-season turfgrass. This presentation will discuss the results of two projects designed to improve our understanding of goosegrass emergence and effective options for pre-emergence weed control.

The objective of the first project was to determine goosegrass seedling emergence patterns in New Jersey turfgrass and develop a temperature-based model to predict seasonal emergence. Experiments conducted in 2017 and 2018 at the Rutgers Horticultural Research Farm No. 2 in North Brunswick, NJ. Off-site experiments were conducted on golf courses in East Brunswick, NJ in 2017 and Manalapan, NJ in 2018. Goosegrass seedlings were counted and removed approximately weekly from April through October within fixed circles, replicated four times for each treatment and location. Treatments at the Rutgers site consisted of three different ground cover treatments; bare ground, perennial ryegrass (PRG; *Lolium perenne*) mowed at 1.3 cm, and PRG mowed at 6.4 cm. Fixed circles were sized 1000 cm² within each 1.0 by 1.0 m plot. At the golf course locations plots were maintained as a golf course fairway at a 1.25 cm mowing height and fixed circles were sized 500 cm² within each 1.0 by 1.0 m plot. Soil temperature was monitored at a 5.0 cm depth at both locations and used to calculate growing degree-days. Soil volumetric water content was measured at the Rutgers site. The total number of seedlings that emerged at each site and the number of days during which seedlings emerged were determined at all sites. These data from the research farm site were subjected to ANOVA in SAS (v 9.4) to determine the effect of ground cover treatments. The percent cumulative emergence was calculated for each location and subjected to non-linear regression using a Weibull function in Prism (v 7.0). Cumulative emergence was regressed over day of year and growing degree-days.
In 2017 and 2018, more seedlings emerged in bare soil than in PRG plots maintained 1.3 or 6.4 cm. The emergence duration was longer in bare soil plots compared to PRG plots in both years. For individual sites, the Weibull model described the data very well (R^2 >0.9). However, growing degree-day models did not predict goosegrass emergence better than day-of-year. Emergence patterns were variable across sites and years and these differences were not explained by soil temperature.

A second set of experiments was initiated to evaluate various pre-emergence herbicide programs for goosegrass control. In 2017, preliminary research conducted on a golf course in East Brunswick, NJ demonstrated that pre-emergence herbicides prodiamine and dithiopyr provided poor goosegrass control, while oxadiazon provided excellent control. The lack of goosegrass control provided by dithiopyr and prodiamine was unexpected and, thus, warranted a more thorough investigation. In 2018 we initiated two field experiments to investigate the efficacy of various pre-emergence herbicide programs for goosegrass control. One experiment was initiated at the Rutgers Hort. Farm No. 2 and the other was initiated on a golf course fairway in Manalapan, NJ. Treatments consisted of single or sequential applications of prodiamine (0.36 kg ha⁻¹), dithiopyr (0.28 to 0.56 kg ha⁻¹), or oxadiazon (2.2 to 3.4 kg ha⁻¹). Both experiments were conducted using methods typical of replicated small plot field research. The initial applications were made on 1 and 2 May at the golf course and research farm, respectively. Sequential

applications were made on 20 and 19 June at the golf course and research farm, respectively. Goosegrass control was visually estimated on a 0 (no control) to 100 (complete control) percent scale using the non-treated control for comparison.

At the research farm site, single and sequential oxadiazon treatments controlled goosegrass ≥85% while prodiamine and dithiopyr programs provided ≤65% control. At the golf course location oxadiazon treatments controlled goosegrass ≥90% while prodiamine and dithiopyr programs provided <10% control. Sequential applications of oxadiazon controlled goosegrass more than single applications at the golf course location. Dithiopyr and prodiamine less goosegrass control at t the golf course compared to the research farm, which suggests the goosegrass population at the golf course is resistant to mitotic-inhibiting herbicides. Future glasshouse research will determine if the golf course populations evaluated in 2017 and 2018 experiments are resistant to mitotic-inhibiting herbicides.

Not Your Grandfather's Dollar Spot:
Resolving the Taxonomy of this Important Turfgrass Disease

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Dollar spot is one of the most common and widespread fungal diseases of cool- and warm-season turfgrass species. Yet, the true identity of the causal agent has remained a mystery until recently. The disease was first reported in 1927 by the USDA scientist Dr. John Monteith, however, Monteith did not provide a valid Latin binomial for the causal agent [1]. In 1937, F.T. Bennett examined the reproductive structures of the fungus and concluded that the structure from which the sporophores arose resembled aggregates of microsclerotia, and thus formally named the pathogen as *Sclerotinia homoeocarpa* [2]. In 1945, the mycologist H.H. Whetzel reviewed the taxonomy of the Sclerotiniaceae family and restricted the genus *Sclerotinia* to include only fungi that produce apothecia from tuberoid sclerotia [3], a characteristic not found in the dollar spot pathogen. Whetzel suggested that the dollar spot fungus may be more accurately placed within the genus *Rutstroemia*, though he never formally reclassified the pathogen.

For the next seventy years, the taxonomic placement of the causal agent of dollar spot disease would be the subject of much debate. Morphological reviews of the dollar spot pathogen continued throughout much the 1970s, with the genera *Lanzia*, *Lambertella*, *Moellerodiscus*, *Poculum*, or *Rutstroemia* being suggested as alternative genera [4,5]. However, due to limited observations of sexual structures to support taxonomic reclassification, the name remained *S. homoeocarpa*. With the arrival of DNA-based technologies, the dollar spot pathogen was subjected to a number of genetic analyses in the 1990s. Molecular analysis of the rDNA internal transcribed spacer (ITS) region confirmed that the dollar spot pathogen was not a true member of the genus *Sclerotinia*, however, due to genetic variation within the ITS region, small sample size, and limited outgroup species represented, taxonomic placement for this fungus was still not clear [6,7].

