

## **Symposium Organizing Committee**

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Stacy A. Bonos  
Bruce B. Clarke  
Matthew Elmore  
Barbara Fitzgerald  
William A. Meyer  
James A. Murphy  
Phillip L. Vines

## **Proceedings of the Thirtieth Anniversary Rutgers Turfgrass Symposium**

Phillip L. Vines and Barbara Fitzgerald, Editors

*Rutgers Cooperative Extension educational programs are offered to all without regard to race, religion, color, age, national origin, gender, sexual orientation, or disability.*

## **Director's Remarks**

Welcome to the Thirtieth Annual Rutgers Turfgrass Symposium. Established in 1991, the Symposium provides a forum for Rutgers faculty, students, staff and guests with diverse expertise and backgrounds to exchange ideas and encourage collaboration on a wide range of topics in turfgrass science. I thank our invited speakers, Dr. Cristobal Uauy (John Innes Center, United Kingdom) who will present a keynote on “Unlocking the polyploid potential of crops through genomics,” as well as Mr. Naveen Singa (Siemens Corporation), Dr. Josh Friell (The Toro Company) and Dr. J. Scott McElroy (Auburn University), and all the Center faculty and students who have agreed to present at this year’s symposium. I also thank Drs. Bingru Huang, Stacy Bonos, Rong Di, and William Meyer for serving as session moderators and the Symposium Planning Committee comprised of Drs. Rong Di (Symposium Chair), Bruce Clarke, William Meyer, Stacy Bonos, and Matt Elmore as well as Dr. Phillip Vines and Ms. Barbara Fitzgerald (co-editors of the Symposium Proceedings) for their contributions in preparing this year’s program. We appreciate the technical support of Mr. Bernard Ward and Ms. Alanna Perez who made it possible to live stream this year’s virtual Symposium.

Our faculty, students and staff continue to be recognized for excellence. Five of our students were recognized during graduate student poster and oral paper competitions at the annual meeting of the Crop Science Society of America. William Errickson received first place in the Turfgrass Science—Turf Management poster session and second place in the Society-Wide Graduate Student Competition; Stephanie Rossi took first place in the oral sessions of two divisions, Crop Physiology and Metabolism and Turfgrass Science—Golf; Cathryn Chapman was awarded first place in the Turfgrass Science—Turf Management oral session and second place in the Turfgrass Science—Turf Management poster session; Pingyuan “Bay” Zhang was awarded first place in the Turfgrass Science—Industry poster session. Katherine Diehl placed first in the graduate oral presentation contest at the Northeastern Plant Pest and Soils Conference.

Dr. William Meyer was awarded the 2020 United States Golf Association Green Section Award for his work in sustainability through agronomic advancements. Dr. Bruce Clarke received the Nebraska Turfgrass Association Presidential Award for significant contributions to the turfgrass industry in Nebraska and the nation as well as the 2020 RCE Extension Specialist of the Year Award. Dr. Bingru Huang was invited by the publisher Maximum Academic Press to create a new journal called Grass Research and serve as editor-in-chief. Dr. Stacy Bonos was awarded Fellow by the American Society of Agronomy and was appointed director of the Turfgrass Breeding Program and

associate director of the Center for Turfgrass Science. Dr. James Murphy was appointed the Ralph Geiger Endowed Chair in Turfgrass Science and director of the Center for Turfgrass Science.

We owe a debt of gratitude to Drs. Bruce Clarke and William Meyer for their leadership as director and associate director of the Center. Their wisdom and commitment to excellence has yielded tremendous growth and productivity over nearly three decades – they built a foundation to perpetuate success. I am sincerely thankful for all the support that both have provided me and the turfgrass program at Rutgers. I look forward to both continuing to share their insights and talents as the Center pursues its mission of generating and disseminating knowledge and providing training and education in the turfgrass sciences.

We are indeed fortunate for the outstanding partnership with our turfgrass industry colleagues across the state, region, and nation. The industry's sharing of intellectual and material support is a truly appreciated. The Center is so much better for it. The Turfgrass Symposium has become a long and valued tradition at Rutgers. We are glad that you chose to spend time with us and hope that you enjoy the many opportunities that this Symposium has to offer.

Sincerely,

A handwritten signature in cursive script that reads "James A. Murphy". The signature is written in black ink and is centered below the word "Sincerely,".

James A. Murphy, Director

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**THIRTIETH ANNIVERSARY RUTGERS TURFGRASS SYMPOSIUM**  
**Advances in Turfgrass Science: Looking to the Future**

School of Environmental and Biological Sciences, Rutgers University

March 18, 2021  
 Virtual Event

**Thursday, March 18, 2021**

- 9:00 AM**                    **Welcome – Laura Lawson** (*Interim Executive Dean, School of Environmental and Biological Sciences*)
- 9:10 - 10:40 AM**        **SESSION I: New Technologies for Turfgrass Breeding and Management** (Moderator: Bingru Huang)
- Rong Di** (*Department of Plant Biology, Rutgers University*)  
 Application Of CRISPR-Gene Editing and Tissue Culture to Improve Creeping Bentgrass
- Josh Friell** (*Senior Research Scientist, The Toro Company*)  
 New Technologies for Optimizing Turf Management
- Phillip Vines** (*Department of Plant Biology, Rutgers University*)  
 Applications of High-throughput Plant Phenotyping in Turfgrass Breeding
- Naveen Singa** (*Research Professional, Siemens Technology*)  
 Decision Support System - Collect, Analyze, Deploy, and Integrate Edge Solutions for the Food and Beverage Industry
- 10:40 - 11:00 AM**        **Discussion, e- Posters, and Break**
- 11:00 - 11:45 AM**        **Keynote Address** (Moderator: Stacy Bonos)
- Cristobal Uauy** (*John Innes Center, United Kingdom*)  
 Unlocking the Polyploid Potential of Crops Through Genomics
- 11:45 - 12:00 PM**        **Discussion Session**
- 12:00 – 1:00 PM**        **Lunch Break**

1:00 – 2:00 PM

**SESSION II: Poster Session** (Moderator: Rong Di)

**Physiological Effects of Plant-Health Products for Improving Drought Tolerance and Post-Stress Recovery in Creeping Bentgrass**

Cathryn Chapman and Bingru Huang

*Department of Plant Biology, Rutgers University*

**Herbicide Application Timing Affects Deer-tongue Grass (*Dichanthelium clandestinum*) Control in Native Areas**

Katie H. Diehl, Matthew T. Elmore, and Phillip L. Vines

*Department of Plant Biology, Rutgers University*

**Rhizobacteria Inoculation and Colonization for Promoting Plant Growth in Cool Season Turfgrass**

William Errickson<sup>1</sup>, Bingru Huang<sup>1</sup>, and Ning Zhang<sup>1,2</sup>

<sup>1</sup>*Department of Plant Biology, Rutgers University*, <sup>2</sup>*Department of Biochemistry and Microbiology, Rutgers University*

**Applications of the Fungal Endophyte *Epichloë festucae* Antifungal Protein *Efe-AfpA***

Patrick Fardella, Bruce B. Clarke, and Faith C. Belanger

*Department of Plant Biology, Rutgers University*

**Spatial Distribution of Dollar Spot Fungus in Asymptomatic and Symptomatic Turfgrass**

Glen Groben<sup>1</sup>, Bruce Clarke<sup>1</sup>, James Murphy<sup>1</sup>, Patrick Purdon<sup>1</sup>, Paul Koch<sup>2</sup>, Ning Zhang<sup>1,3</sup>

<sup>1</sup>*Department of Plant Biology, Rutgers University*, <sup>2</sup>*Department of Plant Pathology, University of Wisconsin-Madison*, <sup>3</sup>*Department of Biochemistry and Microbiology, Rutgers University*

**Organic Lawn Clippings Can Feed Livestock and Produce Food for People**

Joseph Heckman<sup>1</sup> and Mike Westendorf<sup>2</sup>

<sup>1</sup>*Department of Plant Biology, Rutgers University*, <sup>2</sup>*Department of Animal Science, Rutgers University*

**Differential Physiological Responses to Heat and Drought Stress for Annual Bluegrass and Creeping Bentgrass**

Sean McBride, James Murphy, and Bingru Huang

*Department of Plant Biology, Rutgers University*



**Genetic Control of Eastern Filbert Blight Resistance in F<sub>2</sub> Generation Hybrid Hazelnut (*Corylus americana* × *C. avellana*) Populations**

Thomas J. Molnar<sup>1</sup>, David Hlubik<sup>1</sup>, Shawn Mehlenbacher<sup>2</sup>, and John M. Capik<sup>1</sup>

<sup>1</sup>*Department of Plant Biology, Rutgers University,* <sup>2</sup>*Department of Horticulture, Oregon State University*

**Kentucky Bluegrass Tolerance to Traffic During Summer and Autumn**

Bradley S. Park and James A. Murphy

*Department of Plant Biology, Rutgers University*

**Improvements to an Endophyte Detection Kit: A Story of Milk and Phosphatase**

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**Metabolic Regulation of  $\gamma$ -Aminobutyric Acid During Heat-induced Leaf Senescence in Creeping Bentgrass**

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**Understanding and Optimizing Sampling Methods for the Annual Bluegrass Weevil**

Anna Luiza Sousa, Ryan Geisert, and Albrecht M. Koppenhöfer

*Department of Entomology, Rutgers University*

**Viruses in Fungi That Cause Dollar Spot Disease of Turfgrass**

Trini Taccad<sup>1</sup>, Alanna Cohen<sup>1,2</sup>, Limei Du<sup>1</sup>, and Bradley Hillman<sup>1,2</sup>

<sup>1</sup>*Department of Plant Biology, Rutgers University,* <sup>2</sup>*Graduate Program in Microbial Biology, Rutgers University*

**Genome Wide Association Study of Anthracnose Disease in Switchgrass**

Christopher Tkach<sup>1</sup>, Jeremy Sutherland<sup>2</sup>, Stacy A. Bonos<sup>1</sup>, John E. Carlson<sup>3</sup>, Terrence H. Bell<sup>2</sup>, Jesse R. Lasky<sup>4</sup>, Julie L. Hansen<sup>5</sup>, Ryan V. Crawford<sup>5</sup> and Donald Viands<sup>5</sup>

