THE USGA®

1998 Turfgrass and Environmental Research Summary

1998
Turfgrass and Environmental Research Summary

Submitted By:

The United States Golf Association
Golf House
Far Hills, New Jersey 07931

The user of the 1998 Turfgrass and Environmental Research Summary is responsible for determining that the intended use of pesticide information is consistent with the label of the product being used. Use pesticides safely. Read and follow the label directions. The information given herein is for research and educational purposes only. Reference to commercial products or trade names is made with the understanding that no discrimination is intended and no endorsement by the United States Golf Association is implied.

This summary is prepared each year by the USGA Green Section. Dr. Michael P. Kenna, Research Director, and James T. Snow, National Director, assemble and edit the annual progress reports submitted by USGA-sponsored researchers. If you would like additional information about the research program, please write to:

USGA Green Section
Research Office
P.O. Box 2227
Stillwater, OK 74076
(405) 743-3900
FAX: (405) 743-3910
Email: mkenna@usga.org

Table of Contents

Integrated Turfgrass Management *(continued)*

Turfgrass Germplasm Enhancement 33

Turfgrass Germplasm Enhancement *(continued)*

Environmental Impact of Golf ... 53

Wildlife Links Program ... 63

List of Tables

List of Figures

Turfgrass and Environmental Research Program

In 1998, the United States Golf Association embarked on a new five-year Turfgrass and Environmental Research Program. This program employs science as the foundation to benefit golf in the areas of turfgrass and resource management, sustainable development, and environmental protection. This report summarizes the results from the first year of research effort.

There are two primary goals of the research program. The first is to develop turfgrasses and cultural systems with enhanced stress tolerance and reduced supplemental water requirements, pesticide use and other costs. Fifty research projects initiated in golf course construction practices, integrated pest management, and turfgrass germplasm enhancement address the USGA's first research goal.

The second goal is to investigate environmental issues and sustainable resource management for golf courses. Fifteen research projects that investigate the environmental impact of golf courses and wildlife management issues are underway.

The USGA Turfgrass and Environmental Research Program actively coordinates and supports research, associated educational programs, and other partnerships to benefit golf, the environment, and people. For example, the USGA cooperates with the National Fish and Wildlife Foundation to conduct the Wildlife Links program. Wildlife Links projects focus on the research concerning wildlife management and habitat issues. The USGA and Golf Course Superintendents Association of America (GCSAA) co-fund five projects involving putting green construction. The USGA, GCSAA and National Turfgrass Evaluation Program (NTEP) together have developed turfgrass variety testing programs conducted on golf courses throughout the United States.

If you have any questions about the projects summarized in this annual report, please contact:

Dr. Michael P. Kenna
Research Director
PO Box 2227
Stillwater, OK 74076
mkenna@usga.org

The USGA Green Section Turfgrass and Environmental Research Program

Vision

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

Mission

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

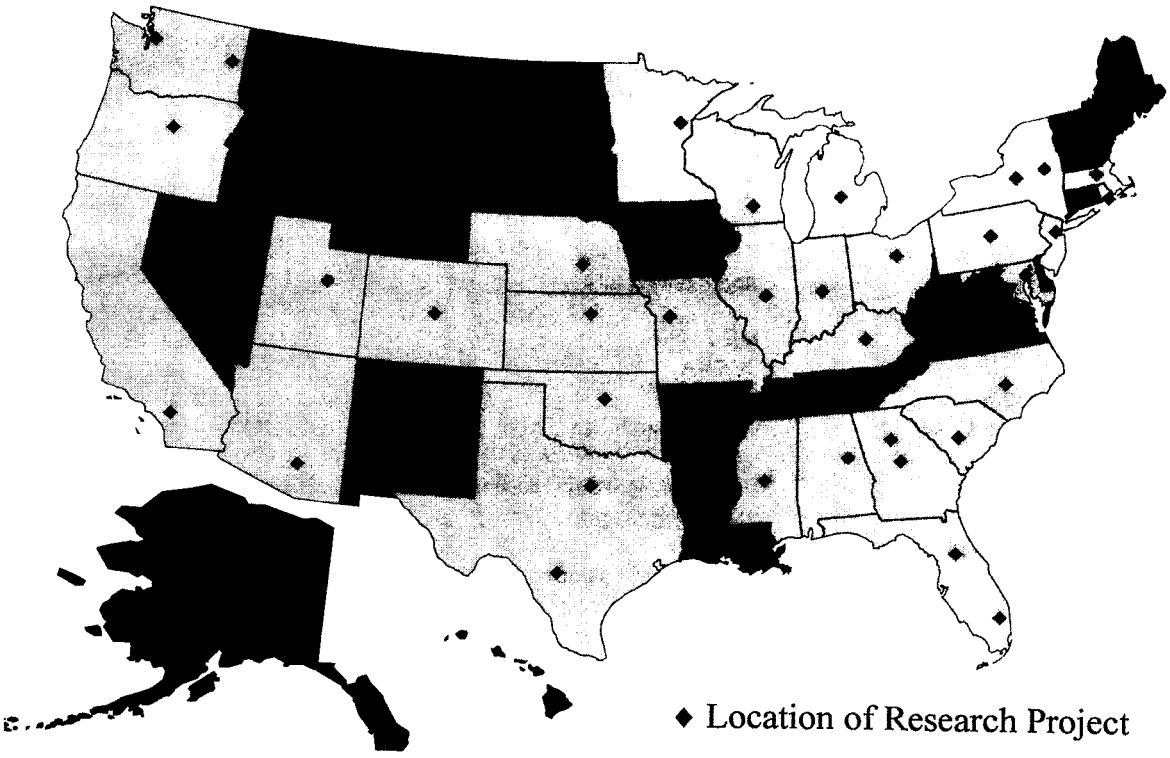
Goals

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

- *Course Construction Practices*
- *Integrated Turfgrass Management*
- *Turfgrass Germplasm Enhancement*

Investigate environmental issues and sustainable resource management for golf courses.

- *Environmental Impact of Golf Courses*
- *Wildlife And Habitat Management*



The United States Golf Association

Turfgrass and Environmental Research Program

Research Goal	Projects		Funding	
	Number	% of Total	Grant $	% of Total
Course Construction	14	21.5%	178,982	14.4%
Integrated Turf Management	14	21.5%	321,409	25.8%
Germplasm Enhancement	22	33.8%	520,656	41.8%
Pesticide/Nutrient Fate	10	15.4%	231,540	18.6%
Wildlife Links	5	7.7%	170,778	13.7%
Total	**65**		**1,244,383**	

1999 USGA Turfgrass and Environmental Research Committee

Patricia P. Cobb
2104 Executive Park Drive
Opelika, AL 36801
v: 334-745-4358 f:
patcobb@mindspring.com

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

Joe England
P.O. Box 58
Hungtington, WV 25706
v: 304-526-4720 f: 304-526-4747
mcdrules@aol.com

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

Ali Harivandi
University of California
Cooperative Extension
1131 Harbor Bay Parkway, #131
Alameda, CA 94502
v: 510-639-1271 f: 510-567-6813
maharivandi@ucdavis.edu

Rees Jones
Rees Jones, Inc.
P.O. Box 285
Montclair, NJ 07042
v: 973-744-4031 f: 973-744-1044
rjonesinc@aol.com

Michael P. Kenna
United States Golf Association
Green Section Research
P.O. Box 2227
Stillwater, OK 74076
v: 405-743-3900 f: 405-743-3910
mkenna@usga.org

Jim Latham
P.O. Box 2130
Whitney, TX 76692
v: 254-694-6252 f: 254-694-6273
jlatham@hillsboro.net

James Moore
United States Golf Association
Green Section Construction Education
720 Wooded Crest
Waco, TX 76712
v: 254-776-0765 f: 254-776-0227
jmoore@usga.org

Jeff L. Nus
GCSAA
Research
1421 Research Park Drive
Lawrence, KS 66049-3859
v: 785-832-4429 f: 785-832-3665
jeffnus@gcsaa.org

Jamie Ortiz-Patino
80 Grosvenor Street
London W1X 8DE
United Kingdom
v: 011-44-171-753-0073
f: 011-44-171-753-0074
joylopez@redestb.es

Charles Peacock
North Carolina State University
Crop Science Department
1215 Williams Hall
Raleigh, NC 27695
v: 919-515-7615 f: 919-515-7959
peacock@cropserv1.cropsci.ncsu.edu

Gerald Pepin
Pickseed West, Inc.
P.O. Box 888
33149 Highway 99 East
Tangent, OR 97389
v: 541-926-8886 f: 541-928-1599
Jerry@PickSeed.com

Paul Rieke
Michigan State University
Department of Crop & Soil Sciences
East Lansing, MI 48824
v: 517-355-0266 f: 517-355-0270
rieke@pilot.msu.edu

Robert Shearman
University of Nebraska
Department of Horticulture
377 Plant Sciences
Lincoln, NE 68583-0724
v: 402-472-0022 f: 402-472-8650
rshearma@unlinfo.unl.edu

James T. Snow
United States Golf Association
Green Section
P.O. Box 708
Far Hills, NJ 07931-0708
v: 908-234-2300 f: 908-781-1736
jsnow@usga.org

David Stubbs
European Golf Assoc., Ecology Unit
51 South Street
Dorking, Surrey RH4 2JX
United Kingdom
v: 011-44-01306-743288
f: 011-44-01306-742496
ega.golf.ecology@dial.pipex.com

James Watson
3 Larkdale Dr.
Littleton, CO 80123
v: 303-794-5346 f: 303-794-5346
ellen.watson@toro.com

Tommy Witt
StillWaters
1816 StillWaters
Dadeville, AL 36853-5630
v: 256-825-5549 f: 256-825-4469
tomwitt@ix.netcom.com

Teri Yamada
Royal Canadian Golf Association
Golf House, R.R. #2
Oakville, Ontario L6J 4Z3
Canada
v: 9058499700 f: 905-845-7040
yamada@rcga.or

Course Construction Practices

To take advantage of new ideas and technologies, and to address the environmental and economic challenges of the coming decades, the USGA in cooperation with the GCSAA, has sponsored research on golf course construction and establishment. Preference was given to research studies that address issues related to:

- Alternative construction methods
- Materials testing procedures
- Calcareous sands
- Effects of irrigation water quality on the selection of construction materials
- Various organic and inorganic matter sources
- Soil-based construction
- Putting green irrigation versus surrounds
- Addition of secondary and micronutrients

An interdisciplinary approach is expected, including the disciplines of soil physics, soil chemistry, soil microbiology, turfgrass physiology and management, and turfgrass pathology. The studies give due consideration to environmental issues, and to the use of alternative water sources for golf course irrigation.

Engineering Characteristics and Maintenance of Golf Putting Greens

Michigan State University

Dr. James Crum

Start Date: 1996
Number of Years: 5
Total Funding: $115,000

Objectives:

1. *Create an experimental design matrix of various sands.*
2. *Determine friction angles for each of the six sands.*
3. *Determine the bearing capacity for each of the six sands.*
4. *Develop trends relating grain size and gradation to friction angle and ulimate bearing capacity.*

This project is a continuation of previous putting green construction project conducted at Michigan State University. This project has allowed for application and expansion of the previous two years of research. Results indicate that putting green stiffness increases with sands that have higher coefficients of uniformity. In addition, it has been shown the turfgrass roots add significant strength and stiffness to the root-zone sand. Field-testing continues to show variation in stiffness for putting greens constructed on sands that meet the USGA gradation guidelines. Further field testing will make it possible to predicted stiffness based on laboratory data within some degree of certainty. From this, we will develop guidelines that superintendents can utilize to design a sand mixture that will achieve desired results and still meet USGA guidelines.

Laboratory Testing: In order to ensure consistency of measurements in the laboratory, six sands were produced rather than selecting market sands. These sands were made from commonly available construction sand (MDOT 2NS) which has a wide range of particle sizes. Three different gradations of sands were designed, a coarse, intermediate and fine. Each of these three classifications was again divided into a high coefficient of uniformity (C_u) and a low coefficient of uniformity (C_u).

A direct measure of a soil's strength against failure under surface compression is its bearing capacity. This can be directly tested in the lab with the Modified California Bearing Ratio (CBR) testing device (ASTM 1883). This device has a small plunger that is forced into a sample volume of sand. Attached to the plunger is a load cell that records the force pushing down on the soil sample. The depth the plunger has punctured into the soil can then be measured to determine the amount of force necessary to cause failure within a soil. Figure 1 indicates the pressure as a function of piston displacement. The peak of the test graph designates the ultimate pressure that the soil can withstand before it fails. The bearing capacity test was run approximately 290 times on the sand samples under all types of conditions.

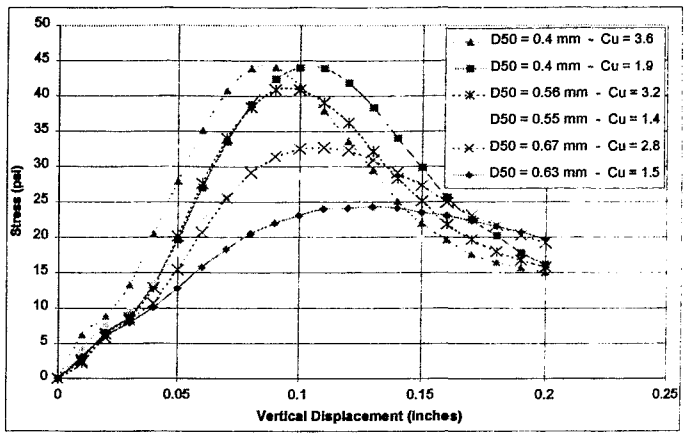


Figure 1. Bearing Capcity of sands with different coefficients of uniformity (cu) meeting USGA Specifications.

The bearing capacity tests also show the benefits of sands with a high coefficient of uniformity (C_u). The well-graded sands were capable of withstanding an ultimate pressure on the order of 45 psi. The poorly graded sands, under the same conditions, could only withstand pressures up to 25 psi. This is below the tire pressure found in some golf course maintenance vehicles and indicates that a golf putting green may suffer deformation during normal servicing. It should be reiterated that although these sands display such a wide variety between their ultimate bearing capacities, they all fall within USGA gradation specifications and would be considered acceptable sands for golf putting green construction.

Field Testing: The field CBR device is designed to model the California Bearing Ratio testing device. The field CBR device can be attached to a three-point hitch or loading bucket of most tractors. The device has a plunger that is forced into the ground. A load cell measures the force on the plunger directly. This force is recorded with the corresponding vertical displacement of the plunger into the ground, measured by a dial gauge on a reference beam.

The force on the load piston divided by the area of the load piston gives us the stress on the surface of the putting green. Force is recorded at every 0.01 inch of displacement for consistency. The stress (force) at each 0.01-inch of displacement is plotted versus the vertical displacement as shown in Figure 2. The initial part of the curve, is the stress on the thatch layer that offers little resistance to deformation. However, the underlying sand-based root zone can take significantly more stress with less deformation than the overlying thatch.

The slope of a line drawn tangent to the curve, k_s, is the stiffness of the green and is referred to in geotechnical engineering as the modulus of subgrade reaction. For example, a green with k_s of 100 pounds per square inch would be displaced 0.10 inches under a load of 10 psi.

As the putting green is loaded and then unloaded, some consolidation of the thatch and sand occurs. For example, one

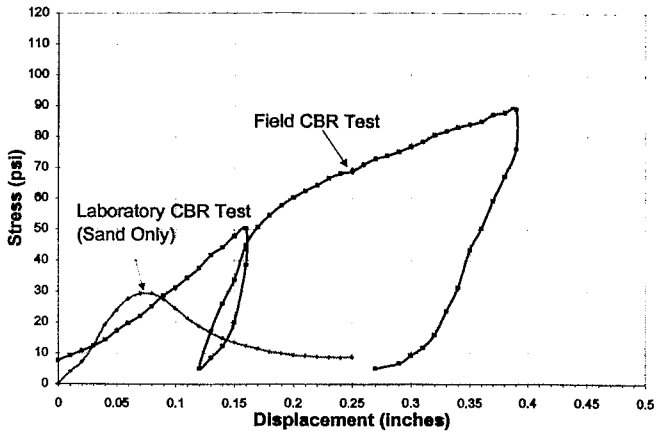


Figure 2. Laboratory and Field California Bearing Capacity (CBR) tests performed at Michigan State University.

sand and thatch consolidated approximately 0.12 inches when subjected to a 50-pounds per square inch load. When reloaded, the stress-displacement curve followed the same line back to 50 pounds per square inch stress since the thatch and sand have already 'felt' that stress. Beyond 50 pounds per square inch, the thatch and sand experience new, higher stresses, and will continue to consolidate until the sand begins to fail. Engineers often refer to the load and reload curve as an elastic rebound curve.

A problem associated with testing existing golf putting greens to evaluate the stiffness of the sand root-zone is separating the contributing strength of the root system. Referring to Figure 2, it is clear that the same soil tested in the field with an established root system has significant reserve strength over the same sand tested with no turf. Regardless of where we evaluate the modulus of subgrade reaction in Figure 2, it is consistently greater than that of the sand measured in the laboratory. This suggests that the root system adds strength and stiffness to the elastic and plastic properties of the root-zone sand. This additional strength and stiffness is most likely due to the tensile strength of the root system that reduces local shear failure within the root-zone sand.

Initial findings suggest that golf putting greens can be modeled as an elastic spring that has some stiffness, k_s. The stiffness or modulus of subgrade reaction of the root-zone sand increases with higher coefficient of uniformity, C_u. The median grain size has no effect on the stiffness of the sand. Field tests show that the stiffness of the green is dependent on soil properties but it also has increased strength and stiffness due to tensile strength contributed by the root structure. The short-term growing season of established root-zones have no effect on the stiffness of the putting green.

Methods for Classifying Sand Shape and the Effects of Sand Shape on USGA Specification Rootzone Physical Properties

The Pennsylvania State University

Dr. Charles Mancino

Start Date: 1996
Number of Years: 2
Total Funding: $38,254

Objectives:

1. *To determine if a simple, inexpensive and quantitative procedure can be used to give a reliable estimate of sand shape without having to examine individual grains.*
2. *Determine the effect of sand shape on the physical properties of rootzone sands and whether particle size distributions of USGA rootzone sands should be modified to account for differences in sand shape.*

A series of experiments were conducted to determine a method for assessing the shape of sand grains in a non-subjective manner. Methods tested in the past have included the direct shear strength method, the rotatable drum method, dense soil angle of repose, and cone penetrometry. These methods have not been capable of separating all classes of sand according to shape. In 1998, sand shape was assessed through computer imaging and analysis, cubical triaxial testing and further evaluation of cone penetrometry.

The use of computer imaging to determine sand shape was performed to compare sphericity ratings for samples of angular, sub-angular, sub-rounded, and rounded sands as calculated by an experienced technician and an image processing and analysis program *ImageTool* is a public domain freeware program. The comparison of measures of sphericity, however, resulted in a low coefficient of determination R^2 0.521. This R^2 value is too small to have confidence that the results produced by *ImageTool* and the technician will be similar. The reason for the low values is due to the failure of the software to properly define the edges of the scanned sand grains. The program tends to make the image more angular. Another inexpensive software program, *ArcView*, will be tested next. The *ArcView* package may be able to produce better results using algorithms to generalize or smooth the shape of the grains before analysis.

A cubical triaxial tester was used with the four sands to measure bulk mechanical behavior and how it relates to grain surface texture. The tester showed substantial differences between the sands with the sub-round sand having the best compaction resistance. The angular sand was the most compressible with the round and sub-angular materials being intermediate. In regards to soil strength, at lower pressures the subround sand was strongest while the round sand was weakest. At higher pressure, it was the angular sand and sub-rounded sand with the highest strength. Overall, the sub-round and

sub-angular sands had the best combination of compaction resistance and strength.

Layers in Golf Green Construction

Sports Turf Research Institute

Dr. Stephen Baker

Start Date: 1996
Number of Years: 2
Total Funding: $28,778

Objectives:

1. *To examine particle migration from the rootzone layer into underlying gravels of increasing size in situations where no intermediate layer is present.*
2. *To assess the effects of different intermediate and drainage layers on moisture retention in the rootzone layer.*
3. *To review the particle size criteria for the selection of intermediate layer and drainage layer materials.*

Particle Migration is being examined for two contrasting rootzone materials placed directly over ten drainage layer gravels of varying sizes. The two rootzones are an 85:15 mix of medium sand and sphagnum peat and a 70:30 mix of medium-coarse sand and peat. Five of the gravels are rounded and the other five are angular. The D_{15} size values range from 2.2 mm to 5.6 mm. Gravel sizes were selected so that, in theory, no migration would occur from the rootzone into the gravel for the finer gravels but the risk of particle migration into the coarser gravels was high. Each profile is receiving 3000 mm of simulated rainfall before particle migration is examined.

A technique was developed to examine whether migration has occurred at the interface of the rootzone layer and the gravel (Figure 3). The profile is stabilized using plaster of Paris. This is then impregnated with an araldite resin containing fluorescent dye. When the resin has hardened, the profile can be sectioned and photographed under ultra-violet light. This will enable examination of pore-space blockage within the gravel due to particle migration from the rootzone.

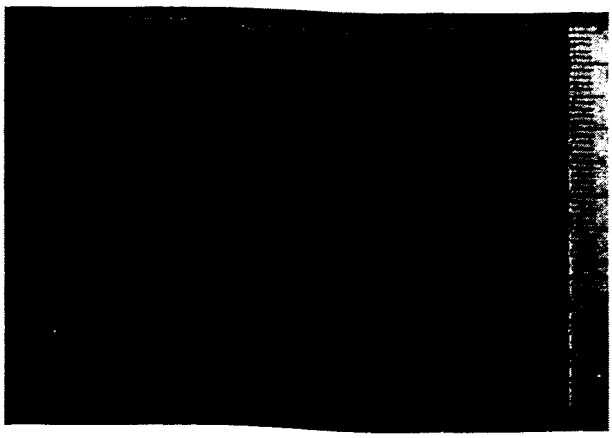


Figure 3. Cross section of intermediate sand layer above the gravel layer observed under UV light.

Moisture Profiles. The vertical distribution of moisture within the profiles discussed above is being measured after 48 hours of gravitational drainage to examine whether variations in the in type of gravel influence moisture retention in the profile.