In recent years, scientists from the USDA Agricultural Research Service, Rutgers University, North Carolina State University, and the Ohio State University began a new collaborative effort to reclassify the dollar spot pathogen [8,9]. In this study, sixty-seven fungal isolates, representing members of both the Rutstroemiaceae (*Lambertella*, *Rutstroemia*, *Lanzia*) and Sclerotiniaceae (*Ciboria*, *Monilinia*, *Sclerotinia*) were analyzed. Representative dollar spot isolates from both warm- and cool-season grass hosts were selected from an existing collection of ~1100 dollar spot isolates collected from around the world. DNA was extracted and three genetic regions— calmodulin, Mcm7, and ITS (~1800 base pairs) were PCR amplified and sequenced from all

isolates. Sequence data was subjected to phylogenetic analysis. Individual gene trees, as well as a combined phylogeny that included all three molecular markers, were generated using maximum likelihood and Bayesian approaches. Clades were considered well-supported with posterior probabilities (pp) ≥ 0.95 and bootstrap ≥70%. In addition to molecular analyses, a subset of dollar spot isolates were evaluated for their ability to produce apothecia *in vitro* by performing crosses of isolates with different mating types, however, all apothecia produced were sterile.

Both individual gene trees and the combined phylogeny produced tree topologies consistent with one another. Members of the Sclerotiniaceae family (*Botrytis*, *Ciboria*, *Sclerotinia*) formed their own monophyletic group (pp = 1, bootstrap = 100), separate from members of the Rutstroemiaceae family, which formed their own well-supported clade (pp = 1, bootstrap = 100). Within the Rutstroemiaceae clade, dollar spot isolates clustered within their own group (pp = 1, bootstrap = 73%), distinct from all other isolates within the Rutstroemiaceae family. As a result, a new genus, termed *Clarireedia*, was established to accommodate these fungi. The new genus name recognizes Dr. C. Reed Funk, influential turfgrass breeder, for his efforts to develop resistance to dollar spot disease in cool-season turfgrasses ("Clarus" is Latin for famous; 'reedia' is in honor of Dr. C. Reed Funk). Within the genus *Clarireedia*, four species are now named: *C. homoeocarpa*, *C. bennettii*, *C. jacksonii,* and *C. monteithiana*.

Clarireedia homoeocarpa represents the type species for the genus and has only been found on *Festuca rubra* hosts in the United Kingdom. *Clarireedia bennettii* (named for F.T. Bennett), has been found primarily on cool-season grass hosts in the Netherlands, United Kingdom, and United States. *Clarireedia jacksonii*, named in honor of Dr. Noel Jackson in recognition of his work on dollar spot in the 1970s, is distributed worldwide on cool-season grass hosts. *Clarireedia monteithiana,* named for Dr. John Monteith who first reported dollar spot disease, is also globally distributed and found on warm-season grass hosts. Together, *C. jacksonii* and *C. monteithiana* appear to be responsible for most modern-day outbreaks of dollar spot disease observed around the world.

The resolved taxonomy of the fungi responsible for dollar spot disease marks an important turning point for scientific research within these fungi [8,9]. The data presented here reveals considerable diversity within the genus, however, it is not yet known how this diversity relates to disease development. Regardless, with resolved taxonomy in place, researchers can now begin to examine the individual species of dollar spot fungi and determine the best methods for control and management of this economically important disease.

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Understanding the Microbiome Associated with Turfgrasses

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Microbiomes, the microbial community and their genomes in a system, play important roles in adaptation, stress tolerance and disease resistance of their associated plants. The next-generation sequencing (NGS) technologies enable a holistic view of the microbial diversity compared to the culture-based methods that only capture a fraction of the total diversity. The long-term goal of the project is to describe the microbiomes associated with turfgrasses, understand the causality of microbiomes and the plant health/performance, and to translate this knowledge to develop new sustainable management strategies and other applications.

Turfgrass microbiome research is still in its infancy but has the potential to help solve some of the largest problems in turfgrass management. There is evidence that microorganisms can alleviate abiotic stresses like drought stress and salinity stress. Certain microorganisms have also demonstrated an ability to suppress diseases like dollar spot caused by *Clarireedia jacksonii*. Identifying these beneficial microbes using metagenomics is the first step into manipulating microbiomes to produce desirable phenotypes in the field. The second step will be a functional analysis of the metatranscriptome to decipher which microbial proteins are associated with the enhanced phenotypes. These different analyses will lay the framework for which members of a microbiome should be selected for depending on the desired phenotype. Eventually, scientists will be able to tailor microbiome communities to enhance plant resistance to abiotic and biotic stresses, reducing the quantity of inputs that are needed to manage a turfgrass system.