<sup>1</sup>*Department of Plant Biology, Rutgers University,* <sup>2</sup>*Department of Plant Pathology and Environmental Microbiology, Penn State University,* <sup>3</sup>*Department of Ecosystem Science and Management, Penn State University,* <sup>4</sup>*Department of Biology, Penn State University,* <sup>5</sup>*School of Integrative Plant Science, Cornell University*

**Comparing Relative Virulence Among *Magnaporthiopsis* spp. on Hard Fescue and Kentucky Bluegrass Hosts**

Phillip L. Vines, Kyle M. Genova, Glen Groben, Marcus Rountree, William A. Meyer, and Bruce B. Clarke  
*Department of Plant Biology, Rutgers University*

**Inheritance of Summer Patch Disease Resistance in Hard Fescue (*Festuca brevipila* Tracey)**

Shidi Wu, Austin L. Grimshaw, Yuanshuo Qu, Phillip L. Vines, Eric N. Weibel, William A. Meyer, and Stacy A. Bonos  
*Department of Plant Biology, Rutgers University*

**Potassium Fertilization Effect on Dollar Spot of Annual Bluegrass**

Zhongqi Xu, Daniel Ward, James A. Murphy, and Bruce B. Clarke  
*Department of Plant Biology, Rutgers University*

**Influence of Fungicide Programming and Bentgrass Susceptibility on Dollar Spot Control**

Pingyuan Zhang, Daniel Ward, James A. Murphy, and Bruce B. Clarke  
*Department of Plant Biology, Rutgers University*

**2:00 – 2:30 PM**

**Q & A, Discussion Session, and e- Posters**

**2:30 – 4:00 PM**

**SESSION III: Pest Management** (Moderator: William Meyer)

2:30 – 2:50

**Matt Elmore** (*Department of Plant Biology, Rutgers University*)  
Goosegrass Resistance to Dithiopyr

2:50 – 3:15

**Scott McElroy** (*Department of Crop, Soil, and Environmental Science, Auburn University*)  
Identifying the Mechanism of Oxadiazon Resistance in Goosegrass and Improved Understanding of PPO-Inhibitor Mode of Action

3:15 – 3:35

**Pingyuan Zhang** (*Department of Plant Biology, Rutgers University*)  
Interpretations of a Logistic Regression Model for Fungicide Control of Dollar Spot on Creeping Bentgrass

3:35 – 4:00

**Bruce Clarke** (*Department of Plant Biology, Rutgers University*)  
Developing Turf Disease Control Programs That are Efficacious and Environmentally Sound

**4:00 - 4:15 PM**

**Discussion Session and Closing Remarks**

**PLENARY PRESENTATIONS**

## Application of CRISPR-Gene Editing and Tissue Culture to Improve ‘Crenshaw’ Creeping Bentgrass

Rong Di, Stacy A. Bonos, and William A. 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. Besides being stressed by heat and drought during summer months, many commonly used cultivars of creeping bentgrass are highly susceptible to dollar spot disease caused by *Clariireedia jacksonii*. CRISPR (clustered regularly interspaced short palindromic repeats-associated endonuclease)-gene editing technology has been used to precisely knock-out stress negative regulators to enhance plant stress tolerance and disease susceptible genes to improve plant disease resistance. We have constructed our own CRISPR-gene editing vector with 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 Crenshaw partial genomic DNA (gDNA) sequences of *DREB* (dehydration responsive element binding 2)-like gene, a negative stress regulating transcription factor, and *CPK12* encoding calcium-dependent protein kinase (CDPK), a proven negative regulator for rice blast disease resistance, were identified by bioinformatics analysis and cloned by PCR (polymerase chain reaction). The 20-nucleotide *AsDREB-EcoRV* target sequence and the 26-nucleotide *AsCPK12-SacI* target sequence were chosen and the CRISPR-gene editing vectors pRD303 and pRD302 were 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. The gDNA fragments spanning the *AsDREB* and *AsCPK12* target sites from each transgenic plant were PCR-amplified, analyzed by RFLP (restriction fragment length polymorphism), DNA sequencing and ICE (inference for CRISPR editing) analysis. Selected *AsDREB* mutant and non-edited Crenshaw plants were tested for their drought and salt 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. Some *AsDREB*-gene edited mutant Crenshaw plants were shown to be less stressed by drought and salt compared to the non-gene edited plants. We are in the process of analyzing the putative *AsCPK12*-edited transgenic Crenshaw plants. Supplementing chemicals in tissue culture media and manipulating culture conditions presents an alternative method to produce stress tolerant turfgrass plants, negating the utilization of transgenes. Some of our progresses in tissue culturing method will be presented. Our developed CRISPR-gene editing platform can be applied to other turfgrass species and other agronomically important traits.

## **New Technologies for Optimizing Turf Management**

Joshua Friell

*Center for Technology, Research, and Innovation, The Toro Company*

Technology is enabling rapid advancements in turfgrass management equipment design and capability. This in turn allows turfgrass managers to re-imagine their maintenance practices, thereby helping to address industry priorities such as Labor and Training, Environmental Stewardship, and Resource Use Optimization. In this presentation, examples of technology that are driving these changes are explored and areas for potential collaboration between turf managers, researchers, and manufacturers are identified.

Electrification of equipment, driven primarily by improved battery technology, is a growing trend in the turf equipment industry. While lithium batteries can achieve several times greater energy density than traditional lead-acid batteries, they are still orders of magnitude less energy dense than fossil fuels. However, greater efficiency of other system components helps to overcome this difference and provide opportunity for cost savings and environmental stewardship. Electric technology implementation and overall energy needs are functions of the load profiles and use cases of the equipment. Researchers, turf managers, and industry must work together to characterize the energy requirements for turf maintenance operations, quantify impacts of agronomic practices on that requirement, and develop a clear definition of the environment in which the equipment is used.

Smart and Connected products are driving advanced decision support and more intelligent machines powered by smaller, less expensive, and more powerful processors and controllers. Smart systems built into equipment allow for features such as user customization, precise application of inputs such as on GPS-enabled application equipment, and implementation of machine learning approaches for myriad other applications. Connecting these devices to one another, to the internet, and to external data streams allows for even broader capabilities. However, because countless implementation strategies for these technologies are possible, application-specific insights are needed to inform technology and design choices.

By combining the technologies discussed above, many types of equipment and machinery have great potential to become robotic and autonomous. Equipment may perform any number of specialized tasks of varying levels of precision. Most commonly, today, autonomous rotary mowers are used in residential and light commercial applications. A growing area of interest is to add value through niche applications of robotics technology to perform low precision, yet specialized, tasks. Understanding which tasks may be automated to provide the greatest value in terms of time, cost, and labor savings while achieving sufficient levels of precision and quality is an area of ongoing study across the industry.

Advancements in electrical, mechanical, and computer technology are driving change in the turfgrass industry. While technology evolves, industry, academia, and practitioners must work together to identify feature sets that add the greatest value and determine how best to implement those features. Application-specific information should inform technology and design choices

thereby ensuring development of equipment that enables a bright and sustainable future for turfgrass management.

## **Applications of High-Throughput Plant Phenotyping in Turfgrass Breeding**

Phillip L. Vines and Ryan M. Daddio

*Department of Plant Biology, Rutgers University*

The general approach to plant breeding is to make as many crosses as possible and evaluate progeny of those crosses for specific phenotypic traits across diverse environments with an overarching goal of identifying superior, broadly-adapted plant material. For turfgrass breeding, the focus is to improve turfgrass quality characteristics (color, uniformity, texture, and density), increase seed yield, develop resistance to abiotic and biotic stress factors, and reduce input (water, nutrients, light, etc.) requirements. Both labor and time demands associated with manual phenotyping for these types of traits limit the number of genotypes and locations that can be tested. Thus, the ability of a turfgrass breeder to develop improved varieties is largely constrained by this phenotyping bottleneck. Recent improvements in remote sensing technologies for high-throughput phenotyping offer opportunities to mitigate these limitations, obtain more data points than would be humanly possible, and improve breeding efficiency in modern plant breeding programs. The objective of this study was to compare unmanned aerial system (UAS)-mounted light reflectance sensors with visual plant evaluations for turf health and performance. Data was collected from field trials during the 2020 growing season for establishment rate of fine fescue and tall fescue, percent ground cover of Kentucky bluegrass after summer stress, and gray leaf spot disease resistance and white grub tolerance of tall fescue. Multispectral and digital images were captured using UAS-mounted sensors, and visual ratings were conducted for ground truthing purposes. Color and vegetation indices including normalized difference vegetation index (NDVI), normalized difference red edge index (NDRE), optimized soil adjusted vegetation index (OSAVI), ratio vegetation index (RVI), transformed vegetation index (TVI), and excess green index (EXG) were calculated using remote sensing data. Results show a strong relationship between visual evaluations for fine fescue and tall fescue establishment rate and ExG, NDRE, and NDRE705 indices. A strong relationship was also observed between visual assessments for percent ground cover and NDVI on Kentucky bluegrass. With respect to gray leaf spot disease resistance of tall fescue, the strongest relationships with visual ratings were observed with OSAVI and TVI. None of the indices evaluated in this study were closely related to visual ratings for white grub tolerance on tall fescue. These findings indicate that remote sensing-based high-throughput phenotyping data has potential applications in evaluation of turfgrass germplasm in turfgrass breeding programs. Studies are underway to further evaluate these and other indices for use in mowed turf plot trials and spaced-plant nurseries.

## **Decision Support System – Collect, Analyze, Deploy, and Integrate Edge Solutions for the Food and Beverage Industry**

Naveen Singa

*Siemens Technology*

A decision support system (DSS) is an interactive and intelligent system of hardware and software components primarily used by decision makers to (i) compile useful information from multi-dimensional data comprising raw sensor streams, documents, and personal knowledge; (ii) identify and predict problems (simulation models, economic models); and (iii) make an optimized decision (mathematical optimizations, data-driven). The final output of a DSS is a recommendation, interpretation, or prediction regarding the situation of interest, such as irrigation management, crop treatment, and food safety and quality.

Maintaining quality with minimized resource utilization (thereby the cost) is a global problem statement in the food and beverage industry. The main advantages of using a DSS here includes examination of multiple alternatives, better understanding of the processes, identification of unpredicted situations, enhanced communication, cost effectiveness, and better use of data and resources. Few examples are (i) tracking and tracing application, part of DSS for food and beverage industry, helped derive quality tracking and monitoring of key performance indicators derived from various data sources; (ii) using the block chain technology helped optimized the transportation routines, minimizing the time and transportation food wastage; (iii) improving irrigation scheme management in arid climates; and (iv) implementing for precision farming gathering satellite data, field moisture sensor data, and generating irrigation and fertilization profiles.

