PROCEEDINGS OF THE TWELFTH ANNUAL RUTGERS TURFGRASS SYMPOSIUM

Bruce B. Clarke, Director
William A. Meyer, Associate Director

January 9-10, 2003
Cook College

Symposium Organizing Committee

Bingru Huang, Chair
Bruce B. Clarke
Barbara Fitzgerald
Albrecht Koppenhöfer
James Murphy
James F. White, Jr.

Proceedings of the Twelfth Annual Rutgers Turfgrass Symposium

James Murphy, Diana Corrington and Barbara Fitzgerald, Editors

Director's Opening Remarks:

Welcome to the Twelfth Annual Rutgers Turfgrass Symposium at Cook College/NJAES. The Symposium was established in 1991 to provide Rutgers faculty, students, and staff with an annual forum for the exchange of ideas on a wide range of topics in turfgrass science. The format was expanded several years ago to include presentations by colleagues at other institutions. I would like to thank Dr. Thomas Hsiang, (University of Guelph) for giving this year's keynote address, as well as Dr. Randy Kane (Chicago District Golf Association) and the Center faculty who have agreed to present their research at this year's meeting. I would also like to thank the Symposium Planning Committee, comprised of Drs. Bingru Huang (Chair), Jim Murphy (editor of the Symposium Proceedings), Albrecht Koppenhöfer, Jim White, Diana Corrington and Barbara Fitzgerald, for their hard work in the preparation of this year's program. Without their efforts, this year's Symposium would not have been possible.

This is indeed an exciting time for the Rutgers Turfgrass Program. The Center faculty continue to be recognized for excellence in turfgrass breeding, management, biotechnology, physiology, and pest science. We have also expanded our efforts to provide undergraduate, graduate, continuing professional education, and service programs in support of students and turfgrass managers throughout the United States.

During the past two decades, the Turfgrass Industry has donated over 2.5 million dollars in grants, scholarships and gifts to our turfgrass program. This includes the construction of the new Ralph Geiger Turfgrass Education Complex, as well as several research greens and the 5,000 sq. ft. C. Reed Funk equipment storage facility at Hort Farm II. The Industry has also donated their time and effort to ensure the success of our research field days and educational conferences in the state.

In 2002, the Center and the Department of Plant Biology and Pathology embarked on an ambitious plan to expand our undergraduate teaching program in Turfgrass Management at Cook College. With the help of Dr. Richard Hurley and a $30,000 grant from a private donor, we developed two new recruitment brochures, conducted several open houses for interested students and guidance counselors, and lead numerous tours of our teaching and research facilities. We also attracted four new scholarship donors in 2002 and provided over $70,000 in scholarships to deserving students including, for the first time, targeted scholarships for several outstanding high school seniors. We look forward to continuing our recruitment efforts in 2003.

Thank you for coming to this year's symposium. I hope that you will find it an interesting and worthwhile experience.

Sincerely,

Bruce B. Clarke, Director
Center for Turfgrass Science

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TWELFTH ANNUAL RUTGERS TURFGRASS SYMPOSIUM

Cook College, Rutgers University
January 9 - 10, 2003
Foran Hall - Room 138

Thursday, January 9, 2003

7:00 - 7:30 PM	Registration
7:30 - 7:40 PM	Welcome and Introduction: **Dr. Bruce Clarke, Director - Center for Turfgrass Science**
7:40 - 8:30 PM	Keynote Address: **Dr. Tom Hsiang** (Environmental Biology Department, University of Guelph, Ontario, Canada) *Development of an Effective Biological Control Agent for Turfgrass Snow Molds (From the Laboratory to a Commercial Product)*
8:30 - 10:00 PM	Wine and Cheese Reception

Friday, January 10, 2003

8:30 - 9:00 AM	**Registration, Coffee and Donuts**
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9:00 - 9:20	**Dr. Randall Kane** (Chicago District Golf Association, Oak Brook, IL) *InteractiveTurf.com -- IPM in Action*
9:20 - *9:40*	**Dr. Stacy Bonos** (Department of Plant Biology and Pathology, Rutgers University*) Genetic Analysis of Dollar Spot Resistance in Creeping Bentgrass Crosses and Populations*
9:40 - 10:00	**Dr. William Meyer** (Department of Plant Biology and Pathology, Rutgers University) *Breeding Progess for Resistance to Gray Leaf Spot in Perennial Ryegrass*
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10:50 - 11:10 **Dr. Bingru Huang** (Department of Plant Biology and Pathology, Rutgers University) *Physiological Responses of Cool-Season Turfgrasses to Drought and Heat Stress*

11:10 - 11:30 **Dr. James Murphy** (Department of Plant Biology and Pathology, Rutgers University) *Assessing Cool-Season Turfgrasses for Performance Under Simulated Traffic Stress*

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12:00 - 1:30 PM Lunch and Poster Session

1:30 - 2:30 PM SESSION 2A: TURF AND PEST MANAGEMENT
(Moderator: Dr. James F. White, Jr.)

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1:50 - 2:10 **Dr. Faith Belanger** (Department of Plant Biology and Pathology, Rutgers University) *A New Approach to Dollar Spot Resistance in Creeping Bentgrass*

2:10 - 2:30 **Gengyun Zhang** (Department of Plant Biology and Pathology, Rutgers University) *Use of Somaclonal Variation in Kentucky Bluegrass Breeding*

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3:00 - 4:00 PM SESSION 2B: TURF AND PEST MANAGEMENT

3:00 - 3:20 **Dr. Albrecht Koppenhöfer** (Department of Entomology, Rutgers University) *Wonderworm and No Sex – Bad Times for Grubs*

3:20 - 3:40 **Dr. Randy Gaugler** (Department of Entomology, Rutgers University) *In vivo Production of Entomopathogenic Nematodes*

3:40 - 4:00 **Dr. Stephen Hart** (Department of Plant Biology and Pathology, Rutgers University) *Potential Use of V-10029 for Weed Control in Cool-Season Turfgrass*

4:00 - 4:30 PM Discussion/Closing Remarks

Pre-registered Participants

Ms. Melissa Aa
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Dr. Michael Agnew
Syngenta
302 Rose Glen Lane
Kennett Square, PA 19308

Mr. Ronald Bara
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Dr. Faith Belanger
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Joseph Bischoff
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Dr. Stacy Bonos
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Chris Carson
Echo Lake Country Club
P.O. Box 399
Westfield, NJ 07091

Mr. Jim Castagno
Office of Corp. Licensing & Tech.
Martin Hall
88 Lipman Drive
New Brunswick, NJ 08901

Dr. Richard Caton
New Jersey Turfgrass Association
41 Lupton Avenue
Woodbury, NJ 08096

Mr. Steven Chirip
N.J. Turfgrass Association
HCI Box 54B
Swiftwater, PA 18370

Dr. Bruce Clarke
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Ms. Jo Anne Crouch
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Ms. Michelle DaCosta
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Bill Dickson
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Mike Fidanza
Penn State University - Berks Campus
Division of Science
Tulpehocken Road
Reading, PA 19610

Dr. C. Reed Funk
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Pre-registered Participants

Mr. Eugene M. Fuzy
Dept. of Entomology
Blake Hall - 93 Lipman Drive
New Brunswick, NJ 08901

Dr. Randy Gaugler
Dept. of Entomology
Blake Hall - 93 Lipman Drive
New Brunswick, NJ 08901

Dr. Thomas Gianfagna
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. David Gibson
NaturTek
5013 Miller Trunk Highway
Duluth, MN 55811

Dr. Daniel Giménez
Dept. of Environmental Sciences
20 Ag Way
New Brunswick, NJ 08901

Ms. Bernadette Glasheen
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Alan Habiak
Rutgers University
Plant Science Research Center
594 Halls Mill Road
Freehold, NJ 07728

Mr. Yuanhong Han
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Dr. Stephen Hart
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Dr. Joseph Heckman
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Gerald Henry
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Ms. Priscilla Hockin-Brown
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Dr. Tom Hsiang
University of Guelph
Environmental Biology Dept.
Bovey Bldg. – Room 3227
Guelph, ON N1G 2W1 Canada

Dr. Bingru Huang
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Dr. Richard Hurley
RD #6, Box 6803
East Stroudsburg, PA 18301

Dr. Randall Kane
Chicago District Golf Association
Turf Programs
Midwest Golf House
11855 Archer Avenue
Lemont, IL 60439

Pre-registered Participants

Mr. Matthew Kinkade
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Ms. Polina Kogan
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Dr. Albrecht Koppenhöfer
Dept. of Entomology
Blake Hall - 93 Lipman Drive
New Brunswick, NJ 08901

Mr. Mark J. Krupka
Ecological Laboratories, Inc.
256 Oakshade Road
Tabernacle, NJ 08088

Ms. Cindy Laramore
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. T. J. Lawson
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Ms. Elizabeth Lewis
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Huaijun (Mike) Li
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Dr. Xiaozhong Liu
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Dan Loughner
Dow Agrosciences
497 Leonard Road
Huntingdon Valley, PA 19006

Mr. Darren Lycan
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Dr. Pradip Majumdar
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Marcello J. Mangano
New Jersey Dept. of Agriculture
New Jersey Plant Laboratory
P. O. Box 330
Trenton, NJ 08625

Mr. Dean Marzocca
Dean's Lawn & Landscape
P. O. Box 2128
East Millstone, NJ 08875

Dr. William Meyer
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Ms. Melissa Mohr
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Pre-registered Participants

Dr. James Murphy
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Larry Norton
Bayer Environmental Science
739 Blair Road
Bethlehem, PA 18017

Mr. Mark Peacos
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Pedro Perdomo
Agricultural & Resource Management
P. O. Box 900
Morristown, NJ 07963-0900

Mr. Frank Perrone
Dept. of Plant Biology & Pathology
Foran Hall-59 Dudley Road
New Brunswick, NJ 08901

Mr. Frank Petersen
Rutgers – Food Science
The Nutraceuticals Institute
65 Dudley Road
New Brunswick, NJ 08901

Dr. Karen Plumley
Rutgers - OCPE
Hort Farm No. 2 Laws House
102 Ryders Lane
New Brunswick, NJ 08901

Mr. John Pote
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Dr. James A. Quinn
Dept. of Ecology,Evolution, &
Natural Resources
Plant Physiology Bldg. - 1 College Farm Rd.
New Brunswick, NJ 08901

Mr. Ralph M. Reedy
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Dr. Hiranthi Samaranayake
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Chuck Silcox
Bayer Environmental Science
512 Concord Place
Perkasie, PA 18944

Mr. Dirk Smith
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. James Snow
USGA
P. O. Box 708
Far Hills, NJ 07931

Ms. Monica Torres
Dept. of Plant Biology & Pathology
Foran Hall -59 Dudley Road
New Brunswick, NJ 08901