In a separate study, the influence of particle size of the intermediate layers on moisture retention within an 80:20 sand/peat rootzone has been examined. The underlying gravel was predominantly a 6 to 9 mm material while the intermediate layer was based on 1 to 4 mm grit but with increasing proportions of medium (0.25-0.5 mm) and coarse sand (0.5-1.0 mm). Moisture profiles were assessed after saturation followed by 48 hours gravitational drainage.

Increasing proportions of coarse and medium-coarse sand had significant effects on the moisture content of the intermediate layer. For example volumetric moisture content increased from 7.5 percent when the 1 to 4 mm. grit included no sand to 18.4 percent when 50 percent coarse sand was added to the grit. However, no strong relationships were found between the composition of the intermediate layer and moisture retention with the rootzone. These data suggest that it should be possible to increase the proportion of material between 0.25 mm and 1 mm in the intermediate layer without a significant reduction of water retention in the rootzone. However, the work on moisture profiles directly over a gravel base must be completed before firm recommendations are made.

Understanding the Hydrology of Modern Putting Green Construction Methods

The Ohio State University - OARDC

Dr. Edward McCoy

Start Date: 1996
Number of Years: 5
Total Funding: $100,000 (co-funded with the GCSAA)

Objectives:

1. *Examine the effects of rootzone composition and putting green construction method on water drainage and redistribution within the profile.*
2. *Examine the effects of rootzone compostition, soil depth and degree of water perching on turf water use and irrigation management.*
3. *Examine long-term changes in physical, biochemical and microbiological properties of the rootzone; and relate these changes to the long-term hydrologic behavior of modern putting green designs.*

The overall program investigates the influence of green construction method on hydrologic processes including water infiltration, redistribution within the rootzone, drainage, and uptake by the turf. The two most prevalent, modern putting green construction methods are the United States Golf Association (USGA) and the California (CA) green construction techniques. The principal differences between these construction methods are the presence of a gravel drainage blanket in the USGA design and a higher recommended root zone permeability in the modified CA design. The one modification in the CA greens for these experiments was the addition of organic matter (sphagnum peat) to the rootzone mixture. However, this is commonly done throughout the United States where these greens are constructed.

Phase I. Earlier reported research on these systems examined water drainage and redistribution as influenced by root zone composition and green slope. The results of this study showed that putting green profile design, root zone permeability, and slope all yielded distinct hydrologic behaviors. Given equal root zone permeability, the experimental USGA greens yielded a more rapid drainage. Indeed, even a rainfall rate of about 4.5 inches per hour failed to overwhelm drainage of the USGA profiles as evidenced by equivalent drainage rates for both the low and high permeability root zones. Further, this same rainfall rate exceeded the drainage capacity of the modified CA green containing a rootzone mix initially tested to have a permeability of 20 inches per hour. For equivalent drainage performance, therefore, it seems that a modified CA style green would need a root zone mix permeability at least 20 inches hour greater than a USGA green.

Drainage rate represents an intensity factor. The capacity factor of the drainage process is the completeness of excess water removal from the root zone. Here, it is commonly thought that a USGA green would be less completely drained than a modified CA green. Our results showed that for equivalent root zone mix permeability the USGA green was drier after 48 hours (interpreted as more completely drained) than a modified CA green. This appears to be principally due to the need for water to move laterally through the root zone in a modified CA green before reaching a drain line. Again, for more complete drainage, a modified CA green would appear to need a higher root zone permeability. Nearly equal soil moistures were found after 48 hours drainage in the modified CA high permeability profile and the USGA low permeability profile.

All greens are sloped somewhat. This contouring clearly has an effect on water redistribution following rainfall. Prior to this study we believed that the perched water table in a sloped, USGA green would lead to strong lateral movement of water to down slope locations. We did not believe this would occur largely in a modified CA green. Again, our results showed our prior beliefs to be somewhat incorrect. While lateral water movement was observed in sloped USGA greens, it was also observed in modified CA greens. Thus, for equal rootzone permeability, there was a much greater lateral difference in water contents after 48 hours drainage in the modified CA greens than in the USGA greens.

The current findings of our Phase I research (as summarized above) are incomplete without a physical characterization of the root zones for the respective experimental greens. Thus, we have conducted lab and field measurements of root zone physical properties from *fresh* mixes and from the Phase I root zones on a yearly basis. We will continue these measurements for the 5-year duration of the overall project.

Table 1. Mean ponded infiltration rates and soil water contents with depth as influenced by putting green soil profile and root zone mix composition.

Profile	Root Zone	Infiltration Rate (40 mm head)	Water Content 3 in depth	Water Content 6 in depth	Water Content 9 in depth
		cm min^{-1}	m^{3}m^{-3}	m^{3}m^{-3}	m^{3}m^{-3}
USGA	High Perm.	1.91	0.47	0.46	0.44
	Low Perm.	1.20	0.43	0.43	0.41
California	High Perm.	1.31	0.44	0.40	0.44
	Low Perm.	0.96	0.45	0.41	0.41
Analysis of Variance					
Profile		*	NS	**	NS
Root Zone		**	**	*	NS
Profile x Root Zone		NS	**	*	NS
LSD (0.05)		0.41	0.013	0.013	0.06

*, **, *** indicates significance at $p<0.05$, 0.01 and 0.001 respectively.
LDS (0.05) given for the highest order interaction.

Ponded infiltration rates measured 20 months after turf establishment (Table 1) yielded some surprising results but were consistent with our observations reported in November, 1997. While we expected and observed greater infiltration rates from the high-permeability root zone, we also observed greater infiltration rates from the USGA profile than in the modified CA profile. Apparently, the presence of the gravel drainage blanket below the root zone in the USGA profile allowed for more rapid water infiltration regardless of the rootzone mix composition. This behavior was inferred in our previous study where a reduction in total drainage from the modified CA profile was associated with visually observed runoff that was not apparent for the USGA profile. Thus, soil profile features at 12 inches depth have an influence of water entering the soil surface in modern putting green designs.

In addition, the Phase I research showed lateral patterns of root zone moisture after 48 hours drainage. These patterns were influenced by drain line spacing in modified CA greens and slope in both modified CA and USGA greens. The question, therefore, arose whether turf drought symptoms would be observed with lateral position across these greens if further irrigation was withheld. Consequently, we conducted an initial replication of a dry-down study on the Phase I experimental units to help address this concern.

We anticipated an earlier onset of drought stress in the high permeability rootzones than in the low permeability rootzones. We also anticipated earlier drought over drain lines in the modified CA green with no slope, and at further distances up slope for both green systems at 4 percent slope. The strongest realization of these expectations was the response to slope in the high permeability modified CA and USGA greens. These greens at 10 days contained from 6 to 7 percent surface soil moisture at up slope locations and from 15 to 16 percent moisture at extreme down slope locations. This gradient in soil moisture yielded progressively increased stress symptoms from the down slope to the up slope locations. This was confirmed by our clipping yield measurements where fewer clippings were collected at up slope locations than down slope locations. Some of the more subtle turf responses to stress during this dry down study (if they exist) will require further data analysis before they are revealed.

Finally, one caveat of our Phase I results to date is that these greens were just one year old and had not experienced foot traffic. During this past year, we have applied simulated foot traffic to the greens by using a weighted roller. The roller is 4 feet in length, 8 inches in diameter and has a weight of about 325 lbs. The 'rolling factor' for this roller is about 1.2, which we estimate to simulate the heel pressure of an average human. Consequently, the water drainage and redistribution study conducted in the fall of 1997 will be repeated in the spring of 1999.

Phase II Progress. We have recently completed construction and established turf on an additional experiment to assess turf water use as influenced by root zone depth, root zone composition, and water perching in a USGA profile. The study employs six root zone mixes and two root zone depths constructed as a 2-tier USGA soil profile. Two of the root zones are 100 percent sand where the sands are relatively coarse and fine as based on USGA specifications. Two root zones are sand:sphagnum peat blends using the coarse and fine sand materials, and the final two root zones are sand:soil:peat blends again using the coarse and fine sands. Each root zone is placed in a 2-tier USGA profile with root zone depths of 9 or 12 inches. Each root zone mix and profile depth treatment combination is replicated three times for a total of 36 experimental greens.

To study turf water use, a complete accounting must be made of all water inputs and outputs from the root zone. For this reason, the greens soil profile is constructed within 6-ft diameter non-weighing lysimeters where drainage from individual greens is collected in an adjacent service pit. Additionally, TDR probes for soil moisture measurements are located at 3 and 6 inches depth for the 9-inch root zone and 3, 6 and 9 inches depth for the 12 inch profile. Use of the TDR probes will allow measurement of water loss from the turf by evapotranspiration. Water for the entire area is provided by an overhead irrigation system. The greens were seeded to Penncross creeping bentgrass in the spring of 1998.

Bacterial Populations and Diversity withing New USGA Putting Greens

University of Florida
Dr. M. Elliott

Auburn University
Dr. E. Guertal

Clemson Universtiy
Dr. H. Skipper

Start Date: 1996
Number of Years: 5
Total Funding: $66,667

Objectives:

1. *Determine bacterial populations associated with putting green root-zone mix materials.*
2. *Determine bacterial populations of the root-zone mixes before and after fumigation.*
3. *Compare rhizosphere bacterial populations on two different turfgrasses, bentgrass and bermudagrass.*
4. *Compare rhizosphere bacterial populations of bentgrass in two different locations, Alabama and South Carolina.*
5. *Compare rhizosphere bacterial populations of bermudagrass in two differrent locations, southern Florida and northern Florida.*
6. *Compare thatch development, rooting and bacterial population of bentgrass in relation to rootzone mix and nitrogen fertilization.*
7. *Compare soil and rhizosphere bacterial populations of root-zone mixes containing various clay sources.*
8. *Document rhizosphere bacterial population dynamics on bentgrass and bermudagrass over a four year time period.*

The overall objective of this project is to develop baseline data concerning bacterial composition (populations and diversity) of new USGA putting greens, both during and after construction. During 1996, the best methods for enumerating specific groups of bacteria were determined. These were incorporated into the research accomplished during the past two years.

University of Florida. For the project solely associated with Florida, the work completed thus far in 1998 was a repeat of the experiments carried out in 1997, with some modifications. Bacterial groups associated with putting green construction materials, prior to and after fumigation, and the bermudagrass sprigs used at planting were enumerated.

Trenches were dug at the FLREC for placement of 100-gallon size Lerio™ tree containers. These containers are 36-inch square and 18-inches deep. A 6-inch layer of non-calcareous washed river gravel was placed in the bottom of each container. No intermediate layer was added as the gravel and root-zone mixes met USGA specifications. Two peat materials were used to make the mixes, either sphagnum peat or reed sedge peat. The Canadian sphagnum peat was mixed with the sand to obtain an 85:15 mix. The Dakota reed sedge peat was mixed with the sand to obtain a 93:7 mix. The root-zone mixes are the two main treatments. The subplots or second factor is fumigation type. The containers are either not fumigated (control) or are fumigated with methyl bromide or metam sodium.

Samples were obtained for enumeration of seven different bacterial groups from: 1) individual root-zone components prior to blending, 2) each root-zone mix after blending, 3) prior to fumigation, 4) 9 days post-fumigation, 5) 23 days post-fumigation, and 6) each month after planting of bermudagrass for five months total. Samples were also obtained of the bermudagrass sprigs prior to planting. Only three monthly samples have been obtained to date.

The sand and sphagnum peat contained the lowest number of bacteria, with two groups not detected at all (fluorescent *Pseudomonads* and *Stenotrophomonas maltophilia*). All bacterial groups were detected in the reed sedge peat. Only *Actinomycetes* were not detected in the two root-zone mixes. For both root-zone mixes, there were significant differences within each fumigation treatment among the different bacterial groups at both sampling periods. The only consistent results obtained were: 1) the inability to detect fluorescent *Pseudomonas* in the methyl bromide treated containers 14 days after the plastic was removed; and 2) the lack of effect of the fumigants on *Actinomycetes* populations 14 days after the plastic was removed (Table 2).

All the bacterial groups were present when the *TIFDWARF* bermudagrass was sampled prior to planting. Throughout the next three months of sampling, all groups continued to be detected on plant material and in the root-zone mix. Data have not been analyzed to date concerning differences among root-zone mixes or fumigation treatments after planting.

Results this year indicate that bacterial numbers for most of the bacterial groups enumerated are actually increased by fumigation with either methyl bromide or metam sodium. Overall, the study would thus far indicate that bacteria certainly are not absent from root-zone mixes, even after fumigation, and that planting of the bermudagrass will introduce even more bacteria into the putting green.

Auburn. At Auburn University, treatments include nitrogen rate (1x or 2x normal rate) and construction materials (pure sand putting green or 80:20 sand:peat mix). Sixteen

Table 2. Bacterial groups present when materials delivered to UF/FLREC.

	Colony Forming Units per gram dry weight[y]						
Bacterial group	Sand	SP[z]	RSP[z]	SP- mix	RSP-mix	P.F	F value
Total	5.0 A e	6.6 A b	8.4 A a	5.8 A d	6.3 A c	0.0001	398.1
Fl. Pseudomonads	0.0 E c	0.0 E c	3.7 D a	1.6 D b	1.3 D b	0.0001	14.1
S. maltophilia	0.0 E c	0.0 E c	1.3 E b	2.1 CD ab	2.4 C a	0.0003	10.6
Gram positive	2.4 D b	1.2 D c	4.2 D a	0.5 E cd	0.0 E d	0.0001	18.1
Gram negative	3.4 B d	5.8 A b	6.4 C a	4.5 B c	4.5 B c	0.0001	293.4
Actinomycetes	2.7 C b	2.3 C b	7.5 B a	0.0 E c	0.0 E c	0.0001	75.3
Heat tolerant	2.7 C d	4.1 B c	7.1 BC a	3.0 C d	5.2 B b	0.0001	42.6
P>F	0.0001	0.0001	0.0001	0.0001	0.0001		
F value	456.2	43.6	70.2	37.4	75.3		

[x] Values are means of four replicate samples. Means in the same column followed by the same capital letter or means in the same row followed by the same small letter are not significantly different ($P \leq 0.05$), according to the Waller Duncan *k*-ration *t* test.
[z] SP, sphagnum peat; RSP, reed sedge peat

containerized greens were constructed at the Auburn University Turfgrass Research Unit, (four replications of each fertility/soil mix combination). Greens were sodded in January 1997 with washed bentgrass sod (*CRENSHAW*). Greens are 1 m long x 0.5 m wide, and each drains to an individual collection chamber. Total leachate from each green is collected as needed, volume recorded and a subsample is analyzed for NO_3-N and NH_4-N concentration.

In February, May, August and November, root and soil samples (0-4 inch depth) are collected from each green. These samples are shipped to the University of Florida, where they are subject to dilution plating and identification. Selected isolates are returned to Auburn University, where identification at the species level is conducted via GC FAME analysis. Nitrogen rates applied at the Auburn University site were originally 1 or 2 lbs. N/1000 ft^2/month (granular fertilizer source). Excessive loss of N through leachate and burning of turf at application resulted in a shift of application times and amounts to 1/5 or 1/10 lb N/1000 ft^2/week applied via a CO_2 backpack sprayer.

Year 1 analysis of nitrate and ammonium leachate indicated that both N rate and mix type affected NO_3-N and NH_4-N concentration in leachate, and there was rarely a significant N rate by mix type interaction. During Year 1 leaching of NH_4-N was greater in the pure sand green than USGA-type green. Leaching of NO_3-N from the USGA green was greater than that from the sand green, but only in the first few months after construction (January - April).

Clemson. Rhizobacteria are being evaluated for promotion of plant growth and for biological control of weeds, insects, diseases, and nematodes in a number of ecosystems. A critical research need in putting green management is to understand the bacterial interactions in the rhizosphere of turfgrasses. A database on turfgrass rhizobacteria from newly constructed bentgrass putting greens was initiated in December 1996. Each quarter, 160 bacterial isolates growing on tryptic soy-broth agar (TSBA) are randomly selected and identified by GC FAME analyses. Broad classes of rhizo-bacterial populations were successfully separated on selective media. Numerical differences of rhizo-bacterial populations in bentgrass rhizosphere over eight sampling periods were observed (Figure 1).

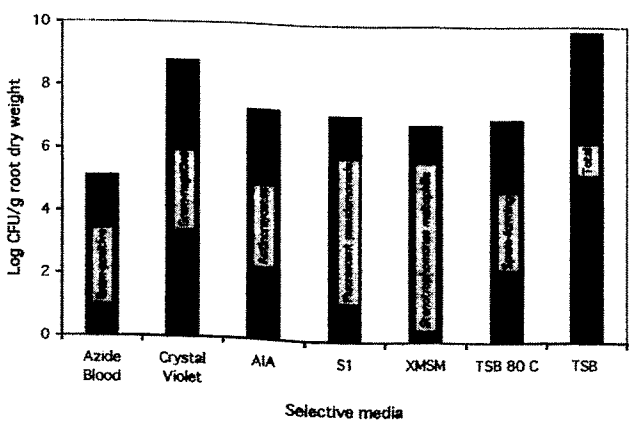


Figure 4. Rhizobacteria populations were averaged over eight sampling periods from bentgrass greens. Samples were collected from December 1996 to September 1998 from Charlotte Country Club Golf Course, NC.

In the samples of December 1996, isolates identified from bentgrass rhizosphere belonged to 23 genera and 34 species. *Acidovorax*, *Burkholderia*, and *Pseudomonas* were the major genera. However, in the samples of June 1998, isolates identified from bentgrass rhizosphere belonged to 23 genera and 43 species. *Pseudomonas* and *Arthrobacter* were the major genera. Based on the KOH method, 83% of the bentgrass isolates were Gram-negative over eight sampling periods.

Chemical and Physical Stability of Calcareous Sands Used for Putting Green Construction

Washington State University

Eric Miltner

Start Date: 1998
Number of Years: 3
Total Funding: $55,342

Objectives:

1. *To examine changes in particle size distribution, hydraulic conductivity, and calcium carbonate chemistry in calcareous sands used for putting green construction.*
2. *To qualitatively examine mineralogical properties of calcareous sands with scanning electron microscopy, both prior to and following weathering.*
3. *To survey existing golf courses of varying ages for variations in physical and chemical attributes of the greens mix.*

Calcareous sand can be defined as any sand that contains at least one-percent calcium carbonate (calcite) on a weight basis. In areas where they exist, they are often used for construction of golf course putting greens and other sand-based root zone media. Because of either perceived or real problems associated with these sands, which are not well defined or understood, their use is discouraged. In general, the types of problems that may occur are related to undesirable soil-physical properties (aeration, hydraulic conductivity, and water holding capacity) as compared to USGA recommendations. It is the objective of this research project to determine if performance characteristics of putting greens decline because of weathering of calcareous sands, and to determine the mechanism of this weathering and the subsequent performance decline. Ultimately, we hope to provide guidelines concerning suitability for use of various sands for putting green construction.

Two primary approaches are being used in this project. The first is a column study that investigates the weathering potential of various sands under controlled conditions. The second is a field survey that involves collection of intact cores from putting greens constructed of calcareous sands.

The column study includes three different sands. Silica sand, to serve as a control, will contain no calcite. The other two sands will have varying calcite contents so that a range of materials will exist. We have collected over thirty sands from various sources throughout the country, and have found most contain less than 10 percent calcite. Many of these samples were previously thought to be highly calcareous based on pH and reaction to acid. However, these samples had never actually been analyzed for calcite content.

We have expanded our search and are currently collecting new samples and characterizing them. One sample from Hawaii contains in excess of 85 percent calcite, but is too fine in gradation. In addition, this coral derived sand is atypical of most sands used in this country. We have recently solicited samples from suppliers in Florida that should be derived primarily from limestone, but have not yet received these. Once the sands to be used are identified, they will be subjected to acidifying conditions (as caused by fertilization) and bicarbonates (usually present in irrigation waters), both important factors in weathering of calcite. Changes in chemical and physical characteristics will be studied.

For the field survey portion of the project, cores have been collected from three golf courses to date. The first two courses, located in northern Utah, were constructed using the same sand (approximately 6% calcite). One course was built in 1989 and the other in 1993. We hope to see different physical characteristics in these samples based on their ages. Samples from a Texas course that has experienced problems that may be due to calcareous sand were also collected. These samples will be analyzed after more golf course samples are collected.

Evaluation of New Technologies in Construction and Maintenance of Golf Course Greens

North Carolina State University

Dr. Daniel Bowman

Start Date: 1996
Number of Years: 5
Total Funding: 100,000

Objectives:

1. *Survey golf courses throughout North Carolina to determine putting green aeration as a function of depth.*
2. *Develop and characterize a soil mix providing optimal moisture and aeration throughout the soil profile.*
3. *Measure the response of turf to the various mixes, and the impact of the turf on soil physical and chemical properties.*
4. *Conduct a field study examining turf response to promising soil mixes under natural environmental conditions.*

The physical properties of three sands (fine, medium, and coarse) and inorganic amendments (Ecolite, Greenschoice, Isolite, Profile, and sphagnum peat moss) were evaluated in this study. The 0, 10, and 20 percent (v:v) amendment were combined with each sand to produce nine rootzone mixtures. Also, 0, 10, 20 percent amendment (v:v) was added into the top 15 cm of the three sands. Physical properties measured included: hydraulic conductivity, bulk density, moisture retention (with depth), and pore size distributions/water retention at 0 to 200 cm tensions. Inorganic amendment evaluations included; particle size analysis, and pore size distributions/water retention at 0 to 15,000 cm tensions.

Nutrient retention of inorganic and organically amended sand rootzones was measured for 30-cm deep rootzone mixtures over 10 cm of suitable gravel. One application of 50 kg ha^{-1} of ammonium nitrate in liquid solution was applied and leached with 2.5 pore volumes distilled water. Leachate was analyzed for ammonium and nitrate by the rapid diffusion method. Four different experiments included: all amendments at 20 (v:v) material; Ecolite and Profile at 1, 5, 10, 20 percent; Ecolite and Profile at 10 percent (v:v) incorporated in the top 2.5, 15, and 30 cm depths; and an incubation study at 0, 12, and 24 hours in pure sand, 10 percent Ecolite and Profile.