One of our current projects is evaluating the microbiome associated with tall fescue genotypes grown in a rainout shelter at the Rutgers Plant Science Research and Extension Farm in Adelphia, NJ after prolonged periods of drought stress. Twelve plants were selected for analysis, comprised of six sets of siblings, one exhibiting a drought tolerant phenotype and the other a susceptible phenotype. The microbiome associated with the shoot, root, and rhizosphere soil was evaluated for each tall fescue half-sib pair. Microbiome analysis was preformed utilizing an Illumina NGS metabarcoding approach that sequenced the 16S and ITS barcoding region to determine the composition of the bacterial and fungal communities, respectively. Comparative analysis of the microbiomes associated with different tissue types showed clear differences. The two phenotypes showed no significant differences in microbiome composition but certain microbial species differed in abundance. For example, two common root associated Helotiales fungi *Rhexocercosporidium* and *Cadophora* were found to have higher abundances in the drought susceptible phenotypes. To see if there are any significant differences in microbial functions, the metatranscriptome of the same set of siblings is currently being evaluated.

Another project is looking at changes in the microbiome before and after dollar spot symptoms develop for a susceptible and tolerant cultivar maintained at fairway height. Dollar spot is caused by fungal species in the genus *Clarireedia* and is an economically important disease. Understanding what constitutes a healthy microbiome versus a diseased microbiome for this disease may lead to the development of better biocontrols and cultural practices.

What We Know So Far About the *Epichloë festucae* Antifungal Protein

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Many grasses in their natural environment are associated with fungal endophytes from the genus *Epichloë*. Some *Epichloë* spp. have been confirmed to confer enhanced resistance to abiotic and biotic stresses to the plant hosts. Endophyte-mediated resistance to the fungal diseases red thread, caused by *Laetisaria fuciformis* (McAlp.) Burdsall*,* and dollar spot, caused by *Clarireedia jacksonii,* has been well documented in fine fescues infected with *E. festucae* (Bonos et al., 2005; Clarke et al., 2006). Such disease resistance is not a general effect of endophyte infection in other grass species/*Epichloë* spp. interactions. The underlying mechanism of the endophyte-mediated disease resistance has not yet been established.

Festuca rubra L. subsp. *rubra* (strong creeping red fescue) exhibits endophyte-mediated fungal disease resistance. *Epichloë festucae* Leuchtm., Schardl & M.R. Siegel, the fungal endophyte of *F. rubra* subsp. *rubra*, produces an abundant antifungal protein that is secreted into the apoplast of the infected plant (Ambrose and Belanger, 2012). The antifungal protein gene found in *E. festucae* infecting strong creeping red fescue is not present in most *Epichloë* genomes for which whole genome sequences are available (Ambrose and Belanger, 2012). We are therefore pursuing the possibility that the antifungal protein may be a component of the unique endophyte-mediated disease resistance observed in strong creeping red fescue.

We previously expressed the antifungal protein in the yeast *Pichia pastoris* and showed it did have activity against the dollar spot fungus (Tian et al., 2017). We are currently working on expression of the antifungal protein in bacteria since that would be a simpler method of obtaining large quantities of the protein. The antifungal protein was expressed in *E. coli* and had activity against the model test organism *Neurospora crassa*.

We have also generated a knock-out of the antifungal protein gene to determine its effect on disease susceptibility in strong creeping red fescue. The standard approach to generating knock-outs in *E. festucae* relies on homologous recombination of long DNA fragments, generally greater than 1000 base pairs, flanking the gene of interest. This approach was not successful for the *E. festucae* antifungal protein gene, likely because it resides in an unusual genomic region where the gene is flanked by repeated sequences. However, we successfully applied the new CRISPR-Cas9 technology to the non-model fungus *E. festucae*. The Cas9 endonuclease generated a double-strand break at the target site of the antifungal protein gene, and a mutation was introduced when repairing the break. To date, we have identified one mutant with a single base pair insertion at the Cas9 target site, which introduced an early stop codon, and two mutants with large insertions from the fragments of the transformation vector. Next, the knockout isolates will be reintroduced into strong creeping red fescue as a proof of concept to evaluate susceptibility of the host plant to dollar spot disease using an established greenhouse inoculation assay.

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POSTER PRESENTATIONS

Effects of Plant Health Products on Drought Tolerance and Post-Stress Recovery in Creeping Bentgrass

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Drought is a major abiotic stress that has been associated with decreases in leaf water relations and aesthetic qualities. Due to prolonged lack of rainfall, turfgrass managers may need to implement water-use restrictions by imposing deficit irrigation practices (irrigating to replace less than 100% water lost due to evapotranspiration), or in some extreme cases, may be forced to withhold irrigation altogether. Due to these conditions, dormancy or mortality of turfgrass can occur, and finding alternative methods for improving drought stress tolerance, as well as promoting rapid regrowth and post-stress recovery once irrigation can resume, is critical for maintaining sustainable turfgrass stands. The 2017 summer creeping bentgrass (*Agrostis stolonifera*) fairway study investigated the effectiveness of different plant health products for improving summer drought stress tolerance. The products tested included Fluazinam+Acibenzolar alone or in combination with Azoxystrobin+Acibenzolar, and/ or Trinexapac-ethyl, and each were evaluated under different levels of irrigation to determine their effects on drought stress tolerance (replacing 100%, 60%, or 0% water lost due to evapotranspiration). In addition, this study examined how these products could improve post-stress recovery upon re-watering to field capacity. The study found that under a moderate (60% ET replacement) or severe (complete water withholding) drought stress, all treatments had positive effects for improving drought tolerance in terms of turf quality, leaf relative water content, and green canopy density data. Upon re-watering, Fluazinam+Acibenzolar + Trinexapac-ethyl most notably promoted rapid recovery of turf from drought stress by improving upon leaf hydration status and green canopy density. Effective use of these plant health products can facilitate the maintenance of creeping bentgrass with deficit irrigation during periods of drought or water-use restrictions, and can strengthen turfgrass management programs by aiding in post-stress recovery when normal watering conditions are restored.