Decision support systems could be constructed for turfgrass-specific applications such as precision turfgrass management and high-throughput phenotyping. In this case, machine learning-based predictive models would be developed by considering soil and climatic conditions and other micro-scale (plant and soil level) and macro-scale (site level) information to predict turfgrass needs and identify superior turfgrass breeding lines. The DSS could make real-time suggestions of turfgrass management programs and selection of top-performing turfgrass cultivars. The ultimate goal of developing a DSS for turfgrass applications would be to enable the multi-sensor suite of ground and aerial phenotyping platforms to actively obtain images of individual plants, spaced-plants, or contiguous turf areas to acquire on-site information of turfgrass phenotypes and physiological conditions and store images and data in a point cloud platform, which could be easily accessible through any mobile device.



## Unlocking the Polyploid Potential of Crops Through Genomics

Cristobal Uauy

*John Innes Centre, Norwich, United Kingdom*

Developments over the past few years have radically changed the way we work with polyploid wheat. Both hexaploid and tetraploid wheat now have whole genome sequences and reliable gene models. This has expanded beyond the single reference genome to multiple cultivars. These and additional developments (e.g. sequenced mutant resources, expression browsers, speed breeding) have dramatically lowered the barriers to undertake biological research in polyploid wheat. For many purposes, wheat can now be treated (almost) like a model crop species. The next phase will be to start understanding the biological mechanisms underlying the most important traits in polyploid wheat and to design strategies to ensure this knowledge is quickly transferred to the field. In the talk, I will discuss how we have built a series of community resources to help genomics-enabled breeding in wheat. I'll present some of the lessons we have learnt in the process and exemplify how we are using these resources for trait discovery. This will include an update on how we are using the multiple sequenced genomes to define and characterise haplotypes within elite gene pools. I will discuss how we successfully used this approach for focused discovery of novel haplotypes from landrace collections and documented its potential for trait improvement in modern bread wheat. I will also present recent results on how we are using these resources to understand yield components in wheat. I will argue that given polyploidy, breeders have exploited only a fraction of the potential genetic variation in the wheat genome. The recent breakthroughs in wheat genomics now allow us to make a decisive effort towards exploiting this under-utilised variation, thereby unleashing the full potential of the polyploid wheat genome.

## Goosegrass Resistance to Dithiopyr

Matthew T. Elmore<sup>1</sup>, Katherine H. Diehl<sup>1</sup>, Rong Di<sup>1</sup>, Sarah L. Boggess<sup>2</sup>, Robert N. Trigiano<sup>2</sup>, James T. Brosnan<sup>3</sup>, Daniel P. Tuck<sup>1</sup>, and Brandon C. McNally<sup>1</sup>

<sup>1</sup>*Department of Plant Biology, Rutgers University*

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Goosegrass (*Eleusine indica*) is a problematic C<sub>4</sub> annual grassy weed. Previous field experiments demonstrated poor goosegrass control at two golf course sites when mitotic-inhibiting herbicides were applied preemergence. The objectives of this research were the following: 1) to quantify the resistance to mitosis-inhibiting herbicides in these goosegrass biotypes; 2) elucidate the mechanism conferring resistance; and 3) screen goosegrass biotypes from the greater New Jersey region for mitotic-inhibitor resistance. Goosegrass biotypes were collected from golf course fairways in East Brunswick, NJ, Philadelphia, PA and Manalapan, NJ. An Murashige and Skoog medium bioassay was used to determine the response of each biotype to dithiopyr and proflumicarb. Seeds were planted to media containing proflumicarb and dithiopyr at 0, 0.01, 0.05, 0.1, 1.0 and 10.0  $\mu\text{M}$ . Root lengths were measured after three weeks. All proflumicarb concentrations completely inhibited root growth in all biotypes. Root growth in the Philadelphia and susceptible standard biotypes were completely inhibited at 0.01  $\mu\text{M}$  dithiopyr. The dithiopyr GR<sub>50</sub> values for the East Brunswick and Manalapan biotypes were 0.05 and 0.02  $\mu\text{M}$ , respectively. This experiment supported field trial observations and demonstrated that the East Brunswick and Manalapan biotypes are resistant to dithiopyr. To explore potential mechanisms of resistance, additional experiments were initiated. The  $\alpha$ -tubulin (*TUA1*) gene of these putative resistant biotypes was amplified, sequenced and fluoresced using specific TaqMan probes for a threonine to isoleucine substitution at position 239. This target site mutation is a common mechanism of resistance to mitotic-inhibiting herbicides, but was not detected in either population. Additional molecular analysis sequenced *TUA1* amino acids 158 to 275 and found no mutations at other positions known to confer resistance. To examine whether enhanced cP450 metabolism is responsible for resistance, a greenhouse dose-response experiment was conducted to evaluate the response of these biotypes to dithiopyr alone and in combination with the cP450 inhibitor piperonyl butoxide (PBO). The East Brunswick and Manalapan biotypes were seeded to pots filled with sand and peat moss (4:1 v/v) and treated with dithiopyr at 0, 10, 50, 100, 500 and 1,000 g ha<sup>-1</sup> alone or in combination with PBO (1.12 kg ha<sup>-1</sup>). Aboveground dry biomass was measured 19 days after treatment. Data were subjected to non-linear regression and a lack-of-fit F-test to determine GR<sub>50</sub> values and the effect of PBO. Dithiopyr GR<sub>50</sub> values were not reduced by PBO suggesting that cP450 enzymes inhibited by PBO are not responsible for resistance. Dithiopyr GR<sub>50</sub> values were 40, 320, and 140 g ha<sup>-1</sup> for the susceptible, East Brunswick and Manalapan biotypes, respectively. To determine if resistance is widespread across the greater New Jersey region, 20 goosegrass biotypes were collected from golf course fairways and athletic fields in New Jersey and Pennsylvania during summer 2019. Plants were collected from sites where turfgrass managers reported preemergence herbicide failures following mitotic-inhibiting herbicide applications. For a preliminary assessment of herbicide tolerance, these biotypes were seeded to pots filled with sand and peat moss (4:1 v/v) and treated with dithiopyr at 280 and 560 g ha<sup>-1</sup> and proflumicarb at 560 and 1120 g ha<sup>-1</sup>. The number of plants were counted at 37 days after

treatment and compared to the non-treated control. For the two susceptible biotypes all herbicide treatments plants counted were  $<2\%$  of the non-treated control. Plants counted were  $\geq 50\%$  of the non-treated control for 11 biotypes treated with  $280 \text{ g dithiopyr ha}^{-1}$ . This research demonstrated that biotypes with resistance to dithiopyr are prevalent in New Jersey. More research to elucidate the mechanism of resistance and understand the resistance profile of biotypes from the greater New Jersey region is warranted.

## **Oxadiazon-Resistant Goosegrass and a Possible New Classification of the PPO-Inhibitor Mode of Action**

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Oxadiazon is an effective preemergence herbicide for goosegrass control in turf. Consistent use over decades of use has led to the selection of oxadiazon-resistant goosegrass. Two populations were first identified in North Carolina and Virginia with increased tolerance to oxadiazon applied preemergence. Subsequent research determined that a postemergence screen could be used to diagnose oxadiazon-resistant goosegrass which decreased time to resistance diagnosis. All previous resistance to protoporphyrinogen-oxidase (PPO) inhibitors was attributed to target-site mutations in mitochondrial localized PPO2. Sequencing of both chloroplast localized PPO1 and PPO2 of oxadiazon-resistant goosegrass identified an alanine to threonine amino acid substitution at position 212 in PPO1 (A212T) and no changes in PPO2. Cloning of PPO1 from oxadiazon-resistant and susceptible goosegrass biotypes into heme deficient *E. coli* allowed for survival of *E. coli* containing threonine-212 resistant PPO1 in the presence of oxadiazon while *E. coli* with alanine-212 susceptible PPO1 did not survive. Structural modeling of PPO1 revealed a change in the binding pocket of PPO1 which repelled oxadiazon from the binding pocket. These data indicated that A212T amino acid substitution in PPO1 was the causal mechanism of goosegrass resistance to oxadiazon.

In 2020, approximately 30 suspected oxadiazon-resistant goosegrass populations were submitted to the Auburn University Herbicide Resistance Diagnostic Lab for screening for possible resistance to oxadiazon. Of the 30 populations, eleven populations were diagnosed resistant to oxadiazon and ten were determined to contain the A212T substitution. Screening will continue in 2021 of possible resistant populations.

Based on previous findings, it was theorized that oxadiazon could be classified more specifically as a PPO1 inhibitor not simply a PPO-inhibiting herbicide. Cloning of susceptible PPO1 and PPO2 from goosegrass into heme deficient *E. coli* revealed greater inhibition of PPO1 transformed *E. coli* than PPO2 transformed *E. coli* by oxadiazon. These data are an early indication that oxadiazon is a specific PPO1 inhibitor and does not inhibit PPO2. However, such differential inhibition may be specific to goosegrass and may not apply to all species. Understanding the biochemistry of PPO resistance remains an active area of research especially with novel PPO inhibitors being developed. Theories of differential localization of PPO1 and PPO2 in different species and variation between eudicots and monocots currently exist and have not been resolved. The discovery of oxadiazon-resistant goosegrass has increased understanding of the PPO-inhibitor mode of action, but it also has raised new questions.

The work presented here is part of PhD student Bo Bi's dissertation at Auburn University. This research could have not been completed without the research collaboration with Aimone Porri, Jens Lerchl, and Micheal Betz of BASF, generous grant and research support from Bruce Spesard and others at Bayer, and grant support from the EIFG, Alabama Golf Course Superintendents Association, and the Alabama Turfgrass Research Foundation.