Mr. Gabriel Towers
Dept. of Plant Biology & Pathology
Foran Hall -59 Dudley Road
New Brunswick, NJ 08901

Pre-registered Participants

Ms. Jennifer Vaiciunas
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Zhaolong Wang
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Eric Watkins
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Eric Weibel
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Ms. Betty Wiest
NJLCA
P. O. Box 604
Ho-Ho-Kus, NJ 07423

Dr. James White
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Ms. Michelle H. Williams
Lesco, Inc.
11 Halstead Street
Clinton, NJ 08809

Mr. John Zajac
Zseeds
P. O. Box 8
Berlin, MD 21811

Dr. David Zaurov
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. Gengyun Zhang
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Mr. George T. Ziemienski
Rutgers University
Plant Science Research Center
594 Halls Mill Road
Freehold, NJ 07728

Dr. Barbara Zilinskas
Dept. of Plant Biology & Pathology
Foran Hall - 59 Dudley Road
New Brunswick, NJ 08901

Plenary Presentations

Development of an Effective Biological Control Agent for Turfgrass Snow Molds (From the Laboratory to a Commercial Product)

Dr. Tom Hsiang, *Department of Environmental Biology*
University of Guelph, Guelph, Ontario N1G 2W1, Canada

For the past eight years, we have been working on a biological control system for snow molds. Gray snow mold caused by the fungi *Typhula incarnata* and *T. ishikariensis* generally occurs in regions with more than 3 months of continuous snow cover. The fungus *Microdochium nivale*, the cause of pink snow mold, does not require long duration of snow cover, and often occurs with gray snow mold. In the past, these winter diseases have been controlled with high doses of heavy metal fungicides such as cadmium or mercury fungicides, or chlorinated hydrocarbons such as pentachloronitrobenzene. As these fungicides lose their registrations, and with increasing public sentiment against the use of synthetic pesticides, we saw a need to develop alternatives for snow mold control.

We began in 1994 searching for better strains of a fungus that could suppress gray snow mold. In the late 1980's, Dr. L. Burpee, formerly at Guelph, and Dr. Now Matsumoto in Japan had independently found that strains of a related fungus named *Typhula phacorrhiza* could inhibit gray snow mold, but neither continued this work. We collected strains of *T. phacorrhiza* from corn fields throughout southern Ontario, and by 1997, we had identified five strains out of several hundred isolates that worked as well at suppressing gray snow mold as conventional fungicides. During the first three years, this research was supported by the Canadian Turfgrass Research Foundation (CTRF) with matching funds from the Ontario Ministry of Education.

In 1998, we started on a new phase of this research with funding from CTRF and Nu-Gro Corporation. This funding was matched by the Natural Sciences and Engineering Research Council of Canada (NSERC) for 1999 to 2004. As a result of increased funding, we were able to expand the study to sites across Canada and begin more intensive work on biological processes involved in suppression as well as providing data for a biopesticide registration package. The same stringent rules which govern the registration of synthetic pesticides also apply to biopesticides. Although *T. phacorrhiza* can be found in abundance in corn fields after spring snow melt, the large majority of these isolates have little or no effect against gray snow mold. Since 1998, we have also tested and seen suppression of pink snow mold by *T. phacorrhiza* in field tests.

The expanded *T. phacorrhiza* trials across Canada from 1999 to 2002 showed varying results, some with strong suppression of snow molds, and others with limited effect. One of the difficulties with winter disease research is that abiotic winter injury is a confounding factor. Also, there were problems in the production process for fungal inoculum, and we have been working to improve quality assurance methods. The scale-up from research plot to full-scale field trials involves greatly increased production of fungal inoculum, and this has been a major obstacle in attempts to commercialize fungal biological control agents. In winter 2000-2001, we developed a method to mass produce inoculum of *T. phacorrhiza*, but the process was considered too risky with the potential for microbial contamination in the opinion of regulatory agencies.

Our continuing work is to develop ways of growing and formulating the inoculum of a select isolate of *T. phacorrhiza* that is antagonistic to both gray snow mold and pink snow mold. We are hoping for a commercially available product within 2 years formulated as a granular that can be applied with conventional equipment. Before commercialization however, there are the major hurdles of quality assurance of mass-produced inoculum, as well as passing the federal regulatory requirements. These hurdles are being addressed by current research which involves close cooperation between CTRF, Nu-Gro and researchers at the University of Guelph.

Some Research Highlights

• **Suppression of gray snow mold by *Typhula phacorrhiza***: In many years of trials at the Guelph Turfgrass Institute, select isolates of *T. phacorrhiza* have been able to suppress gray snow mold to the same extent of protection provided by fungicides such as Daconil (chlorothalonil) and Quintozene (pentachloronitrobenzene).

• **Residual Efficacy of *Typhula phacorrhiza* on creeping bentgrass**: In an experiment lasting five years, we found that a single application of *Typhula phacorrhiza* in the first year could suppress gray snow mold disease for the subsequent three years.

• **Suppression of pink snow mold by *Typhula phacorrhiza***: In trials in Northern Ontario (natural inoculum), Guelph (pathogen inoculated), and in the Rocky Mountains (natural inoculum), pink snow mold was suppressed by a select isolate of *Typhula phacorrhiza* in several years of field testing.

• **Fungicide sensitivity of disease-suppressive isolates of *Typhula phacorrhiza***: In an integrated disease management program, fungicides may also need to be used either before or along with *Typhula phacorrhiza*. Although *Typhula phacorrhiza* is sensitive to the fungicides used for snow mold control, the differences in sensitivity between it and snow mold pathogens may permit fungicides such as Arrest (carbendazim) to be used for gray snow mold caused by *Typhula incarnata*, or benzimidazoles for pink snow mold.

• **Registration Package**: The package was submitted to the Health Canada Pest Management Regulatory Agency (PMRA) in fall 2000, as well as to the U.S. Environmental Protection Agency (EPA) in winter 2001. In spring 2001, we met in Ottawa with the PMRA and with the EPA via teleconferencing. Although specifics of these conversations are confidential, some of the general aspects of this review will be discussed here.

• **Current *Typhula phacorrhiza* trials**: In fall 2002, we established trials across Canada in Alberta (2), Saskatchewan (1), Ontario (4), and Quebec (3). We are hoping for a good long snowy winter!

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Hsiang, T. and C. Wu. 2000. Genetic relationships of pathogenic *Typhula* species assessed by RAPD, ITS-RFLP and ITS sequencing. Mycological Research 104: 16-22.

Hsiang, T., C. Wu and S. Cook. 1999. Residual efficacy of *Typhula phacorrhiza* as a biocontrol of grey snow mould on creeping bentgrass. Can. Journal of Plant Pathology 21:382-387.

Hsiang, T., N. Matsumoto, S.M. Millett. 1999. Biology and management of Typhula snow molds of turfgrass. Plant Disease 83:788-798.

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Wu, C. and T. Hsiang. 1998. Pathogenicity and formulation of *Typhula phacorrhiza*, a biocontrol agent of gray snow mold. Plant Disease 82:1003-1006.

Wu, C., T. Hsiang, L. Yang and L. X. Liu. 1998. Efficacy of *Typhula phacorrhiza* as a biocontrol agent of grey snow mould of creeping bentgrass. Can. Journal of Botany 76:1276-1281.

If you have questions about this research, I can be contacted at thsiang@uoguelph.ca or (519) 824-4120 ext. 52753, fax (519) 837-0442. In addition to the publications listed above, images and summaries are available on the web at: http://www.uoguelph.ca/~thsiang/snow/snow.htm.

InteractiveTurf.com – IPM in Action

R.T. Kane, *Director of Turfgrass Programs* and G. L. Miller, *Manager of Turfgrass Research, Chicago District Golf Association*, and H.T. Wilkinson, *Professor of Turfgrass Science, University of Illinois at Urbana-Champaign*

An operational integrated pest management (IPM) program has been the goal of most golf turf managers since the environmental activism and protests against pesticide use that occurred in the late 1980's and early 90's. However, as it has been practiced, IPM has consisted of little more than efforts to reduce pesticide use or incorporate improved cultural practices to reduce pest impact. Today, with modern environmental monitoring equipment, telecommunications equipment, and the internet, we have the opportunity to assist turf managers in developing and implementing a fully integrated pest management program that can be customized for their site.

Since 2000, we have been developing a web-based IPM system called Interactive Turf (www.interactiveturf.com) that consists of 'on-site' weather data collection, pest predictions based on computer models and local weather parameters, scouting and communication of actual pest occurrences, and providing up to date pest control recommendations based on current research and testing programs. Our goal is to assist golf course superintendents, sod producers, and other turf professionals in making well-informed pest management decisions. The use of the internet as our primary vehicle for communication will also allow turf professionals increased access to research and education resources at both the University of Illinois and the Chicago District Golf Association. This program will also provide users with documented justification of the need for both cultural and chemical management practices to maintain high value amenity turfgrasses.

Genetic Analysis of Dollar Spot Resistance in Creeping Bentgrass Crosses and Populations

Stacy A. Bonos and William A. Meyer
Department of Plant Biology and Pathology, Rutgers University

The dollar spot disease incited by *Sclerotinia homoeocarpa* F.T. Bennet. is an economically important disease of creeping bentgrass golf greens and fairways. As fungicide resistance and pesticide restrictions increase there will be fewer strategies for turf managers to control dollar spot disease. Genetic resistance to dollar spot is a promising alternative to chemical, cultural and bio-control methods. Previous research in this area indicates that dollar spot may be quantitatively inherited. Isolate/host specificities between the pathogen and creeping bentgrass genotypes have also been identified. The objective of this study was to evaluate the disease response and disease progression of resistant x resistant, resistant x susceptible and susceptible x susceptible creeping bentgrass crosses to one isolate of *S. homoeocarpa*. Additionally, a polycross population of creeping bentgrass plants selected for dollar spot resistance was evaluated to determine narrow-sense heritability estimates and number of loci involved in resistance.

In the spring of 2001, dollar spot resistant and dollar spot susceptible genotypes were crossed to develop full sib F_1 progeny. Seed was harvested, dried then treated with KNO_3 to induce germination. Individual seedlings were put in seedling flats and transplanted to the field on 10 Oct 2001. Approximately 50 progeny of each of seven reciprocal crosses along with eight clonal replicates of each parent were established in a randomized complete block (RCB) design. Additionally, in the same field another study was established. This study, also in a RCB design, contained 50 progeny from each of 13 parents inter-pollinated in an isolated crossing block along with three clonal replicates of each parent. The plants in both studies were maintained as mowed tiller plots at approximately 5 cm with a rotary mower. The plants were inoculated with an isolate of *S. homoeocarpa* isolated from 'Crenshaw' creeping bentgrass. Dollar spot disease was evaluated weekly throughout the growing season using a 1 to 9 scale, with 9 representing no disease and 1 representing completely brown turf.