Laboratory Results. The measured physical properties of three sand sizes amended with inorganic and organic amendments were as follows. Compared to pure sand, amendment addition increased total porosity, macroporosity, and water retained at 20-kPa tension. While, plant available water (water released from 4 kPa to 20 kPa) decreased with amendment addition Only fine sand and amended fine sands met USGA guidelines for total porosity, macroporosity, and capillary water retention. Medium and coarse sands and sand amendment mixtures resulted in rootzone mixtures that had excess macroporosity and lacked adequate water retention. Of the amendments tested, sphagnum peat resulted in the most water retained and had the most dramatic effect in the medium and coarse sands.

Evaluations of the inorganic amendments alone resulted in the observation that indeed these materials have a high degree of internal porosity (> 55%) and retain significant water (> 20%) even at high tensions. There were two clusters of amendment performance. Ecolite was similar to Greenschoice for water retention and release, both measured less than Isolite and Profile that were similar.

Hydraulic conductivity effect was variable between the three sand sizes and related to sand and amendment sizes. Sphagnum peat with its variety of particle sizes had the most consistent effect on K_{sat} and decreased this parameter for all three sands. At no point was a K_{sat} recorded that was less than 6 inches per hour.

Amendment addition decreased bulk density of all rootzone mixtures compared to pure sand.

Ammonium leached rapidly from all mixtures with peak concentrations occurring at approximately 0.5 pore volumes.

Significantly more NH_4^+-nitrogen leached from pure sand than for 20 percent (v:v) amended mixtures. Leaching losses ranked in decreasing order: pure sand > Greenschoice = Isolite > Peat > Profile > Ecolite. The most effective amendments, Profile and Ecolite, reduced NH_4^+-nitrogen leaching compared to pure sand by 75 and 88 percent, respectively.

Further studies with Ecolite and Profile had the following results. Increasing Profile and Ecolite rates from 1 to 20 percent resulted in stepwise decreases in NH_4^+-nitrogen loss. Although 20% amendment may be the most effective rate for retaining NH_4^+, it may not be economically feasible. Amendment at 10 percent significantly reduced NH_4^+-nitrogen leaching by 63 and 79 percent compared to pure sand for Profile and Ecolite, respectively.

Ecolite and Profile (10% v:v with sand) incorporated at three rootzone depths significantly decreased NH_4^+-nitrogen losses by approximately 25 percent, compared to pure sand. Again, there was a step-wise reduction of NH_4^+-nitrogen leaching reduction with increasing amendment depth. Incorporation of 10 percent amendment through the entire 30 cm rootzone resulted in the least NH_4^+-nitrogen leaching loss, with a significant difference noted between Profile and Ecolite. Losses were decreased by 65 and 80 percent for Profile and Ecolite, respectively.

Large quantities of $N0_3^-$-nitrogen(>90%), were recovered in leachate from all treatments under all experimental conditions. Peak $N0_3^-$-nitrogen concentrations of over 70 mg L^{-1} in pure sand leachate were observed.

Grow-in and Cultural Practice Inputs on USGA Putting Greens and Their Microbial Communities

University of Nebraska

Dr. Roch Gaussoin

Start Date: 1996
Number of Years: 5
Total Funding: $100,000 (Co-funded with the GCSAA)

Objectives:

1. *Evaluate grow-in procedure effects on putting green establishment and performance, and develop criteria and recommendations for new putting green readiness for play.*
2. *Determine grow-in procedure impacts on root zone physical and chemical properties.*
3. *Evaluate post grow-in cultural practice effects on putting green long-term performance.*
4. *Determine temporal and spatial (by depth) patterns of rhizosphere community development in golf greens during accelerated and controlled grow-in of select root zone mixes and during long-term green maintenance.*

The five year project is composed of three phases, One: Construction and Grow-in, Two: Microbial Community Assessments, and Three: Grow-in Procedure Impacts on the Long-term performance of the Putting Green. Phases I and II will span three-year periods, while Phase III will involve experiments repeated over the five years of the project.

Two separate USGA-specification root zone mixtures - one composed of sand and peat (80:20 ratio) and one a combination of sand, soil, and peat (80:5:15 ratio) - were developed in 1996. Materials used for construction complied with USGA Greens recommendations for physical characteristics and organic matter content. First year greens (1997 Greens) were constructed in late summer of 1996, allowed to settle over the winter, and were seeded with Providence creeping bentgrass (1.5 lbs/1000 ft^2) in the spring (May 30) of 1997. Second year greens (1998 Greens) were constructed in the summer of 1997, allowed to settle over the winter, and were seeded with Providence creeping bentgrass (1.5 lbs/1000 ft^2) in the spring (May 27) of 1998.

Preliminary results indicate the following trends. Microbial biomass was not affected by root-zone mix or grow-in procedure on plots established in 1997. Microbial biomass increased over 200% from spring to fall and decreased 40-60% as sampling depth increased. Water infiltration from these same plots were- not affected by root-zone mix or grow-in procedure when measured in 1998.

For two consecutive years, it was found that higher inputs would initially increase cover during grow-in. However, this increase may not translate to earlier opening for play if environmental stress conditions occur that result in damage to lush, immature turf.

The root zone mix containing soil established faster and recovered from environmental stress better than a soil-less mix. A soil-containing mix also will be harder and may result in longer ball roll distance. Addition of soil to the root zone mix did not effect water infiltration during the establishment year.

Assessing Differential Root Zone Mixes for Putting Greens Over Time Under Two Environmental Conditions

Rutgers/Cook College

Dr. James Murphy

Start Date: 1996
Number of Years: 5
Total Funding: $100,000 (Co-funded with the GCSAA)

Objectives:

1. *Evaluate grow-in procedure effects on putting green establishment and performance, and develop criteria and recommendations for new putting green readiness for play.*
2. *Determine grow-in procedure impacts on root zone physical and chemical propeties.*

3. *Evaluate post grow-in cultural practice effects on putting green long-term performance.*

Field Research. The primary objective of the 1998 growing season was to evaluate the establishment of creeping bentgrass as affected by the sand size distribution and amendment used in root zone mixes. The 37 root zone treatments constructed in either one or two microenvironments of the field research facility at North Brunswick, NJ were seeded to L-93 creeping bentgrass turf on 31 May 1998.

Location Effect. Environment (location) did affect establishment ratings for a few observation dates; however, there was not a strong influence on the establishment of creeping bentgrass in these two studies. It is expected that environment will have a greater effect on performance of turf maintained at a lower cutting height (< 5/32-inch) and receiving compaction treatment during 1999. No significant interaction between location and root zone treatment was observed during the 60-day evaluation period of bentgrass establishment.

Sand Size Distribution Study. Two finer sand size distributions (not meeting USGA guidelines) had a better rate of establishment than coarser sands. This was likely due to better moisture retention and subsequently better nutrient availability in those finer sands. The coarsest sand size established well; however, after 60 days, the performance of the plots declined. This may be an initial indication of the limitations of coarser sands.

Amendment Study. A greater affect on the establishment of bentgrass was observed in the amendment study compared to sand size distribution study. Generally, increasing the rate of amendment with soil and peat enhanced establishment. This was likely due to increased fertility and/or moisture retention in these mixes. However, establishment ratings for the 20 percent soil or peat treatments 40 days following establishment were similar to respective lower amendment rate plots. This may indicate the development of stresses associated with low air-filled porosity in the root zone. As expected, the greater fertility of ZeoPro plots enhanced establishment. Both ZeoPro and Profile (inorganic) amendments enhanced establishment up to 40 days compared to unamended sand. ZeoPro maintained high establishment ratings up to 60 days following treatment; whereas, Profile plots were more similar to the unamended sand after 40 days. Additional establishment data for all amendment treatments constructed in the enclosed environment are currently being summarized.

Laboratory Research. Research studies in the laboratory have been conducted to evaluate the influence of sample preparation on saturated hydraulic conductivity (K_{sat}). The saturated hydraulic conductivity (K_{sat}) measurement continues to be a highly variable measurement within and among USGA testing. An understanding of the source of this variability would improve testing procedures and benefit the golf course construction industry. A possible source of the variability is the phenomenon of air entrapment within 46 saturated" laboratory packed cores. Four studies assessed the influence of core diameter, antecedent moisture content prior to saturation, and saturation method on K_{sat} variability, as affected by air entrapment.

Effects on K_{sat}. Increasing core sample diameter (2- to 3-inches) resulted in higher sample densities and lower K_{sat} rates. K_{sat} rates for sand:peat and sand samples increased as the sample moisture content at time of saturation decreased. Greater sample moisture content at saturation apparently results in a sufficient amount of "pore necks" being filled with water that subsequently encloses air-filled pores during saturation (entrapped air). Conversely, a relatively dry sample provides open passages for the expulsion of air during saturation. Thus, dry sample cores did not entrap as much air during saturation and have higher K_{sat}.

Vacuum saturation procedure demonstrated the importance of removing entrapped air from core samples. Vacuum saturation of sample cores increased K_{sat} rates compared to saturation at standard air pressure. Temperature affects the solubility of gases in water. Water and room temperature can vary greatly within and between laboratories over time, and consequently could influence the air entrapment and K_{sat} of core samples. These factors are currently being evaluated for their effect on K_{sat} by varying water temperatures relative to ambient air temperatures in the lab.

Organic Matter Dynamics in the Surface Zone of a USGA Green: Practices to Alleviate Problems

University of Georgia

Dr. Robert Carrow

Start Date: 1996
Number of Years: 5
Total Funding: $100,000 (Co-funded with the GCSAA)

Objectives:

1. *Determine the effectiveness of selected fall/spring-applied cultivation on enhancement of bentgrass root development, water infiltration, and soil oxygen status during spring and fall root development periods.*
2. *Determine the effectiveness of selected summer-applied cultivation, topdressing and wetting agent practices on bentgrass root maintenance and viability, water infiltration, and soil oxygen status during the summer months when root decline occurs.*
3. *The best treatments from the above objectives will be combined to develop an integrated year-round program for maximum root development and maintenance during stress periods.*

Organic matter accumulation occurs even under excellent management and regardless of specification (i.e., it is not dependent on specifications) due to the abundance of roots produced by bentgrass within this surface zone along with any

thatch/mat accumulation. A considerable portion of the OM in the surface zone is as root tissue that can contribute to soil macropore plugging or sealing. Organic matter (OM) in the surface 0 to 2 inch zone of a USGA green can accumulate from an initial level of 1.0 to 4.0 percent (by weight) at establishment to 8 to 12 percent or more after two years.

A project was initiated in late spring 1996 to investigate the influence of treatments (summer cultivation, sand topdressing, sand substitutes, wetting agents) on maintaining infiltration, soil O_2 status, and roots. This field study continued until fall 1998. It is proposed that high temperatures, especially in conjunction with high humidity, causes an increase in the rate of summer death of roots. Since many roots reside in the surface 0 to 2 inches, death of a substantial percentage of these roots in a narrow time frame. The organic matter form can change from live roots with structure to gel-like, fresh, dead organic matter that rapidly plugs surface macropores. Any water applied at this point causes a saturated zone due to a low infiltration rate, thereby inducing low soil O_2 levels as gas exchange declines. Turfgrass and soil microorganism O_2 demands are very high under hot, moist weather and severe O_2 stress (similar to wet wilt without necessarily having standing water but with a saturated surface zone) occurs. This triggers very rapid enhancement of summer bentgrass decline and further root death.

Table 3. USGA specification and treatment ranges for bulk density, total porosity, and aeration porosity.

Parameter	Surface 0-3 cm Range for Study Treatments [a]	USGA Specification Range
Bulk density (g cm^{-3})	0.61 to 0.76	1.20 to 1.60
Total Porosity (%)	66.9 to 73.2	35.0 to 55.0
Aeration Porosity (%) (-0.004 M Pa)	17.4 to 24.3	15.0 to 30.0
Moisture Retention (%) (-0.004 M Pa)	42.5 to 54.7	15.0 to 25.0

The initial field study on this problem will continue until fall 1998. Observations to date are:

a. Percent OM by weight was 9.8 percent at 30 months after initiation of treatments for the untreated Control in the surface 0 to 3.0 cm (0 to 1.2 inch) zone. The Control received light, frequent sand topdressing at 0.5 to 1.0 ft^3 per 1000 ft^2 every 3 weeks, as did all treatments but not core aeration. Core aeration (CA) with a heavy topdressing (6.2 ft^3 per 1000 ft^2) in March was the only treatment to reduce percent OM (i.e., by 25% to 7.8% OM) while all other treatments ranged from 8.9 to 10.3 percent OM. Such high OM contents in this surface zone indicates that OM controls soil physical properties more than the sand matrix. Thus, soil physical properties within this zone were substantially different from the specification ranges of a USGA root zone mix. Since OM content is the primary factor affecting these soil physical properties of the surface 0 to 3 cm zone, OM would be expected to influence soil O_2 status and water infiltration.

b. Measurement of soil oxygen diffusion rate (ODR) was made in three treatments (CA, core aeration in Mar; HJR, Toro Hydro-Ject† raised to create a 0.25 inch diameter hole to a depth of 4 to 8 inches every 3 weeks June through September; HJR + (Co-funded with the GCSAA), same as HJR but with wetting agent applied every 3 weeks from mid-May through September. ODR was ≤0.20 μg O_2 cm^{-2} min^{-1} (the ODR rate at which soil O_2 becomes limiting for roots) 38, 43, and 24 percent of readings at 2.5 cm (1.0 inch) for CA, HJR, and HJR + WA, respectively over 3 summers. At 10 cm (4 inch) ODR readings were equal or below the limiting value on 0, 14, and 14 percent of readings, respectively, over 2 summers. Thus, even with these cultivation treatments, limiting ODR values were observed at 3 to 26 hours after irrigation within the surface zone.

c. Treatments that enhanced average water infiltration (as saturated hydraulic conductivity, SHC) at 17 to 26 days after cultivation greater than the Control (128 mm hr^{-1}) were: HJR + Sand (451 mm hr^{-1}) (HJR + additional topdressing at 0.75 ft^3 per 1000 ft^2 every 3 weeks); HJR + WA (406); HJR (400); HJR + B (395) (HJR + Biostimulant, cytokinin); HJR + Sand + WA (371); HJR + Sand + WA + B (361); HJL (331) (Hydro-Ject lowered position for 0.125 inch dia. hole); and HJR + Greenschoice (269 mm hr^{-1}) (HJR + Greenschoice topdressing at 0.75 ft^3 per 1000 ft^2). The normal desirable SHC for a high rainfall region is at least 120 mm hr^{-1} with the Control and CA treatments below this value 50 and 43 percent of readings. High SHC in the summer is essential to allow rapid water movement across the 0 to 3 cm zone that controls field SHC. High SHC also prevents standing water and excessively long periods of saturation and to enhance O_2 movement into the soil.

d. Percent of shoot density ratings significantly greater than the Control was highest for HJL (38% of readings), HJR + WA (29), HJR (24), and HJR + Sand + WA (24), while treatments exhibiting lower shoot density than control were LP + G (LandPride cultivation with vertical injection of Greenschoice into holes) (33%) and CA (29%). Root data are still in progress.

Stimulation of <u>Root Development (in Spring/Fall)</u> from the Zone of High Organic Matter Content. High OM content in the surface zone in cool weather may be due to rapid development of shallow roots. As roots developed in mid-fall to late spring, they may result in sufficient plugging of surface macropores in the surface zone to cause periods of suboptimal soil O_2 and low water infiltration. While adverse shoot responses to low soil O_2 may not be apparent in cool periods (as they are in summer where summer bentgrass decline can occur very rapidly), deep root development from late fall through late spring could be reduced, thereby limiting deep rooting going into the summer. A second project was initiated in winter 1996 to investigate the influence of selected cultivation procedures, that are non-disruptive to turfgrass shoots, on root development. Wetting agent and sand substitute treatments were also included. The objectives were to enhance SHC, soil ODR, and root development. Observations to date are:

a. Percent OM by weight in the surface 0 to 3 cm zone for the Control (received light, frequent sand topdressing at 0.75 ft^3 per 1000 ft^2 every 3 weeks as did all treatments but no core aeration) at 30 months after study initiation was 16.1 percent. Core aeration (CA) treatment in March and September with 6.2 ft^3 per 1000 ft^2 had OM of 9.3 percent while other treatments ranged from 9.8 to 16.8 percent. As in Study 1, OM in the surface dominated the physical properties of the root zone.

b. For the three treatments where soil ODR was determined, ODR at 2.5 cm (1.0 inch) was ≤ 0.20 μg O_2 cm^{-2} min^{-1} (the limiting value) 59 to 62 percent of readings in October to June and 25 to 38 percent at 10 cm (4.0 inch). The three treatments were CA, HJR + WA (Hydro-Ject Raised for 0.25 in diameter hole plus wetting agent, both at 3 week intervals); HJR + G + WA (where G = Greenschoice topdressing at 70 percent sand plus 30% G every 3 weeks at 0.75 ft^3 per 1000 ft^2 above the base sand topdressing of all treatments). Lowest ODR and SHC values occurred in December/January and May periods and values were lower than summer ODR and SHC values of an adjacent study (i.e., Study 1).

c. Maintaining high SHC across the 0 to 3 cm zone should increase soil O_2 and minimize periods of standing water or surface saturation that may inhibit rooting. Treatments that increased average SHC compared to the Control (71 mm hr^{-1}) at 24 to 41 days after cultivation were: HJR + WA (221 mm hr^{-1}); HJR (214); HJR + G + WA (183); HJR + G (152) and AW (Aerway Slicer, 100 tines, 4 inch penetration) (145 mm hr^{-1}). Lowest average SHC at 24 to 41 DAC were exhibited by QT + G (Solid quad tine of 0.25 inch dia. with Greenschoice) (53) and AW + G (63); where both of these treatments were not significantly different from the Control.

d. Treatments with 0 to 6 percent of shoot density rating less than the Control and 0 to 22 percent ratings greater than the control were AW + G, HJR, and HJR + G + WA. Treatments with highest percent of readings less than the Control were QT (28%), LP + GI (LandPride cultivation with Greenschoice vertical injection) (19), and QT + G (17). Root data are in progress.

Results from these two projects will be used in Phase II to formulate potential annual management programs (cultivation, topdressing, wetting agents, etc.) that a) would allow maximum root growth development in spring/fall without the decrease in rooting depth now observed on high sand golf greens a couple years after grass establishment, and b) would maintain root viability in the summertime and minimize summer bentgrass decline caused by low soil O_2 exchange.

Integrated Turfgrass Management

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

1. Determination of the range of adaptability and stress tolerance of turfgrasses (new and old varieties).
2. Evaluation of direct and interacting effects of two or three cultural practices, such as watering, fertilizing, cultivating, mowing, and programs to control disease, insects, and weeds.
3. Evaluation of direct and indirect cultural and environmental effects on population dynamics of turfgrass species, ecotypes, and/or cultivars.
4. Development of cultural practices that allow efficient turfgrass management under unique conditions, such as poor quality soils, shade, severe air pollution, and use of marginal quality water.
5. Investigation of pest management practices such as biological, cultural, and mechanical controls, and application of turf management practices utilizing IPM and reduced inputs.
6. Site specific management that utilizes remotely sensed data as a powerful graphic and quantitative tool to aid in the development of cultural practices. Such remotely sensed data can also provide synoptic evaluations of turfgrass response to changes in management and help with pest modeling or forecasting

The results of these studies will lead to the development of turfgrass management programs that conserve our natural resources and reduce costs, with minimal impairment of playing quality conditions or aesthetic appeal. Regional cooperation among researchers where similar climatic and soil conditions exist was encouraged in this research effort.

The Biology and Management of Spring Dead Spot in Bermudagrass

Kansas State University

Ned Tisserat

Start Date: 1998
Number of Years:
Total Funding: $65,874

Objectives:

1. *Determine the distribution of the three pathogens (Ophiosphaerella herpotricha, O. korrae, and Leptosphaeria narmari) associated with spring dead spot on bermudagrass.*
2. *Test the aggressiveness of each of the three spring dead spot pathogens in field tests at Manhattan and Wichita, KS, and Stillwater, OK.*
3. *Develop techniques to rapidly screen bermudagrass selections for resistance.*
4. *Monitor development of spring dead spot fungi on bermudagrass roots during the growing season in order to better understand the seasonal colonization and more accurately time fungicide applications.*

Spring dead spot (SDS) is a serious disease of bermudagrass along the northern range of its adaptation in the United States. Three distinct root-rotting fungi called *Ophiosphaerella herpotricha, 0. korrae,* and *0. narmari* (formerly *Leptosphaeria korrae* and *L. narmari)* cause this disease. The purpose of our research is to learn more about the distribution and biology of these SDS pathogens, and based on this understanding, to develop more effective strategies for managing this disease.

Diseased bermudagrass stolons and roots were sampled from golf courses in Kansas, Oklahoma, and Kentucky. Isolates of *0. herpotricha* and *0. korrae* were recovered from samples in all states, with *0. herpotricha* being slightly more abundant. The species *0. narmari,* previously reported only in Australia, was detected for the first time in North America from samples collected in Oklahoma and Kansas.

Little is known about the population structure of SDS pathogens on a local and regional scale. Similarities among isolates of *0. herpotricha* are being analyzed by amplified fragment length polymorphism (AFLP) analysis. Preliminary analysis suggests that on individual fairways, there is a mosaic of clones of *0. herpotricha.* However, on a regional scale there appears to be significant genetic heterogeneity among isolates. Less diversity has been detected among isolates of *0. korrae* and *0. narmari*

Field and greenhouse studies are being conducted to evaluate the resistance of seed and vegetatively propagated bermudagrass selections to spring dead spot. Field trials in Oklahoma indicated that several bermudagrass entries including *GUYMON, SUNDEVIL, MIDLAWN, MIDFIELD, FT. RENO,* and *MIRAGE* were more resistant to spring dead spot. We are currently developing greenhouse and laboratory methods to more rapidly screen bermudagrass selections for disease resistance. Furthermore, we are determining whether there are differences in pathogenicity to bermudagrass selections among the three SDS pathogens.

Various cultural and chemical control strategies have been proposed to control spring dead spot. We established a trial in 1998 to evaluate the effects of some of these control recommendations, alone and in combination, for suppression of SDS. Treatments include summer aerification and verticutting, soil acidification with ammonium sulfate, and fungicide treatments.