## Action Thresholds of a Logistic Regression Model for Fungicide Control of Dollar Spot on Creeping Bentgrass

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A logistic regression model that produces a risk index (RI) based on 5-d moving averages of air temperature and relative humidity is currently used by turf managers to guide fungicide applications to control of dollar spot caused by *Clariireedia jacksonii*. Alternate action thresholds of the RI may be needed, particularly for cultivars that are more tolerant of the disease. A field trial was initiated in May 2019 to assess multiple interpretations of the RI output (action thresholds) from a logistic regression model for the control of dollar spot on two creeping bentgrass (*A. stolonifera*) cultivars in North Brunswick, NJ. Each cultivar ('Declaration', more tolerant and 'Independence', more susceptible) was subjected to a calendar-based preventive fungicide schedule, disease-threshold curative schedule, or 15 ( $3 \times 5$  factorial) action threshold schedules based on the RI output from the logistic regression model. The 15 action thresholds included three RI levels (20, 30, and 40%) and five RI-slope levels to apply fungicides. RI-slope was the change in RI over the previous and/or forecasted 5-day period. Fungicide was applied if the RI-slope was positive over the previous 5-day period (PS); positive over the forecasted 5-day period (FS); positive over both the previous and forecasted 5-day periods (PS-and-FS); positive over the previous or forecasted 5-day periods (PS-or-FS); or no RI-slope was considered. Dollar spot disease developed naturally during 2019 and 2020. The number and diameter of infection centers were assessed every 1 to 3 days from May through Nov. each year and used to calculate the area under disease progress curve.

All action thresholds used to schedule fungicide applications on Declaration provided a high level of dollar spot control that was as effective as the calendar-based schedule, in both years of the study. Thus, an RI action threshold as high as 40% (20% is the current model standard) was feasible for the disease tolerant cultivar. The disease response to action thresholds on Independence differed between 2019 and 2020. Disease control on Independence was equally effective at 20, 30, and 40% RI action thresholds in 2019. However, the 20% action threshold controlled the disease better than 30 and 40% RI action thresholds in 2020. Adding RI-slope to the RI action threshold did not improve disease control on Declaration or Independence in either year and intensified disease severity of some action thresholds on Independence in both years. The 15 action thresholds resulted in 4 to 9 fungicide applications per year compared to 9 applications with the calendar-based schedule. There were 2 to 5 and 4 to 7 disease-threshold applications on Declaration and Independence in 2019 and 2020, respectively. Increasing the RI action threshold reduced the number of applications and the level of reduction differed between years. Increasing the RI threshold from 20 to 30% reduced the number of applications by 2 in 2019 but did not reduce applications in 2020. Increasing the RI threshold from 30 to 40% did not change the number of applications in 2019 but reduced 2 applications during 2020. Adding the FS RI-slope condition to a RI action threshold had the greatest impact on annual fungicide use, reducing the number of applications by 2 to 3. When using an action threshold of 40% RI, PS-and-FS RI-slope resulted in the fewest (4) applications each year. This was noteworthy for Declaration where very good disease control was achieved with all action thresholds.

## **Developing Turf Disease Control Programs That are Efficacious and Environmentally Sound**

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Good disease control programs incorporate proper fungicide selection and use with best management practices to optimize plant health, decrease pathogen populations, and reduce fungicide inputs. Recent advances in the development of best management practices for economically important turfgrass diseases such as anthracnose, dollar spot, gray leaf spot, and summer patch have helped turf managers improve disease control while reducing their reliance on fungicides.

There is a systematic approach for developing fungicide programs that are efficacious and environmentally sound. This approach relies on identifying key diseases on a turfgrass sward and the time of year that they typically appear. Understanding the biology and etiology of turfgrass diseases is also important when seeking to optimize fungicide efficacy. Knowing the history of epidemics on the site, as well as disease hot spots, can further aid in targeting fungicide applications rather than spraying indiscriminately over the entire golf course, athletic field, or landscape. When developing a disease control plan, it is important to build the core fungicide program around key diseases. For example, anthracnose, brown patch, dollar spot, and summer patch are typically the focus of core fungicide programs for golf courses in the mid-Atlantic region. Once this framework is in place, modifications can be made in product selection and timing to broaden the spectrum of control to include diseases that periodically occur on the course such as brown ring patch, fairy ring, *Pythium* blight, take-all patch, and yellow tuft.

When constructing a fungicide program, it is important to recognize the strengths (efficacy and diseases controlled) and potential weaknesses (limited efficacy for specific diseases, risk of resistance, potential to cause phytotoxicity) of the available products.

and to incorporate them into a program where they will be most effective. Unbiased product evaluations are available from many universities highlighting the relative efficacy of fungicides against important turfgrass diseases in a region. For example, the Chemical Control of Turfgrass Diseases [PPA-1; PPA-1: Chemical Control of Turfgrass Diseases, 2020 ([uky.edu](http://uky.edu))] is an online publication developed by Extension specialists at the University of Kentucky, Rutgers University, and the University of Wisconsin that provides efficacy ratings of single- and multi-active ingredient (combination) fungicides for the control of important diseases on cool- and warm-season turf. Such information is critically important for the development of an effective fungicide program.

To optimize a disease control program, select fungicides that are efficacious against the key diseases at the site, as well as other diseases that historically occur at various times throughout the year. In the spring, for example, dollar spot and anthracnose are often the only major diseases that occur on golf courses in the mid-Atlantic and northeastern U.S, and they can be easily controlled with well-timed applications of selective fungicides. However, during hot, humid summers, five or more major diseases may occur in these locations requiring the

application of broad-spectrum products consisting of one or more active ingredients to prevent serious disease damage. Disease control can be further enhanced by applying fungicides at the optimum rate, timing, and application criteria (water volume, nozzle type, pressure). Fungicides applied with air induction (AI) or extended range (XR) nozzles that produce medium to medium-coarse droplets are often the most efficacious nozzles for the control of foliar diseases; whereas, nozzles that product large droplets, such as the turf jet nozzle, are typically the best choice for suppressing root diseases. Moreover, optimum control of foliar diseases is often obtained when fungicides are applied in 405-810 L water/ha (1-2 gal. water/1,000 sq ft), whereas fungicides are most efficacious when applied at 810-2,000 L water/ha (2-5 gal. water/1,000 sq ft) for the suppression of root diseases.

Many turfgrass managers apply fungicides on a calendar-basis (every 7, 14, 21, or 28 days). This is particularly true on high value areas such as golf course putting greens. Although this approach may prevent disease outbreaks, it can also result in excessive fungicide use. Using disease predictive models or disease severity thresholds to schedule applications can reduce fungicide inputs and effectively limit disease outbreaks, particularly on more disease tolerant turfgrass cultivars. Disease predictive models have been developed for both root (fairy ring, spring dead spot, summer patch, and take-all patch) and foliar diseases (brown patch, dollar spot and Pythium Blight) and many are currently being used in cloud-based apps and onsite weather stations.

Fungicide resistance has been reported for many chemical classes used to control turfgrass diseases. Fungi can develop resistance to fungicides after repeated use, particularly for products with single-site modes of action including the benzimidazole, demethylation inhibitor, quinone outside inhibitor, and succinate dehydrogenase inhibitor fungicide groups. To reduce the potential for developing fungicide resistance, alternate or apply tank mixtures of fungicides with different modes of action, avoid sequential applications of moderate- and high-risk chemistries, and follow best management practices to reduce disease pressure. Adding multi-site fungicides that have a low risk of fungicide resistance, such as the aromatic hydrocarbon, chloronitrile, dithiocarbamate, mineral oil, phosphonate, pyridinamine, and salicylate chemistries, can also reduce the potential for developing resistance to medium and high-risk fungicides in a disease control program. However, some multi-site fungicides have drawbacks including phytotoxicity during periods of environmental stress, short residual activity, use restrictions and, in some cases, the potential for negative environmental and health impacts. The potential for developing resistance is also affected by the type of turfgrass disease being controlled. The literature is replete with examples of fungicide resistance for diseases such as dollar spot, anthracnose, gray leaf spot and Pythium blight, while few if any cases have been reported for brown patch or diseases caused by ectotrophic, root-infecting fungi. Turf managers should therefore be sure to follow good resistance prevention strategies when using moderate- to high-risk fungicide chemistries to control diseases with a history of fungicide resistance issues.

Finally, before a disease control program is implemented, it should be evaluated for potential impact on the environment. Several integrated models are available to access environmental effects of pesticides. A model developed at Cornell University called the Environmental Impact Quotient (EIQ) measures the impact of pesticides on workers, consumers, and the environment.

Estimates for field use EIQ can be used by turf managers to make informed decisions regarding their pesticide selection.



**POSTER PRESENTATIONS**

## **Physiological Effects of Plant-Health Products for Improving Drought Tolerance and Post-Stress Recovery in Creeping Bentgrass**

Cathryn Chapman and Bingru Huang

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Drought stress due to lack of rainfall and a decreased supply of water available for irrigation can severely limit turfgrass growth and performance. Implementation of deficit irrigation practices, which replace less than 100% water lost due to evapotranspiration (ET), or complete water withholding practices have caused a demand not only for improving drought stress tolerance of turfgrass but also for promoting rapid post-stress recovery. The goal of the summer creeping bentgrass (*Agrostis stolonifera*) fairway study was to evaluate the effectiveness of plant-health products, such as fungicides and plant growth regulators, targeted for improved drought stress tolerance under either complete water withholding (drought) or deficit irrigation (replacing 60% of the water lost due to ET) for 28 d. This study also examined whether or not the products could promote rapid post-stress recovery through resumption of growth upon rewatering. The plant-health products enhanced drought tolerance and post-stress recuperative potential during both a moderate (60% ET replacement) or severe drought stress, as manifested by increased leaf relative water content, turf quality, green canopy density, and leaf area index, as well as reduced stress index. Most notably, the combination of Fluazinam+Acibenzolar with Azoxystrobin+Acibenzolar and Trinexapac-ethyl promoted rapid recovery of turf from drought stress by improving leaf hydration status and green canopy density. The overall improved physiological health and performance observed in this study is a critical component for maintaining sustainable turfgrass stands. Such knowledge highlights the significance of using plant-health products to facilitate the maintenance of creeping bentgrass under sub-optimal irrigation management programs without sacrificing turfgrass function or productivity.

## Herbicide Application Timing Affects Deer-tongue Grass (*Dichanthelium clandestinum*) Control in Native Areas

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Weed control for no-mow, or “naturalized areas” on golf courses can be difficult and often requires extensive manual labor. Deer-tongue grass is a broad-bladed perennial with deep roots and thick rhizomes that is becoming an increasing problematic weed to manage in no-mow fine fescue (*Festuca* spp) areas. Deer-tongue grass is native to the eastern United States and grows copious amounts of lateral branches throughout the summer months, forming a dense canopy that competes with desirable species and reduces the playability of golf course roughs.