Resistant x resistant crosses resulted in more dollar spot resistant progeny compared to resistant x susceptible and susceptible x susceptible crosses. Variation in disease response and different levels of resistance were observed depending on the particular resistant parents used in the crosses. This indicates that careful selection of parents is necessary in order to assure improved stability of dollar spot resistance in subsequent generations.

Breeding Progress for Resistance to Gray Leaf Spot in Perennial Ryegrass

W.A. Meyer, S. A. Bonos, B. B. Clarke, C. R. Funk, R. F. Bara, D. A. Smith,
M. M. Mohr, E. Watkins, Y. Han
Department of Plant Biology and Pathology, Rutgers University

Since 1996, over 550 collections of perennial ryegrasses and their associated endophytes from Central and Eastern Europe have been evaluated at Rutgers University. Each year the most promising lines were topcrossed with the best Rutgers' Germplasm available being used as the recurrent parent. The Rutgers' Germplasm originated from plants selected out of old turfs in the Northeastern US between 1962 and 1977. This material combined with new germplasm added from other US collections over the years has been cycled each year since the early 1970's as part of a population improvement program for perennial ryegrass. Continued improvements in turf quality, overall disease resistance and persistence have been made in the populations of perennial ryegrass released by Rutgers.

Gray leaf spot (caused by *Pyricularia grisea* (Cooke) Sacc) has become a devastating new disease of perennial ryegrass since the early 1990's in the Northeastern US and other humid parts of the USA. This disease was never observed on the Rutgers Adelphia Research Station in Freehold, NJ before the fall of 2000, making it impossible to breed for resistance. Individual progenies of perennial ryegrass with improved resistance to gray leaf spot were identified in the fall of 2000. Fifteen of the 36 new sources of resistance trace their maternal origin to European collections. The other sources were from the original Rutgers' Germplasm sources. Most of the commercial varieties in this trial were devastated (up to 90%) by gray leaf spot while the most attractive gray leaf spot resistant progenies were damaged less than 5%.

In the spring of 2001 the maternal clonal parents of the most resistant progenies in the 2000 test were placed in isolated crossing blocks according to their plant type and maturity. The individual progenies and composites of the different crossing blocks were seeded on August 17, 2001 at the Adelphia farm. The three most recent populations had gray leaf spot ratings of over 7.7 out of 9 = no disease, while most of the commercial varieties rated from a 5.0 down to 1.5. The 2001 results were similar to the 2000 results for the commercial cultivars but the progeny plots exhibited better resistance as a result of the selection for resistance in 2000. The realized heritability = 0.98 indicates that selection for gray leaf spot resistance should be very effective in improving resistance in subsequent generations.

During the late fall of 2001, the 32 single-plot progenies showing the highest level of resistance to gray leaf spot in turf trials, were sampled and placed in spaced-plant nurseries for another cycle of population improvement in 2002. An additional 33 single-plot progenies were selected and established for backcrossing with the top gray leaf spot resistant populations of ryegrass from the breeding program, using a field-based modified backcrossing program.

Another population of 13 single progenies of perennial ryegrass were selected from the 1999 turf trial. These progenies exhibited significant increases in resistance to brown patch (caused by *Rhizoctonia solani*) during the summer of 2000 and 2001 compared to over 2000 other perennial ryegrass selections. These were also put into spaced plant nurseries for 2002 harvest.

During the spring of 2002 over 993 single progenies were harvested from 12 different populations of perennial ryegrass previously selected for resistance to gray leaf spot. Three different populations consisting of eighty-eight clones were developed from parents exhibiting either gray leaf spot or brown patch resistance, in an attempt to incorporate both traits.

All of these progenies along with over 110 replicated entries were seeded at the Adelphia Research Farm on August 7, 2002. A uniform epidemic of gray leaf spot again occurred in late September. The results of disease resistance on the new sources of gray leaf spot resistant lines compared to susceptible commercial lines were similar to the results of 2000 and 2001. An additional cycle based on the most resistant lines has been started for harvest in 2003.

Suppression of Anthracnose Basal Rot with Nitrogen and Selected Fungicide Chemistries

Bruce B. Clarke, James A. Murphy, and Pradip Majumdar
Department of Plant Biology and Pathology
Rutgers University

Anthracnose basal rot, incited by the fungus *Colletotrichum graminicola,* is a destructive disease of weakened or senescent turf. It occurs throughout the world on almost all turfgrass species but is particularly severe on annual bluegrass and creeping bentgrass. During the last few years, there has been an increase in the incidence and severity of anthracnose basal rot on golf course greens throughout the east coast and mid-western states. In many cases, epidemics have been so severe that fungicides have proven ineffective when used at labeled rates or at recommended intervals of application. Moreover, the trend in the industry toward reducing nitrogen rates on greens to increase playability has further enhanced disease severity.

Prior to the current study, only fungicides within the benzimidazole, strobilurin (QoI), nitrile, and demethylation inhibitor (DMI) classes have been shown to be efficaceous against anthracnose basal rot in the field. Within the past few years, however, several turf managers have reported difficulty controlling this disease with benzimidazole or QoI fungicides, and laboratory studies have recently identified isolates of *C. graminicola* with reduced sensitivity to these two chemical groups. Due to the increased incidence of anthracnose basal rot on golf course greens in the region and the paucity of effective chemical and cultural controls, a field study was established in 2002 to evaluate experimental and currently labeled fungicides for their ability to suppress this disease. A 37 year-old commercial putting green at the Ridgewood Country Club in Paramus, NJ was selected as a study site because it had a previous history of anthracnose and the superintendent had reported difficulty controlling outbreaks with benzimidazole and QoI fungicides. Fungicides representing 12 different chemical classes were applied on a preventive basis at various rates and mixtures every two weeks from 16 May to 19 August. The entire study received a total of 1.5 lb N/1000 sq. ft. in periodic increments throughout the growing season. In addition, the nitrogen source (urea) was applied to one-third of each 3 x 9 plot as a separate split-plot treatment every two weeks at a rate of 0.125 lb N/1000 sq. ft. All chemicals were applied in water equivalent to 2 gal/1000 sq. ft. with a CO_2-powered sprayer. Data was collected for disease severity and turf quality from mid-July to mid-September.

The disease developed naturally on 4 July and became severe and uniformly distributed by 18 July. Disease severity peaked on 10 September (83% turfgrass area infected on non-fungicide treated turf). In general, fungicides within the nitrile (Daconil Ultrex 82.5SDG at 3.2 oz/1000 sq. ft.) and the antibiotic polyoxin-D (Endorse 2.5W at 4.0 oz/1000 sq. ft.) chemical classes provided excellent control of anthracnose (96-100%), compared to non-fungicide treated turf. Of the DMI fungicides, only propiconazole (Banner MAXX 1.3MC at 1.0 fl oz/1000 sq. ft.), tebuconazole (Lynx 45W at 1.11 oz/1000 sq. ft.), and triticonazole (Chipco Triton 1.67SC at 1.0 fl oz/1000 sq. ft.) adequately controlled the disease (98-100% control), whereas myclobutanil (Eagle 40W 1.0 oz/1000 sq. ft.) provided moderate control (80-100% control) and triadimefon (Bayleton 50W at 1.0 oz/1000 sq. ft.) proved ineffective at the rate tested.

The phosphonate fosetyl-Al (Chipco Signature 80WG at 4.0 oz /1000 sq. ft.), the dicarboximide iprodione (Chipco 26GT 2SC at 4.0 fl oz/1000 sq. ft.), and the phenylpyrrole fludioxonil (Medallion 50W at 0.25 oz/1000 sq. ft.) provided good to excellent suppression of this disease (78-100%). As a group, fungicides within the QoI chemical class including pyraclostrobin (Insignia 20WG at 0.5 oz/1000 sq. ft.), azoxystrobin (Heritage 50WG at 0.2 oz/1000 sq. ft.), and trifloxystrobin (Compass 50W at 0.25 oz/1000 sq. ft.) provided relatively poor control of anthracnose basal rot (3-46% control) at this site. The carboximide flutolanil (ProStar 70WG at 2.2 oz/1000 sq. ft.), the dicarboximide vinclozolin (Vorlan 50DF at 1.0 oz/1000 sq. ft.), the dithiocarbamate mancozeb (Fore Rainshield 80W at 8.0 oz/1000 sq. ft.), and the benzimidazole thiophanate-methyl (Cleary 3336 50W at 4.0 and 6.0 oz/1000 sq. ft.) also did not significantly control this disease. In fact, on 55% of the rating dates, flutolanil intensified anthracnose basal rot 17-36%, compared to non-fungicide treated turf.

Although date and fungicide dependent, nitrogen (urea) significantly reduced disease severity. On non-fungicide treated turf, the addition of 0.125 lb N/1000 sq. ft. every two weeks reduced the severity of symptoms 18-36%. Tank mixtures and rotational programs (i.e., applying products from different chemical classes every two weeks) provided excellent disease control that was equivalent to or better than single product entries. Turf quality was closely associated with the severity of anthracnose basal rot. Other than a dark green color induced by the DMI fungicides, no phytotoxicity was observed.

Physiological Responses of Cool-season Turfgrasses to Drought and Heat Stress

B. Huang, J. Larkindale, and M. DaCosta
Department of Plant Biology and Pathology, Rutgers University

Drought and heat stress are two major environmental factors limiting growth of cool-season grasses. Understanding how plants adapt to those stresses would help develop better stress tolerant grasses and effective management programs to maintain quality turfgrass during the stress period. Several studies were conducted to investigate major physiological factors involved in heat tolerance for creeping bentgrass and drought tolerance for Kentucky bluegrass. Specifically, some signaling components such as salicylic acid, abscisic acid (ABA), ethylene, Ca, and hydrogen peroxide were involved in heat responses in creeping bentgrass. All compounds had antioxidant effects. ABA accumulated in roots and shoots in Kentucky bluegrass in response to soil drying, which may serve as a chemical signal to cause stomata closure and water conservation during drought stress.

Assessing Cool-Season Turfgrasses for Performance Under Simulated Traffic Stress

James A. Murphy, Hiranthi Samaranayake, T.J. Lawson,
Joost Den Haan, William Meyer, and Stacy Bonos
Department of Plant Biology and Pathology, Rutgers University

Improved bentgrass cultivars that have greater shoot density and finer leaf texture have been released for use as golf course turfs during the past decade. The objective of this research was to identify bentgrass cultivars that exhibit an ability to resist annual bluegrass invasion under traffic stress. A field experiment was initiated on a sandy loam, which used a two factor split plot design with 4 replications. The traffic factor (non-traffic, wear, compaction, and wear plus compaction) was arranged as main plots. The 15 subplot treatments were cultivars of creeping (*Agrostis stolonifera* L.) or velvet bentgrass (*A. canina* L). The turf was maintained as a putting green turf under a 3 mm cutting height. Plots were evaluated for turf quality, density, and bentgrass population. A significant increase in annual bluegrass invasion was observed in the traffic treatments compared to non-trafficked treatment. Cultivars that produced denser, higher quality turf exhibited good to excellent tolerance to both wear and compaction and suppressed annual bluegrass invasion. Velvet bentgrass has considerably better tolerance of traffic stresses than previously reported.