Determining Best Management Practices to Convert a Putting Green from Penncross to a New Variety

North Carolina State University

Dr. Daniel Bowman

Start Date: 1996
Number of Years: 3
Total Funding: $12,000

Objectives:

1. *To develop molecular methods that will allow us to determine for a mixed population of bentgrasses the relative percentages of the component varieties.*
2. *To determine appropriate management practices for converting an existing golf green from Penncross to one of the new varieties, using the method developed in the first objective.*
3. *To compare the competitive ability of the new bentgrass varieties during Penncross conversion.*

For several decades, *PENNCROSS* was the creeping bentgrass of choice for putting greens. This has changed over the past several years with the introduction of new bentgrass varieties with improved stress tolerance or better shoot density, and the trend will continue as more new varieties are released. Many superintendents are interested in replacing *PENNCROSS* with one of the new bentgrasses, but are uncertain of how to accomplish this without resorting to complete renovation. This study is designed to evaluate several methods by which an existing *PENNCROSS* green may be converted to one of the new bentgrass varieties.

One of the challenges in addressing the question of conversion is to develop methods to quantify the success of conversion. Simple visual examination is not possible, since bentgrass varieties are essentially indistinguishable based on morphological characteristics. We are using an existing molecular technology, Restriction Fragment Length

Polymorphism (RFLP) to determine population composition. This method can be used to determine the relative percentages of two (or more) varieties in bulk tissue samples (clippings).

The following fall treatments, applied each September, are being evaluated for their effect on bentgrass conversion:

1. Control, no interseeding
2. Broadcast interseeding with *L93* and *A4*
3. Cultivation with JobSaver tines plus broadcast interseeding
4. Verticutting plus broadcast interseeding
5. Primo plus JobSaver cultivation plus broadcast interseeding
6. Primo plus verticutting plus broadcast interseeding

Primo was applied at a rate of 0.3 ounces per 1000 ft^2 three days before interseeding. Success of the treatments are being evaluated annually for at least three years by sampling clippings from each plot and subjecting them to molecular genetic analysis.

Leaf tissue was sampled from the plots in late August. DNA was extracted and Southern analysis was conducted using a RFLP probe which distinguishes *A4* from *PENNCROSS*.

Computer imagery and data analysis indicates that conversion from *PENNCROSS* to *A4* during the first year occurred to the greatest extent with the JobSaver plus Primo treatments (Table 4). Conversion was approximately 20 percent. The least effective treatments were verticutting and verticutting plus Primo. These results led us to cultivate the plots more aggressively in year two, hoping to open-up the canopy more and provide a more favorable environment for the seedlings.

The data indicate that conversion from *PENNCROSS* is probably feasible, but that it will take a number of years. Further, it seems likely that complete conversion, in which *PENNCROSS* is completely eliminated, may not be possible.

Table 4. Conversion of *PENNCROSS* to *A4* bentgrass.

Treatment	Conversion to A4
	--------- % --------
Control, no interseeding	0.0 c
Broadcast seeding	13.8 abc
JobSaver Tines	16.2 ab
JobSaver Tines + Primo	21.2 a
Verticutting	2.5 bc
Verticutting + Primo	2.5 bc

Values followed by the same letter are not significantly different at P = 0.10.

The Distribution, Characterization and Management of Gray (*Typhula incarnata*), Speckled (*T. ishikariensis* complex) and *T. phacorrhiza* snow molds of Wisconsin Golf Courses

University of Wisconsin

Steve Millett

Start Date: 1998
Number of Years: 1
Total Funding: $18,225

Objectives:

1. *Determine the distribution and population structure of T. incarnata, the T. ishikariensis complex and T. phacorrhiza in Wisconsin golf courses.*
2. *To investigate the genetic variation within the nuclear ribosomal DNA (rDNA) among isolates of T. incarnata, the T. ishikariensis complex and T. phacorrhiza.*
3. *To determine the relative aggressiveness of T. incarnata, the T. ishikariensis complex and T. phacorrhiza on PENNCROSS creeping bentgrass.*
4. *To determine if fungicides and alternative tactics have different efficacies for control of gray (T. incarnata), speckled (T. ishikariensis complex) and T. phacorrhiza snow molds.*
5. *To determine the in vitro sensitivity of Typhula incarnata, the T. ishikariensis complex and T. phaxorrhiza to standard fungicides.*

A systematic random sampling technique was used to estimate the distribution of *Typhula* snow molds in Wisconsin golf courses. The sampling frame divided the State into three climate zones. Within these zones, seven golf courses that are within a 70 kilometer radius of Madison (southern), Stevens Point (central) and Woodruff (northern) were randomly selected to survey. Samples were air dried, crushed and sieved to collect sclerotia. The sclerotia were identified as either *Typhula incarnata* (TIN), *T. ishikariensis* complex (TISH) or *T. phacorrhiza* (TP). TIN was the most frequently collected species in the southern zone and TISH was the most frequent in the central and northern zones. Also, TP was found associated with distinctive patches in the central and northern zones. The DNA sequence of the complete internal transcribed spacer region (CITS) of the nuclear ribosomal DNA (rDNA) was used to genetically characterize the three *Typhula* species. Also, the relative aggressiveness of TIN, TISH and TP on creeping bentgrass was also determined using a growth chamber assay.

Survey Results. In general, the snow mold pressure was mild to moderate in the southern zone and moderate to severe in the central and northern zones. *T. incarnata* was the most frequently collected *Typhula* species in the southern zone. *T.*

ishikariensis was found as far south as Christmas Mountain Resort, Wisconsin Dells in the southern zone. *T. ishikariensis* was the dominant species in the central and northern zones. *T. phacorrhiza* was found as far south as Waupaca in the central zone. Complexes were common in the central and northern zones but not in southern zone. *T. phacorrhiza* was found more frequently in the northern zone than in the central but not in the southern zone. The survey results are illustrated in Table 5.

Characterization of rDNA ITS regions. The sequences of selected *Typhula ishikariensis*, *T. incarnata* and *T. phacorrhiza* isolates were analyzed for percent identity of the pairwise comparisons of the CITS and are presented in Table 6.

Aggressiveness assay. The relative aggressiveness of Wisconsin isolates of TIN, TISH and TP on creeping bentgrass was determined in a growth chamber assay at 5 and 10 C. The average aggressiveness ratings taken 21 days after inoculation are presented in Table 7.

Discussion. Typhula incarnata was the most frequently collected fungus in southern zone while *T. ishikariensis* was the most frequently collected fungus in northern two-thirds of the State. The CITS regions of the Wisconsin isolates *T. incarnata, T. ishikariensis* and *T. phacorrhiza* are different enough to

Table 5. Percentage of Typhula snow mold fungi collected from Wisconsin golf courses.

	TIN	TISH	TP	TIN/TISH	TIN/TISH/TP	TIN/TP	TISH/TP	unknown
Southern	94	4	0	2	0	0	0	0
Central	26	56	6	11	0	1	0	0
Northern	8	56	7	15	3	1	9	2

TIN=*T. incarnata*, TISH=*T. ishikariensis* and TP=*T. phacorrhiza*.

Table 6. Percent identity of the pairwise comparisons of the complete internal transcribed spacer regions (CITS) of Wisconsin Typhula *ishikariensis, T. incarnata* and *T. pharcorrhiza* isolates.

	TIN 100B dikaryon	TISH 100A dikaryon	TISH 105 dikaryon	TISH 105.1 monokaryon	TISH 105.2 monokaryon	TP 7 dikaryon
TIN 100B Dikaryon	100	70	70	70	70	61
TISH 100A Dikaryon	X	100	100	100	99	66
TISH 105 Dikaryon	X	X	100	100	99	66
TISH 105.1 Monokaryon	X	X	X	100	100	66
TISH 105.2 Monokaryon	X	X	X	X	100	66
TP 7 Dikaryon	X	X	X	X	X	100

Table 7. Relative aggressiveness of Wisconsin *Typhula* isolates on creeping bentgrass at 21 days after inoculation.

	TIN 1.31	TIN 1.35	TIN 2.100	TIN 3.113	TISH 2.97	TISH 2.105	TISH 3.124	TISH 3.114	TISH 3.122	TP 3.1	TP 3.117	TP 3.120
5 C	3.0	3.1	3.0	4.0	3.0	3.1	3.6	4.0	3.6	1.0	1.0	0.6
10 C	2.9	2.7	3.4	4.0	2.9	2.1	3.2	4.0	3.9	1.0	1.9	0.8

Rating scale used: 0=0%, 1=1.25%, 2=26-50%, 3=51-75%, 4=76-100% of turf.

continue exploring the molecular differences between and within these species. Wisconsin isolates will be tested for mating reactions by di-mon pairings with tester isolates from Japan, Russia, Norway, Canada and the United States of America. The percent similarities of the CITS regions will then be compared with the di-mon pairings.

Increasing the Nitrogen Use Efficiency of Cool-Season Turfgrasses by Regulating Nitrate Metabolism

University of Rhode Island

Richard J. Hull

Start Date: 1998
Number of Years: 3
Total Funding: $75,000

Objectives:

1. *To quantify each step in nitrate metabolism for ten Kentucky bluegrass (Poa pratensis L.) and five creeping bentgrass (Agrostis palustris Huds.) genotypes.*
2. *To determine which of these steps correlates best with nitrogen use efficiency under field conditions.*
3. *To assess the potential for increasing nitrogen use efficiency by optimizing the activity and location of those steps which are limiting.*

A long-standing paradox confronting turfgrass managers is the simple fact that high quality turf cannot be maintained without annual applications of nitrogen even when clippings are retained on the turf and no nitrogen is removed. This paradox is made all the more intriguing by recent research that shows very little nitrogen is lost from turf through nitrate leaching, ammonia volatilization or denitrification (usually less than 15% of that applied as fertilizer). Our research has shown that more than 2000 lbs. of nitrogen per acre can be recovered within the turf-soil ecosystem of long established turf. With that much nitrogen present in the turf environment, it would appear unlikely that additional applications are unnecessary.

The answer to this paradox lies in the fact that in the spring when nitrogen is most needed by turfgrasses, soils are too cold to mineralize much of the organic nitrogen available. During the summer, when the soil is warm and nitrogen becomes available, turfgrass roots are starved for energy because of high respiratory demands by the shoots due to elevated air temperatures. Warm-season turfgrasses do not experience this problem because their leaf respiration does not increase as much during hot weather.

This research project is intended to find means for making cool-season turf more efficient in recovering soil nitrogen. One obvious approach would be to promote greater root development and less shoot growth. This would make the grass better able to absorb nitrate from a larger volume of soil while less energy is committed to rapid shoot growth. Plant growth regulators have been used to achieve this goal but their action is not long lasting and while they inhibit shoot growth they often fail to stimulate root development. We believe the location of nitrogen metabolism in turfgrasses may be the key to this problem.

Most soil nitrogen is available to turfgrass roots as nitrate and is readily absorbed in that form. However, before nitrate-nitrogen can be assimilated into amino acids and proteins it first must be reduced to ammonium. This reduction of nitrate occurs in two steps or reactions: nitrate reduction (NR) and nitrite reduction (NiR). In roots, the eight electrons (e^-) required for these two reactions come from the reduction of sugars produced during photosynthesis and translocated to the roots. In leaves, most of the eight electrons come directly from photosynthetic reactions. The ammonium (NH_4^+) is assimilated directly into the amide-nitrogen of glutamine, which is a five carbon amino acid.

$$N0_3^- + 2H^+ + 2e^- \text{----------NR----------} > N0_2^- + H_20$$
$$N0_2^- + 8H^+ + 6e^- \text{----------NiR----------} > NH_4^- + 2H_20$$

The first reaction in this process is catalyzed by the enzyme nitrate reductase and this has been determined to be the rate limiting step in the chain of reactions leading to nitrogen assimilation. If nitrate is reduced in the roots, amino acids are produced there and root growth is promoted. If roots are unable to reduce nitrate as rapidly as it is absorbed from the soil, the nitrate can be transported to leaves where it will be reduced, assimilated into amino acids and stimulate shoot growth. When nitrate is reduced in leaves, photosynthetic products are diverted to shoot growth and away from roots. This lowered carbon flow to roots makes them even less able to reduce nitrate so more is transported to leaves and shoot growth is further stimulated and roots are not. This is what normally happens when turf receives nitrate from the soil or fertilizers.

The research conducted in this project will determine if the nitrate stimulation of shoot growth can be minimized by promoting nitrate reductase activity (NRA) within roots. We have found that in all Kentucky bluegrass cultivars studied, NRA is often ten times more active in the leaves than it is in roots. However, some cultivars (*LIBERTY*) did exhibit significantly greater NRA in their roots. We will determine if such cultivars produce greater root growth and if this contributes to more efficient nitrogen use. Currently we are extending this investigation to include diverse cultivars of perennial ryegrass and creeping bentgrass. Similar comparisons will be made and we will determine if greater root NRA correlates well with increased nitrogen use efficiency and field performance. We are also examining management practices that promote greater root growth (higher mowing heights, lower nitrogen fertilization, infrequent but thorough irrigation, etc.) to determine if they contribute to greater root NRA.

If this relationship between NRA in turfgrass roots and increased root growth is substantiated, efforts will be made to alter turfgrasses genetically to produce cultivars that have a more active nitrate reductase enzyme in their roots. This may produce turfgrasses that will utilize soil nitrate so efficiently that

little if any fertilizer nitrogen will be required. This would all but eliminate nitrate leaching from turf and produce turf with a larger, stronger root system that would be more tolerant of drought, and root-feeding insects. This research could greatly increase the over-all efficiency of turfgrass management.

Management Practices for Golf Course Roughs, Fairways, and Tees using Buffalograss

University of Nebraska

Terrance Riordan

Start Date: 1998
Number of Years: 3
Total Funding: $75,000

Objectives:

1. *Develop fertilization, mowing, irrigation, and pesticide recommendations for new buffalograsses.*
2. *Evaluate effect of cultivation on buffalograss.*
3. *Evaluate management for wear and divot recovery on buffalograss.*
4. *Use quantitative measures of turfgrass quality and recovery.*
5. *Study population changes in seeded cultivars due to management changes.*

Evaluation for Low-mowing and Wear tolerance. Under low mowing and no wear the female clone *92-135*, which outperformed all other entries in 1997, performed very well again in 1998 along with the female clone *92-31*. However, two male clones, *92-141* and *92-116*, had the best overall performance in 1998. All seed established experimentals exhibited average color and quality characteristics. The trial had a number of promising male and female clones. Wear results indicated that male and monoecious clones exhibited the most damage, while wear tolerance of female cultivars was significantly better than males, but not as good as for mixed seeded types.

Fertility and Mowing Effects on Buffalograss. At the Nebraska site, *NE 91-118* and *378* had the highest quality ratings at the 2.5 cm mowing heights for years 1996-1998. *CODY* and *TEXOKA* had poor quality ratings at the 2.5 cm mowing height for all years. In 1998, *NE 91-118*, *378*, and *CODY* had the highest quality ratings at the 5.1 cm mowing height. At the 7.6 cm mowing height, *CODY* and *TEXOKA* had the highest quality rating in 1997 but *CODY* and 378 had the highest quality ratings in 1998.

From 1997 to 1998, several trends were evident. First, turfgrass quality decreased from 1997 to 1998 for all cultivars at the 0, 2.4, and 5.0 g N m^{-2} rates. At 10 g N m^{-2}, *NE 91-118* and *378* had higher quality in 1998 than in 1997. All cultivars had improved quality ratings in 1998 at the 20 g N m^{-2} rate. Quality ratings in 1998 were poor (< 6) for all cultivars at 0, 2.4, and 5.0 g N m^{-2} rates. At 10 g N m^{-2} *NE 91-118, 378*, and *CODY* had good turfgrass quality. Management recommendations for 378 and *NE 91-118* are 2.5 or 5.1 cm mowing heights and a nitrogen rate of 10 g N m^{-2} $year^{-1}$. Recommendations for *CODY* and *TEXOKA* are 5.1 or 7.6 cm mowing heights and a nitrogen rate of 10 g N m^{-2} $year^{-1}$.

Nitrogen Partitioning in Turfgrasses. Field experiments to determine the fate of nitrogen fertilizer applied to three turfgrass species were initiated in 1997 at the John Seaton Anderson Turfgrass Research Facility near Mead, Nebraska. Fate of fertilizer nitrogen will be followed in buffalograss, Kentucky bluegrass, and tall fescue. Established turfgrass plots of two cultivars of buffalograss, *NE 91-118* and *NE 86-120*, a blend of Kentucky bluegrass, and a blend of tall fescue. The total amount of actual nitrogen that will be applied each year to a 9 m^2 plot is 0, 10, and 20 g N m^{-2}. For Kentucky bluegrass and tall fescue 80 percent of evapotranspiration will be returned every four days and for buffalograss 60 percent of evapotranspiration will be returned weekly. Plots will be randomly sampled prior to each fertilizer application to analyze for nitrogen content in plant and soil fractions. A Giddeon Soil Probe will be used to extract six cores (5 cm diameter) to a depth of 62 cm. Cores will be divided into thatch, verdure, roots, and soil components. The soil cores will be partitioned to four depths: 0 to 8, 8 to 16, 16 to 32, and 32 to 62 cm. After partitioning the cores by depth, the six samples will be composited, mixed thoroughly, and analyzed for total N, NH_4^+-N, NO_3^--N, and N-isotope ratio.

Buffalograss Resistance to Chinch Bugs. The initial phase of this research involved developing screening methods and evaluating selected buffalograss germplasm for resistance to *Blissus occiduus*. Eleven buffalograss cultivars/selections (*CODY, BONNIE BRAE, TATANKA, TEXOKA, NE 91-118, NE 86-120, NE 86-61, 315, 378, 609*, and *NE 84-45-3*) were screened for resistance to *B. occiduus* in two greenhouse trials. Using chinch bug numbers and plant damage ratings to assess levels of resistance, the 11 buffalograss cultivars/selections were separated into categories of resistance. *CODY* and *TATANKA* consistently exhibited high levels of resistance to chinch bug feeding, while *BONNIE BRAE* and *NE 91-118* showed high to moderate levels of resistance. Other cultivars/selections, including *378, 315, NE 84-45-3*, and *NE 86-61*, were moderately to highly susceptible. *CODY* and *TATANKA* maintained acceptable turf quality although both became heavily infested with chinch bugs. This suggests tolerance may be a mechanism of the resistance. Studies designed to characterize the mechanisms of resistance are currently underway. Antixenosis experiments have revealed chinch bug preference for *TEXOKA, NE 86-120*, and *BONNIE BRAE*. Other cultivars/selections such as, *609* and *NE 91-118* are rarely preferred.

Integrating Natural Enemies, Cultural Control, and Plant Resistance for Sustainable Management of Insect Pests on Golf Courses

University of Kentucky

Daniel A. Potter

Start Date: 1998
Number of Years: 3
Total Funding: $105,000

Objectives:

1. *Evaluate the role of ants as beneficial predators in golf turf: determine the predominant species inhabiting golf courses; and develop tactics for managing mound-building pest ants on putting greens with reduced environmental risk or impact on beneficial species.*
2. *Investigate synergism between endophyte-enhanced, resistant turfgrasses and bio-rational insecticides for improved management of white grubs and black cutworms.*
3. *Examine the main and interacting effects of cultural practices (mowing height, irrigation, and N fertilization) on nutritional and defensive characteristics of creeping bentgrass on relative susceptibility to white grubs and black cutworms.*

Conservation of naturally occurring biological controls is important for reducing need for insecticide usage on golf courses. Ants, the most abundant insects inhabiting turfgrass, are highly efficient predators of eggs and larvae of cutworms, grubs, and other pest insects. On golf courses, however, the positive aspects of ant predation must be weighed against the fact that some species build nests and mounds on putting greens and tees. This research seeks to identify beneficial and harmful ant species, document their significance, and develop effective means for controlling pest ants while conserving useful, predatory species.

Surveys of ants inhabiting roughs, fairways, tees, and putting greens of central Kentucky golf courses revealed that virtually all of the mound-building problems in close-cut creeping bentgrass are caused by one species, *Lasius neoniger*. *Lasius* appears to be the major nuisance ant on golf courses throughout much of the United States. Surface insecticides usually won't eliminate these ants because they fail to reach the ground nesting queen.

We evaluated two novel approaches for suppressing mounding activity on tees and greens. The first involved use of target-selective ant baits, some of which already have revolutionized ant control tactics used by the structural pest control industry. After testing seven candidate baits for acceptability to *Lasius*, we selected the three most attractive ones for evaluation on golf courses. These baits contained as active ingredients either avermectin (Advance Granular Carpenter Ant Bait; WhitMire Micro-Gen, Inc.) hydramethylnon (Maxforce granular ant bait; Clorox, Inc.), or spinosad (NAF-464; Dow AgroSciences). Each has a different, insect-specific mode of action, low mammalian toxicity, and favorable environmental characteristics. Advance and MaxForce already are labeled for use on turfgrass sites. Field trials on golf tees showed that use of these baits will provide rapid, 80 to 95 percent elimination of *Lasius* mounds and nests, either by broadcast, or by selective application from a shaker can. In another study, fipronil (Chipco Choice, Rhone-Poulenc, Inc.) was found effective for season-long suppression of *Lasius* nests and mounds on putting greens. This novel phenyl pyrazole, characterized by low mammalian toxicity and very low use rates, is a promising candidate for ant management on golf courses.

Field experiments demonstrated that *Lasius neoniger* and other ant species are very important in suppressing other insect pests. In trial after trial on roughs, fairways, or putting greens, ants eliminated large numbers of eggs and young larvae of black cutworms, and eggs of Japanese beetle. This underscores the wisdom of selective, rather than fence-to-fence, management of nuisance ants where mound building becomes a problem. Fortunately, our related work with halofenozide (Mach 2) and imidacloprid (Merit) has shown that these powerful new insecticides are compatible with preservation of beneficial species, including ants.