Currently, deer-tongue grass must be removed manually or with applications of the non-selective herbicide, glyphosate. However, studies evaluating glyphosate safety for fine fescue injury are inconsistent. The selective herbicide fluazifop has also been shown to provide deer-tongue grass control, but only if applied sequentially on three-week intervals. No research to date has evaluated the effect of application timing on herbicide efficacy for deer-tongue grass control. The objective of this research was to evaluate the efficacy of fluazifop and glyphosate at various application timings for deer-tongue grass control in naturalized areas.

Research was conducted in 2020 from April to October at the Mendham Golf and Tennis Club (Mendham, NJ). Treatments were arranged in a two-by-five factorial, with fluazifop (280 g ha<sup>-1</sup>; with NIS 0.25% v/v) and glyphosate (560 g ha<sup>-1</sup>) applied singly at five different application timings. Treatments were replicated five times and arranged in a randomized complete block design with 2.0 by 3.0 m plots. Herbicides were applied using a CO<sub>2</sub>-powered sprayer and four-nozzle boom equipped with 1103VS nozzles (TeeJet AIXR) and with a carrier volume of 410 L ha<sup>-1</sup>. Application timings were selected using a combination of growing degree-days (GDD; base 10°C), cooling degree-days (CDD; base 20°C), and weed developmental stages visually evaluated on site. Each herbicide was applied singly at 75 and 175 GDD (28 April and 26 May 2020), during spring flowering (18 June), in mid-July (22 July), and at 25 CDD (22 September). Plots were mowed once in October, two weeks after the final treatments were applied. To determine percent deer-tongue grass control, deer-tongue grass injury was visually assessed on a 0 (no control) to 100 (complete necrosis) scale every two to three weeks from May through October. Deer-tongue grass cover was also visually evaluated from 0 (no cover) to 100 (complete canopy cover) in each plot. All data were subject to ANOVA as a factorial using the GLIMMIX procedure (P=0.05) in SAS (v. 9.4).

The effect of herbicide treatment on deer-tongue grass control was significant on each rating date from 3 to 5 WAT and application timing was significant from 9 to 15 WAT, and at 21 WAT. A herbicide-by-application timing interaction for deer-tongue grass control was detected from 14 to 21 WAT and trends were similar on each date. Glyphosate applied at 175 GDD and spring flowering provided greater control (89 and 97%) than glyphosate applied at 75 GDD and 25 CDD (36 and 62%) and all fluazifop treatments (11 to 59%) on 22 October 2020. Glyphosate applied at 25 CDD (22 September) and glyphosate and fluazifop applied mid-July provided 59 to

79% deer-tongue grass control by the final rating in October. Both herbicides were least effective when applied at 75 GDD (26 April), with glyphosate and fluazifop providing 36 and 11% control, respectively at the final rating in October.

## **Rhizobacteria Inoculation and Colonization for Promoting Plant Growth in Cool Season Turfgrass**

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Cool season turfgrass, such as creeping bentgrass (*Agrostis stolonifera*), can be severely affected by drought stress, resulting in leaf senescence, a thinning of the turf canopy, and a reduction in turf quality. These symptoms are caused by an increase in the hormone ethylene in response to drought stress. Plant growth promoting rhizobacteria (PGPR) can co-exist within the roots of plants and in the soil rhizosphere to enhance turf performance and stress tolerance. Some rhizobacteria can produce an enzyme called ACC deaminase, which effectively reduces ethylene levels in plants, thus limiting the impacts of abiotic stress. Our previous research has demonstrated success in using PGPR to inoculate turfgrass in controlled environment conditions; however effective field inoculation is more challenging due to the presence of native soil organisms and fluctuating environmental conditions. These factors can prevent efficient colonization and establishment of the new strains of bacteria, thus limiting their ability to improve plant growth and stress tolerance. This study used a novel combination of growth promoting *Burkholderia* bacteria strains (ACCdR23+14) to inoculate creeping bentgrass field plots maintained at fairway height (1.2 cm). This novel combination of PGPR was mixed into a 0.01% humic acid solution and applied as a soil drench. Non-inoculated control plots and plots treated with a commercially available inoculant were used for comparison. After inoculation, the plots were subjected to 49 days of drought stress (60% ET) using a rain-out shelter, followed by re-watering and a 28-day recovery period. Weekly physiological measurements and digital images were collected throughout the growing season. Treatment with ACCdR23+14 resulted in higher turf quality, percent green cover, normalized difference vegetation index (NDVI), and dark green color index (DGCI) during the drought and recovery periods. These results suggest that this novel combination of PGPR strains has the potential for development as a biofertilizer to improve drought tolerance and reduce water use in creeping bentgrass.

## Applications of the Fungal Endophyte *Epichloë festucae* Antifungal Protein *Efe-AfpA*

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The Class I Clavicipitaceous endophyte *Epichloë* is a genus of fungi that infects turfgrasses, resulting in a variety of benefits for the grass host. Besides the anti-herbivory compounds produced and the altered physiology of the grass, *E. festucae* has been shown to provide fungal disease resistance, a unique benefit of its association with the grass host *Festuca rubra*. Specifically, *E. festucae* provides resistance to dollar spot disease caused by *Claviceps jacksonii*, a very detrimental disease of turfgrasses throughout the world (Clarke et al., 2006).

The *E. festucae* antifungal protein, designated *Efe-AfpA*, is highly expressed in the infected grass host *Festuca rubra* subsp. *rubra* (Ambrose and Belanger, 2012), but purification from plant tissue is not practical since it is a minor protein in the mixed fungal/plant tissue. Moreover, it is not expressed when the fungus is grown in culture. Therefore, we have explored producing the antifungal protein in several established protein expression systems. The antifungal protein has been successfully expressed in the yeast *Pichia pastoris* (Tian et al., 2017), in the bacterium *Escherichia coli*, and now in the fungus *Penicillium chrysogenum*. The objective was to identify a protein expression system that can generate a large amount of active antifungal protein in the simplest way. The *E. festucae* antifungal protein is similar to a protein from *P. chrysogenum*, which is designated PAF (*Penicillium* antifungal protein), and which also has antifungal activity (Marx, 2004). We obtained an engineered PAF overexpression strain of *P. chrysogenum* from Dr. F. Marx (Medical University of Innsbruck, Innsbruck, Austria) so that we could directly compare the activities of PAF and the *E. festucae* antifungal protein.

The antifungal activity of PAF and the *E. festucae* antifungal protein produced in fungi, yeast, and bacteria was compared against *Neurospora crassa* conidia, a model fungus used in such systems. The *P. chrysogenum* produced *Efe-AfpA* and PAF had nearly identical inhibition profiles with peak inhibitions at 1.2 to 5  $\mu\text{g mL}^{-1}$ . The *Penicillium* expression system was the most convenient for purification and produced the highest quantity of the *E. festucae* antifungal protein. To initially confirm activity of *Efe-AfpA* expressed in *P. chrysogenum*, it was tested against *C. jacksonii* (dollar spot) mycelium utilizing Evan's Blue stain. Although *Efe-AfpA* and PAF had similar activities against *N. crassa*, only *Efe-AfpA* had activity against *C. jacksonii*. We are currently working on establishing a greenhouse system for testing the activity of the purified *Efe-AfpA* protein against dollar spot on fine fescue and creeping bentgrass.

With the goal of determining if *Efe-AfpA* could be used as a biocontrol agent, it was first tested against the well-characterized grey mold system in apple. The transgenic *Efe-AfpA* producing *P. pastoris* was tested for efficacy on apple fruit challenged with the postharvest pathogen *Botrytis cinerea*. While both the *Efe-AfpA* and empty vector transgenic yeast strains inhibited grey mold on apples, there was no enhanced inhibition with the strain producing the antifungal protein. However, a high degree of inhibition was observed when *B. cinerea* conidia were challenged with the pure *Efe-AfpA* protein. Additional biocontrol studies with the pure protein will be conducted using the grey mold system.

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## Spatial Distribution of Dollar Spot Fungus in Asymptomatic and Symptomatic Turfgrass

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Dollar spot is one of the most economically important diseases of turfgrasses. A recent taxonomic revision has identified four distinct fungal species within the new genus *Clarireedia* as the causal agents of dollar spot. We developed a quantitative real-time polymerase chain reaction (qPCR) molecular detection assay specific to the *Clarireedia* genus. The qPCR assay was able to detect all *Clarireedia* species tested and did not cross react with non-target fungi and oomycetes. It is capable of detecting as little as 38.0 fg ( $3.8 \times 10^{-14}$  g) of *Clarireedia* genomic DNA in three hours and identified *Clarireedia* in both symptomatic and asymptomatic creeping bentgrass (*Agrostis stolonifera*) tissue. The goal of the project was to determine the distribution of *Clarireedia* in asymptomatic and symptomatic creeping bentgrass maintained at 9.5 mm over two years using our qPCR assay. The abundances of *Clarireedia* were measured in the leaves and crown tissue in May 2019, August 2019, May 2020, and July 2020 from three replicates of 30, 1 cm dia. x 2.5 cm deep cores spaced 10 cm apart. The number of positive detections for *Clarireedia* ranged from 37% (May 2019) to 69% (May 2020) in asymptomatic turfgrass and 77% (July 2020) to 95% (August 2019) for symptomatic turfgrass. The high number of positive detections in both asymptomatic and symptomatic tissues suggests that *Clarireedia* may have an endophytic phase as part of its life cycle. The fact that the pathogen was detected in asymptomatic tissue further suggests that creeping bentgrass may be able to tolerate a certain quantity of the pathogens in leaves (a biotrophic phase) before disease symptoms appear; however, further research is needed to confirm this hypothesis.



## Organic Lawn Clippings Can Feed Livestock and Produce Food for People

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Lawns are valued for beauty, recreation, and protection of soil and water quality. Home lawns are also sometimes vilified as mere “status symbols” with “no agricultural value” that contribute to chemical runoff and water pollution. The trend towards organic lawn care is an attempted correction – to manage lawns and landscapes as an ecological system – much like an idealized organic farming system.

Livestock and pasture are vital components of a well-functioning organic farm plan. They are integral to building soil fertility and producing healthy food to nourish people. Before modern lawn care with mechanical lawnmowers, grazing sheep were employed to manicure grass covered landscapes. The traditional lawn had much in common with the grazing of pasture. Consequently, the same plant species are often used for pasture and turf.