Numerous species were assessed for turfgrass performance under simulated traffic stress using a wear simulator and compacting roller in 2002. Results with Kentucky bluegrass (*Poa pratensis* L.) indicate there is considerable potential to selection cultivars with good to excellent performance under traffic. It was also evident that the proper selection of a Kentucky bluegrass cultivar for traffic tolerance is dependent on understanding the expected management and mircoenvironmental conditions where the cultivar will be grown and utilized. Data for perennial ryegrass (*Lolium perenne* L.) demonstrated that improved North American cultivars performed better under simulated traffic than European cultivars. Tall fescue (*Festuca arundinacea* Schreb.) cultivars grown for less than one year had good traffic tolerance, however, recovery was slow during summer months. Colonial (*A. capillaris*) and velvet bentgrass have generally exhibited better performance under simulated traffic compared to creeping bentgrass in a number of evaluation trials.

The Possible Roles of Alkaloids and Other Natural Products in Insect Resistance of Turfgrasses

Thomas J. Gianfagna, Rongqi Sun, Jamel Bishop and William A. Meyer
Department of Plant Biology and Pathology, Rutgers University

Endophyte-infected turfgrasses often exhibit superior resistance to insects and disease and may be more tolerant of environmental stress. Turfgrasses that contain endophytes may require fewer pesticide treatments, and usually maintain their appearance longer during periods of drought and heat stress. Insect resistance of endophyte-infected (E+) grasses has been correlated with the production of alkaloids. Peramine, ergovaline, and lolitrem B reduce insect feeding and survival, and these compounds are not produced by endophyte-free (E) grasses. Lolitrem B and ergovaline are harmful to mammals and may be responsible in part for the various toxic syndromes affecting grazing animals that feed on E+ grasses. Peramine, however, is reported to have little affect on mammals.

We analyzed the alkaloid content of ryegrasses collected in Poland, focusing on selections representing six taxonomic groups, as determined by Jim White's lab using DNA sequencing data. We are interested in determining if there may be useful endophytes in these selections, and if the different taxonomic groups also have different alkaloid profiles. All of the selections from each taxonomic group produce significant levels of peramine, however group VI endophytes produced 2- 10X the level of peramine than any of the other taxonomic groups. There were notable differences in lolitrem B and ergovaline levels among endophytes, but these differences were unrelated to taxonomic group. Two of the most interesting of the ryegrass selections are 9828 and 9576. Both of these endophytes are low lolitrem B producers, and have undetectable (less than 5 ng/g) or barely detectable levels of ergovaline. 9576, in taxonomic group I, produces moderate levels of peramine whereas 9828 in group VI had the highest level of peramine. These endophytes should be good candidates for producing ryegrass genotypes that are safe for grazing animals, but are still insect resistant.

Insect feeding trials were conducted with fall army worm (*Spodoptera frugiperda*) second instar larvae (1.5- 2.5 cm in length). All of the E+ grasses inhibited insect feeding compared to E grass controls. There were also significant differences in insect feeding behavior among the E+ grasses. 9828, 9820A, 9733 and 9630 were the most inhibitory: caterpillar growth was poor, mortality high, and frass production low among the survivors compared to control. Selections 9576, 9677 and 9820B were moderately inhibitory: caterpillar growth was limited, there was some insect mortality, and frass production was moderate. All selections from taxonomic group IV were significantly inhibitory to insect feeding whereas the other strongly inhibitory endophytes were scattered among the different taxonomic groups.

There may be other lines of defense against insects in addition to alkaloids. Some insects locate a suitable host by identifying and following the volatile organic compounds

(VOCs) that are emitted by plants. VOCs, however, may also play a role in the repulsion of insects or in the attraction of insect predators. We have characterized the VOC profiles from ryegrasses. The major volatile compound from intact leaves is 3-hexen-1-ol acetate. From mechanically damaged leaves a large number of five to nine carbon saturated and unsaturated alcohols, ketones and aldehydes are produced. When leaves are mechanically damaged and treated with jasmonic acid, in addition to the above named compounds, there were significant amounts of monoterpenes, particularly ocimene and its isomers. Some of the VOCs such as 1-octen-3-ol, 1-hexen-3-ol and trans-2-hexenal were particularly inhibitory to insect feeding compared to controls and other VOCs including 3-hexen-1-ol acetate.

We are currently investigating the VOC profiles of each of the ryegrass selections to determine if the VOCs that are inhibitory to insect feeding are correlated to endophyte taxonomic group.

A New Approach to Dollar Spot Resistance in Creeping Bentgrass

Faith C. Belanger, Stacy Bonos, and William Meyer
Department of Plant Biology and Pathology, Rutgers University

We are investigating the possibility of improving dollar spot resistance in creeping bentgrass through interspecific hybridization with colonial bentgrass. Interspecific hybridization between crop species and related species followed by backcrossing is commonly used to introgress desirable genes into the crop. This method has been used in breeding improved cultivars of numerous species, but has not been used in creeping bentgrass breeding and may offer new opportunities for cultivar improvement. Colonial bentgrass is quite resistant to dollar spot and we may be able to introgress that trait into creeping bentgrass. We have found that interspecific hybridization between colonial bentgrass and creeping bentgrass can occur at low frequencies and the hybrids can be fertile. Some of our interspecific hybrids showed essentially complete dollar spot resistance in the 2001 and 2002 field tests. Backcrosses to creeping bentgrass were carried out in the summer of 2002. From one large cross we have recovered over 1000 progeny. These progeny will be field-tested in 2003. Those progeny continuing to show dollar spot resistance could be used in additional crosses aimed at developing an improved cultivar.

In addition to introgression of the dollar spot resistance trait into creeping bentgrass, the dollar spot resistant hybrids offer opportunities to investigate the basis of resistance to the pathogen. We are pursuing subtractive cloning to see if we can identify genes that are specifically associated with the resistant phenotype. Information from this approach may help in understanding the biological basis of susceptibility or resistance.

Use of Somaclonal Variation in Kentucky Bluegrass Breeding

Gengyun Zhang, Shaoyun Lu, Tseh An Chen, William A. Meyer, and C. Reed Funk
Department of Plant Biology and Pathology, Rutgers University

Somaclonal variation refers to variations occurring in the process of tissue culture and regeneration. Thus, somaclonal variation provides a protocol to generate variability without sexual recombination. Furthermore, if a biotic or abiotic stress factor can be applied in tissue culture process, the somaclonal variation provides a protocol for orientated mutation selection in the laboratory. On the other hand, similar to other mutation breeding protocols, somaclonal variation also has the disadvantages of aberrant variation, progeny segregation and the desired variation usually accompanying other deleterious changes. These disadvantages make somaclonal variation of limited value in most plant breeding projects.

Kentucky bluegrass is a facultative apomictic species. Apomixis makes backcross breeding and recurrent selection, which are the most popular procedures to integrate a new trait into current elite cultivars, impossible. However, apomixis provides the ability to fix variation in one generation. This is an obvious advantage for applying somaclonal variation in Kentucky bluegrass breeding.

In order to explore the potential of somaclonal variation in Kentucky bluegrass breeding, 558 independent regenerated lines from the embryogenic callus of the cultivar "Shamrock" were developed and transferred to a spaced-plant nursery in 2001(436 lines in the spring and 122 lines in the fall, respectively). Nursery results in 2002 indicated that the range of variability was very broad and the frequency of mutation was very high. The variability covered many traits related to turfgrass quality, such as vigor, leaf texture, leaf color, disease resistance, fertility and seed yield. Total somaclonal variation rate (number of R_1 variants / total number of R_1) was over 60%. The somaclonal variation ratio was about 32% on leaf color, 55% on fertility and 22% on resistance to powdery mildew. Very high somaclonal variation ratio on fertility indicates that the successful use of somaclonal variation in Kentucky bluegrass requires large population. From nursery work, 14 lines of plants potential for further selection were selected and put into nursery and turf tests in the fall of 2002. Besides better turfgrass quality, the seed yield of some potential lines was 10-30% higher than our highest seed yielding control.

Lack of good salt tolerant germplasm has largely delayed the genetic improvement of salt tolerance of elite cultivars. Somaclonal variation has been proven effective in obtaining salt resistant mutants. Through somaclonal variation, 40 cell lines of Kentucky bluegrass (cv. Shamrock) which could grow stably on medium supplemented with 0.7% NaCl were selected and 30 regenerated lines were obtained. Ten regeneration lines showed significant improvement for salt tolerance in greenhouse at 1.0-1.2% NaCl concentration. Eight of these regeneration lines also showed significant improvement for drought tolerance. Eleven salt and/or drought tolerance improved lines were put into nursery this fall for additional evaluations.

These results indicate that somaclonal variation promises to be a very useful method in the genetic improvement of Kentucky bluegrass, especially in the improvement of abiotic stress tolerance.

Wonderworm and No Sex – Bad Times for Grubs

Albrecht M. Koppenhöfer and Eugene M. Fuzy
Department of Entomology, Rutgers University

A complex of white grub species are the major turfgrass insect pest in the northeastern United States. The oriental beetle, *Exomala* (=*Anomala*) *orientalis*, has become the most important white grub species in New Jersey and some neighboring areas. Other species that reach damaging population levels include the Japanese beetle, *Popillia japonica*, the northern masked chafer, *Cyclocephala borealis*, and the primarily low-maintenance turf pests, European chafer, *Rhizotrogus majalis*, and Asiatic garden beetle, *Maladera castanea*.

Wonderworm

Steinernema n. sp. (taxonomic description in press) was isolated from epizootics in Japanese beetle and oriental beetle larvae in turfgrass areas in New Jersey. Laboratory studies indicated that this species is rather specialized to scarab larvae as hosts. Thus, *Steinernema* n. sp. was highly pathogenic to and reproduced very well in oriental beetle and Japanese beetle larvae but its pathogenicity to and reproduction in the larvae of four lepidopteran species was mediocre and variable. Pathogenicity to and reproduction in larvae or adults of species from various other families of Coleoptera and other insect orders was generally low to non-existent. *Steinernema* n. sp. is well adapted to infecting sedentary hosts below the soil surface but poorly performs against mobile hosts on the soil surface. *Steinernema* n. sp. caused significant mortality to and reproduced in oriental beetle larvae at 15 to 27.5°C (optimum 17.5 to 25°C).