Poa pratensis Selection Dependent Responses to Mesotrione Under Controlled Conditions

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Mesotrione (2-(4-Mesyl-2-nitrobenzoyl)-1,3-cyclohexanedione) is a relatively new selective herbicide marketed as Tenacity (Syngetnta, 2001). Mesotrione is a 4-hydroxyphenylpyruvate dioxygenase (HPPD) enzyme inhibitor that prevents the production of carotenoids in susceptible plants resulting in a bleaching response. It can be applied pre- and post-emergent and effectively controls clover (*Trifolium sp.*), creeping bentgrass (*Agrostis sp.*) and annual bluegrass (*Poa annua*) in Kentucky bluegrass (*Poa pratensis*) turf stands. In this experiment, we assessed the responses of Kentucky bluegrass cultivars and selections to mesotrione herbicide at seeding in a growth chamber. All eighty-nine entries from the 2017 National Turfgrass Evaluation Program Kentucky bluegrass test plus one additional selection were evaluated for their responses to a high label rate (230 g ha^{-1}) treatment of mesotrione at the time of seeding and 31 days after seeding (DAS). Entries were seeded in 10.2 cm square pots filled with a fine silica sand and potting mix blend at a 50/50 ratio by volume. Utilizing randomized complete block design, each selection was present twice in each of the four blocks with one treated (water, mesotrione and a non-ionic surfactant) and one un-treated control (water and non-ionic surfactant). Applications were made using a track sprayer with water carrier at 410 L ha^{-1}. Digital images were captured using a light box at days 14, 18 and 22 DAS of the experiment for a total of 3 image sets for digital analysis. Percent green cover was determined using imageJ and the results were subjected to statistical analysis. After bleaching subsided, 55 DAS, biomass samples were taken, excluding the roots, to evaluate the difference in establishment from the treated control. Cultivar dependent responses to mesotrione were detected among Kentucky bluegrass entries in percent green cover but not in biomass. There were significant differences between runs, however, PPG-KB 1131 and Barvette HGT exhibited very little injury in both runs while Selway, A06-8 and PST-K13-143 were susceptible to the mesotrione application in both runs when all measurements were considered.

Annual Bluegrass Weevil (*Listronotus maculicollis*), Paclobutrazol and Interseeding Effects on Annual Bluegrass (*Poa annua)* in Creeping Bentgrass (*Agrostis stolonifera*) Fairways

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In the Northeastern US, the annual bluegrass weevil (ABW; *Listronotus maculicollis*) is a pest of annual bluegrass (*Poa annua*). The ABW showcases strong ovipositional preference to annual bluegrass and its larvae can cause substantial injury to the plant. We hypothesized that allowing ABW to injure annual bluegrass will control annual bluegrass in a creeping bentgrass (*Agrostis stolonifera*) fairway alone or in combination with paclobutrazol and creeping bentgrass interseeding.

Research was conducted in 2017 at the Rutgers Horticulture Farm No. 2 in North Brunswick, NJ on a simulated fairway that consisted of creeping bentgrass and annual bluegrass. Whole plots consisted of creeping bentgrass interseeding. Whole plots were split into three ABW control programs. The first program (preventative) consisted of an industry-standard insecticide program that controlled for larvae at all stages to prevent any turfgrass injury. The second program (threshold) consisted of a larvicide applied only after visual ratings determined that ABW damage resulted in unacceptable turfgrass quality. The third program (no-insecticide) was a non-treated control with no insecticides applied for ABW control. Insecticide programs were split with monthly applications of paclobutrazol (280 g ha^{-1}). Once the damage threshold was met, the appropriate treatments were interseeded with '007' creeping bentgrass.

Percent annual bluegrass cover was evaluated visually on a monthly basis. Grid counts were also taken at the start and end of each growing season. Turfgrass quality, annual bluegrass quality and creeping bentgrass quality were all evaluated monthly on a 1-9 scale, where 6 was considered acceptable. Lightbox photos were taken each month and subjected to digital image analysis to determine percent green cover (Turfanalyzer software). Data were subjected to ANOVA in SAS (v9.4) as a 2 by 2 factorial RCBD. (α=0.05). Fisher's Protected LSD (α=0.05) was used to separate means.

In 2017, the damage threshold was met on June 8th. The threshold and no-insecticide programs (250 larvae m^{-2}) reduced annual bluegrass quality compared to the preventative (7 larvae m^{-2}) in June only. Turfgrass quality of the threshold and no-insecticide programs did not differ from the preventative program from July to November. Bentgrass quality was not affected by insecticide program on any rating date. The no-insecticide program reduced annual bluegrass cover by 8% in July, but otherwise there were no differences between programs through November 2017. Regardless of ABW or interseeding treatment paclobutrazol reduced annual bluegrass cover by 67% as of November 2017.

In 2018, the damage threshold was met on July 1st. The no-insecticide and the preventative plots averaged 83 and 3 larvae m^{-2} , respectively. By September, the preventative program had the more

annual bluegrass cover (16.8%) than the threshold and no-insecticide programs (9.7% and 8.3% respectively). The threshold program reduced annual bluegrass cover in July and the no-insecticide program reduced annual bluegrass cover from June through September. The PGR program resulted in the annual bluegrass reduction both years. On September 2018, PGR treated plots averaged 5% cover and non-PGR treated plots averaged 18% annual bluegrass cover.