With the advent of the lawnmower grass clippings were no longer harvested by livestock. In modern lawn care clippings are either removed or left shredded in place to recycle organic matter and mineral nutrients. In the case of grazing of pasture, a substantial amount of the minerals once consumed by animals are similarly recycled in place as manure.

Although leaving clippings reduces the need for N fertilization by about half or more there are some disadvantages to this practice. Clipping residue, especially the larger amounts that occur in spring, can distract from the attractiveness of a lawn. Also, walking over recently mowed turf can lead to tracking of clipping residue offsite. On the other hand, harvest of clippings depletes soil fertility, requires extra time and labor, and creates a disposal problem. But harvested clippings may also find beneficial use as compost or garden mulch.

In the case of organic lawn care there may be some advantage to occasional clipping harvest and removal. Because organic lawns typically rely on natural organic fertilizer materials such as compost or manure-based fertilizers, they often supply more P than is needed for lawn maintenance. Many New Jersey soils, including those used for turf, already have soil test P levels above the optimum range. Thus, adding more P from organic fertilizers is sometimes not desirable unless a nutrient management plan can be designed to balance P fertilizer inputs with P harvest by clipping removal.

The objectives of this case study in organic lawn care were to evaluate a proof of concept for spring harvest of an organically managed lawn, fermentation of the fresh clippings, their storage for winter livestock feed, and palatability when offered to goats and beef cattle.

An organic lawn was established in late summer 2014 on the east side of my farmhouse in Ringoes, New Jersey. Since establishment, this lawn continues to be cared for using the principles of organic lawn as would be required for official USDA Certified Organic farming.

On May 10, 2018 and May 15, 2019, lawn clippings were collected by mowing the grass at a height 2 inches when the turf was 4 to 5 inches tall. After the clippings were partially dried for about an hour after mowing, they were raked, collected, weighted, and placed inside plastic bags designed for vacuum seal. A vacuum cleaner with hose was used to withdraw air from the plastic bags. Stored inside these bags, the moist clippings fermented into clipping “silage”. Because the process of fermentation initially off gasses, it was necessary to re-vacuum the plastic bags after two days of fermentation to reestablish the vacuum seal. Once the vacuum seal was permanently established, the lawn clipping silage was stored inside a garage until January when trial feedings were conducted with of goats and cattle.

When the lawn clipping silage was offered to goats and cattle, both species readily consumed and cleaned up the limited amount of the feed that they were offered by free choice. If greater amounts of lawn clipping silage were produced, there is little doubt that goats and cattle would readily consume this feed in the winter months.

In summary, this research project demonstrates proof of concept that it is possible to harvest and store lawn clippings in vacuum bags as fermented feed for winter livestock feed. It also suggests that lawns – sometimes vilified - can in fact be of “agricultural value” when harvested for livestock feed. And furthermore, organic home lawn care can potentially contribute to feeding humanity with help of livestock transformation into nutrient rich meat and milk. Also, from the organic lawn care perspective, the occasional clipping harvest from the spring growth flush, can serve to improve nutrient management balance from organic fertilizer inputs which otherwise tend to oversupply P. Nutrient management planning may use the typical analysis of clippings (N 3%, P 0.4%, K 2% on a dry weight basis) to calculate mineral uptake and harvest. Further research is currently underway to analyze feed value of the lawn clipping silage at a forage testing laboratory.

## Differential Physiological Responses to Heat and Drought Stress for Annual Bluegrass and Creeping Bentgrass

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High temperatures and drought are major limiting factors for the growth of cool-season turfgrasses. Annual bluegrass (*Poa annua*) exhibits more severe summer quality decline than creeping bentgrass (*Agrostis palustris* Huds.) on golf courses where they are co-present. It is unclear whether this decline in *Poa* is attributed more to heat or drought sensitivity. The goal of this study is to better understand the differential physiological responses of *Poa* to heat and drought and compare it to bentgrass so more efficient management strategies can be developed for golf courses with *Poa* greens. Creeping bentgrass, and *Poa* were grown under three treatment conditions: well-watered plants at 22/17 °C (day/night) (control); well-watered plants at 35/30 °C (heat stress); and unwatered at 22/17 °C (drought) in growth chambers for 42 days. Visual turf quality (TQ), canopy temperatures, green canopy cover, leaf chlorophyll content, electrolyte leakage (EL), and leaf relative water content (RWC) were determined weekly during the study. *Poa* exhibited greater extent of decline in TQ, green canopy cover, leaf chlorophyll content, and increases in EL, relative to the control, compared to creeping bentgrass. Additionally, bentgrass recovered faster than *Poa*, having relatively higher TQ, percent canopy cover, RWC, and lower EL upon re-watering following drought stress. Both *Poa* and bentgrass experienced greater signs of physiological damages under drought conditions than under heat stress. Comparing to bentgrass, *Poa* was more sensitive to heat and drought stress, mainly due to stress-induced leaf chlorosis or senescence and membrane damages.

## Genetic Control of Eastern Filbert Blight Resistance in F<sub>2</sub> Generation Hybrid Hazelnut (*Corylus americana* × *C. avellana*) Populations

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Nearly all commercial production of hazelnuts worldwide relies on cultivars of the European species, *Corylus avellana*, and is centered in regions with Mediterranean-like climates. Unfortunately, most European hazelnut cultivars are highly susceptible to the disease eastern filbert blight (EFB), which has largely prevented production in eastern North America. The disease is caused by the fungus *Anisogramma anomala* which is harbored by the wild American hazelnut, *C. americana*. While *C. americana* rarely displays symptoms of EFB, it acts as a natural reservoir of inoculum, spreading the fungus over its native range that spans a wide area east of the Rocky Mountains. Consequently, when a susceptible European hazelnut is planted across much of North America, it inevitably becomes infected by *A. anomala* and later dies from EFB.

Today, EFB resistant European hazelnut cultivars have become available from breeding programs (Botta et al., 2019). They underpin the expansion of acreage in Oregon, and the recent release of cultivars from Rutgers University allows for production in parts of the eastern United States. However, European hazelnuts are limited in their growing range by their lack of severe cold tolerance. They are primarily adapted to moderated climates such as the Willamette Valley of Oregon or the Mid-Atlantic and Great Lakes “Fruit Belt” regions where crops like peaches are grown. In contrast, the wild American hazelnut is EFB-resistant as well as very cold hardy, reaching into Minnesota and Manitoba, Canada, but its small, thick-shelled nuts have little commercial value. Fortunately, it can be hybridized with the European species. Studies have shown that F<sub>1</sub> hybrids between the species can express both EFB resistance and improved cold tolerance, but in general their nut quality traits do not meet commercial standards (Molnar, 2011). Most forms of eastern filbert blight resistance in the wild hazelnut appear to be under multi-genic control (Molnar and Capik, 2012; Revord et al., 2020). In accordance, we have observed that when using a modified backcross breeding approach (European hazelnut as the recurrent parent), most offspring have insufficient EFB tolerance. Thus, an alternative breeding approach is needed to improve EFB-resistance and nut quality traits concurrently with cold tolerance. One possible approach is to intercross unrelated, select F<sub>1</sub> interspecific hybrids with improved nut traits to create what we classify as “F<sub>2</sub>” interspecific hybrid progenies. In these F<sub>2</sub> progenies, given large enough populations, we hypothesize the recovery of a useful proportion of plants that recombine traits for EFB resistance, good nut quality (close to 1.0 gram kernels, >45% kernel, etc.), and improved cold tolerance.

In this study, we evaluated EFB disease response in a large population of F<sub>2</sub> hybrid plants alongside their F<sub>1</sub> parents. Twenty-six different bi-parental crosses were made at Oregon State University (Corvallis, OR) in 2014 and 2015, comprising 30 different F<sub>1</sub> parents selected for improved nut quality in the absence of high EFB pressure. The resulting offspring (n = 1,274) and replicates of their parents (4 trees each) were grown in the field at Rutgers and subjected to

heavy EFB pressure for at least 5 years. Trees were evaluated in winter 2020/21 using a scale of 0 = no EFB to 5 = all stems heavily infected. Data were assembled, with progeny and parent means for EFB response calculated and frequency distribution of EFB scores compiled for each progeny and the population as a whole. Offspring means were then regressed on mid-parent values to provide a preliminary estimate of narrow sense heritability ( $h^2$ ) for EFB response in the  $F_2$  populations.

Results showed that most of the  $F_1$  parent trees expressed moderate to severe EFB with a mean score of 4.01 for the group, although several were shown to be highly tolerant or resistant (EFB score <3.0). Mean EFB scores for the progenies ranged from 0.67 to 4.92 with a total population mean of 2.98. Segregation for EFB response in the  $F_2$  progeny generally followed a frequency distribution curve expected for quantitatively controlled traits, but some progeny skewed toward either high susceptibility or were found to hold an unexpected abundance of disease-free offspring, resulting in a bimodal appearance.  $h^2$  for EFB response was calculated to be 0.64 with an  $R^2$  of 0.234. In general, progeny with one or more parents that had an EFB score  $\leq 3.5$  tended to have a lower mean EFB score than progeny lacking a single EFB tolerant parent ( $\geq 4.0$ ). Transgressive segregation, where individual progeny trees had a much better EFB score than either parent, was widely observed. In many cases, susceptible parents yielded a significant number of resistant offspring; for example, ~15% of the total  $F_2$  population remained EFB free despite only 1 parent of 30, OSU 1299.048, having this same response. The moderately high  $h^2$  estimate supports the wide presence of additive gene action, although to increase accuracy the analysis will be updated with significantly more progeny (in 2021 and 2022) and a regression of weighted family means used to reduce bias in the estimation of  $h^2$  due to uneven progeny numbers and unequal representation of some parents. Overall, this study demonstrates the creation of a large pool of resistant and highly tolerant individuals that can be further selected upon for nut traits and cold hardiness, and thus supports the utility and effectiveness of developing  $F_2$  hybrid hazelnut populations for the future development of more widely adapted hazelnut cultivars.