In a laboratory study (in 30-ml cups with soil and grass), *Steinernema* n. sp. was highly pathogenic to 3rd instars of Japanese beetle, oriental beetle, European chafer, Asiatic garden beetle, and 3 *Phyllophaga* species (May/June beetles). In contrast, *S. glaseri* and *Heterorhabditis bacteriophora* were only very pathogenic to Japanese beetle larvae and showed mediocre to very low pathogenicity to the other above species. All 3 nematodes showed only mediocre pathogenicity to 3 *Cyclocephala* spp. (masked chafers) and low pathogenicity to the green June beetle, *Cotinis nitida*. However, in microplot field trials (at 2.5×10^9 nematodes/ha and 21 DAT), *Steinernema* n. sp. provided good to excellent (71-100%) control of Japanese beetle, oriental beetle, European chafer, Asiatic garden beetle, and northern masked chafer. On the other hand, *H. bacteriophora* provided excellent control only against the Japanese beetle (90%) but no to mediocre (10-50%) control of the other white grub species.

Due to its high pathogenicity and specialization to white grubs, *Steinernema* n. sp. may also be effective for sustainable white grub suppression. To test this hypothesis, we treated 1.5 m^2 turfgrass enclosure containing 10 oriental beetle larvae per 0.1 m^2 at rates of 0, 0.4, 1.0, or 2.5×10^9 *Steinernema* n. sp./ha. At 31 DAT, the oriental beetle populations were examined in 8 turf/soil cores from each plot. Larval populations in the control had not changed, but every single larva recovered in the 3 nematode treatments had been killed by *Steinernema* n. sp. (1.6 to 4.1 infected larvae per 0.1 m^2). Based on our previous field studies, this extremely high efficacy could only have been achieved through additional infections caused by nematodes emerged from hosts infected by the originally applied nematodes. This hypothesis was also supported by the number of *Steinernema* n. sp. that was baited out of soil samples taken from the plots. Compared to the levels observed in samples taken directly after application, the number of *Steinernema* n. sp. baited had remained at the same level at the highest rate, and had increased 4-fold in the medium and lowest rate, even though most of the grub cadavers recovered still contained the nematode progeny. While it is likely that *Steinernema* n. sp. will survive through the winter, the nematodes have already deprived themselves of an opportunity to boost population levels in spring by infecting more grubs. Thus, sufficient survival through summer to infect the next grub generation should be unlikely. Future experiments will therefore have to include even lower nematodes rates.

No Sex

Mating disruption with sex pheromones has been developed as an environmentally safe, non-toxic alternative to broad-spectrum insecticides for several moth species. However, mating disruption technology has not been shown to be effective for insects other than Lepidoptera. The sex pheromone of the oriental beetle consists of a 9:1 blend of (*Z*)-7-tetradecen-2-one and (*E*)-7-tetradecen-2-one. Field studies have indicated that at concentrations > 10 micrograms, (*Z*)-7-tetradecen-2-one alone is as effective as a 9:1 blend containing both. For trapping, rubber septa impregnated with the pheromone were placed in Trécé Japanese beetle traps. The traps were placed in the turf with the funnel near ground level. Experiments in blueberries and ornamentals nurseries have already indicated that application of the pheromone, either as a spray or released from widely spaced macrodispensers significantly reduces catch of male oriental beetles in pheromone-baited traps and oriental beetle larval populations.

In 2002 we conducted the 1st mating disruption study in turfgrass. The plots measured 0.4 to 0.5 ha in size and were separated from each other by at least 90 m distance. Four traps with rubber septa at 300 µg concentration of (*Z*)-7-tetradecen-2-one were placed in each plot to monitor beetle flight and to determine whether pheromone application would reduce the number of male beetles finding the traps. The treatment plots were sprayed with sprayable pheromone at the rate of 50 g a.i./ha applied twice at 14-day interval for a total of 100 g a.i./ha.

Male oriental beetle trap catches compared to the control plots were 98 to 75% reduced at 2 to 15 days after the 1st application and 99 to 84% reduced at 3 to 20 days after the 2nd application. As an additional measure of mating disruption, we placed pots with grass and tethered with virgin female oriental beetles into the plots on 3 separate occasions. The pots were recovered after 3 days, incubated for 10 days in the laboratory before they were searched for eggs. The first 2 sets of virgins were rendered useless due to heavy predation, probably mostly by birds despite bird netting placed over the pots in the 2nd set. The 3rd set was protected with metal cloth cages placed over the pots, and almost all recovered pots contained beetles. The data from this set indicated a 30% reduction in number of females laying eggs. However, these pots had been in the field between 12 and 15 days after the 2nd spray when the effect of the spray, at least with respect to number of male beetles in the traps, had already started to wane. As a third measure of mating disruption we determined grub densities in each plot in September. Unfortunately one of the control plots was treated with insecticide by mistake, making that plot pair useless for this evaluation. However, in the other plot pairs, oriental beetle larval populations in the pheromone-treated plots were reduced by 76% and 60%, respectively.

Oriental beetle mating disruption in turfgrass appears to be feasible. However, the data need to be confirmed. We need to test the effect of typical turfgrass management practices on the pheromone efficacy and whether contamination of shoes or other surfaces that come into contact with the treated turf could result in male oriental beetles being attracted to those surfaces (i.e., ‘bug’ nuisances).

In vivo Production of Entomopathogenic Nematodes

Dr. Randy Gaugler
Department of Entomology, Rutgers University

As the only biocontrol agent available for most turf insects, entomopathogenic nematodes should be poised for wider use as regulatory scrutiny in the USA continues to restrict or ban important chemical insecticides. One component of USA nematode production is a cottage industry of low volume producers using in vivo technology based on a method devised in 1927: the White trap. We report the first scalable system for in vivo nematode mass production. Unlike the White trap, there is no requirement for nematode migration to a water reservoir. The LOTEK system of tools and procedures provides process technology for low-cost, high-efficiency mass production. The system consists of (1) trays to secure hosts during inoculation, conditioning (synchronizes nematode emergence), and harvesting; (2) an automated, self-cleaning harvester with misting nozzles that trigger infective juvenile emergence and rinse the nematodes through the holding trays to a central bulk storage tank; and (3) a continuous deflection separator for washing and concentrating nematodes. The harvester collects 97% of *Heterorhabditis bacteriophora* Poinar that emerged from *Galleria mellonella* (L.) cadavers in 48 h. The separator removes 97.5% of wastewater in three passes, while nematode concentration increased 81-fold. The rearing system offers an increase in efficiency relative to the conventional White trap method with reduced labor and space.

Potential Use of V-10029 For Weed Control in Cool-Season Turfgrass

Stephen E. Hart[1], Darren W. Lycan[1], and Jason Fausey[2]
[1]Department of Plant Biology and Pathology, Rutgers University and
[2]Valent USA Corp., Lansing, MI.

Field experiments were conducted in 2002 in New Jersey to evaluate V-10029 for safety on cool-season turfgrass species and control of annual bluegrass (*Poa annua* ssp. *annua*) and roughstalk bluegrass (*P. trivialis*). All herbicide treatments were applied to mature stands of turf using a CO_2 backpack sprayer delivering 40 gal/A.

Turf tolerance studies were established on June 11, 2002 on Kentucky bluegrass (*Poa pratensis* 'Gnome'), perennial ryegrass (*Lolium perenne* 'Jet'), tall fescue (*Festuca arundinacea* 'Houndog 5'), and Chewings fine fescue (*Festuca rubra* ssp. *falax* 'Shadow II'). Treatments consisted of single applications of V-10029 at 15, 30, 45, 60, and 120 g ai/A. Visual injury data and clipping weights were taken at 35 and 70 d after treatment (DAT). Clipping weights were converted to per cent growth reduction based on the untreated checks. At 35 DAT, injury was evident in Kentucky bluegrass plots treated with 30 g or greater. Visual injury ranged from 8 to 26%, while growth reduction values were 19 to 35%. By 70 DAT growth reduction was still evident in plots treated with higher rates. Perennial ryegrass and tall fescue injury at 35 DAT was minimal and by 70 DAT all plots recovered from initial injury and growth reduction. Fine fescue showed significant growth reduction from rates of 45 g and greater at 35 DAT. A treatment of 120 g resulted in 28% growth reduction at this time. Evaluations at 70 DAT were not taken due to poor fine fescue performance during severe environmental stresses.

Two separate studies were established in the spring of 2002 to evaluate V-10029 for *Poa annua* and *P. trivialis* control in creeping bentgrass (*Agrostis palustris* 'L-93'). Treatments consisted of V-10029 at 15 g ai/A applied once, twice (4 week interval, *P. trivialis* study only), and four times (2 week intervals); 30 g ai/A applied once and twice (4 week interval); and 60 g ai/A applied once. Per cent weed population reduction was calculated per plot from populations present at study initiation and those present at each evaluation date. By 16 d after final application, *P. annua* populations in untreated check plots decreased by an average of 23% due to seasonal fluctuations. Four applications of V-10029 at 15 g or one 60 g application resulted in 34 and 44% *P. annua* population reduction, respectively. Other V-10029 treatments resulted in *P. annua* population reduction values that were not statistically different than the untreated check. *Poa trivialis* population reduction increased with increasing rates of V-10029. Single applications at 15, 30, 45, and 60 g ai/A resulted in 17, 28, 44, and 50% population reduction, respectively, at 8 weeks after treatment. *Poa trivialis* populations in untreated check plots decreased by an average of 14% by this time. Multiple applications of V-10029 resulted in greater population reduction as compared to single applications. Four applications at 15 g resulted in 68% *P. trivialis* population reduction by 16 d after final application. In both studies, creeping bentgrass showed good tolerance to V-10029. Plots receiving two applications within two weeks displayed initial chlorosis levels of 10 to 20% but fully recovered within two weeks.

These studies suggest that V-10029 has the ability to reduce *Poa annua and P. trivialis* populations in established turfgrass. Creeping bentgrass, perennial ryegrass, and tall fescue tolerance to V-10029 applications appears to be acceptable, while the tolerance of Kentucky bluegrass and fine fescue is more questionable.