Our second objective concerns whether use of insect-resistant grasses in combination with reduced-risk insecticides can provide levels of control previously possible only with more broad-spectrum pesticides. In 1998, we studied possible synergistic or antagonistic interactions between endophytic perennial ryegrass and efficacy of *Bacillus thuringiensis* (Bt), *Bacillus popilliae* (milky disease bacteria), and spinosad (Conserve). We sought to determine if the sublethal stress endured by pests feeding on endophytic grass might enhance the activity of these products. Dose-mortality studies with Bt and spinosad were conducted with black cutworms fed on either endophytic (E +) or non-endophytic (E-) grass. Even full label rates of Bt provided no suppression of cutworms, irrespective of endophyte level. Spinosad provided 100 percent control, even at 1/4 label rate, on both E+ and E- grass. Dose-mortality studies with milky disease and Japanese beetle grubs showed significant rate effects, but disease incidence was not affected by endophyte level.

Finally, progress continued toward identifying the female sex pheromone of northern and southern masked chafers, the most destructive native grubs species in the United States. A synthetic bait would be useful for monitoring, fine-tuning treatment schedules, or assessing local grub densities on golf courses. Earlier, we discovered that the pheromone also is present in the grubs. In 1998, approximately 10,000 grubs were dug from Kentucky golf courses for pheromone collection. The grubs were rinsed in hexane to remove the pheromone; extracts were sent to collaborators (A. Attygalle, J. Meinwald, Cornell University) for analysis. The chemical peak representing the

pheromone was pinpointed by gas chromatography and electroantennogram analysis, and its molecular weight was determined. Gas phase IR was used to further characterize the compound's structure. Hopefully, the identification can be completed this winter, so that field testing during beetle flights can begin in 1999.

A Parasitic Fly that Kills Mole Crickets: Its Use in States North of Florida.

University of Florida

Dr. J. Howard Frank

Start Date: 1998
Number of Years: 3
Total Funding: $26,680

Objectives:

1. *To explore farther south in South America (colder climates) to obtain stocks of the fly Ormia depleta, a natural enemy of the mole cricket.*
2. *To culture the captured South American flies in our laboratory and supply them to collaborators in other states for release.*

Ormia depleta is a tachinid fly specialist on some species of *Scapteriscus* mole crickets. It is native to Brazil and Paraguay. A stock of this fly, captured at Piricicaba in subtropical Brazil (about 23° S) was brought to Florida in 1987 and cultured in quarantine. Beginning in 1988, progeny of these flies were released in all areas of Florida in an attempt to establish a population - about 10,000 flies were released. A population became established in peninsular Florida and persists year-round to about 28°N latitude, and seasonally (the fall of each year) in a marginal area extending to about 29°N. Subsequent releases in Georgia, North Carolina, and Alabama did not result in establishment of populations there.

Although the established populations of the fly exhibit strong seasonality in Florida, with much greater numbers trapped in May-June and in November-December than at other times of year, the fly seems capable of breeding throughout the year. That is, there is no dormant period (diapause) in winter. In the laboratory, adult flies need artificial nectar as a dietary item. Thus, it seems that the established stock of the fly, from subtropical Brazil, fares poorly in winter in northern Florida perhaps because it is not adapted to diapause during those months of the winter when plant nectars are in short supply (after freezes).

The fly is known to exist in southern Brazil to 30°S. It is possible that flies at 30°S are adapted to withstand colder winters by entering diapause. Therefore, they might be expected to survive in the southern USA at 30°N, and perhaps much farther north. The objective of this project is to obtain a stock of the fly from extreme southern Brazil, bring it to quarantine in Gainesville, culture it, and provide stock to collaborating turfgrass entomologists in Alabama, Georgia, Louisiana, Mississippi, North Carolina, South Carolina, and Texas.

In November 1998, two entomologists will travel to southern Brazil to spend almost four weeks collecting living *Scapteriscus* mole crickets as hosts for the fly. When scores of mole crickets have been assembled and maintained in containers in a laboratory in southern Brazil, flies will be trapped. Larvae of the flies will be reared on the mole crickets, and brought to the pupal stage. Fly pupae will be brought to quarantine in Florida for establishment of a laboratory culture. The timing of the visit (early summer) is based upon what is known about abundance and seasonality of *Scapteriscus* mole crickets and the fly in subtropical and temperate Brazil.

Work in 1999 will focus on labor-intensive culturing of several of the fly for distribution to other southern states.

Best Management Practices for New Dwarf Bermudagrasses

Texas A&M University

Richard H. White

Start Date: 1998
Number of Years: 3
Total Funding: $69,989

Objectives:

1. *Determine the performance, mowing tolerance, and pest resistance of 15 experimental and commercially available bermudagrass and one zoysiagrass on a golf green.*
2. *Determine the effects of vertical mowing, topdressing, and nitrogen fertility on performance, thatch development, fall and spring overseeding trasition, and turf quality of five dwarf bermudagrasses.*

New dwarf bermudagrasses are, in general, more aggressive thatch producers than *TIFDWARF*. Judicious nitrogen fertilization will be required to slow the rate of thatch accumulation for many of the new bermudagrass cultivars. Nitrogen amounts greater than 10 pounds annually per 1000 square feet improved turf quality but contributed to increased thatch, decreased ball roll distance, and did not substantially increase shoot density. No differences in thatch accumulation have been observed among light (frequent) and severe (infrequent) vertical mowing and topdressing regimes. However, severe, infrequent vertical mowing reduced turf quality for long periods. Overseeded *Poa trivialis* establishment the first season was good for all grasses when light, frequent vertical mowing was applied during the growing season.

Several new-dwarf bermudagrasses provided good to excellent turf quality and were superior to *TIFDWARF* at 0. 125 inch mowing heights. Mean turf quality of *MINIVERDE, TIFEAGLE, CHAMPION, MOBILE, FLORADWARF, MS SUPREME,*

LAKEWOOD, and *TXDB67* was superior to *TIFDWARF* at a mowing height of 0. 125 inch. Only *MINIVERDE* and *TIFEAGLE* produced higher quality than *TIFDWARF* at a mowing height of 0.187 inch. The results of this study indicate that several new bermudagrasses show promise for providing superior surfaces on golf greens.

Cultivar and Traffic Effects on Population Dynamics of Agrostis spp. and Poa annua Mixtures

Rutgers University/Cook College

James Murphy

Start Date: 1998
Number of Years: 3
Total Funding: $74,820

Objectives:

1. *Assessing the population dynamics of turf mixtures comprised of annual bluegrass and individual cultivars of creeping and velvet bentgrass grown on soil and sand-based root zones.*
2. *Evaluating the influence of traffic stresses on the populations dynamics of individual bentgrass cultivars mixed with annual bluegrass.*
3. *Identifying the time of year when it is most effective to establish bentgrass cultivars with minimal annual bluegrass invasion.*
4. *Assessing the effect of environmental conditions at the time of germination on the expression of annual or perennial biotypes in a sward containing annual bluegrass turf.*

Over the past decade, there has been a concerted effort by turfgrass breeders to develop improved cultivars of creeping and velvet bentgrasses that are denser, finer, more aggressive, more stress tolerant, and are more competitive than older industry standards. This affords the opportunity to take advantage of the genetic improvements in competitive ability of these bentgrasses in an annual bluegrass control program. The goals of this research project are to identify bentgrass cultivars that exhibit improved genetic competitive ability against annual bluegrass invasion under the influence of traffic, and to determine if the time of year for establishment affects the competitive posture of bentgrasses against annual bluegrass invasion. Putting green trials established on two dates in 1995 and one date in 1996 have consistently shown differences in the amount of annual bluegrass in mixed stands with bentgrass cultivars.

Data collected in June 1998. Percent annual bluegrass invasion on 8 June 1998 indicated *L-93* had less annual bluegrass invasion than all cultivars in the August 1995 seeded trial. *PENNCROSS* had a higher percent annual bluegrass invasion than all remaining cultivars in the same seeding date. In the September 1995 seeded trial, *A-4*, *L-93*, and *SOUTHSHORE* had less percent annual bluegrass invasion than *PENNCROSS*; *A-4* and *L-93* also had less annual bluegrass invasion than *PROVIDENCE*. *A-4*, *SOUTHSHORE*, and *G-2* showed less annual bluegrass invasion than *PENNCROSS* in the June 1996 seeded trial.

Data collected in October 1998. *A-4* and *L-93* had less percent annual bluegrass invasion than all cultivars except *SOUTHSHORE* in the August 1995 seeded trial. *SOUTHSHORE* had less annual bluegrass invasion than *PROVIDENCE* and *PENNCROSS*. *L-93* had less annual bluegrass invasion than all cultivars except *A-4* and *G-2* in the September 1995 seeded trial. *A-4*, *SOUTHSHORE*, and *G-2* had less annual bluegrass invasion than *PENNCROSS*. *A-4* also had less annual bluegrass invasion than *PENNLINKS* in the same seeding date. *L-93* had less annual bluegrass invasion than *PENNCROSS* and *G-2* in the June 1996 seeded trial. *A4* had less annual bluegrass invasion than *PENNCROSS* in the same seeding date.

A trial was initiated in 1998 to evaluate the time of year for bentgrass establishment that may enhance the competitive ability of bentgrass species and cultivars, against annual bluegrass. Initial data indicates that a June seeding date resulted in less annual bluegrass invasion than seeding dates in May and August. This would be expected because annual bluegrass is a winter annual, and peak seed germination would be late summer to early-fall.

Two additional trials were established in 1998. Both were established on soil and managed under putting green and fairways conditions. Both trials will assess the population dynamics between bentgrass and annual bluegrass under four levels of traffic. A third trial will be established on a sand-based (USGA style) root zone and maintained as a putting green (construction of the root zone was completed in October 1998). This third trial will also evaluate the effect of traffic on bentgrass and annual bluegrass population dynamics. More than a dozen cultivars of two bentgrass species are being evaluated in these three trials. Data will be collected for the percent population of each species as well as turf performance for each cultivar treatment under each level of traffic.

Improved Mole Cricket Management Through the Application of an Enhanced Ecological and Behavioral Data Base

North Carolina State University

Rick L. Brandenburg

Start Date: 1998
Number of Years: 3
Total Funding: $75,069

Objectives:

1. *Develop an effective integrated pest management program for mole crickets that ultimately reduces total pesticide use*

through improved implementation of chemical strategies and effective integration of biological and cultural options

2. *Apply the extensive research findings and validation of biological control strategies based upon our new knowledge of mole cricket ecology and behavior*

Studies during 1998 focused on four specific areas. These include: 1) defining high risk areas for mole cricket infestations, 2) determining the impact of soil moisture on oviposition and mole cricket development, 3) investigating the effect of irrigation and specific environmental parameters on insecticide (both conventional and biological) performance, 4) documenting the repellant response behavior of mole crickets to insecticide applications.

Defining high risk areas for mole cricket infestations: The field portion of this study was completed in 1998 year as a Masters student research project. The results of this study are currently undergoing statistical analysis and the measurement of the soil texture is still underway in the laboratory. However, preliminary findings indicate there is some degree of separation between those sites most commonly inhabited by tawny and southern mole crickets.

Determining the impact of soil moisture on oviposition and mole cricket development: Greenhouse studies to document the impact of soil moisture on oviposition have been completed by a Ph.D. candidate student. These studies were conducted in 7.5 cm diameter by 15 cm deep PVC cylinders filled with a uniform mixture of Kureb fine sand maintained at specific soil moistures. The results conclusively demonstrated that crickets lay eggs more quickly and in higher numbers when soil moisture is maintained above 7 percent (Figure 5). This effect helps explain the annual variation we observe in mole cricket oviposition and egg hatch in the field, on not only a calendar basis, but also based on degree-day accumulations.

Investigating the effect of irrigation and specific environmental parameters on insecticides (both conventional and biological) performance. Soil dissipation studies in association with insecticide rate and irrigation regimen treatment indicate that irrigation may play less of a role in positioning the insecticide than it does in affecting mole cricket behavior. This area is scheduled for additional investigation.

Documenting the repellant response behavior of mole crickets to insecticide applications: Field studies have examined a wide range of insecticide rates, formulations, and soil moisture levels for short and long-term control effects. Similar studies have been conducted for biological control (*Beauveria bassiana*). Results indicate that reverse rate responses often occur with higher rates providing less control. This may be the result of avoidance behavior associated with higher rates.

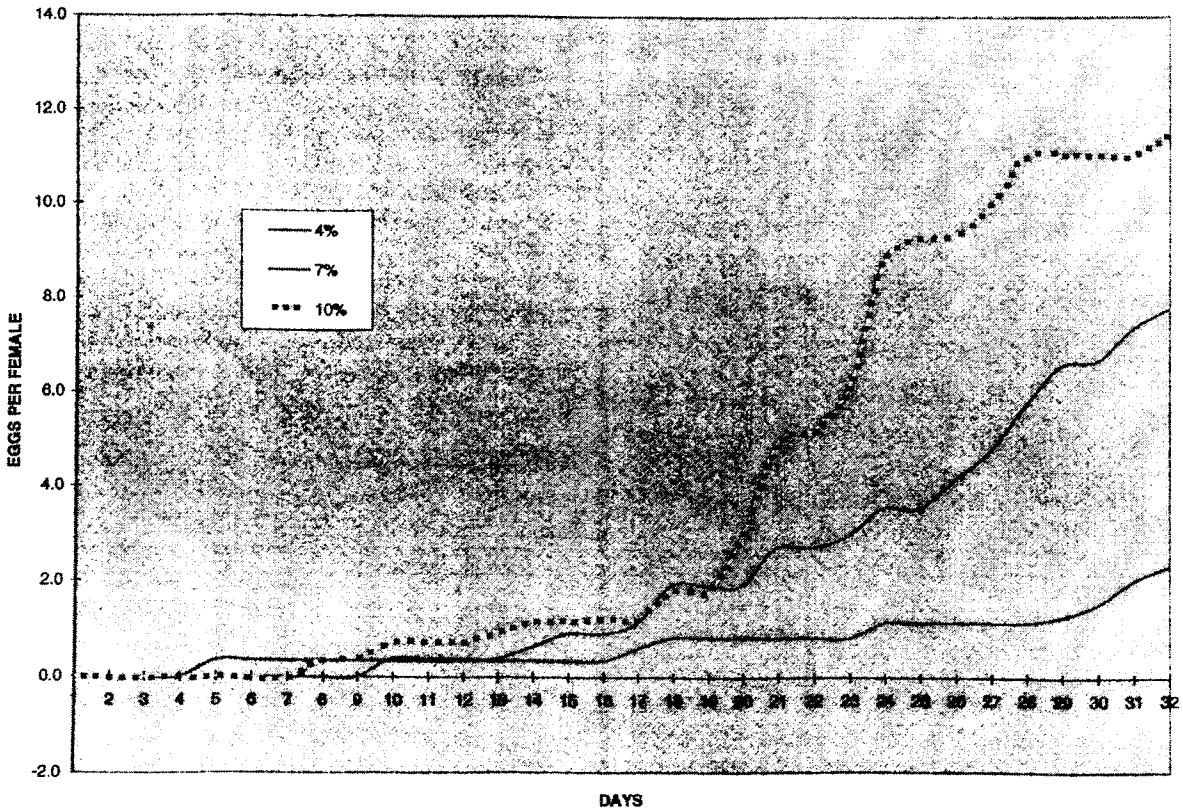


Figure 5. Mean number of eggs per ovipositing female held at different soil moistures in a greenhouse study.

A Disease Management Program to Reduce Pesticide Use on Bentgrass Greens

North Carolina State University

Jack Bailey

Start Date: 1998
Number of Years: 3
Total Funding: $74,752

Objectives:

1. *Verify the utility of using microclimate information for scheduling the use of fans, irrigation and fungicides for disease management.*
2. *Develop the system, hardware and software, to monitor and analyze the microenvironment on golf courses.*
3. *Determine if unnecessary fungicide applications can be reduced by using microclimate-based information for disease management.*

Specially-designed weather stations will be used to monitor the microenvironment and analyze the data regarding the likelihood of disease outbreaks. This information will be constantly updated and displayed on a personal computer. Analyses will be automated using the most current research information on the relationship of the environment and disease. Results of these *disease advisories* will be used to alter the times and duration of fan and irrigation usage to minimize the rate of disease progress. Weather-based thresholds also will be used to time fungicide applications to minimize the unnecessary use of fungicides while maintaining turf quality.

Standard ANOVA and regression statistics will be used to describe the relationship between air speed, total rain/irrigation, and hours of disease favorable conditions. Fungicides applied on *standard* and advisory-based schedules will be compared regarding disease incidence, turf quality and cost of maintenance.

Weather-based disease advisory models can be utilized to minimize the unnecessary use of fungicides while minimizing the risks to turfgrass quality associated with reduced pesticide input. Golf course fans and irrigation can have positive and negative impacts on turf quality. This work would create an objective method of determining when and how long fans and irrigation systems should operate to maximize their efficiency while reducing the likelihood of disease outbreaks.

Progress to Date. Funds were issued this spring and a thorough search was made to find a graduate student candidate that had expertise both in agriculture and engineering sciences. Jasson Latta was selected and trained during the summer on soft money (non-USGA funds) to familiarize him with the turfgrass research being conducted by Paul Lyford. Jasson started his MS program this fall in the Mechanical and Aerospace Engineering (MAE) Program at NCSU. Dr. Bailey will serve as his principal advisor in the Department of Plant Pathology, with Dr. Chuck Hall serving as a co-advisor in MAE. In addition to his engineering training, Jasson will be taking his second plant pathology course this spring in preparation for collecting the biological information necessary for this work.

Jasson has built a seven-foot wingspan, electric, radio-controlled airplane to be used in this work. A camera has been mounted in the body of the aircraft to remotely sense turf stress, quality, and foliage moisture patterns. Work is underway to locate the digital analysis hardware and software to be used in evaluating the images collected in this manner. This device will be used for data collection on fan design and to assess the success of each fan design on experiment stations and golf courses. The airplane is necessary to obtain images at right angles without the use of heavy machinery (i.e. cherry pickers) normally used for these types of studies.

The Importance of Carbon Balance and Root Activity in Creeping Bentgrass Tolerance to Summer Stresses

Kansas State University

Bingru Huang

Start Date: 1998
Number of Years: 3
Total Funding: $75,000

Objectives:

Investigate the physiological factors that cause summer bentgrass decline, and specifically, examine how carbohydrate metabolism influences the decline in creeping bentgrass root activity and turf quality under low mowing and high temperatures.

It was proposed that imbalanced photosynthesis and respiration process and carbohydrate depletion could be the primary physiological factors contributing to bentgrass quality decline under high temperature and close mowing conditions. The overall objective of the project was to test this hypothesis in creeping bentgrass cultivars grown under close mowing and high temperature stresses. This project involved two studies, in which responses of turf quality, root growth, viability, and carbohydrate metabolic activities for four creeping bentgrass cultivars to high temperatures and close mowing conditions were examined in controlled environment growth chambers.

The first study investigated effects of differential shoot/root temperatures and mowing frequency on turf and root growth and carbohydrate metabolic activities to determine whether turf quality and carbon balance could be improved by modifying root temperatures. In this study, two widely grown bentgrass cultivars *CRENSHAW* and *PENNCROSS*, and two relatively new cultivars with promising summer performance under close mowing, *L-93* and *PENN A-4*, were examined. Grasses were exposed to differential shoot/root temperatures, including low

shoot/root (20/20 C; control), low shoot/high root (20/35 C), high shoot/low root (35/20 C), or high shoot/root (35/35 C) conditions. Grasses were mowed at a 3 to 4 mm height daily or on alternate days.

It was found that turf quality and root activity were much lower at high root (20/35 C) or high shoot/root (35/35 C) temperatures than those of their respective controls for all four cultivars. Reducing root temperature to 20 C while maintaining shoots at 35 C improved turf quality and root growth to levels similar to those of the control treatment. High shoot/root temperatures reduced canopy photosynthetic rate and caused an imbalance between photosynthesis and respiration (carbon deficit) whereas reducing root temperatures reversed, to some extent, the adverse effects of high shoot/root temperature on carbon balance. The decline in turf quality was more severe for *PENNCROSS* than *CRENSHAW, L-93* and *PENN A-4* under high root or shoot/root temperatures. Similarly, daily carbon consumption to production ratio was higher for *PENNCROSS* than other cultivars under high root or shoot/root temperatures when grasses were closely mowed daily. Extending mowing frequency from daily to every other day improved turf quality and root growth, especially under high root or shoot/root temperatures, which was accompanied by enhanced photosynthetic rate and reduced carbon consumption to production ratio.

The second study examined whether declines in shoot and root growth with increasing temperatures (20, 24, 30, 34, and 38 C) were related to changes in carbohydrate metabolisms in *PENNCROSS* under close mowing conditions. Turf quality, root growth and viability of *PENNCROSS* declined significantly with increasing temperature to 30 C and higher. The imbalance between photosynthesis and respiration, carbon deficit, and reduced carbohydrate availability occurred as temperatures exceeded 30 C.

Results from both studies clearly demonstrated that first, carbohydrate depletion was a major physiological cause of summer bentgrass decline under high temperatures and close mowing. This was related to the imbalance between photosynthesis and respiration, which was caused by severe decline in photosynthesis capacity under high temperatures and low mowing. Second, roots played an important role in the regulation of creeping bentgrass tolerance to high temperature stress. Therefore, reducing root-zone temperature improved turf quality.

Two manuscripts describing the results of the project are currently being prepared for submission to Crop Science by the end of 1998.

The Basic Biology and Etiology of Sclerotinia Homoeocarpa, The Causal Agent of Dollar Spot

Cornell University

Gary E. Harman

Start Date: 1998
Number of Years: 3
Total Funding: $75,000

Objectives:

1. *Examine the development, including possible apothetical production, of the pathogen in creeping bentgrass greens and fairways when present in leaf tissue, in root tissue or as isolated stroma and to determine the length of survival of the pathogen in infected tissue or as stroma.*
2. *Measure the genotypic variation of the pathogen from similar and diverse geographical locations using RAPD analysis and anastomosis groupings.*

A summary of observations and tentative conclusions from the first field season are provided below together with action plans for the upcoming months.