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## Kentucky Bluegrass Tolerance to Traffic During Summer and Autumn

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The quality of natural turfgrass playing surfaces can be improved with the establishment of traffic stress tolerant turfgrasses. Kentucky bluegrass (*Poa pratensis* L.) is among the most frequently established cool-season turfgrasses on sports fields and other highly trafficked recreational sites throughout temperate climates in the United States. The objective of this field trial was to assess Kentucky bluegrass traffic stress tolerance during summer and autumn. Three replications of 90 entries (including the 2017 NTEP Kentucky bluegrass test) were seeded in September 2017 on a loam in North Brunswick, NJ. During summer 2019, thirty-two traffic passes were applied as a strip across entries using a combination of the Rutgers wear simulator and the Cady traffic simulator during 1 to 24 July (4 pass wk<sup>-1</sup> with each machine during a 4 wk traffic period). The autumn 2019 traffic period consisted of 28 traffic passes; fourteen passes with the RWS and fourteen passes with a pavement roller (1135 kg) during 11 September to 8 October. Uniformity of turf cover (1 to 9 scale) was visually evaluated and digital images were captured after each seasonal traffic period on no-traffic and traffic plots; digital image analysis was used to determine green cover (%). Data were analyzed as a 2 (no traffic and traffic) × 90 (entries) factorial strip-plot design. Traffic reduced uniformity of turf cover and green cover of all entries compared to no-traffic during both seasons. Entries with the best uniformity of turf cover and green cover after summer traffic were A16-17, BAR PP 7K426, BAR PP 7309V, BAR PP 71213, DLFPS-340/3549, PST-K15-172, Jersey (NAI-A16-3), KH3492, PPG-KB 1131, DLFPS-340/3552, PST-K15-167. Entries with the best uniformity of turf cover and green cover after autumn traffic were Barvette HGT, BAR PP 71213, A16-17, and RAD 553.

## Improvements to an Endophyte Detection Kit: A Story of Milk and Phosphatase

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Tall fescue (*Schedonorus arundinaceus* Schreb.) and perennial ryegrass (*Lolium perenne*) are the most important cool-season grasses in the United States, providing the primary ground cover on over 35 million acres. They have versatile uses for livestock feed, various turf purposes, and for erosion control. Most selections contain fungal endophytes, which are well known to improve environmental and biological stress tolerance in grasses, but the alkaloids they produce may be toxic to grazing animals. Fortunately, not all the alkaloids produced by endophytes are toxic to mammals. There is sufficient genetic diversity in the grass endophyte so that plant-endophyte combinations can be selected that produce low levels of the toxic ergopeptine and indolediterpene alkaloids, but still produce adequate peramine and loline alkaloids that are toxic only to insects.

Endophytes can be detected in turfgrass visually by staining and microscopic examination, by immunoblot screening, or by molecular biology techniques such as PCR. Currently, there are two commercially available immunoblot kits (Cropmark Seeds, Rolleston, NZ and Agrinostics, GA USA), which we have used satisfactorily, but are expensive when used to evaluate the endophyte status of the large number of selections produced in a breeding program. Unfortunately, the antibodies used in both commercially available kits cross-react with *Claviceps* spp, and *Sarocladium strictum* (formerly *Acremonium strictum*) resulting in false positives. Our objective was to make the kit more accurate by determining the reasons for this non-specificity. One reason for this non-specificity is that plant and fungal extracts contain endogenous phosphatase activity that reacts with the substrate used by the secondary antibody in the kit to produce a colored spot. Secondly, the concentration of blocking solution provided in the kit is too low. Application of the phosphatase inhibitor levamisole effectively inhibits endogenous phosphatase activity, and an increase of non-fat dry milk from 0.5% (w/v) to 5% (w/v) reduces non-specific protein binding to the nitrocellulose membrane, greatly improving the accuracy of the kit. The addition of 10 mM levamisole plus 5% (w/v) non-fat dry milk reduces the non-specific signal intensity of *Claviceps purpurea* and *Sarocladium strictum* by 97% in the Phytoscreen Immunoblot kit #ENDO7973, from Agrinostics.

## **Metabolic Regulation of $\gamma$ -Aminobutyric Acid during Heat-induced Leaf Senescence in Creeping Bentgrass**

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Prolonged exposure to high temperatures leads to premature leaf senescence in cool-season crops such as creeping bentgrass (*Agrostis stolonifera* L.) and accounts for reductions in turf development and performance. The objectives of this study were to investigate the effects of exogenously applied  $\gamma$ -aminobutyric acid (GABA) on leaf senescence and to determine metabolic factors that are regulated by GABA contributing to the mitigation of heat-induced leaf senescence in creeping bentgrass. Creeping bentgrass was subjected to non-stress (22/18 °C day/night) or heat-stress (35/30 °C day/night) conditions for 35 d in environmentally controlled growth chambers and foliar-sprayed with GABA. Physiological parameters, such as turf quality, leaf chlorophyll content, and photochemical efficiency were quantified, and the content of amino acids was measured. The activity of a key chlorophyll-degrading enzyme, chlorophyllase, was determined. In GABA-treated plants under heat stress, the enzymatic activity of chlorophyllase decreased, while endogenous levels of glutamic acid, threonine, and GABA increased. Our study found that GABA-mediated regulation of leaf senescence under heat stress could mainly be due to regulation of chlorophyll catabolism and amino acids in the GABA shunt pathway.



## Understanding and Optimizing Sampling Methods for the Annual Bluegrass Weevil

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The annual bluegrass weevil (ABW) is the most important and difficult to control insect pest of short-mown golf course turf in eastern North America. Golf course superintendents have relied on synthetic insecticides for ABW management, but excessive insecticide use has led to widespread insecticide resistance to insecticides from several classes. It can be expected that overuse of any remaining effective synthetic insecticides will desensitize ABW to these compounds as well. Ultimately, golf course superintendents have to delay resistance development by applying control products only when and where necessary. That requires monitoring and sampling methods that are easy enough to use and fit into their busy schedules while still having a high predictive power. Currently available monitoring methods monitor the adult or the larval stage.

The quickest and most likely to be used monitoring methods involve sampling adults by vacuuming, soap flushing, or clippings examination. However, various factors are likely to influence the efficiency of these methods, particularly that of vacuuming and clippings examination including temperature and mowing height. The method least likely to be affected by environmental conditions and mowing height is soap flushes where water mixed with liquid dish washing detergent is applied to a specified area which irritates the adult to the surface and up the grass blades where they can be counted. For soap flushing, the effect of water volume and concentration of the detergent on extraction efficiency has yet to be examined.

*Effect of mowing height on extraction efficiency by vacuuming and from mower clippings.* In the first year of study, sampling methods were examined under warm conditions to allow for the optimization of extraction methods. Color-marked adults were released into turf plots about 1 hr before extractions started to allow the adults to settle in and distribute naturally. In lab observations it had been found that the color powder adhered for several days to the adults without interfering with their behavior. In all experiments, adult recovery was tested in areas consisting of mix stands of annual bluegrass and creeping bentgrass mown at fairway (9 mm) and greens (3 mm) heights. After the plots were either mown or vacuumed, adult ABW were extracted from the plots with soap flushes. 500 ml water with 0.4% lemon scented dish washing detergent was distributed within a 30.5 cm × 30.5 cm sampling square at 0 and 5 minutes and adults collected for 20 min.

Recovery of adults in mower clippings from a Toro flex 21" mower was significantly affected by mowing height. Adults were recovered only sporadically in the fairway, and significantly more adults were recovered from the green. However, the total number recovered from clippings and soap extraction was about twice as high from the fairway as from the green. Adults clearly dispersed more quickly from the release area on the green and some may have left the sampling area before sampling started. Relative to the total recovery, recovery from the clippings was only 0.2% from the green irrespective of attachment of a brush in front of the mower basket. At

fairway height, significantly more adults were recovered with the brush (24% of total recovery) than without the brush (15%).

Recovery of adults by vacuuming was also significantly affected by mowing height. Adult recovery did not differ significantly between the treatments with one or two passages with the vacuum. But significantly more adults were recovered from the green than the fairway. As in the mowing experiment, about half as many adults were recovered in total (including soap extraction) from the green as from the fairway, whether plots were vacuumed or not before soap extraction. Soap flushing alone recovered 83% of adults from the fairway but only 42% from the green. This was likely again because of faster dispersal of the adults out of the sampling area on the green. Relative to the total recovery, recovery by vacuuming was 4.5% from the fairway and 31% from the green.

*Optimizing soap extraction efficiency* To optimize the efficacy of the soap-flushing method, we extracted adults of a natural ABW population from fairway height turf using 0.2%, 0.4%, or 0.8% soap solution (lemon scented dish washing detergent). The solution was applied once at the beginning of the 20-minute observation period (0.5 or 1.0 Liter solution per 0.1 m<sup>2</sup>) or at the beginning and again 5 minutes later (both times 0.5 Liter). Adults were collected from the turf surface every 5 minutes.

Extraction efficacy increased with soap concentration, being highest at 0.8% whether applied once or twice at 0.5 Liter per 0.1 m<sup>2</sup>. Soap extraction tended to be more effective when applied twice at 0.5 Liter than applied once at 1.0 Liter; it was also all but impossible to apply the higher volume without significant run-off. In all treatments, at least 75% of the total recovery was reached after 15 minutes, but additional weevils were recovered by 20 min in all treatments. The most effective soap-flush protocol therefore is to apply 0.8% twice at 0.5 Liter after 0 and 5 min, and to observe for at least 15 minutes, better for 20 minutes.

*Effect of temperature on extraction efficiency.* In additional experiments we investigated the effect of environmental temperature on the recovery of natural populations of ABW adults from greens height turf by mowing and soaping. As in previous experiments, much higher (10–45x) numbers of adults were recovered by soap-flushing than by mowing. Temperature (range 44–71 °F) had no effect on soap-flushing extraction efficacy. The number of adults picked up in mower clippings tended to increase with temperature, albeit not statistically significant due to low and highly variable adult counts. Additional experiments will have to be conducted in spring 2021 to attempt to solidify the trend in temperature effect on weevil extraction in mower clippings and investigate the effect of temperature on extraction efficacy by vacuuming.

## Viruses in Fungi That Cause Dollar Spot Disease of Turfgrass

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The objective of this research project is to discover mycoviruses that might perturb development of the dollar spot fungus and cause it to be hypovirulent to various turfgrass species. Dollar spot is a destructive and globally distributed fungal disease that affects a variety of C3 and C4 grass hosts. The fungal genus *Clarireedia* contains species formerly classified as *Sclerotinia homeocarpa* that infect many of the C3 grass hosts and cause dollar spot disease, which is among the most important in commercial turfgrasses in New Jersey.