Poster Presentations

Breeding for Drought Avoidance Characteristics in Tall Fescue and Perennial Ryegrass Populations

Stacy A. Bonos[1], Debra Rush[2], Kenneth Hignight[2] and William A. Meyer[1]
[1]Department of Plant Biology and Pathology, Rutgers University
[2]Advanta Seeds Pacific Inc., Albany, OR

The identification and selection of germplasm with improved drought tolerance will play an important role in developing turfs with better performance and persistence during drought stress periods. The objectives of the study were to: 1) determine the feasibility of simultaneously selecting plants with low shoot: high root ratios and increased root mass in lower soil profiles using selection within flexible tubes under greenhouse conditions, and 2) determine gain from two cycles selection for increased root production within the flexible tubes. Seeds from two populations of tall fescue and perennial ryegrass were germinated on 29.5% (-1.4 MPa osmotic potential) and 28.5% (1.2 MPa osmotic potential) PEG respectively, for each cycle of selection. The vigorous seedlings were transferred to flexible root tubes, 63.5 cm long filled with silica sand, to evaluate for deep root production in the greenhouse. Clippings were collected weekly and root weights determined after approximately 5 weeks of growth in the flexible root tubes. The top 2-4% of the populations were selected for the following characteristics: clipping weights at or below the mean of the population and root weights (in the bottom 30 cm) at least 1 s.d. above the mean of the population. After two cycles of selection, results indicate that gain from selection for plants with deeper root production was approximately 41% in a narrow population and 81% in a broad population of tall fescue and 130% in a turf-type and 79% in a forage-type perennial ryegrass. Root:shoot ratios did not increase in the narrow tall fescue population but did increase in the broad tall fescue and turf-type perennial ryegrass populations. This technique should be very successful in developing turfgrasses with improved dehydration avoidance characteristics. Future studies are underway to see if the improved root growth observed in these selected populations result in improved drought tolerance under field conditions.

Water Use and Requirements for Bentgrasses Under Fairway Conditions

Michelle DaCosta and Bingru Huang
Department of Plant Biology and Pathology, Rutgers University

There is an increasing demand for the use of bentgrasses on golf course fairways. However, water use and minimum water requirements for different bentgrass species under fairway conditions have not yet been determined. Because fairways occupy the majority of the acreage in a golf course, proper irrigation management of these areas is a significant challenge as water becomes increasingly limited for irrigation. Consequently, this study was designed to examine and compare the water use characteristics and minimum water requirements for maintaining quality fairways established to creeping bentgrass (*Agrostis palustris*), colonial bentgrass (*Agrostis capillaris*) and velvet bentgrass (*Agrostis canina*). In order to quantitatively control irrigation and soil moisture under field conditions, this study was conducted in a fully automated, mobile "rainout" shelter that excluded rainfall from test plot areas. The three species were maintained under typical cultural practices for fairways in the Northeast, with irrigation replacement being the main variable manipulated. Water use and drought responses under four irrigation regimes (100, 80, 60, and 40% ET replacement) were evaluated by measuring evapotranspiration rates (ET), canopy photosynthesis rates, water use efficiency, leaf relative water content, and water uptake rates using time domain reflectometry (TDR). Rooting characteristics throughout the experimental period were monitored nondestructively using the minirhizotron imaging technique. In addition, visual quality rating of general turf performance was also evaluated. Soil moisture of control plots, where 100% of ET was replaced by irrigation, did not differ significantly between the three species. In the other treatments where water quantity was reduced, a similar trend for soil moisture depletion rates was observed: velvet bentgrass plots generally had the highest soil moisture, while colonial bentgrass had the lowest soil water content. Based on visual turf quality, it seems that 100% ET replacement is necessary for colonial bentgrass in warmer, summer months. Creeping bentgrass, however, maintained adequate turf quality even at 60% ET replacement. This was also seen in velvet bentgrass, but quality at this irrigation level was maintained at higher levels and for greater duration than for creeping bentgrass. Under cooler temperatures of the fall, the 40% irrigation replacement was sufficient to maintain adequate turf quality for creeping and velvet bentgrass, while 60% replacement was necessary to maintain similar levels in colonial bentgrass. In general, results show that complete replacement of ET may not be necessary to sustain plant growth and physiological processes, and that this depends on species and time of year. This has important implications for water conservation and development of effective irrigation management programs. Other physiological parameters will be discussed.

Using Transposons as Molecular Markers to Examine Variation Among Isolates of *Colletotrichum graminicola*

Bernadette M. Glasheen, Jo Anne Crouch, Bruce B. Clarke, and Bradley I. Hillman,
Department of Plant Biology and Pathology, Rutgers University

Colletotrichum graminicola is the causal agent of anthracnose, a serious disease of turfgrasses in the United States. We have used molecular analysis of several repetitive genetic elements, including a *gypsy*-like retrotransposon and a *pot*-like DNA transposable element, to begin to examine variability among isolates of *C. graminicola*. A genomic library was made from *C. graminicola* isolate Coll-98-5000-5. Screening the library by hybridization and selected sequence analysis led to the identification of clone DBP#16 with homology to Cgret, a long terminal repeat (LTR) retrotransposon of the *gypsy* family from *C. gloeosporioides*. Southern blots were performed using a 476 bp PCR product representing the predicted 5' end of the putative gag (coat protein) gene to probe total *Hind*III digested genomic DNA of 21 isolates of *C. graminicola*. Analysis of the blots indicated either the presence or absence of this element in different isolates. Based on these data, a genomic library was made from Coll-99-5023-1, an isolate that presumably did not contain the Cgret homolog. This library was screened in a similar manner for repeat elements. Sequence analysis revealed one clone, A15, with sequence similarity to both the pol (polymerase) and gag genes of Cgret. Primers were made to amplify a 576 bp fragment to the 5' end of the pol gene. PCR results confirm that this element is present in all 21 isolates of *C. graminicola* and Southern blot analysis using several isolates shows that this transposable element is present in varying number within the genome. Clone1-29 from the Coll-98-5000-5 library was found to have homology to the Pot3 transposon from *Magnaporthe grisea.* A 330 bp PCR fragment representing the predicted 3'end of the putative transposase gene was used to probe Southern blots. Isolates that hybridized to the 476 bp Cgret-like probe also hybridized to the pot3-like probe. PCR reactions on genomic DNA with the primers used to synthesize probe fragments were used to confirm these hybridization results. Based on these experiments, repetitive elements present within the *C. graminicola* genome may provide valuable clues to the evolutionary events that have given rise to the genetic diversity and distribution of this important fungal pathogen.

Nitrogen Mineralization of Grass Clippings – A Case Study in Fall Cabbage Production

Joseph R. Heckman, *Department of Plant Biology and Pathology, Rutgers University*
Uta Krogmann and Lisa S. Boyles, *Department of Environmental Sciences, Rutgers University*

In this three-year field study, grass clippings were applied to fall cabbage to determine the effectiveness of using this common yard waste as a nutrient supplement for crops. Objectives of this research were: 1) To determine the characteristics of grass clippings used in this study; 2) To monitor soil NH_4-N and NO_3-N dynamics in the upper 30-cm soil layer over time; and 3) To determine fall cabbage yield at various grass clippings application rates. The grass clippings used were heterogeneous in their chemical and physical characteristics. Even at the same application rate, soil NH_4-N and NO_3-N concentrations and yields were very different for each year. To avoid over- and underfertilization, targeted N supply from grass clippings should be less than necessary to grow fall cabbage. Additional N fertilizer can be applied based on the PSNT (presidedress soil nitrate test).

Nutrient Management of Land Applied Grass Clippings

Joseph R. Heckman, *Department of Plant Biology and Pathology, Rutgers University*
Uta Krogmann, *Department of Environmental Sciences, Rutgers University*

Research has clearly shown that leaving clippings on the lawn improves turfgrass quality. Nevertheless, grass clippings are sometimes collected for various reasons. The collected grass clippings create an urgent disposal problem for the turfgrass manager because clippings that remain in a pile heat up quickly and cause unpleasant odors. Like other types of yard waste, clippings may be applied to farmland, provided permission is obtained from the NJDEP. The relatively high nutrient content of grass clippings (Table 1) requires that the material be managed as a significant source of nutrients for crop production. The application of clippings to farmland must be accounted for in a farm's nutrient management plan.

In comparison to shade tree leaves, grass clippings are a very different material that require a higher level of management. The low carbon to nitrogen ratio of grass clippings encourages rapid decomposition and release of nutrients. Land spreading of grass clippings that have been stock piled for only a few days can release odors. Ideally, the clippings should be spread on the land the same day as they are collected or at least as soon as possible after collection. Unless the grass clippings are specifically being applied for use as a mulch, they should be incorporated into soil with tillage.

Nitrogen contained in grass clippings is released very quickly when the material is land applied and incorporated. Crops that can utilize this N must be planted shortly after land application. The greatest volume of grass clippings is produced in May and June. Land application of these clippings permits late plantings of field corn, sweet corn, and possibly some late season vegetable crops. When planting these crops, do not apply any broadcast N fertilizer at the time of planting, but an application of starter fertilizer at the rate of 25 lbs N/acre may be applied with the planter. Later in the growing season, when plants are about six inches tall, use the presidedress soil nitrate test to determine if any supplemental N fertilizer is needed.

Grass clippings should be applied at moderate rates to land where soils and crops can benefit from the applied nutrients. If additional nutrients are needed to grow the intended crop, fertilizer can be applied to make up the balance. Application rates of grass clippings should not exceed 12.5 wet tons/acre or 6 dry tons/acre. This is equivalent to spreading 0.5 inch layer of grass clippings over the soil surface. This amount of material would add about 365 lbs/acre of total nitrogen to the soil resulting in an estimated 110-150 lbs/acre of N becoming available to the crop in the first year after application. This amount of clippings would also add about 110 lbs/acre of P_2O_5, 273 lbs/acre of K_2O and 45 lbs/acre S to soil along with organic matter.

Growers who land apply grass clippings should also be aware of some other concerns. Herbicides that are applied to turfgrass may be present in the clippings and have the potential to damage crops. Another concern is the possible presence of pet droppings that may carry pathogens. Research is needed about these concerns.

Table 1. Moisture and nutrient concentrations in municipal collected grass clippings.

	Minimum	Maximum	Average	Average
	--------------------------------- Water % ----------------------------------			
Moisture	34	71	53	
	------------------- Macronutrients, % (dry wt. basis) -------------------			lb/dry ton
Nitrogen	2.34	3.80	3.04	60.8
Phosphorus (P_2O_5)	0.23	0.56	0.40	8.0 (18.3)
Potassium (K_2O)	1.44	2.32	1.88	37.6 (45.5)
Sulfur	0.003	0.005	0.0038	7.6
Calcium	0.67	1.02	0.85	17.0
Magnesium	0.28	0.39	0.33	6.6
	------------------- Micronutrients, ppm (dry wt. basis) -----------------			
Sodium	500	700	500	1.0
Iron	2730	8280	5190	10.38
Aluminum	1760	5710	2860	5.72
Manganese	96	161	128	0.256
Copper	16	55	24	0.048
Zinc	58	155	85	0.17

(Krogmann, et al. 2001)

Fungal Chitinolytic Enzymes are Expressed in Endophyte-Infected P. ampla

Huaijun Michael Li and Faith C. Belanger
Department of Plant Biology and Pathology, Rutgers University

Neotyphodium endophyte-grass associations are naturally occurring mutualistic symbioses. Endophyte-infection generally confers insect resistance, and in some cases disease resistance, to the infected plant. Because of these benefits, endophytes are often incorporated into turfgrass cultivars of several species. The factors that are involved in the establishment of these mutualistic interactions and the mechanisms underlying the endophyte-enhanced traits are the targets of continuing research interest. Apoplastic secreted proteins, both plant and fungal, are likely to be important components of the mutualistic interaction since they are located at the interface of the two species. We have therefore begun investigating some of the proteins secreted in culture and apoplastic proteins isolated from infected plants. An endochitinase was identified by peptide sequencing of an abundant extracellular 52 kD protein expressed in culture. Activity gel analysis using the substrates 4-MU-(GlcNAc)2 and 4-MU-(GlcNAc)3 indicated the 52 kD protein had endochitinase activity.