Physiological Effects of Endophytic Rhizobacteria on Creeping Bentgrass (*Agrostis stolonifera*) Tolerance to Heat and Drought Stress

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High temperature and drought stress are the primary abiotic stresses limiting the growth of cool-season grass species, such as creeping bentgrass (*Agrostis stolonifera*). Leaf senescence or chlorosis associated with summer bentgrass decline are symptomatic of heat and drought injury. Plant growth promoting rhizobacteria (PGPR) that produce ACC deaminase can help reduce the impacts of abiotic stress in plants. Four strains of endophytic *Burkholderia sp.* bacteria were isolated from the roots of native grasses from the New Jersey Pine Barrens and were found to possess ACC deaminase activity. Each individual strain, as well as a mixture of all four strains, were used to inoculate the roots of creeping bentgrass (cv. 'Penncross'). The plants were subjected to optimum conditions, heat stress, or drought stress treatments in growth chamber trials. Physiological parameters of inoculated plants were compared to non-inoculated controls by measuring turf quality (TQ), relative water content (RWC), leaf chlorophyll content (CHL), electrolyte leakage (EL), and photochemical efficiency (Fv/Fm) as indicators of leaf senescence. At the conclusion of the stress treatments, root morphological parameters (root volume, surface area, and diameter) were analyzed. While the plant response differed among individual PGPR strains, several strains were effective in improving physiological and morphological traits in creeping bentgrass under heat and drought stress conditions. Future studies focused on optimizing the bacteria/host/stress interaction and understanding the mechanisms involved in the bacteria-mediated stress tolerance response will facilitate the adaptation of PGPR inoculants for field application.

Anthracnose Response to Potassium Under Two Nitrogen Levels

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Anthracnose (caused by *Colletotrichum cereale* Manns sensu lato Crouch, Clarke and Hillman) is a prevalent disease of annual bluegrass [ABG; *Poa annua* L. f. *reptans* (Hausskn) T. Koyama] grown on golf course putting greens in the northeast region of the United States. Outbreaks of this fungal disease can occur any time of the year but are most destructive during warmer temperatures in the summer. A 2-yr field study assessed the response of anthracnose to potassium and nitrogen applications on ABG turf maintained at a 2.8-mm on a sandy loam in North Brunswick, NJ. A randomized complete block design with four replications was used to evaluate four potassium rates (0, 4, 8 and 16 kg K_2O ha^{-1}) applied as a potassium sulfate solution every two weeks under two levels of N. Nitrogen was applied as a urea solution at 4.9 kg ha^{-1} every 28 days (29 kg N ha^{-1} yr^{-1}) or seven days (117 kg N ha^{-1} yr^{-1}) for a total of 24 weeks beginning 28 and 13 April in 2016 and 2017, respectively. Increasing both potassium and nitrogen reduced disease severity measured as area under the disease progress curve (AUDPC) in 2016 and 2017. The N x K interaction was not significant for AUDPC in either year but was for three ratings dates in 2016. Two rating dates in June during the onset of disease indicated an effect size difference where all K fertilization rates were more effective at reducing disease compared to no K fertilization under lower N fertility than higher N. However by 16 August 2016, none of the K fertilization rates were effective at reducing anthracnose compared to no K under lower N, whereas K fertilization reduced disease compared to no K under greater N rate. During 2016 and 2017, plots treated with K (all rates) had a 55 to 59% and 49 to 54% reduction in disease severity, respectively, compared to plots that received no potassium. Anthracnose was lowest in plots treated with K every 2 weeks and N every seven days in 2016 and 2017.

Heritability of Summer Patch Resistance in Hard Fescue

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The summer patch disease incited by *Magnaporthiopsis poae* and *Magnaporthiopsis meyeri-festucae* is an important disease of hard fescue (*Festuca brevipilia*). Genetic resistance is an important control strategy and could reduce fungicide use. The objectives of this study were to (i) determine narrow-sense heritability summer patch resistance in hard fescue and (ii) evaluate inheritance characteristics of summer patch disease resistance. Inheritance characteristics were determined by evaluating the disease severity of progeny from crosses between resistant and susceptible hard fescue clones by both visual and NDVI ratings. Parental clones and progenies from reciprocal crosses were established in a field trial at horticultural farm 2 in North Brunswick, NJ in 2016. A trial was conducted in a randomized complete block design and inoculated with isolates of *Magnaporthiopsis poae* and *Magnaporthiopsis meyeri-festucae* applied at a rate of 3 cc per plant of prepared inoculum. Differences in progeny means between crosses were observed. Progeny from resistant × resistant crosses had less disease severity than resistant × susceptible and susceptible × susceptible crosses. Medium narrow-sense heritability estimates of 0.56 for visual ratings and 0.37 for NDVI were determined in 2018. Overall, this indicates a level of genetic control for summer patch resistance in hard fescue that can be utilized by breeders.