1. Small (2 x 4 cm) porous nylon bags were prepared, inoculum of *S. homoeocarpa,* in the form of infected grass or grown on sterile wheat, was placed in the bags, the bags were heat-sealed and they were buried vertically in bentgrass greens. The upper edge of the bags was even with soil line.
2. The bags containing the wheat-based inoculum caused low levels of disease shortly after burial. Conversely, the bags with the turf-based inoculum rarely, if ever, caused disease. Disease was attributed to the bags since the natural epiphytotic had not occurred yet in this area.
3. At the time of the natural epiphytotic in August and September, no disease from the bags occurred from either inoculum type.
4. Re-isolation of *S. homoeocarpa* from the internal region of the bags resulted in slow-growing colonies that were almost overlooked due to the great difference in the growth patterns and morphologies of the laboratory culture.
5. *S. homoeocarpa's* normal growth type is rapid and floccose. This morphotype occurs in laboratory-adapted cultures and is obtained if the pathogen is isolated from infected turf. The slow-growing phase of the organism and the rapid-growing phase are very different.
6. After a week of incubation, the slow-growing, *S. homoeocarpa* colonies from the buried inoculum suddenly began to grow rapidly and become indistinguishable from the rapid-growing phase. The sudden explosive development is the only way that we could recognize the pathogen on the plates. We are still attempting to isolate

the pathogen from the dark stromal area on the surface of the bags.

These observations have permitted us to develop some concepts regarding how *S. homoeocarpa* may survive and cause epiphytotics. These are provided below.

1. Data suggest that *Sclerotinia homoeocarpa* in soil have a slow-growing near-dormant phase that may not be infective. The lack of infection is suggested by the fact that the pathogen in our buried bags, which was in the slow-growing phase, did not cause disease during the time when natural epiphytotics were occurring. It is very difficult to isolate the fungus in this phase; this difficulty has no doubt interfered with research on the presence, etiology and epidemiology of this disease.

2. It may well be that *Sclerotinia homoeocarpa* has two phases - a near dormant, heretofore undescribed, phase and the expected rapidly growing phase described by other researchers. It is tempting to speculate that the slow-growing (near-dormant) phase may be a survival mechanism and that the rapid-growing phase is the infective one. If so, then the mechanisms that cause the shift between the two phases could be the trigger for the onset of epiphytotics that are typical of the disease.

Upcoming Work

Turf Bags. Some of the bags buried in 1998 will be allowed to overwinter and will be recovered in the spring. We will examine the effects of overwintering and determine what, if any, dormancy structures are present in the bags and the surrounding turf. This fall, more bags containing inoculum from our strain, as well as bags containing small cores of naturally diseased turf will be placed in the field similar to last year. Some bags will be recovered in 1999 and some will be left to overwinter to 2000.

Genetic Diversity. - Genetic assays will be performed during the winter on the isolates on various turf species were obtained from Massachusetts, Michigan, Nebraska, New Jersey, New York, Pennsylvania, and Canada. Assays will include RAPDs (Randomly Amplified Polymorphic DNA) and anastomosis groupings, as well as any new techniques that are applicable. We will compare the similarities and differences between isolates from the same area and host species relative to ones from different geographical areas or hosts.

Greenhouse Assays. Flats of creeping bentgrass and possibly other species of turfgrass will be grown in the greenhouse and inoculated with the isolates of *S. homoeocarpa* to assess pathogenicity of the isolates. Dead or infected turf will be inspected for structures related to infection by means of microscopic examination following clearing and trypan blue staining. We will observe how different strains might behave differently.

Selective Medium for *S. homoeocarpa*. We will continue to investigate new methods for recovering and enumerating the dollar spot pathogen. Due to competition from *Trichoderma*, *Gliocladium* and *Penicillium* spp., the dollar spot pathogen rarely shows up even when we know it is present. Therefore, we need to eliminate these faster growing species in order to recover and enumerate *S. homoeocarpa*. This may be useful, perhaps, for predicting epiphytotics.

The Impact of Golf Courses on Soil Quality

Kansas State University

Steve J. Thien

Start Date: 1998
Number of Years: 5
Total Funding: $50,000

Objectives:

1. *Study the construction of a golf course in a grassland ecosystem.*
2. *Quantify indicators of soil quality and follow their change during the construction and establishment of a golf course on a natural grassland site.*
3. *Changes to soil quality indicators will be described, quantified, and used to predict areas where future golf construction and/or management actions may require special attention to minimize their negative environmental impact.*

This project is monitoring some soil quality criteria needed to assess the long-term impact and sustainability of golf courses on the environment. The research was initiated on native grassland destined to become Colbert Hills Golf Course, near Kansas State University in Manhattan, Kansas. Colbert Hills has been designated as a *living laboratory* by KSU to highlight its utility for research in environmental resources and turf management. This situation presents a unique opportunity to characterize site resources prior to construction and follow the long-term impacts and changes brought on by construction, use, and management of the facility. The golf industry needs this information to realistically understand its environmental impact, to formulate knowledgeable responses to public inquiries, to establish management strategies for new courses, and to provide knowledge for future planning and growth.

Relevance of Soil Quality to Golf Courses. Golf courses are only as sustainable as their weakest natural component, which can often be soil quality. The inherent sustainability of managed areas can be viewed as inversely proportional to the level of management needed to maintain it. Golf courses that diverge the most from their natural surroundings require the highest levels of management inputs to remain sustainable.

Soils play a central role in determining the sustainable land use potential of golf courses. Soil influences such critical properties as; leaching, aeration, fertility, water relations,

rooting, microbiological activity, and chemical use, detoxification, and effectiveness. A carefully selected set of properties, matched to the intended use of the soil, can be monitored as indicators of soil quality change. As these soil quality indicators degrade, they become the primary factors preventing superintendents from achieving course conditions expected by their management and players. The influence of siting, constructing, developing, and using a golf course on these indicators will ultimately determine both the sustainability of a course and the level of management necessary for day-to-day operations. This project is initiating a process of tracking changes to soil quality indicators during the life of a golf course.

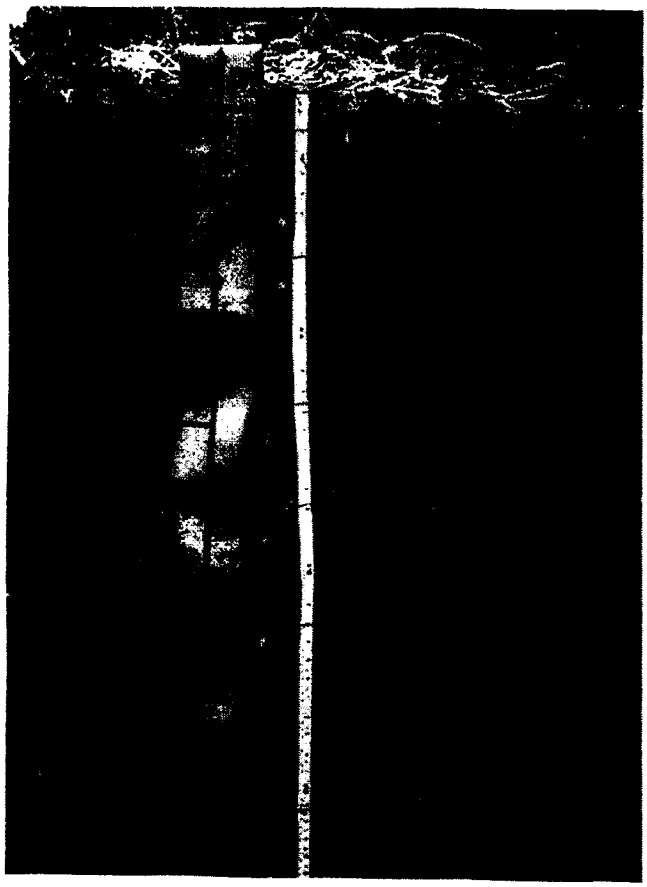


Figure 6. For each soil type identified, a pit was dug and the soil profile was fully characterized according to NRCS field standards. Loose samples for further analysis were collected from each horizon down to bedrock or to a depth of at least 2 meters

To evaluate the relative sustainability of different soils, soil scientists use indicators of soil quality. Selection of soil quality indicators should be related to soil use but reliable indicators for golf course soils have not yet been studied. This study will extend the concept of soil quality by identifying those indicators specifically important to the construction and sustainable use of a golf course.

1998 Research. Construction on Colbert Hills began in May 1998. Prior to construction, we made field observations and collected soil samples to establish base-line values for critical indicators of soil quality.

Starting in the spring and summer of 1997, soil scientists from Kansas State University and the USDA Natural Resources Conservation Service (NRCS) identified and mapped nine soil series on the site. By combining soil maps and architectural drawings of the course, we have located soil types according to their fairway and rough locations. Two sites for each soil type were selected and marked by global positioning for future referencing. One site was located in a fairway and one in an adjacent rough. The fairway site will be subject to the expected disturbances accompanying course construction. The site in the rough will be undisturbed.

For each soil type identified, a pit was dug and the soil profile was fully characterized according to NRCS field standards. Loose samples for further analysis were collected from each horizon down to bedrock or to a depth of at least 2 meters.

Sampling and Analyses Began and/or Completed.

1. Soil map of Colbert Hills A soil map has been nearly completed at a scale = 1:7920 (8 inches per mile). At this scale, the map is 4 times more detailed than maps typically found in soil survey reports.
2. Soil characterization open pit identification of horizons, depth, texture, color, structure, consistency, and pore and root distribution as per the USDA-NRCS Soil Survey Investigations Report No. 42, Version 3.0, Soil Survey Methods Laboratory Manual (1996).
3. Soil series identification of surface texture, slope, depth, drainage, permeability, physiographic location, and parent material.

Soil sample collection and laboratory analysis. Operations performed using standard methodology, as published in the American Society of Agronomy Monograph No. 9, Methods of Soil Analysis (1965). For each horizon present we have determined: depth, bulk density, pH, (1:1 water), pH (2:1 CACL2), total nitrogen (%), total carbon (%), microbial biomass nitrogen ($\mu g\ g^{-1}$, microbial biomass carbon ($\mu g\ g^{-1}$).

Turfgrass Germplasm Enhancement

The quality and stress tolerance of turfgrass is a product of the environment, management practices and genetic potential of the grass plant. In many cases, major limitations to quality turf are stress effects, many of which can be modified or controlled through plant improvement. These projects are directed toward the development of turfgrasses that conserve natural resources by requiring less water, pesticides, and fertilizers. Conventional breeding and new bio-technological methods are used to improve turfgrass varieties for golf courses. Among the characteristics most desirable in the new turfgrasses are:

1. Ability to survive high and low temperature extremes
2. Reduced need for pesticides by increasing resistance to disease, insects, nematodes, and weed encroachment
3. Tolerance of intensive traffic
4. Reduced requirements for mowing, irrigation, and fertilization
5. Tolerance of non-potable water
6. Stability of hereditary characteristics
7. Tolerance of acid, alkaline or saline soils
8. Increased shade tolerance
9. Tolerance of smog and other pollutants

The species of primary interest for turfgrass germplasm enhancement are bentgrass, bermudagrass, native grasses, seashore paspalum, zoysiagrass, and other turfgrass species for golf course use. The research effort encompasses the fields of Genetics, Cytogenetics, Cytology, Biotechnology, Phytopathology, Entomology, Nematology, Plant Physiology, Microbiology, and other sciences that support the project objectives and provide improved germplasm enhancement techniques.

Breeding and Evaluation of Turf Bermudagrass Varieties

Oklahoma State University
Charles M. Taliaferro

Start Date: 1998
Number of Years: 5
Total Funding: $124,978

Objectives:

1. *Assemble and evaluate cynodon germplasm accessions for important descriptors. Incorporate descriptor information and accessions into the National Plant Germplasm System. Maintain a working collection of germplasm accessions with breeding value and utilize in turf bermudagrass breeding program.*
2. *Improve bermudagrass breeding populations for seed production potential, cold tolerance, and other traits conditioning turf performance.*
3. *Identify bermudagrass parental plants with superior combining ability for use in producing inter- and intra-specific F1 hybrid.*
4. *Develop, evaluate and release seed- and vegetatively propagated turf bermudagrass varieties.*

The principal objective of the turf bermudagrass breeding program at Oklahoma State University is to develop improved seed- and vegetatively propagated cultivars for the transition zone. The development of seeded turf bermudagrass cultivars for the transition zone requires combining into breeding populations cold hardiness, economic seed yield potential and acceptable turf quality. Recurrent selection (RS) for these traits in broad genetic base *C. dactylon* population has resulted in incremental improvement with each cycle of selection.

The cold tolerant, seeded, synthetic variety OKS 91-11, was released in January 1997. Current synthetic varieties under evaluation as candidates for commercial release are OKS 91-3 and OKS 95-1. Additional plants were selected from recurrent breeding nurseries over the past year to generate new populations. The most elite of the selected plants will also serve as parents in narrow genetic base synthetic varieties. Breeding improvement in the broad base populations has now reached threshold levels that will allow more rapid progress in seeded turf bermudagrass cultivar development.

Intra- and inter-specific crosses were made to generate F_1 progeny populations for evaluation as potential vegetatively-propagated, hybrid bermudagrass cultivars. One thousand F_1 hybrid progeny from crosses made in 1997 were transplanted into field nurseries in spring 1998 for initial screening Approximately 50 select hybrid plants selected over the past 3 years are now in advanced stages of evaluation. Potentially valuable fertile hybrid plants from *C. dactylon* (2n = 6x = 54 chromosome) x *C. transvaalensis* (2n = 2x = 18 chromosome) crosses have been obtained. These tetraploid (2n = 4x = 36 chromosome) plants have one full genome (9 chromosomes) from *C. transvaalensis* and three full genomes (27 chromosomes) from *C. dactylon*. Open-pollinated and hybrid progeny from these plants have shown desirable turf characteristics.

Breeding and Evaluation of Kentucky Bluegrass, Perennial Ryegrass, Tall Fescue, Fine Fescues, and Bentgrass for Turf

Rutgers University
Dr. Reed Funk

Start Date: 1998
Number of Years: 5
Total Funding: $40,000

Objectives:

1. *Collect and evaluate potentially useful turfgrass germplasm.*
2. *Collect and evaluate endophytes associated with cool-season turfgrass species.*
3. *Continue the breeding and development of new cool-season turfgrasses.*
4. *Develop and apply several new tools designed to improve the ability to discriminate among endophyte isolates from nature and to synthesize new grass-endophyte combinations for experimental testing and possible commercial use.*

Promising germplasm of turfgrasses and *Neotyphodium* endophytes were collected from Bulgaria, Poland, the Slovic Republic, Inner Mongolia, Uzbekistan, and the United States. This included a number of endophyte-containing bentgrasses with a very small percentage of panicles showing choke stroma.

Germplasm collections and current turf trials indicate opportunities for substantial genetic improvements in a number of grass species which have received limited attention in turfgrass breeding programs in the United States. These include velvet bentgrass, colonial bentgrass, dryland bentgrass, *Koelaria* spp., *Deschampsia* spp. and interspecific hybrids between Texas bluegrass and Kentucky bluegrass. We are also seeing continued genetic improvement in perennial ryegrass, turf-type tall fescue, fine fescues, creeping bentgrass, and Kentucky bluegrass.

Broad sense heritability of dollar spot resistance in creeping bentgrass is being studied through the evaluation of 500 creeping bentgrass clones replicated six times in two environments. This study was initiated in May of 1998 at the Horticultural Farm 11 located on Ryders Lane in North Brunswick, NJ. The clones were heavily inoculated with the dollar spot pathogen (*Sclerotinia homoeocarpa*) using infested Kentucky bluegrass seed in July of 1998. These clones were

evaluated for resistance to dollar spot throughout the summer and into the fall of 1998. Initial results indicate that only 8 out of 500 clones show high levels of resistance to dollar spot disease.

Nearly 11,000 new turfgrass evaluation plots were seeded in field trials at Adelphia, North Brunswick, and Pittstown New Jersey. They included 4,500 plots of Kentucky bluegrass; 1,890 plots of fine fescues; 1,750 plots of tall fescue; 1,700 plots of perennial ryegrass; and 1,040 plots of bentgrass. These tests will be evaluated for a number of years for establishment vigor, turf quality, pest resistance, stress tolerance, texture, density, vertical growth rate, and persistence.

Approximately 100,000 plants were added to our spaced-plant nurseries for selection of promising clones, evaluation of seed yield, disease resistance, growth characteristics, stress tolerance, and breeding behavior. An additional 25,000 seedlings were established in mowed, clonal evaluation trials. These came from promising breeding lines of perennial ryegrasses, fine fescues, *Deschampsia spp.* and *Koelaria spp.*

Research is currently underway to identify and characterize microsatellites in perennial ryegrass. These molecular markers are short DNA sequences that contain short di- or tri-nucleotide repeats that are randomly dispersed throughout the genome in most organisms. Their abundance makes them particularly useful for DNA fingerprinting which can aid in cultivar identification and ultimately cultivar protection. Microsatellites can also be useful for population genetics studies and for marker assisted breeding. Twelve polymorphic or variable microsatellite loci have been identified and used to study ryegrass cultivars, clones and other species within the genus *Lolium*. We find these markers provide reasonable insights into genetic relationships and are excellent for cultivar identification in perennial ryegrass. Seventy-five more potentially useful microsatellite loci that have been identified but need to be investigated further to determine if they are polymorphic. Since 20 to 30 polymorphic or variable loci are ideal for precise cultivar identification and chromosome mapping, future studies will concentrate on identifying more of these loci in perennial ryegrass.

We participated in the development of a number of new turfgrass cultivars. These included Jefferson, *CHAMPAGNE, CACHE, MOONLIGHT, DRAGON, BLACKSTONE, H94-301, SR-2100* and *SR-2109* Kentucky bluegrasses; *BRIGHTSTAR 11, CALYPSO 11, MONTEREY, PANTHER, PRELUDE III, PRIZIN, CATALINA, WINDSTAR, EXACTA, CHURCHILL*, and *PARAGON* perennial ryegrasses; *SHADOW II, AMBASSADOR, VICTORY 11*, and *TIFFANY* Chewings fescues; *OXFORD* and *NORDIC* hard fescues; *FLYER II, TRAPEZE, PATHFINDER, FENWAY* and *AUDUBON* strong creeping red fescues; and *REMBRANDT, RENEGADE, PICASSO, MASTERPIECE, WOLFPACK, PLANTATION, MILLENNIUM*, and *SHENANDOAH II* tall fescues.

Field trials of transgenic creeping bentgrass have been established to evaluate promising transformed genes for herbicide resistance, stress tolerance, and pest resistance. Crosses have been made to initiate population backcrossing and recurrent selection programs to incorporate these genes into elite varieties and breeding synthetics. Work is progressing on the transformation of other turfgrass species.

We continue to make crosses of elite Kentucky bluegrass parents and screen large numbers of seedlings for promising hybrids under greenhouse conditions and in spaced-plant field nurseries.

Seashore Paspalum Ecotype Tolerance to Root Limiting Soil Stresses and Traffic Stresses

University of Georgia

Robert N. Carrow

Start Date: 1998
Number of Years: 3
Total Funding: $75,000

Objectives:

1. *Develop and implement a salinity tolerance screening procedure that a) provides salinity tolerance of seashore paspalum ecotypes under well-watered and drought stress, b) allows 3 salinity tolerance screening protocols to be assessed for efficiency in separating seashore paspalum ecotypes and for establishing a "standard" protocol, and c) provides detailed data on seashore paspalum ecotype root tolerance data on the edaphic (soil) stresses of salinity, drought, and drought+salinty.*
2. *Determine seashore paspalum ecotype tolerance to the multiple root stresses in the acid soil complex (soil strength, drought, nutrient deficiencies, element toxicities, high soil/air temperatures) that strongly influence drought resistance via drought avoidance from deep rooting.*
3. *For nine seashore paspalum ecotypes with the greatest potential for release, to determine relative to TIFWAY bermudagrass overall drought resistance, rooting, shoot performance, and water use (ET)/soil extraction patterns under close-cut fairway conditions.*

The breeding/genetics paradigm of Dr. R. R. Duncan's program for seashore paspalum (*Paspalum vaginatum*) is to systematically determine ecotype tolerance to important stresses. Of particular interest is genetic-based resistance to soil chemical and physical factors that limit root development and longevity. For grasses, these are: 1) high soil strength, 2) soil drought where soil drying causes death of roots that varies considerably with ecotype, 3) high soil salinity limiting root growth through physiological drought and specific ion toxicity, 4) acid soil complex which is common on kaolinitic and Fe/Al oxide soils, 5) low soil oxygen, and 6) high air and soil temperatures, especially for cool-season species.

In this project, seashore paspalum ecotypes are screened for root responses to 4 of the 6 edaphic factors that limit rooting. This *multiple stress approach* provides important

information for seashore paspalum resistance to individual and multiple soil stresses. This approach also is highly effective in identifying seashore paspalum ecotypes with *high nutrient uptake efficiency* and *drought resistance* via possessing a deep, extensive, viable root system. Root tolerance assessment to the major edaphic stresses has been a *missing ingredient* in most breeding programs targeted to improve drought resistance, water-use efficiency, or nutrient-use efficiency.

Study 1. Eighty-four seashore paspalum ecotypes and three control grasses (Common bermudagrass, *TIFWAY* bermudagrass, and Meyer zoysiagrass) were plugged (3.5 in. diameter x 3.0 in. deep) on 30 June 1998 into two adjacent sites at 4.5 feet centers. Both sites were a Cecil kaolinitic clay soil with 23 percent clay (A-horizon) and 45 percent (B-horizon). Site A was at pH 4.2 to create the acid soil complex stress which consists of aluminum/magnesium toxicities and potential deficiencies of manganese, potassium, calcium, and phosphorous. Site B was at pH 6.5. Both sites imposed the root stresses of high soil strength in a non-cracking soil, drought stress, and high soil temperatures.

At 24 days after plugging, irrigation was stopped and all grasses experienced periods of 8, 15, and 12 days without water from 24 July to 15 September. Mowing was at 1.25 inches and fertilization was at 1.0 lb N per 1000 ft^2 as 10-10-10 on 8 July.