A full-length cDNA of the endochitinase, Nchi52, had an open-reading frame of 1377 bp. The deduced amino acid sequence of the clone had 37-40% identity to other fungal chitinases, including those of several mycoparasitic and entomopathogenic fungi. The native enzyme was purified from the culture filtrate by binding to colloidal chitin and filtering through a hydrophobic interaction column. The mature enzyme was a 421 aa protein starting with GIHKGKLDG determined by N-terminal sequencing. RNA gel blot analysis revealed Nchi52 is highly expressed in infected P. ampla leaf sheaths. The peptide sequences of NCHI52 were identified as a major component of a 52 kD apoplastic protein band isolated from infected leaf sheaths.

We have also identified another chitinolytic enzyme, an N-acetyl-glucosaminidase that is expressed in infected P. ampla. The enzyme was was partially purified from the culture filtrate and a cDNA clone was isolated. Sequence alignment showed homology with N-acetyl-glucosaminidses/N-acetyl-hexosaminidases from other fungal species. RNA gel blot analysis revealed Nnag60 is expressed in infected P. ampla leaf sheaths. Activity gel analysis using the substrates 4-MU-GlcNAc revealed an N-acetyl-glucosaminidase activity was associated with endophyte infection in both plant crude proteins and apoplastic proteins.

DNA gel blot analysis indicated there is a single copy of Nchi52 and Nnag60 in the fungal genome. Transcripts homologous to both Nchi52 and Nnag60 were also detected in endophyte-infected perennial ryegrass, tall fescue and Chewings fescue indicating that expression of these enzymes may be a general feature of endophyte infection.

With the evidence for expression of fungal endochitinase, N-acetylglucosaminidase, endo--1,6-glucanase, and proteinase within the infected plant we are investigating the hypothesis that these Neotyphodium sp. hydrolytic enzymes located in the apoplastic space of infected plants may function as a mycolytic system for the endophyte, perhaps contributing to the disease resistance observed in some endophyte-infected plants.

Physiological Responses of Creeping Bentgrass to High Soil Temperature

Xiaozhong Liu and Bingru Huang
Department of Plant Biology and Pathology, Rutgers University

Growth and turf quality of creeping bentgrass (*Agrostis palustris* Huds.) often decline during summer season, due to high temperatures. Determination of the primary physiological processes which cause heat injury in shoots and roots would help us to understand the mechanisms of heat tolerance. Our objectives of this study are to compare the sensitivity of water status, nutrient uptake, and cytokinin/ABA synthesis of roots in response to high soil temperature; and to determine the major root processes regulating leaf senescence. Roots of creeping bentgrass (cv. Penncross) were exposed to high soil temperature (35^{o}C) regime while shoots were kept at optimum temperature regime (20^{o}C). High soil temperature significantly decreased root biomass and increased root mortality in 10 days. High soil temperature also decreased turf quality, shoot growth, shoot photochemical efficiency, and shoot relative water content in 15 d while decreased shoot chlorophyll content in 10 d. Results indicated that high soil temperature caused root injury first, and then shoot injury. The disruption of root functions would lead to leaf senescence and decline in turf quality.

Identification and Isolation of Unique Seed Storage Proteins in *Lolium perenne* and *Lolium multiflorum* for the Creation of an ELISA Diagnostic Assay

Marcello J. Mangano and Glenn W. Freeman, Ph.D.
New Jersey Department of Agriculture, P.O. Box 330, Trenton, NJ 08625
New Jersey Plant Laboratory

Reversed-Phase High-Performance Liquid Chromatography (RP-HPLC) of cereal grain proteins has received much attention in recent years as a quick and efficient means of cultivar identification. A recent study of perennial ryegrass (*Lolium perenne* L.) and annual ryegrass (*Lolium multiflorum* Lam.) demonstrated that RP-HPLC could be adapted for the identification of *L. perenne* and *L.multiflorum*. Chromatograms of different cultivars of the two species studied all had distinct peak patterns, which correlated with each kind. Seed storage protein analysis of single seeds accurately differentiated annual ryegrass seeds from perennial ryegrass seeds. Mixtures of ryegrass seeds were analyzed and compared. A lower limit of 10% annual ryegrass in perennial ryegrass was detected by monitoring changes of area under the peaks of the chromatograms. RP-HPLC was shown to be a quick, repeatable and reliable method of ryegrass species identification for a general screening of seed lots. Further work using polyacrylamide gel electrophoresis (PAGE) was used to differentiate blends of annual and perennial ryegrass. In this research, seed storage prolamin proteins were specifically targeted. The results show that the prolamin zymogram patterns of annual and perennial ryegrass seeds exhibited interesting taxonomic variations. In addition, prolamin protein electrophoresis also provides a means of isolating and purifying specific peptides that can be used in the production of highly specific monoclonal antibodies needed for use in an ELISA immunoassay An ELISA test for determining the presence of annual ryegrass contamination in perennial ryegrass may provide greater sensitivity than current methods and would become a useful tool for seed testing laboratories.

Protein Changes in Response to Increasing Temperatures in Agrostis Species

John Pote and Bingru Huang
Department of Plant Biology and Pathology, Rutgers University

Turfgrass stands decline during extended hot periods in the summer months. This study was conducted to compare responses of protein sysnthesis and expression to high temperatures between bentgrass species, geothermal bentgrass (Agrostis scabra), creeping bentgrass (*Agrostis palustris*) and velvet bentgrass (Agrostis canina), and different cultivars of creeping bentgrass. The genothermal bentgrass was collected from the geothermal areas in Yellowstone National Park that grow in soil temperatures over 40 C. Creeping bentgrass cultivar 'L-93' and geothermal bentgrass leaves were subjected to 40 C for 3 h and resulting protein expression was visualized by SDS-PAGE. Roots from Penncross and L-93 were subjected to 40 C for different times. Radiolabeled amino acids were added to the root assays and incorporated into newly synthesized proteins. Seasonal protein content of velvet and creeping bentgrasses were sampled and analyzed by SDS-PAGE and Bradford assay. Velvet bentgrass had higher protein content during summer stress. Commassie stained gels indicate creeping bentgrass proteins are degraded during periods of thermal stress. Heat shock proteins were expressed in creeping bentgrass root extracts. Geothermal grasses were heat tolerant and had distinct protein expression relative to creeping bentgrass. Further study of geothermal *Agrostis scabra* may provide heat tolerance mechanisms and gene(s) for breeding purposes.

Predicting Saturated Hydraulic Conductivity for Putting Green Root Zone Mixes

Sanju Raturi[1,2], Daniel Giménez[1], James A. Murphy[2], H. Samaranayake[2],
and T.J. Lawson[2], *Departments of Environmental Sciences[1] and Plant Biology and Pathology[2], Rutgers University*

Prediction of saturated hydraulic conductivity (Ksat) is needed for turfgrass management. Models have been developed for soil Ksat, but there is a paucity of information on their performance in putting green root-zone mixes which are typically coarse material dominated by large inter-particle pores. A power law to effective porosity (air-filled porosity at -33 kPa) has been used to predict soil Ksat, with the exponent (pore size distribution index, PDI) determined from a water retention curve in the range between -33 kPa and -1500 kPa. The model is completed with an empirical linear constant. The objective of this study was to test and adapt the power law model to turfgrass root-zone mixes. Five sand materials, one of them amended with organic and inorganic materials at various rates were investigated. Values of PDI were empirically derived from soil water retention data in the range from -2 kPa to -6 kPa and effective porosity was defined as air-filled porosity at incremental values from -2 kPa to -6 kPa. Linear constants were calculated at the various effective porosities using measured values of Ksat. Values of the linear constant as a function of particle size distribution, amendment type, and the choice of the lower limit of effective porosity will be discussed.

Root Mass Relationships with Physical Properties of Sand Root Zones

Hiranthi Samaranayake[1], James A. Murphy[1], Josh A. Honig[1], T. J. Lawson[1], and
Daniel Giménez[2], *Departments of [1]Plant Biology and Pathology and*
[2]Environmental Sciences, Rutgers University

Physical property criteria are used to select construction materials for golf course putting greens; however, relationships between physical properties and root mass are not well known especially across microenvironments. The objective of this field study was to investigate the relationship between root mass and physical properties of root zones over several years in two microenvironments. Root zones with different sand particle size distribution were arranged in a randomized incomplete block design nested over two microenvironments. All root zones were amended with sphagnum peat at 9:1 (v/v) and established to a creeping bentgrass (Agrostis stolonifera L. 'L-93') putting green turf in 1998. Cultural management of plots was typical for putting green turf in the northeastern United States. Irrigation was applied as needed to avoid severe drought stress on an individual plot basis.

Plots were sampled in the fall of 1999, 2000, and 2001 for soil physical properties and root mass at the 0- to 76-mm depth zone. Relationships between root zone physical properties and root mass were assessed using regression and principle component analysis.

Root mass development has not responded to the range of root zone physical properties assessed in the study. Microenvironment had a more effect on root mass development than physical properties of root zone over time

Influence of Nitrogen and Liming on the Incidence and Severity of Gray Leaf Spot in Perennial Ryegrass

Gabriel W. Towers and Bruce B. Clarke
Department of Plant Biology and Pathology, Rutgers University

Gray leaf spot, caused by the fungus *Pyricularia grisea*, has emerged as one of the most devastating diseases of perennial ryegrass in the United States. Though recent studies have shown that increased nitrogen rate can result in greater disease severity, little is known about the impact of nitrogen source and the frequency of fertilizer applications on disease development. In two concurrent studies conducted in 2001 and 2002, the effect of nitrogen source alone and in combination with lime on the incidence and severity of gray leaf spot, soil pH and turf quality was assessed on three-month old 'Palmer II' perennial ryegrass maintained at North Brunswick, NJ.