Microbiome Associated with Tall Fescue Under Drought Stress

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The effects microbiomes have on physiological traits in turfgrasses are poorly understood. Drought tolerance is an economically important trait, which can be influenced by certain microbes. One example is the mycorrhizal association in plants, which has been shown to improve drought tolerance of many plant species. In this study, we evaluated the microbiome associated with tall fescue genotypes grown in a rainout shelter at the Rutgers Plant Science Research and Extension Farm in Adelphia, NJ after prolonged periods of drought stress. Twelve plants were selected for analysis, comprised of six sets of siblings, one exhibiting a drought tolerant phenotype and the other a susceptible phenotype. The microbiome associated with the shoot, root, and rhizosphere soil was evaluated for each tall fescue half-sib pair. Microbiome analysis was preformed utilizing an Illumina next-generation sequencing metabarcoding approach that sequenced the 16S and ITS barcoding region to determine the composition of the bacterial and fungal communities, respectively. Comparative analysis of the microbiomes associated with different tissue types showed clear differences. The two phenotypes showed no significant differences in microbiome composition but certain microbial species differed in abundance. For the fungal microbiome, two common root associated fungi *Rhexocercosporidium* and *Cadophora* were found to have higher abundances in the drought susceptible phenotypes. For the bacterial microbiome, the *Actinobacteria* were found to be significantly higher in the resistant phenotype when comparing the root tissues. The bacterial soil microbiome had three common soil bacteria *Opitutae*, *Chitinophagaceae*, and *Devosia* that were all higher in abundance in the resistant phenotype. To see if there are any significant differences in microbial functions, the metatranscriptome of the same set of siblings is currently being evaluated.

Eastern Filbert Blight of Hazelnuts: A Complicated Nemesis

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Eastern filbert blight (EFB) is a stem canker disease caused by the ascomycete fungus *Anisogramma anomala*, which is native to the eastern U.S. and found associated with our wild hazelnut *Corylus americana*. While our native hazelnut is highly tolerant of EFB and rarely impacted by the disease, the commercial European hazelnut, *C. avellana*, is highly susceptible. When exposed to *A. anomala*, most European hazelnut cultivars develop large infections that compromise stems and can cause tree death in 4-5 years under high disease pressure. Fortunately, a significant germplasm collection and evaluation effort made at Rutgers University and Oregon State University over the past two decades has identified a large number of plants from a wide diversity of genetic backgrounds that are resistant to and/or highly tolerant of the disease (Capik and Molnar, 2012; Molnar et al., 2018; Muehlbauer et al., 2014). However, breeding for resistance in the eastern U.S. is complicated by *A. anomala*'s very high level of genetic diversity (Muehlbauer et al., 2019) and its unexpected large genome size of around 350 Mb, which is nearly seven times the size of an average ascomycete and consists of >85% repetitive elements (Cai et al., 2013; Hillman, unpublished). Further, many of the new sources of resistance to EFB identified appear to be conferred by single dominant genes (Leadbetter et al., 2016; Sathuvalli et al., 2011a, 2011b, 2012). This may present long-term problems if protection from disease is based on an overreliance on only one or two *R*-genes.

The 'Gasaway' source of resistance, the first identified single dominant *R*-gene used in breeding, provides an example of the challenges hazelnut breeders may face when using only single genes to protect plants from EFB. The *R*-gene has been used in breeding since the 1970s and currently protects a significant portion of the U.S. hazelnut industry, which is centered in the Willamette Valley of Oregon (Mehlenbacher et al., 1991, 2016) – a place outside the native range of *A. anomala* – where the fungus is much less genetically diverse (Muehlbauer et al., 2019; Tobia et al., 2017). While the *R*-gene provides a high level of protection in Oregon, it has had inconsistent results in New Jersey and elsewhere in the eastern U.S. depending on location and the cultivar carrying it. In past studies in New Jersey, different cultivars carrying this *R*-gene were shown to express a range of phenotypes from tolerant (moderate amounts of EFB) to resistant (free of EFB) (Capik and Molnar, 2012; Molnar et al., 2010a, 2010b). A study based on large populations of trees segregating for the *R*-gene showed that this varied response was likely related to uncharacterized modifying factors or supporting genes present or absent in the individual genotypes, with the *R*-gene alone providing only a moderate level of tolerance. It is hypothesized that cultivars developed in Oregon were not able to be selected for presence or absence of the modifying factors necessary for protection in the eastern U.S., since the *R*-gene alone continues to provide resistance in that region (Muehlbauer et al., 2018).

Very recent observations and study at Rutgers suggests the scenario described in New Jersey might be more complicated and/or is changing. In the winter of 2016, multiple 'Gasaway' protected plants previously deemed highly tolerant (few cankers of reduced size) were found to express many large cankers more typical of plants considered susceptible. The following two years confirmed this apparent change in response to EFB with many trees, including clones of 'Gasaway' itself, now showing significant amounts of EFB. To examine this field observation more critically, controlled inoculations were performed using *A. anomala* collected from these new large cankers ("Gasaway" inoculum) and applied following published methods (Molnar et al., 2010a) to replications of five cultivars (Yamhill, Jefferson, Dorris, McDonald, and Felix) that carry this *R*-gene. A control population consisting of a set of the same trees was exposed to *A. anomala* inoculum collected from non-'Gasaway' protected trees ("general" inoculum). 'Sacajawea', a tree un-related to 'Gasaway' that expresses tolerance of EFB under field conditions, was included in both sets of inoculations.