Multiple soil stress response was evaluated based on a *Stress Index* that was a combination of two factors, a) the rank of the grass according to the degree of spread over 77 days after establishment at pH 4.2, and b) the ratio of growth (area covered) at pH 4.2 divided by growth at pH 6.5. Grasses that exhibited high growth under the severe soil stress of pH 4.2 situation should be able to grow and persist under a variety of irrigated and non-irrigated field conditions. The ratio of growth (area covered) at pH 4.2 divided by growth at pH 6.5 allows for identification of grasses with the highest tolerance to the acid soil complex stress. This stress is common in the Piedmont Region of the United States and very prevalent in tropical climates. Grasses with the highest ratio (1.0, equal growth at both pH's; < 1.0, less growth at pH 4.2 than pH 6.5) were ranked highest. Performances of selected grasses are in Table 8.

Study 2. Nine fairway type seashore paspalums and *TIFWAY* bermudagrass were stolonized on 16 July 1998 at a normal rate (0.75 bushels per 1000 ft^2). These will be evaluated in 1999 through 2000 under fairway conditions for shoot/root performance, evapotranspiration (ET) or water use, and overall drought resistance.

Table 8. Performance of selected grasses to multiple soil stresses (high soil strength, drought, acid soil complex, high soil temperature) that limit root development, viability, and persistence.

Grass	Stress Index Value	Stress Index Rank	Tolerance to Multiple Root-Limiting Soil Stresses
Hi 32 SP	15 (best)	1	Superior
HYB 7 SP	16	2	
HI 34 SP	17	3	
AP 4 SP	27	4	Very High
COMMON Bermuda	29	5	
PI 28960 SP	29	5	
TCR 6 SP	30	6	
96 HI 10 SP	31	7	
TIFWAY Bermuda	34	8	
AP 15 SP	34	8	
PI 509023	36	9	
Taliaferro SP	36	9	
TCR 3 SP	37	10	
AP 10 SP	38	11	
K 1 SP	38	11	
K 2 SP	39	12	
HI 101 SP	39	12	
Fwy 1 (PI 509019-1) SP	48	20	High
ADALAYD SP	67	32	Moderate
MEYER zoysiagrass	75	38	Low
Mauna Key SP	113 (worst)	54	Very Low

[a] Greens type, projected release 2000; very high salinity tolerance.
[b] Fairway type, projected release 2000; high salinity tolerance; high drought tolerance.

Selection of Turf Type and Seed Production in Inland Saltgrass (*Distichlis spicata*)

Colorado State University
Harrison G. Hughes

Start Date: 1998
Number of Years: 5
Total Funding: $125,000

Objectives:

1. *Determine turf performance of 7 elite CSU-USGA lines, 7 elite University of Arizona lines, 7 Great Basin lines (check lines from the University of Arizona).*
2. *Determining the range of stress tolerance (drought, salinity) present in inland saltgrass.*
3. *Determining seed production of 7 elite CSU-USGA lines.*
4. *Evaluate Kopec collection and Northern Great Plains collection.*
5. *Evaluation of seed germination and seedling vigor of all crosses.*
6. *Evaluate RAPD as a means of identifying unique genotypes of saltgrass.*
7. *Determine the relative chromosome number of elite clones.*
8. *Study the viability and germination requirements of inland saltgrass seed.*
9. *Evaluate seed priming as a possible method by which germination can be improved.*

Initially, elite lines from the University of Arizona collection and the CSU-USGA lines were established in both Arizona and Colorado. This initial year was a grow-in year with data on turf quality and seed production to be observed in future years. The material in Arizona will be used for drought studies in the field as well. CSU-USGA elite lines previously established in Colorado were observed for flowering and seed set. Seed production was evident but shattering was a problem. An extensive nursery consisting of the Arizona lines as well as additional lines from a collecting trip to Utah, Nevada, South Dakota and Nebraska with approximately 200 accessions was established at the CSU Horticulture Research Center outside of Fort Collins.

In order to understand seed fertility of inland saltgrass, a study of chromosome numbers for genotypes found throughout the region was initiated. Variation in chromosome number can lead to low pollen or egg viability resulting in poor seed set. Root tip smears of 40 genotypes were observed with the most common chromosome number being 2n = 38. However, 39, 42, 40, and 74 chromosome counts were observed. Coastal saltgrass was determined to have 40 chromosomes, as previously published and in our observations as well. This would indicate that our most commonly observed chromosome number of 38 is likely an aneuploid, probably a nullisomic.

Studies to determine pollen viability via examination of pistils demonstrated that pollen readily germinated in those clones examined. This was seen via microscopic examination of pistil structure. Furthermore, pollen tube growth reached the egg sac as well. Therefore, pollen viability is not apparently a problem in those clones observed. However, crossing among clones of different chromosome number may still influence successful seed production. In crosses among plants with 38 chromosomes and between 38 and 42 chromosomes, successful seed set was apparent. These seed have been germinated and will be examined in future studies. Due to high pollen viability and observable seed set, we believe that poor fertility as reported elsewhere is probably due to pollen availability rather than pollen quality or genetic deficiencies in most cases.

Three seed lots of inland saltgrass were examined for viability and germination. Seed viability for these lots was 15, 62 and 92 percent as determined by the tetrazolium (TZ) test. This low viability, in some cases, was probably due to a combination of extreme age, early harvest and poor storage conditions.

Inland saltgrass seed at maturity appears to be dormant and this dormancy is apparently due to the seed coat. Recently harvested seed lots that were scarified readily germinated in excess of 90 percent viable seed. For unscarified seed, alternating extremes of temperature prompted greater germination than moderate temperatures. Old seed exhibited low viability but seed that were determined to be alive via the TZ test germinated without scarification.

A Multigene-Transfer Strategy to Improve Disease and Environmental Stress Resistance in Creeping Bentgrass

Michigan State University

Mariam B. Sticklen

Start Date: 1998
Number of Years: 3
Total Funding: $75,000

Objectives:

1. *Enhance the expression (increase level of pest resistance) of the American elm chitinase gene in creeping bentgrass.*
2. *Transfer two drought-resistance genes controlled by either a constitutive or an ABA-inducible promoter into creeping bentgrass.*
3. *Determine disease resistance of transgenic plants expressing different levels of the chitinase gene and transgenic plants containing single-versus multiple-inserted genes under green house and field conditions.*
4. *Determine environmental stress resistance of transgenic plants containing single-versus multiple-inseted genes grown under greenhouse and field condiditons.*
5. *Evaluate transgenic creeping bentgrass clones for turf quality characteristics under field conditions.*
6. *Release transgenic creeping bentgrass germplasm with combined improvements in turf quality and pest and stress resistance to Pure Seed Testing, Inc. and/or other sectors for use in their field testing and commercial breeding program.*

Major biotic and abiotic problems associated in the management of creeping bentgrass turf include several pathogenic disorders and certain environmental extremes such as drought, heat, and cold stress. In addition, environmental extremes such as drought can influence the health of the plant and its ability to resist infection by biotic agents.

Resistance to biotic and abiotic stress in plants has been reported to be associated with relatively complex genetic factors. Most biotechnological approaches of the last two decades, especially those related to the control of insects and diseases, have concentrated on transferring a single gene into plants. The single gene approach may sound attractive over a short period; however, this approach may result in more serious problems over a longer period as populations of biotic agents (i.e., insects or pathogens) develop resistance the single gene. Our long-term goals include development of transgenic turfgrasses with improved resistance to pathogens and drought tolerance.

Previously, the research team developed creeping bentgrass clones that contain the glufosinate ammonia resistant herbicide, a chitinase gene, a proteinase inhibitor gene, and a drought and salt tolerance mannitol dehydrogenase (mt1D) gene. So far, it

glufosinate ammonia was confirmed to have fungicidal as well as herbicidal properties. Therefore, we have been able to simultaneously control weeds and turfgrass pathogens (mainly *Sclerotinia ulnocarpal* and *Rhizoctonia solani*) by spraying this herbicide on transgenic creeping bentgrass expressing the gene under greenhouse conditions.

Studies have shown that the chitinase genes can make transgenic plants resistant to pathogenic fungi such as *R. solani*, etc. Research in Dr. Vargas' laboratory has shown that our transgenic creeping bentgrass clone 711, transcribing the elm chitinase gene controlled by the cauliflower mosaic virus 35S promoter, has improved resistance of plants to *R. solani* under controlled environmental conditions. Recently, Dr. Sticklen's laboratory has constructed a plasmid containing the elm chitinase gene controlled by rice actin promoter (shown to provide greater gene expression in grass family than the 35S promoter) and transformed creeping bentgrass with this construct. Theoretically, using this grass-specific promoter, we could improve the level of expression of the chitinase gene, and the degree of resistance to *R. solani* in transgenic creeping bentgrass plants.

The mannitol 1-phosphate dehydrogenase (mt1D, known for its drought tolerance) gene that we have used to transform creeping bentgrass is also confers salt tolerance, A preliminary experiment performed by Dr. Baird's laboratory has not shown any drought tolerance of transgenic plants. More studies are needed to confirm whether these plants have any tolerance to drought and/or salt.

Determining the Genetic Stability of Triploid Bermudagrasses

Mississippi State University

Michael Goatley, Jr.

Start Date: 1998
Number of Years: 3
Total Funding: $72,790

Objectives:

Determine the origin of the off-types that arise in hybrid (triploid) bermudagrass golf greens through a combination of cytological and molecular analyses.

Off-types in hybrid bermudagrass (*Cynodon dactylon* x *C. transvaalensis* Burtt-Davy) putting green varieties are a persistent problem in southeastern golf courses. They disrupt green uniformity and interfere with ball roll; their effects sometimes necessitate green replacement. Our current goal at Mississippi State is to learn if their formation has a genetic and/or cytological basis. To minimize contamination to the smallest practical extent, we are conducting a greenhouse study using sterilized growth medium.

The genetic detection phase involves cross-species hybridization between bermudagrass DNA and RFLP clones from maize (*Zea mays* L.), and chromosome counts of bermudagrass root-tip cells for the cytology. We chose to use maize clones for two reasons. First, maize has a well characterized genetic map with many markers to choose from, so we know the chromosomal location of the clones we selected. Second, there is considerable evidence that gene order among the grasses is strongly conserved. Therefore, we can select clones from maize with reasonable assurance that we are monitoring a large portion of the bermudagrass genome, rather than using markers that are potentially biased towards small regions of the genome. Additionally, we are attempting to learn if there is a relationship between off-type formation in bermudagrass green varieties and chronic application of mitotic inhibitor herbicides such as pendimethalin and oryzalin.

Off-types in other grasses, most notably the cereal grain species, are frequently due to absence of one or more chromosomes, a condition known as aneuploidy. This chromosome loss may occur spontaneously, but it also may be induced with the application of mitotic inhibiting compounds. Oryzalin is now frequently used in place of colchicine to induce chromosome doubling for production of doubled haploids in lab experiments, mainly because it is much less toxic to humans than colchicine. When a plant's exposure to oryzalin is inadequate, chromosome doubling is incomplete, and aneuploidy sometimes results. If this occurs in bermudagrass putting green varieties, we hope to correlate it with the formation of off-types.

To meet this goal, six varieties are being subjected for one month to weekly drench applications of a 0.5X rate of oryzalin or pendimethalin in a replicated greenhouse experiment, designed as a randomized complete block. This is intended to expose the plants to a cumulative 2X rate application. The varieties are *TIFGREEN, TIFDWARF, TIFEAGLE, MS-SUPREME, CHAMPION,* and *FLORADWARF*. The grasses were established from small stolon pieces (2 nodes in length) in horticultural flats containing an approximate 75:25 masonry sand:peat moss mix, and are maintained at about 0.25-inch mowing height to encourage lateral growth in the flats. As a safeguard against latent contamination, the flats are irrigated for 10 days prior to stolon planting to encourage germination of seed or other dormant propagules so that they can be eliminated.

When the grass reaches 75 percent coverage of the flat, herbicide treatment is commenced. At the conclusion of the herbicide applications, the flats are left unmowed to detect any morphological off-types that might arise. Presence of differences will be determined by comparing leaf blade length and width, as well as internode length and width, between untreated checks and the treated clones using a two-tailed Dunnett's test (following a significant ANOVA). Stolons from these treated flats are then sampled to establish new flats. To date we have completed one cycle of the experiment and are initiating a new cycle with stolons from the previous round of treatment.

To test the efficacy of the herbicides, leachate from the final herbicide application at the end of the first treatment cycle

was used in a bioassay. Herbicide effectiveness was measured as suppression of annual ryegrass seed germination. Oryzalin suppressed germination about as effectively (54.9% of the untreated control) as pendimethalin (55.7% of the untreated control). This will allow us to adjust the treatments to achieve similar *efficacy rates* for both herbicides.

Concurrently with the greenhouse experiment, we are using selected RFLP markers taken from maize (*Zea mays* L.) to check for appearance of DNA polymorphism that might stem from chronic exposure of the grasses to the herbicide. It is unknown if these compounds are mutagenic; however, many organic compounds, including some herbicides, have mutagenic activity at high rates and/or chronic exposure levels). RFLPs will allow us to monitor phenotypically silent mutations. Although these do not result in observable off-types, appearance of new RFLP band polymorphism due to the treatments will guide us in determining application rates that will enhance the odds in favor of producing an off-type.

To date, 71 maize cDNA clones have been tested in cross-species hybridizations against bermudagrass DNA to identify those that show an adequate signal in Southern blots of bermudagrass. Roughly 75 percent of those tested are usable, and there are three subclasses within this category: a) those showing strong signals on bermuda, b) those with moderate signals, and c) those that show a weak signal (these three subclasses are present in fairly equal proportions). These selected probes will be used to probe Southern blots of bermudagrass genomic DNA samples from flats subjected to the herbicide treatments.

Cytological examinations of the six varieties to this point have revealed only the expected number of chromosomes for triploid bermudagrass ($2n = 3x = 27$). ⌶

Genetic Enhancement of Paspalum for Recreational Turf

University of Georgia

Ron R. Duncan

Start Date: 1998
Number of Years: 5
Total Funding: $125,000

Objectives:

1. *Ecotype evaluations off-site and industry collaboration.*
2. *Creation of additional genetic diversity within the species.*
3. *Genetic profiling of ecotypes.*

AP-10 (greens) and Fwy-1 (PI 509018-1: fairway/tees) ecotypes are slated for submission to the University of Georgia germplasm release committee during early 1999. Sufficient vegetative material will be available if the releases are successful. These seashore paspalums have exhibited excellent aggressiveness and performance on golf courses and under sod production. The darker green genetic color and turf quality traits are parallel to or better than most dwarf bermudagrasses. Genetic analysis research involving simple sequence repeats (SSRs or microsatellites) has progressed to the point of effectively profiling individual ecotypes for plant variety protection using trinucleotide repeats.

Wear tolerance mechanisms differ between paspalum and bermudagrass. Recoverability rates were identical between the two species. Wear tolerance in paspalum was attributed to leaf total cell wall contents (50% of the variability) while tolerance in bermuda was due to stem moisture (41%) and stem cellulose (32%).

Fertility studies have revealed that paspalum is more highly responsive to $CaNO_3$ than NH_4NO_3, NH_4SO_4, or urea. These highly soluble fertilizers appear to be critical for rapid establishment and recoverability, and may be important during long-term management in stressed environments. ⌶

Long-Term Preservation of Clonally Propagated Turfgrass Species

Colorado State University

Harrison G. Hughes

Start Date: 1998
Number of Years: 2
Total Funding: $49,701

Objectives:

1. *Develop suitable micropropagation procedures for selected genotypes of bermudagrass, zoysiagrass, saltgrass and buffalograss.*
2. *Develop suitable shoot tip culture media (STCM) for the four species.*
3. *Examine cryopreservation of the four species using vitrification methodologies.*

Clones of saltgrass (6), buffalograss (3), bermudagrass (1), and zoysiagrass (1), were established in the greenhouse and grown for a source of materials to put into tissue culture. It is important to establish *in vitro* protocol for each clone because cryopreservation requires very small growing points which will need to be established *in vitro* after freezing. If the tissue contains bacteria or fungal contaminates, they will likely overgrow any plant tissue thus obscuring positive results.

Various techniques involving different bleach treatment times and *PPM* (a commercially patented compound with antibiotic activity) concentrations, were used to disinfect tissue samples of buffalograss, bermudagrass, and saltgrass. Basal medium used was half strength MS and Nitsch and Nitsch vitamins plus 5 mg L^{-1} thiamine, 2 mg L^{-1} glycine and 30 g L^{-1} sucrose. Best results were obtained when small sections (1 to 2 cm) were used. In addition, either a bleach soak for 20 minutes for buffalograss, or 10-minute soak in bleach containing 5 mg

L^{-1} of *PPM* in the medium for bermudagrass provided better results.

Clean cultures of clones of buffalograss, bermudagrass, and saltgrass have been established and are being propagated for use in the cryopreservation studies.

Germplasm Development for Buffalograss Varieties

University of Nebraska

Terrance Riordan

Start Date: 1998
Number of Years: 5
Total Funding: $125,000

Objectives:

1. *Acquire additional germplasm through collection and recombination of germplasm already in our collection.*
2. *Evaluate germplasm with superior turfgrass characteristics including mowing tolerance, color, length of growing season, insect resistance, establishment and recovery of vigor, sod strength, combining ability, and seed production.*
3. *Obtain inheritance data on important traits, conduct genome size and molecular marker analyses, and evaluate the impact of inbreeding and genetic diversity on variety development.*

Seeded Releases. Native Turf Group (NTG) is considering the possibility of selling *NTG-5*, which was included in the 1991 National Turfgrass Evaluation Trial, and they are looking at *NTG-7* and *FW-3* (a low mowing tolerant experimental) for future release and production.

Vegetative Releases. Patents were filed for new releases *NE 86-61, NE 86-120* and *NE 91-118*, but have not been granted. Official UNL release statements have been completed and these cultivars are included in the 1996 National Turfgrass Evaluation Program Buffalograss Trial. *NE 91-118* has been vegetatively increased at Crenshaw Turf (CT) located at Poteet, Texas. Todd Valley Farms located at Mead, NE, bought a new farm and planted 35 acres of *NE 86-61*.

Sod Production. Crenshaw Turf (CT) has purchased Ellsberry Sod in Florida and Milberger Sod in Bay City, Texas. They continue to grow and have positive growth plans for production of buffalograss and other southern turf species. Todd Valley Farms (TVF) continue to increase sales of *378*, but TVF now has a greater role in developing the buffalograss market in the Northern United States. UNL, CT and TVF are working cooperatively on the development of new releases.

Summary of Breeding Work. Overall, the levels of performance continue to improve with each generation of selection. Newly released cultivars continue to show their superiority over older varieties with improved sod strength, color, quality, and density. Accessions from fairway maintained areas look very promising and show continuing improvements towards a high quality, low maintenance fairway turf. The top performers in the Nebraska National Buffalograss trial were *91-118* and *86-61*, which are being commercialized. The seeded varieties *CODY* and *TATANKA* showed little differentiation during the first year of this study. However, in 1998 the advance-seeded types began to show better performance than the common types like *TEXOKA*.

Evaluation for Low-mowing and Wear tolerance. Under low mowing and no wear the female clone *92-135*, which outperformed all other entries in 1997, performed very well again in 1998 along with the female clone *92-31*. However, two male clones, *92-141* and *92-116*, had the best overall performance in 1998. All seed established experimentals exhibited average color and quality characteristics. The trial had a number of promising male and female clones. Wear results indicated that male and monoecious clones exhibited the most damage, while wear tolerance of female cultivars was significantly better than males, but not as good as for mixed seeded types.

Fertility and Mowing Effects on Buffalograss. At the Nebraska site, *NE 91-118* and *378* had the highest quality ratings at the 2.5 cm mowing heights for years 1996-1998. *CODY* and *TEXOKA* had poor quality ratings at the 2.5 cm mowing height for all years. In 1998, *NE 91-118*, *378*, and *CODY* had the highest quality ratings at the 5.1 cm mowing height. At the 7.6 cm mowing height, *CODY* and *TEXOKA* had the highest quality rating in 1997 but *CODY* and 378 had the highest quality ratings in 1998.

From 1997 to 1998, several trends were evident. First, turfgrass quality decreased from 1997 to 1998 for all cultivars at the 0, 2.4, and 5.0 g N m^{-2} rates. At 10 g N m^{-2}, *NE 91-118* and *378* had higher quality in 1998 than in 1997. All cultivars had improved quality ratings in 1998 at the 20 g N m^{-2} rate. Quality ratings in 1998 were poor (< 6) for all cultivars at 0, 2.4, and 5.0 g N m^{-2} rates. At 10 g N m^{-2} *NE 91-118, 378*, and *CODY* had good turfgrass quality. Management recommendations for 378 and *NE 91-118* are 2.5 or 5.1 cm mowing heights and a nitrogen rate of 10 g N m^{-2} $year^{-1}$. Recommendations for *CODY* and *TEXOKA* are 5.1 or 7.6 cm mowing heights and a nitrogen rate of 10 g N m^{-2} $year^{-1}$.

Nitrogen Partitioning in Turfgrasses. Field experiments to determine the fate of nitrogen fertilizer applied to three turfgrass species were initiated in 1997 at the John Seaton Anderson Turfgrass Research Facility near Mead, Nebraska. Fate of fertilizer nitrogen will be followed in buffalograss, Kentucky bluegrass, and tall fescue. Established turfgrass plots of two cultivars of buffalograss, *NE 91-118* and *NE 86-120*, a blend of Kentucky bluegrass, and a blend of tall fescue. The total amount of actual nitrogen that will be applied each year to a 9 m^2 plot is 0, 10, and 20 g N m^{-2}. For Kentucky bluegrass and tall fescue 80 percent of evapotranspiration will be returned every four days and for buffalograss 60 percent of evapotranspiration will be returned weekly. Plots will be randomly sampled prior to each fertilizer application to analyze for nitrogen content in plant and soil fractions. A Giddeon Soil Probe will be used to extract six cores (5 cm diameter) to a

depth of 62 cm. Cores will be divided into thatch, verdure, roots, and soil components. The soil cores will be partitioned to four depths: 0 to 8, 8 to 16, 16 to 32, and 32 to 62 cm. After partitioning the cores by depth, the six samples will be composited, mixed thoroughly, and analyzed for total N, NH_4^+-N, NO_3^--N, and N-isotope ratio.