Many phytopathogenic fungi are known to harbor mycoviruses, some of which measurably affect the biology of the fungal host. Earlier studies in the lab of Dr. Greg Boland, University of Guelph, identified viruses of *S. homeocarpa* that reduced virulence and could potentially be useful for biological control of the fungus. Dollar spot disease has been an emphasis in Rutgers turfgrass research, both from a breeding and management perspective, and studies from JoAnne Crouch and colleagues resulted in the reclassification of dollar spot-associated fungi. This reclassification and associated collection of fungal isolates allowed us to revisit the presence of viruses that may affect biology and virulence of dollar spot fungi.

We are initiating virus discovery by examining isolates of *Clarireedia* spp. used by Crouch and colleagues in the fungus reclassification. We began with a subset of the 40 fungal isolates used in those studies. We first purified double-stranded (ds) RNA, which is the replicative form of single-stranded (ss) and dsRNA viruses and thus indicates presence of those virus types, from 19 isolates of *Clarireedia* spp. The dsRNA was analyzed by gel electrophoresis and putative viral dsRNAs were identified in six of the isolates.

A computational pipeline was then developed for discovery of viral RNA sequences of *Clarireedia* isolates based on small RNA sequencing. RNAs were isolated from 10 fungal isolates, and paired-end RNA libraries were made for total small RNA sequencing. Nine of the 10 libraries were sequenced. Seven of the sequenced fungal isolates belong to *C. jacksonii* and two isolates belong to *C. monteithiana*. Genome sequences are available for both *C. jacksonii* and *C. monteithiana* in the public database, allowing us to map fungal genome sequences from our total RNA sequence reads to the downloaded fungal genome sequences and subtract those mapped sequences. Remaining unmapped sequences are possible viral sequences. *De novo* assemblies and annotations of the assembled unmapped reads was performed by running BLASTx and BLASTp searches to obtain any known or possible viral hits.

Sequences representing three genera of fungal viruses, *Mitovirus*, *Ourmiavirus*, and *Barnavirus* have been identified. Sequence analysis is proceeding, and the putative virus-containing isolates identified to date are being analyzed in the lab for growth characteristics and colony morphology.

## Genome Wide Association Study of Anthracnose Disease in Switchgrass

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Switchgrass (*Panicum virgatum*) has been identified as a model bioenergy crop by the US Department of Energy due to its potential for high biomass yield and wide range of adaptability throughout the United States. A major road block to the widespread use of switchgrass in the Northeastern US is the prevalence of anthracnose disease caused by a novel fungal pathogen, *Colletotrichum navitas*. A genome wide association panel of 528 distinct switchgrass genotypes previously developed by Lu et al., 2013, has been replicated across three field location sites: Philipsburg PA (marginal land), Ithaca NY, and Freehold NJ. This population has been screened by our lab and our collaborators for severity of anthracnose disease ratings and growth parameters during the 2019 growing season. The data presented here is the first of three years of phenotypic data that is to be collected for the association panel. Preliminary data collected from 2019 indicates that the 528 genotype panel approaches a normal frequency distribution in disease response and has substantial genetic variability. There are individuals within this population that appear to have a high degree of anthracnose disease resistance. The identification of markers associated with anthracnose disease resistance and the identification of superior genotypes will improve our understanding of disease resistance and generate improved, anthracnose disease-resistant cultivars that can achieve high biomass yields on marginal land.

## Comparing Relative Virulence Among *Magnaportheopsis* spp. on Hard Fescue and Kentucky Bluegrass Hosts

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Summer patch is a destructive root disease of hard fescue (*Festuca brevipila*) and Kentucky bluegrass (*Poa pratensis*) cool-season turf. *Magnaportheopsis poae* is a well-documented causal organism of summer patch disease, but in recent years, two new fungal species, *M. cynodontis* and *M. meyeri-festuciae*, have been identified from diseased roots of hard fescue and Kentucky bluegrass turf exhibiting typical summer patch symptoms. The objective of this study was to investigate the disease virulence among isolates of *M. cynodontis*, *M. meyeri-festuciae*, and *M. poae* to better understand which of the pathogens, and more specifically, which isolates affect hard fescue and Kentucky bluegrass turf the most. In this study, four isolates of *M. cynodontis*, eleven isolates of *M. meyeri-festuciae*, and eleven isolates of *M. poae* were used. ‘Beacon’ hard fescue and ‘A16-20’ Kentucky bluegrass were seeded into containers inoculated with the fungi and maintained in cool and warm environments. Data was collected weekly and summarized as disease severity index for each container throughout the study. Disease severity index data was used to generate area under the disease progress curve values, which were subjected to analysis of variance. Our results indicate that *M. poae* is more virulent to both hard fescue and Kentucky bluegrass than *M. cynodontis* or *M. meyeri-festuciae*. *M. poae* isolates 37S, BalB\_24, C3, FFSP1\_2, HF2\_2, Lisa9, OakA\_5, P5, StdA\_7, and WilA\_3 and *M. cynodontis* isolate d29740\_4 significantly affected mean AUDPC in this study. Altogether, these findings suggest that *M. poae* contributes the most to summer patch disease in hard fescue and Kentucky bluegrass, but these findings also underscore the activity of *M. cynodontis* and *M. meyeri-festuciae* on these hosts as well. Future studies should consider the potential synergistic relationship among these fungi and investigate the impact that multiple species would have, together, on these host plant species.

## **Inheritance of Summer Patch Disease Resistance in Hard Fescue (*Festuca brevipila* Tracey)**

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Hard fescue (*Festuca brevipila* Tracey) is a cool-season turfgrass known for exceptional performance under low-maintenance conditions but is damaged by summer patch disease. Summer patch is a root disease caused by *Magnaportheopsis poae*. Recently, *Magnaportheopsis meyeri-festucaae* has been identified as another causal pathogen of the summer patch. The objective of this study was to investigate the inheritance of summer patch resistance in controlled crosses of hard fescue populations. The experimental populations were created by crossing three summer patch resistant parents and three susceptible parents in a diallel crossing design. One hundred progenies from each of the 15 crosses and reciprocals were established in a mowed spaced-plant trial in 2017 (Trial 1) and 2019 (Trial 2). All populations, as well as selected parental genotypes, were arranged in a randomized complete block design with four replications. A mixture of an *M. meyeri-festucaae* isolate (SCR9) and an *M. poae* isolate (C11) served as inoculum for both trials. Hard fescue individual health levels, which may be affected by summer patch disease, were assessed by visual rating during the summer of 2018, 2019, and 2020. Estimated narrow-sense heritability of summer patch severity resistance was  $0.67 \pm 0.01$ . The estimation was in the moderate range, indicating the potential of summer patch resistance to be improved via selection and breeding. As the first report of heritability estimates for summer patch resistance in any turf species let alone hard fescue, this research will help to determine the more efficient selection procedures to this resistance.

## Potassium Fertilization Effect on Dollar Spot of Annual Bluegrass

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Potassium is an essential nutrient for the growth and stress tolerance of plants. However, the effect of potassium on dollar spot (caused by *Clarireedia jacksonii*) of annual bluegrass (ABG) [*Poa annua* L. forma *reptans* (Hauskn.) T. Koyama] is largely unknown. A field trial was initiated in 2019 to determine the effect of K fertilization on dollar spot of ABG turf mowed at 2.8-mm. A 4 × 2 factorial arranged as a randomized complete block design with four replications evaluated four levels of potassium (0, 3.4, 6.9, 13.8 kg K ha<sup>-1</sup> applied every two weeks) and two levels of N (urea) applied at 4.9 kg ha<sup>-1</sup> every 7 days or 28 days over 28 weeks. Each plot was inoculated with the dollar spot pathogen in mid-September of 2019 and 2020. Infection centers were counted every 2 to 6 days over 2 weeks and used to calculate the area under disease progress curve for each plot. There was a quadratic increase in the dollar spot response to increased K fertilization rate, regardless of the N level in 2019. However, the K fertilization response depended on the N fertilization level in 2020. Dollar spot did not respond to K fertilization when turf was fertilized with N every 28-d; whereas there was a quadratic increase in dollar spot as K fertilization increased on turf fertilized with N every 7-d. This study will be continued in 2021 to determine if the interaction observed in 2020 can be reproduced.

## Influence of Fungicide Programming and Bentgrass Susceptibility on Dollar Spot Control

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Bentgrass (*Agrostis* spp.) cultivars have variable levels of host susceptibility to dollar spot caused by *Clariireedia jacksonii*. A  $3 \times 6$  factorial arranged as a randomized complete block design was used to assess the effectiveness of curative threshold-based fungicide applications to control dollar spot on six bentgrass cultivars maintained as a fairway turf (0.375-inch, 0.95mm) during 2018, 2019 and 2020. The fungicide factor included a calendar-based program (21-d interval, nine applications) and two threshold programs that applied fungicide at a threshold of 314-mm<sup>2</sup> symptomatic leaf tissue over 3 m<sup>2</sup> observational area either within 24-h or the next spray-day (Monday). The six cultivars evaluated were ‘Capri’ colonial bentgrass (*A. capillaris*), and ‘Declaration’, ‘007’, ‘Shark’, ‘Penncross’ and ‘Independence’ creeping bentgrass (*A. stolonifera*). Dollar spot was measured every 1 to 3 days as the active infection area, which was then log<sub>10</sub> transformed to correct for heteroscedasticity. Transformed data was used to calculate area under each outbreak period (AUOP). The sum of all AUOP for each treatment was then calculated to represent dollar spot severity for a given year. Calendar-based scheduling resulted in better control of dollar spot across all cultivars compared to threshold-based schedules. There was no difference in disease severity between the 24-hours and next spray-day threshold-based schedules for the more tolerant cultivars (Declaration and 007). For Capri and the more susceptible cultivars (Shark, Penncross and Independence), disease was more severe when threshold-based applications were made the next spray-day compared to the 24-hours schedule. Within the calendar-based and 24-hours threshold-based schedules, there were almost no differences in disease severity among cultivars; except for Declaration, which had less disease than Independence (calendar and 24-hour schedules) and Penncross (calendar-schedule). Within the next spray-day threshold-based schedule, disease was more severe on susceptible than tolerant cultivars. Both threshold schedules reduced the annual number of fungicide applications compared to the calendar-based schedule (nine applications) especially on cultivars that were less susceptible to dollar spot. Declaration required as few as 3 and 2 threshold-based applications in 2018 and 2019, respectively.



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