Preliminary results showed that 87% of the total 'Gasaway' protected trees in the study exposed to the "Gasaway" inoculum expressed EFB. This is in contrast to 20% of trees exposed to the "general" inoculum. Further, the average proportion of disease wood of the infected trees in the "Gasaway" inoculum group was 21.7% of the total tree growth, whereas infected wood from trees of the other group was only 6.7%. Finally, the average canker length of the "Gasaway" and "general" group was 30.1 cm and 9.7 cm, respectively. Interestingly, the tolerant 'Sacajawea' showed fewer trees infected by the "Gasaway" inoculum compared to the "general" inoculum (50% vs. 70%), a reduced proportion of diseased wood (12.5% vs 15.9%), and smaller average cankers size (30.0 cm vs. 36.8 cm). The results of the study are striking, especially when comparing to results of Molnar et al. (2010a) where very few 'Gasaway'-protected trees expressed EFB when inoculated in a similar manner using isolates of the fungus obtained from New Jersey and more than 10 additional locations across its native range. Overall, these results suggest a difference in pathogenicity between the two sources of inoculum. When combined with the field observations, this points to the possibility that a new race of the fungus has emerged. A second replication of the study is underway, in addition to genetic fingerprinting of the fungus to help elucidate the current findings.

Considering the long timelines included in breeding perennial tree crops and the expectation of hazelnut orchards being in production for many decades, experiences with the 'Gasaway' *R*-gene in New Jersey provide impetus for the need to explore resistance breeding approaches shown to enhance durability of disease response. These include examining gene pyramiding and horizontal resistance, and possibly a combination of approaches to develop improved cultivars expected to help manage disease over extended periods of time.

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A New Cooperative Extension Fact Sheet: Tall Fescue Cultivars for High Traffic Turf Surfaces

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Turf managers in the cool temperate and transitional climatic regions of the United States such as New Jersey are increasingly establishing tall fescue (*Schedonorous arundinaceus* [Schreb.] Dumort.) on high traffic turf surfaces such as sports fields. The seasonal traffic tolerance (2014-2017) and non-trafficked turf quality (2013-2017) of tall fescue entries comprising the 2012 National Turfgrass Evaluation Program (NTEP) Tall Fescue Test were assessed in North Brunswick, NJ. Turf professionals frequently request research-based recommendations for turfgrass cultivars for both trafficked and non-trafficked sites. Traffic tolerance, turfgrass quality, and susceptibility to brown patch disease (caused by *Rhizoctonia solani)* are important selection criteria for tall fescue cultivars. A new Cooperative Extension Fact Sheet summarizing field trial results at Rutgers University and other trial locations in the northeast and transition zone is being developed to aid in the selection process. Information concerning purchasing seed, establishing tall fescue turf, and maintenance of tall fescue will be included in the Fact Sheet. Entries with the best average traffic tolerance during spring, summer, or autumn in North Brunswick, NJ, best average non-trafficked turf quality in North Brunswick, NJ during 2013 to 2017 and least brown patch disease across eight NTEP trial locations (2015-2016) will be tabularized and marked with an 'X'. Turf managers will be able to visually associate a greater quantity of 'X' marks across these selection criteria with better performing cultivars. Entries that ranked as best for traffic tolerance during spring, summer, and autumn in North Brunswick, NJ (2014-2017), best average non-trafficked turf quality in North Brunswick, NJ (2013-2017) and least brown patch disease across eight NTEP trial locations (2015-2016) were Regenerate, Raptor III, 4th Millennium SRP, F711, B23, Hemi, and Technique.

NTEP Screening for *Epichloe coenophialum* in 132 Tall Fescue Selections

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Tall fescue (S*chedonorus arundinaceus* Schreb.) is a deep-rooted cool season turf and pasture grass that often contains a fungal endophyte, *Epichloë coenophiala*. The fungus produces alkaloids but does not cause plant disease. The presence of the endophyte results in improved insect resistance and stand establishment, although depending upon the alkaloid profile for each strain of the fungus, the grass may be toxic to grazing animals and suitable only for turf. It is therefore very important to determine the endophyte status for turfgrass, and both the endophyte status and alkaloid profiles for pastures. We have screened 132 tall fescues selections for the presence of the endophyte using a modified version of a commercial endophyte testing kit. We will be presenting the results of this study.

Variance Components and Heritability Estimation for Drought Tolerance of Tall Fescue Using a Bayesian Approach

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Heritability is a fundamental concept in plant breeding and genetics, which measures the fraction of variability in observed traits that can be explained by genetic variability. It provides a measurement of a population's genetic potential in response to selection. The partitioning of variance and estimation of heritability have a long history in genetics dating back to R.A. Fisher in the early 20th century. Numerous methods have been developed over the years, such as parent-offspring regression, nested full-sib or half-sib analysis, and ANOVA. All of them require special experimental designs and represent special cases of local analysis on parts of the pedigree that breeders might have in hand. A general solution is offered by estimating variance components using maximum likelihood or restricted maximum likelihood methods, which allows exploitation of all information included in the pedigree. The objective of this project is to implement this method under the Bayesian framework using phenotypic data and pedigree record. The phenotypic data were Normalized Difference Vegetation Index(NDVI) collected from rainout shelter studies conducted in summer of 2017 and 2018. Heritability of drought tolerance was found to have a posterior mean of 0.23, and a 95% credible interval of 0.12 to 0.34, indicating the difficulty to increase drought tolerance in tall fescue.