In the first study, ammonium sulfate, calcium nitrate, urea, IBDU, and milorganite fertilizers were applied at weekly (12.2 kg N ha^{-1}), bi-weekly (24.4 kg N ha^{-1}), or monthly (48.8 kg N ha^{-1}) intervals each year from June to September. In 2001 and 2002, plots treated with ammonium sulfate or urea sustained significantly more disease than untreated plots. On turf treated with calcium nitrate, IBDU and milorganite, gray leaf spot was not significantly different from the untreated check. Although the total amount of nitrogen applied per growing season was the same, only weekly applications of urea in 2001 yielded a greater incidence and severity of gray leaf spot than either bi-weekly or monthly applications of the same fertilizer. No such response was evident in 2002. When applied on a monthly basis in 2001, only turf treated with IBDU exhibited more disease at the high (48.8 kg N ha^{-1}) versus the low N rate (24.4 kg N ha^{-1}); while in 2002, there was no significant rate response for any N source. Turf quality was not significantly different among treated plots.

In the second study, the acidifying fertilizer ammonium sulfate and the alkalinizing fertilizer calcium nitrate were applied bi-weekly (24.4 kg N ha^{-1}) alone or in combination with a single application of lime (99, 198, and 396 kg CCE ha^{-1}) or elemental sulfur (845 kg S ha^{-1}) each year from July to September. In 2001, disease severity was significantly lower on turf that received calcium nitrate versus ammonium sulfate. Compared to calcium nitrate alone, calcium nitrate + lime (at any rate) had no effect on pH or disease. However, compared to AS alone, ammonium sulfate + the high rate of lime (396 kg CCE ha^{-1}) significantly reduced disease severity and increased pH to levels comparable to the untreated check. Sulfur treated plots sustained very little disease, but sulfur significantly reduced soil pH and lowered turf quality, compared to the untreated turf. Although turf was inoculated four times with *P. grisea* (50,000 conidia/ml^{-1}), disease did not develop in 2002 on this site. It appears that gray leaf spot is enhanced by low pH (<6.5), however, further research is needed to confirm this hypothesis.

Response of Eight Bentgrass Cultivars Maintained Under Selected Mowing, Nitrogen and Fungicide Practices to Dollar Spot

Jennifer Vaiciunas, James A. Murphy, and Bruce B. Clarke**,**
Department of Plant Biology and Pathology, Rutgers University

The objective of this study was to assess the susceptibility of eight bentgrass cultivars (Crenshaw, L-93, Penn A-4, Penn G-2, Penncross, Southshore, SR 1020, and SR 7200) to dollar spot, caused by *Sclerotinia homoeocarpa*, and to identify management factors that can reduce fungicide inputs in the field while maintaining acceptable turf quality. The main plots included two cutting heights (0.356 and 0.953 cm) and two nitrogen levels (10.1 and 25.2 g N m^{-2} yr^{-1}) and were split into eight randomized subplots (cultivar) and six fungicide (chlorothalonil) sub-subplots (untreated, 7, 14, 28, or 56 d intervals, or an economic threshold of 0.3% disease). The fungicide timing and cultivar factors and their interaction had the greatest influence on disease development and accounted for the majority of the variation within the 3 yr study. There was an inverse association between the frequency of fungicide application and disease severity. Penn G2, SR 7200, and L-93 were least susceptible to dollar spot under most nitrogen, cutting height, and fungicide treatments, whereas Crenshaw and SR 1020 usually sustained the most disease.

In 1999, disease severity was high and dollar spot was more severe at the low rate of nitrogen and the high height of cut. In 2000, the dollar spot epiphytotic was low and disease incidence was generally independent of nitrogen rate and cutting height, except for Crenshaw which consistently had more disease at the low cutting height. Turf quality was greatest at the high nitrogen rate and was inversely associated with the incidence and severity of dollar spot. SR 7200, Penn A-4 (1999 and 2000 only), Penn G2, and L-93 exhibited the highest turf quality. Turf density was greatest at the low cutting height, high nitrogen, and more frequent fungicide treatments, whereas turf color was best at the high cutting height and high nitrogen levels. Although the level of disease control afforded by the threshold schedule was often similar to or better than the 14 d calendar schedule, it resulted in an average yearly reduction in fungicide usage of 18% in 1999, 70% in 2000, and 40% in 2001, compared to the 14 d spray interval.

Influence of Foliar Absorption, Post-treatment Irrigation, and Application Methodology on the Control of Turfgrass Diseases with Trifloxystrobin

Eric N. Weibel, Steve E. Hart, and Bruce B. Clarke
Department of Plant Biology and Pathology, Rutgers University

Trifloxystrobin (Compass 50WG) is a new strobilurin-based fungicide that controls a broad range of turfgrass diseases. It belongs to the Q_OI group of fungicides which block electron transport at the quinol-oxidizing (Q_O) site in the bc_1 complex of the fungus, ultimately limiting ATP production. Several experiments were performed to examine the activity and efficacy of trifloxystrobin in cool-season turfgrasses.

Laboratory studies were conducted to determine the rate of foliar absorption of trifloxystrobin in Kentucky bluegrass. To assess foliar absorption, individual blades (five to seven wk old) were spotted with a 2 micro-L spray solution containing Carbon-14, radio-labeled trifloxystrobin. Blades were rinsed with 80% acetonitrile and rinsate was collected (0, 0.17, 0.5, 1, 2, 4, 8 and 24 h post-treatment) and used to calculate percent foliar absorption. In all experiments, there was a rapid increase in foliar absorption between 0.17 and 1 h. Further increases in the rate of absorption were not observed beyond 4 h. Foliar absorption peaked between 28-32% of the total product applied. In the greenhouse, Kentucky bluegrass was grown in 3.8 cm diameter plastic conetainer pots for five to seven wk and sprayed with trifloxystrobin at a rate of 300 g a.i. ha^{-1}. Treated turf was irrigated with 400 ml H_2O pot^{-1} at 0, 0.08, 0.17, 0.5, 1, 2 and 4 h post-treatment to determine the influence of foliar wash-off on the control of powdery mildew (*Erysiphe graminis*). Conetainers of untreated plants were placed around irrigated plants to assess volatilization of trifloxystrobin. Post-treatment irrigation did not affect fungicide efficacy. Even plants irrigated 0.08 h post-treatment provided nearly complete control of this disease. However, untreated plants directly surrounding trifloxystrobin-treated tissue (i.e., up to 2 cm from treated foliage) sustained significantly less powdery mildew than did untreated checks.

Field studies were conducted from 1999 to 2001 to assess the impact of nozzle type (Flat Fan, Rain Drop Drift Reduction, and Turbo Flood Jet) and water volume (200, 400, 800, 1600 and 3200 L ha^{-1}) on the control of brown patch (Rhizoctonia solani) with trifloxystrobin in ‘SR7100’ colonial bentgrass. Trifloxystrobin (230 g a.i. ha^{-1}) was applied every two weeks from June to August. In all years, efficacy was generally independent of nozzle type. Moreover, the lowest water volume (200 L ha^{-1}) was always the least efficacious. Over this same time period, the impact of clipping management on brown patch was studied for both untreated turf and turf treated with trifloxystrobin. Trifloxystrobin (0, 150, and 380 g a.i. ha^{-1}) was applied to ‘Coronado’ tall fescue every three weeks from June to August and clippings were either collected or returned to plots every three to four days. In 1999 and 2000, returning treated clippings to plots significantly reduced disease severity, compared to the removal of treated or untreated clippings. Moreover, returning fungicide treated clippings reduced disease severity 25 to 49%, compared to returning untreated clippings. In 2001 disease severity was unaffected by clipping management.

Turfgrass Collection from Central Asia

David E. Zaurov[1], James A. Murphy[1], C. Reed Funk[1], William A. Meyer[1], Khasan Ch. Buriev[2], Ruslan A. Astanov[2], Usman Norkulov[2], and Ishimbai Sodombekov[3]

[1]*Dept. of Plant Biology & Pathology, Cook College, Rutgers University*
[2]*Tashkent State Agrarian University, Uzbekistan*
[3]*National Academy of Sciences of the Republic of Kyrgyzstan, Kyrgyzstan*

The genetic improvement of turfgrass species by the introduction, evaluation, and incorporation of desirable traits from unique accessions from around the world has proven a valuable strategy. Germplasm collection from Central Asian region has been in the past largely coordinated by Dr. David Zaurov. These efforts have been very valuable, as we have been able to collect many turgrass accessions from this region. Rutgers University now has formal ties with the Republic of Uzbekistan and the Republic of Kyrgyzstan, and with the Tashkent Agrarian University, the Shreder Institute, the Plant Industry (Vavilov) Institute, the Vegetable Institute, the Forestry Institute, the Botany Institute and the Kyrgyz Agrarian Academy, which includes five Institutes.

Our focus continues to be on collecting potentially shade tolerant grasses, grasses that appear productive in otherwise marginal, overgrazed lands, and grasses resistant to heat, diseases and insects. Such an effort not only provides an excellent opportunity to collect turfgrass beat also allows Rutgers to take a leading role in procuring a vast array of other promising germplasm.

Currently, germplasm from Central Asian region is not well represented in the US collections.

From last year trip (2002), we have collected and brought back to Rutgers 956 accessions of turfgrass from Central Asia (Table 1).

Table 1. Turfgrass species collected from Central Asia, 2002.

Country	Species	Number of Accessions
Kyrgyzstan	*Poa pratensis*	*2*
Kyrgyzstan	*Lolium perenne*	4
Kyrgyzstan	*Festuca rubra*	7
Kyrgyzstan	*Festuca pratensis*	*7*
Kyrgyzstan	*Poa angustifolia*	*3*
Kyrgyzstan	*Festuca sulcata*	*8*
Uzbekistan	*Poa Pratensis*	*439*
Uzbekistan	*Lolium perenne*	*489*
	Total accessions collected in 2002: 956	

As part of this grant, we also co-sponsored turfgrass field trials in Uzbekistan. This is the first turfgrass program in Central Asia, and the field plots were well received. We were able to provide to our Tashkent partners turfgrass seeds for these trails and provided technical assistance in setting up the field plots. Results of this trial are published in an international agronomy journal of Uzbekistan.

Drs. W. Meyer, R. Funk, J. Murphy, and D. Zaurov hosted small Uzbekistan as well as Kyrgyzstan delegations (in 1999, 2000, 2001, and 2002). During this period, a special mini-training program was developed and tours were set-up to show them how to establish a turfgrass collection, nursery, and breeding program. This visit also provided Rutgers with the opportunity to develop potential new programs in Uzbekistan and Kyrgyzstan.

The joint turfgrass project between Rutgers University and Tashkent State Agrarian University was approved by the Ministry of Agriculture and Water Resources of the Republic of Uzbekistan and USDA. Duration of the project is 2003-2005. The total funding of the project is $60,000.00.

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