Physiological Effects of Seaweed Extracts for Alleviating Summer Bentgrass Decline

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High temperature stress negatively impacts the health and performance of cool-season turfgrasses during summer months. Physiological effects of the seaweed extracts XP and Stress Rx, developed by Ocean Organics, were examined on creeping bentgrass (*A. stolonifera* L. cv. 'Penncross') putting greens exposed to prolonged heat stress. Creeping bentgrass field plots were maintained according to a putting green program typically employed on golf courses prior to initiation of the study. Products were applied individually or in combination as foliar spray treatments every 14 d through the duration of the summer in 2016 and 2017 and all plots were maintained under well-irrigated conditions. The physiological parameters of visual turf quality (TQ), normalized difference vegetation index (NDVI), leaf area index (LAI), stress index (SI), canopy coverage, and greenness were measured. In both the 2016 and 2017 summer trials, plots treated with XP and Stress Rx applied individually or in combination had higher turf quality ratings, canopy coverage and greenness, NDVI, LAI, and lower SI when compared to untreated controls under prolonged heat stress. The superior quality of creeping bentgrass plots treated with these seaweed extracts suggests that applying XP and Stress Rx alone or in combination during prolonged heat stress may have mitigated heat-induced decline by offering the plants stress defense and recovery mechanisms.

Physiological Effects of Iron for Promoting Salt Tolerance in Creeping Bentgrass

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Salt-induced oxidative stress may inhibit root growth and accelerate leaf senescence while iron nutrition plays roles in regulating these processes. The objectives of this study were to examine whether additional iron in the growing solution of roots would enhance salt tolerance of creeping bentgrass, and to investigate whether the positive effects of iron on salt tolerance is associated with its regulatory functions in antioxidant metabolism in roots exposed to prolonged salt stress. The present study investigated the impact of iron (0, 20 and 40 μM concentrations of Fe(II)-EDTA) on physiological parameters of creeping bentgrass under salt stress (0 and 150 mM concentrations of NaCl) for 35 days in growth chambers, using salt-tolerant *A. scabra* ('NTAS') and salt-sensitive *A. stoloniferous* ('Penncross'). Physiological analysis demonstrated that salt stress accelerated leaf senescence, shown by the decline in chlorophyll content, photochemical efficiency and membrane stability, and induced oxidative damage in roots for both bentgrass species. The adverse physiological damage due to salt stress was less pronounced in 'NTAS' than in 'Penncross'. However, those adverse effects were significantly ameliorated by increasing iron levels in the rooting medium, which was associated with increasing activities of superoxide dismutase, catalase, ascorbate peroxidase and peroxidase, particularly in 'Penncross'. The results demonstrate that exogenously applied iron effectively reduced salt-induced cellular damage by suppressing oxidative damage in bentgrass species.

Dollar Spot Control as Affected by Fungicide Programming and Bentgrass Susceptibility

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Dollar spot epidemics, caused by *Clarireedia jacksonii* (formerly known as *Sclerotinia homoeocarpa* F.T. Bennett), differ in incidence and severity among bentgrass (*Agrostis* spp.) cultivars. A field trial was conducted in North Brunswick, NJ to assess the effectiveness of using threshold-based fungicide applications to control dollar spot among bentgrasses ranging in susceptibility to dollar spot. A 6 × 3 factorial arranged in a randomized complete block design with four replications was used for this field trial. The six bentgrass cultivars ranging in susceptibility to dollar spot included 'Capri' colonial bentgrass (*A. capillaris*), and 'Declaration', '007', 'Shark', 'Penncross' and 'Independence' creeping bentgrass (*A. stolonifera*); cultivar plots were established in 2015. Three fungicide programs were applied to all cultivars and included a calendar-based and two curative programs based on a damage threshold of 314-mm^2 over all four replications. Fungicide was applied every 21-days from 21 May through 4 Nov. 2018 for the calendar-based program. The two curative programs differed in the timing of fungicide application: one program applied fungicide within 24-hours of reaching the damage threshold and the other applied fungicide on the next scheduled spray-day (Monday) after the damage threshold was reached. Disease severity was measured as the active dollar spot infection area within a 0.6- by 1.2-m area of each plot every 1 to 3 days and summarized for the growing season by calculating the area under disease progress curve (AUDPC) from 15 May to 6 Nov. 2018. Fungicide programs had a strong influence on disease response to bentgrass cultivar. In general, the most effective dollar spot control was achieved by the calendar-based fungicide program while the least effective disease control was observed on threshold-based applications on the next spray-day. The AUDPC was not different between cultivars for either the calendar or 24-hour threshold programs; and the calendar-based and 24-hour threshold programs did not differ in AUDPC, albeit AUDPC was numerically greater for the 24-hour threshold program. However, the number of sprays for the 24-hour threshold program was lower than the calendar-based program and varied with cultivar. Curative fungicide applications on the next spray-day also reduced the total number of fungicide applications compared to the preventive calendar-based program. However, the level of disease control under the next spray-day program was not as effective as either the calendar-based program or curative applications within 24-hours of the damage threshold for most cultivars. Penncross had the greatest disease severity whereas Declaration had the lowest severity under the next spray-day program; approximate disease severity ranking among other cultivars (high to low) was Shark ≥ Capri ≥ Independence ≥ 007. Declaration (least susceptible) was the only cultivar that had a similar level of disease control under all three fungicide programs. Nine fungicide applications were made in the calendar-based program while curative applications resulted in two to three sprays (next spray-day vs 24 hours, respectively) on Declaration; four sprays on Capri, 007 and Shark; four to five sprays on Penncross (next spray-day vs 24 hours, respectively); and six sprays on Independence. This trial will be continued in 2019.