Buffalograss Resistance to Chinch Bugs. The initial phase of this research involved developing screening methods and evaluating selected buffalograss germplasm for resistance to *Blissus occiduus*. Eleven buffalograss cultivars/selections (*CODY, BONNIE BRAE, TATANKA, TEXOKA, NE 91-118, NE 86-120, NE 86-61, 315, 378, 609*, and *NE 84-45-3*) were screened for resistance to *B. occiduus* in two greenhouse trials. Using chinch bug numbers and plant damage ratings to assess levels of resistance, the 11 buffalograss cultivars/selections were separated into categories of resistance. *CODY* and *TATANKA* consistently exhibited high levels of resistance to chinch bug feeding, while *BONNIE BRAE* and *NE 91-118* showed high to moderate levels of resistance. Other cultivars/selections, including *378, 315, NE 84-45-3*, and *NE 86-61*, were moderately to highly susceptible. *CODY* and *TATANKA* maintained acceptable turf quality although both became heavily infested with chinch bugs. This suggests tolerance may be a mechanism of the resistance. Studies designed to characterize the mechanisms of resistance are currently underway. Antixenosis experiments have revealed chinch bug preference for *TEXOKA, NE 86-120*, and *BONNIE BRAE*. Other cultivars/selections such as, *609* and *NE 91-118* are rarely preferred.

Hybrid Bermudagrass Improvement by Genetic Transformation

North Carolina State University

Rongda Qu

Start Date: 1998
Number of Years: 5
Total Funding: $125,000

Objectives:

1. *Develop and optimize tissue culture conditions in order to obtain embryogenic calli and to regenerate plantlets of hybrid bermudagrass.*
2. *Develop a procedure to transform the embryogenic calli by the biolistic (particle bombardment) method and to recover transgenic plants.*
3. *Obtain transgenic plants of hybrid bermudagrass that express nematode resistant genes.*

The ultimate goal of this research direction is to improve bermudagrass cultivars for the golf courses through biotechnology. The specific goals of the project include: to optimize tissue culture conditions for inducing embryogenic calli and regenerating plantlets of bermudagrass; to develop procedure to transform the embryogenic calli by the biolistic method and to recover transgenic plants, and to obtain transgenic plants of bermudagrass that express potential nematode resistant genes. Bermudagrass is a recalcitrant species for plant tissue culture. Thus, most of the efforts have been concentrated on optimizing tissue culture conditions, especially at the callus induction stage to improve the callus quality and the regeneration ability.

Various tissues, culture media and supplements to the media have been tested in order to optimize tissue culture conditions of bermudagrass. Approximately 20 percent of the calli induced from young inflorescence (0.5 to 1 cm) of *TIFGREEN* and *SAVANNAH* (a common bermudagrass cultivar) had an embryogenic structure when cultured in MS medium (1 mg L^{-1} 2,4D) supplemented with 0.01 mg/L 6-benzylarninopurine (BAP). No such structures were found in media without BAP. The calli were slow growing, compact, pale or off-white in color, and highly regenerable. The regeneration rate of the calli with embryogenic structure was higher than 50 percent while calli without this structure had the regeneration rate about 1 to 5 percent. In addition, the callus induction rate was raised from 21 to 33 percent to over 60 percent by excising young inflorescence into pieces before the culture inoculation.

It was very difficult to induce callus from the young inflorescence of *TIFWAY* due to the quick browning of the explants in culture medium. The situation can be improved by pretreatment of explants with 0.2 percent ascorbic acid, an anti-oxidant.

Pilot experiments were performed to determine the pressure parameter of the biolistic apparatus. It was found by transient assay of GUS reporter gene that bombardments at 1550 psi on osmotically treated calli were the best for genetic transformation experiment.

Callus growth inhibition assays were performed with three potential selection agents at various levels. Effective selection was found at 250 mg L^{-1} of kanamycin and hygromycin B, while 5 mg L^{-1} was appropriate for selection with bialaphos, an herbicide.

Bermudagrass Cold Hardiness: Characterization of Plants for Freeze Tolerance and Character of Low Temperature-Induced Genes

Oklahoma State University

Charles M. Taliaferro

Start Date: 1998
Number of Years: 5
Total Funding: $125,000

Objectives:

1. *Quantify cold hardiness of advanced breeding lines, recently released varieties, and established standard varieties using laboratory-based methodology.*
2. *Isolate and characterize genes corresponding to low temperature-induced and antifreeze proteins by constructing and screening a representative genomic library from MIDIRON with both homologous and heterologous gene probes.*
3. *Characterize the low temperature induced expression of the cloned genes by northern blot analysis.*
4. *Sequence the cloned genes and characterize gene structure and function based on nucleotide sequence data.*

Injury to bermudagrass turf caused by freezing temperatures during winter is a persistent problem throughout its geographic area of use in the United States. This research seeks to reduce risk of freeze injury to bermudagrass grown in temperate regions. The research focuses on accurately assessing the freeze tolerance of bermudagrass cultivars, isolating genes responsible for enhanced freeze tolerance, and enhancing knowledge of the fundamental mechanisms associated with cold hardiness. Specific objectives are to: 1) quantify cold-hardiness of advanced breeding lines, recently released varieties, and established standard varieties and 2) isolate and characterize cold regulated (*Cor*) genes responsible for conferring freeze tolerance.

Experiments were initiated to determine the low temperature tolerance (LT_{50}) of turf bermudagrasses using laboratory-based methodology. The LT_{50} values will be determined sequentially for selected bermudagrasses in each of four groups. The groups are: 1) vegetatively-propagated fairway types, 2) seeded fairway types, 3) vegetatively-propagated putting green types, and 4) experimental fairway breeding lines. Experiments with the vegetatively propagated fairway types are underway.

Substantial progress has been made toward the goal of isolating and characterizing cold regulated (*Cor*) genes. A *Cynodon* genomic library was constructed from *MIDIRON* (*C. dactylon* x *C. transvaalensis*) turf bermudagrass. Screening the library using a 300-bp cDNA bermudagrass clone provided by Mr. Stephen McMaugh from the University of Sydney, Australia, identified nine putative chitinase genes. Sequencing and homology studies completed for three of the clones provided strong evidence that they are indeed chitinase genes, which we designate as *CynCht-1, CynCht-2,* and *CynCht-3*. We expect all of the clones to also be chitinase genes.

Northern blot analyses indicated chitinase gene expression in *MIDIRON, UGANDA*, and MSU turf bermudagrasses to be strongly affected by acclimation temperatures (4-8 °C). Substantial increases (75-100%) in gene activity in crown and root tissues occurred after 24 hours of cold acclimation. Increases of gene activity in crown and root tissues were proportional to the LT_{50}'s and ploidy levels of the three cultivars. Cold acclimation for 28 days caused an approximate three-fold increase in chitinase gene activity in leaves of *MIDIRON* and *UGANDA*, but had little effect on MSU. Leaves of MSU remained relatively green during the 28 day acclimation, while those of *MIDIRON* and *UGANDA* strongly senesced. Different *Cor* gene regulatory mechanisms may be involved in leaf and crown/root tissues.

Determining the Heritability of Salt Gland Density: A Salinity Tolerance Mechanism of Chloridoid Warm Season Turfgrasses

University of Arizona

Kenneth B. Marcum

Start Date: 1998
Number of Years: 3
Total Funding: $55,815

Objectives:

Determine the broad and narrow sense heritabilities of salt gland density in zoysiagrass. Zoysiagrass is the ideal model system to determine salt gland heritability, as salt gland densities and relative salt tolerances of a large number of genotypes have already been determined. As salt gland density had been found to be an important salt tolerance mechanism in other turfgrasses in the Chloridoideae subfamily (i.e. bermudagrass, buffalograss) results should be applicable to these breeding programs as well.

For this project, Greg Wess was selected for the M.S. student position in the Department of Plant Sciences, having received his B.S. from our department last spring. After consulting with turfgrass breeders, and review of the literature, it was decided to limit the main polycross study to a wide selection of *Zoysia japonica* types. This was decided, based on recent findings from Sharon Anderson (Ph.D. student, Texas A&M University). First, interspecific crossing between *Z. matrella* and *Z. japonica* types in zoysiagrass is possible, but often difficult and unpredictable. Second, flowering

requirements differ between species: *japonicas* behave as long day plants, while *Z. matrellas* and *Z. tenuifolias* tend to be day-neutral.

Because a polycross nursery will be utilized to determine heritability, uniform cross-compatibility and simultaneous flowering among genotypes is necessary. However, in addition to the *Z. japonica* polycross nursery, individual crosses will be attempted between several *Z. matrella* accessions and the *Z. japonicas*. The progeny resulting from these crosses will be evaluated for salt gland density.

Fifteen *japonica* genotypes have been selected, representing a broad range of salinity tolerance and salt gland density (previously determined from work at Texas A&M University). These are: *CROWNE, K162, J2-1, K157, PALISADES, BELAIR, EL TORO, J3-2, J94-5, KOREAN COMMON, JS23, P58, SUNRISE, MEYER*, and *K12*. These genotypes have been increased from single sprigs in a greenhouse (a slow process), and the replicated polycross nursery will be established next month, with 10 replications per genotype. Growth is being accelerated by supplemental lighting and liquid fertilization. Flowering of the *Z. japonicas* is expected in the late spring at which time polycrosses will be made.

Identification of Genetic Insect and Mite Pest Resistance in Turfgrasses

Texas A&M University

James Reinert

Start Date: 1998
Number of Years: 5
Total Funding: $125,000

Objectives:

1. *Establish a Regional Center to identify genotypes of Cynodon, Zoysia, Buchloe, Paspalum, Agrostis, and Poa with genetic resistance to insects and mites (fall armyworms, black cutworm, sod webworms, greenbug and host specific eriophyid mites) for use in cooperating turf breeding programs.*
2. *Bioassay resistant line with insect diets to characterize the mechanisms of resistance and determine their biochemical nature.*
3. *Develop effective and efficient procedures to accommodate screening and identify typical breeding populations-heretofore unavailable to the plant breeder.*

The Project has established a Regional Center to screen and evaluate turfgrass germplasm for resistance to insect and mite pests. The primary goal of the project is to identify genetic lines of bermudagrass, zoysiagrass, buffalograss, seashore paspalum, bentgrass, and bluegrass with resistance to the primary pests; caterpillars (fall armyworms, black cutworms, sod webworms) and host specific eriophyid mites, and characterize the mechanisms of resistance.

Work was initiated on elite bermudagrass germplasm *(Cynodon spp.)* from Dr. Charles Taliaferro's breeding program at Oklahoma State University, and with commercial cultivars under culture at the TAMU-Dallas Center.

Thirty-two bermudagrass hybrids and nine commercial cultivars were evaluated by feeding four day-old larvae on them in no-choice feeding studies. Among the hybrids, *4200W 49-17, 4200W 53-1*, and *4200W 55-5* (Table 9) produced the highest mortality with from 42 to 52 percent mortality, identified as failure of the individuals to emerge as adults from the pupa stage. Also, *3200W 70-18* provided 37 percent mortality at adult emergence with *3200W 94-2, 4200W 38-2, 3200W 18-11* and *3200W 30-20* each producing 33 percent mortality. These same grasses with the exceptions of *4200W55-5* and *4200W 38-2*, usually produced the smaller larvae when weighed at 10 days. At the other end on the gradient, *CCB 24-4* and *3200W 6-12* were the most susceptible hosts and produced only 4 percent mortality of the fall armyworm larvae.

Among the commercial bermudagrass cultivars, mortality was 8.25 percent or less at 17 days for all of the cultivars and 20.6 percent or less at adult emergence. Fall armyworm development was slowest with the smallest larvae and pupa on Common, but this experiment supports previous experiments that Common is not resistant and is a relatively good host for this insect pest. None of the cultivars of bermudagrass in this experiment or in the above experiments exhibits an acceptable level of resistance to the fall armyworm. These experiments support the hypotheses that new cultivars may be developed that are superior to existing cultivars in pest resistance.

Residential landscapes are frequently invaded by large populations of grasshoppers that develop in adjacent landscapes or in agricultural lands. These invasions occur annually in late summer to autumn in some areas, but high populations tend to cycle every three to five years across the southern or southwestern states. Representative cultivars of cool and warm season turfgrasses (Tall fescue, *REVEILLE* hybrid bluegrass, Syn1 Texas bluegrass, *TIFWAY* bermudagrass, *COMMON* bermudagrass, *RALEIGH* St. Augustinegrass, *MEYER* zoysiagrass, *CAVALIER* zoysiagrass, *PRAIRIE* Buffalograss and Johnsongrass) were evaluated for feeding preference or resistance to adult feeding by the differential grasshopper *(Melanoplus differentialis)*. The degree of feeding was ranked (rank = 0 – 5, 0 = no feeding during the test period, 5 = near complete consumption of ration) and measured by the number and weight of fecal pellets produced during the feeding period.

Based upon ranked feeding and the number and weight of fecal pellets after 2-days of feeding, tall fescue was the most preferred host evaluated. *REVEILLE* hybrid bluegrass, *TIFWAY* and *COMMON* bermudagrass, Syn1 Texas bluegrass and *MEYER* zoysiagrass were also highly preferred hosts based upon fecal pellet weights. *PRAIRIE* buffalograss and *CAVALIER* zoysiagrass were resistant to the grasshoppers and exhibited very low feeding damage, and fecal pellets. These trends held true throughout the 8-day feeding period of the test.

Table 9. Mortality of life stages, weight of larvae and pupa, days-to-pupation and adult emergence for fall armyworms fed as 4-day-old larvae on clippings of bermudagrass cultivars in Spring 1998.

Bermudagrass	% Mortality[1,5]				Weight (mg)[2,3]		Days to[4]	
Entry	7 d	10 d	Pupa	Adult	10 d	Pupa	Pupa	Adult
4200W 74-3	0[ns]	0[ns]	18.2 abc[6]	27.3 bcd	25.5 abc	104.8 fgh	36.7 a	50.6 a
4200W 49-17	0	0	28.6 ab	52.4 a	28.9 bcd	109.9 e-h	35.3 ab	49.3 abc
Greg Norman-1	0	4.2	17.6 abc	23.5 bcd	20.2 a	108.2 e-h	35.2 abc	49.5 abc
CCB 10-8	0	0	4.6 c	9.1 d	31.4 cde	144.0 bc	34.4 bcd	49.2 abc
4200W 53-1	0	0	39.1 a	52.2 ab	22.6 ab	109.4 e-h	34.2 cd	48.6 a-d
4200W 51-14	0	0	4.6 c	13.6 cd	34.2 d-g	120.5 def	33.8 cde	48.1 c-f
Midlawn	4.2	4.2	13.0 abc	21.7 bcd	38.5 fg	143.8 bc	33.6 de	48.3 bcd
4200W 47-7	0	0	0.0 c	0.0 d	30.7 cde	149.1 b	33.5 de	48.7 abc
Tifton 94	0	12.5	20.9 abc	25.0 bcd	40.5 g	155.7 b	33.0 def	48.2 cde
ERS-Turf	0	4.2	16.7 abc	16.7 cd	51.6 h	145.1 bc	32.6 ef	47.8 c-f
CCB 25-6	0	0	16.7 abc	16.7 cd	39.2 fg	138.6 bcd	32.4 efg	47.6 c-f
4200W 56-14	4.2	8.3	13.6 bc	22.7 cd	52.1 h	127.6 cde	31.7 f	46.4 d-g
4200W 47-1	0	0	5.9 c	11.8 d	49.9 h	118.1 ef	31.1 g	46.0 efg
4200W 55-5	0	0	31.6 abc	42.0 abc	48.4 h	116.7 efg	31.0 g	45.9 fg
CCB 24-4	4.2	4.2	4.2 c	4.2 d	70.5 I	189.9 a	28.6 h	44.6 g

[1] Mean % of larvae alive at 7 and 10 days after egg hatch, % pupation and % that emerged as adults.
[2] Mean weight of surviving 10-day-old larvae after feeding on each genotype for 6 days.
[3] Mean pupa weight for only individuals that pupated (weight taken within one day after pupation).
[4] Mean number of days from egg hatch to pupation and to adult emergence for surviving insects.
[5] Analysis was made on arcsine transformation of the % mortality; % mortality is presented.
[6] Means in a column not followed by the same letter are significantly different by Waller-Duncan k-ratio t test (k = 100, P = 0.05). ns = not significant.

A Turfgrass Genome Project: Integration of Cynodon Chromosomes with Molecular Maps of the Cereals

University of Georgia

Andrew H. Paterson

Start Date: 1999
Number of Years: 5
Total Funding: $125,000

Objectives:

1. *Establish a primary molecular map for the chromosomes of Cynodon.*
2. *Align the chromosomes of Cynodon with those of the major cereals, gaining access to much genetic information.*

We will combine new DNA probes for *Cynodon*, with tools that have been previously mapped in other *Poaceae*, to develop a primary molecular map of the *Cynodon* chromosomes. The map will be useful for investigating many aspects of turfgrass population biology and genetics, and a molecular conduit for turf improvement to benefit from the large body of genetic information now accumulated about cereals and other grasses. *Cynodon* is chosen as a focal point for turf genome analysis due to its importance across the southern United States, and abundance of phenotypic variation. Dr. Wayne Hanna will assist in population development and maintenance.

To our knowledge, this project is the first effort to enable turf improvement to benefit from extensive genetic information available for well-studied grains such as maize and rice. The *comparative approach* will reduce costs, and leverage existing information and tools. Our experience in molecular analysis of complex populations, such as sugarcane and buffelgrass, as well as grain crops such as rice, maize, and sorghum, together with our extensive repertoire of molecular tools, puts us in a strong position to efficiently develop a *Cynodon* molecular map useful for turf improvement.

Progress to Date. While the genetic crosses are being developed for making the maps, we have made significant progress in characterizing DNA clones from bermudagrass and other grasses (especially *Pennisetum* and Sorghum), for their effectiveness in detecting DNA markers in bermudagrass. There exists a high level of DNA polymorphism in bermudagrass, and the establishment of DNA fingerprints unique to individuals will be routine. We have prepared more than 1,000 cDNA clones (mapped in other taxa) to be applied to bermudagrass. DNA extraction protocols for bermudagrass have been optimized. We have initiated screening of these DNA clones on

genomic Southern blots of DNA from the bermudagrass parents being used in this study. The specific parents used in crossing have been clonally propagated, so that we can greatly increase our supply of DNA and blots, and accelerate accumulation of data.

The data accumulated to date suggests that we will be able to not only meet, but also significantly exceed the proposed goal of 300 mapped loci. The comparative mapping of bermudagrass will draw heavily upon a prior map of sorghum that now includes more than 2,000 DNA loci, and a new map of buffelgrass (Jessup et al., in preparation) that now includes about 400 loci.

Plans for Continuation. The focus of year two will be the scale up of identifying DNA polymorphism, and the beginning of genetic linkage mapping. New lab facilities and personnel will facilitate this. We are anticipating that individual seedlings from most, if not all, of the required genetic populations will be large enough to begin sampling of tissue for DNA during year two. Full-scale genetic mapping will be done in year three. By the end of year three, we expect to meet the formal goals proposed for the full five years (data analysis may continue into year four). We will then apply the map to identify quantitative trait loci (QTLs), identify DNA markers of agriculturally important traits, and develop a small bacterial artificial chromosomes (BAC) library for bermudagrass.

Once the accumulation of genetic differences and DNA markers is proceeding smoothly, as time permits we will begin to explore the development of large-DNA clones of bermuda in BACs. Our prior successes with sorghum (Lin et al, submitted), papaya (Ming et al, in preparation), cotton (Abbey et al, in preparation; Rana et al., in preparation), and peanut (Burow et al., unpubl. results) have led to optimization of BAC technology that should be easily transferable to bermuda.

Leveraging Opportunities Realized. Plant genomics in the United States was recently stimulated by the infusion of nearly $85 million in federal grants through the National Science Foundation. The US Golf Association designated Dr. Paterson's turfgrass genome project to be *matching support of a comparative grass genomics initiative* that Dr. Paterson proposed, together with seven colleagues at three universities. Dr. Paterson's proposal was funded at a level of $3.2 million. This award will provide molecular conduits that will enable improvement of bermudagrass and other turfgrasses to benefit from the rapid progress that is anticipated for grass genomics as a result of the USGA-sponsored project.

Other Significant Events, and their Consequences. Dr. Paterson has recently accepted a Senior Professorship in Plant Biotechnology and Genomics, at the University of Georgia's main campus in Athens, GA. He has the *right of first refusal* to be named the director of the AGTEC Plant Division, to be created in 1999. This will result in a major scale-up of genomics activities in Georgia, and will be a significant expansion of the capabilities that Dr. Paterson's lab can bring to bear on bermudagrass. In the first year (before AGTEC is created), Dr Paterson will occupy about 4,800 square feet of renovated space, fully and newly-equipped for genomics research. When the AGTEC Center is completed in early 2000, around 2,600 square feet of additional lab space, as well as shared core facilities of 5,000 square feet for genomics. About $2.1 million is available for equipping the genomics core facility.

While this move has caused some delays, specifically preventing Dr. Paterson from hiring a person dedicated solely to the bermudagrass project during the first year, by leveraging the activities of other people in the lab they have stayed on schedule. The expanded space and equipment available will greatly accelerate our rate of progress, and we emphasize that we expect to exceed the proposed objectives. Texas A&M has agreed to release Dr. Paterson's extramural grants and funds remaining on these accounts to the University of Georgia. The University of Georgia has agreed to honor the terms of existing contracts.

One advantage of Dr. Paterson's move is that he will be at the same university where Dr. Wayne Hanna is a faculty member. Drs. Paterson and Hanna have already jointly offered an assistantship to Mr. Russell Jessup to focus on the bermudagrass project. Mr. Jessup is presently completing M.S. studies with Dr. Paterson and Dr. Mark Hussey at TAMU, working on *Pennisetum*. The offer remains pending, and contingent on the student's formal acceptance into the University of Georgia graduate school. Consequently, the short-term delays resulting from this move are expected to yield great rewards in long-term progress and capabilities.

Development of Improved Bentgrass Cultivars with Herbicide Resistance, Enhanced Disease Resistance and Abiotic Stress Tolerance through Biotechnology

Rutgers University/Cook College

Faith Belanger

Start Date: 1998
Number of Years: 5
Total Funding: $250,000

Objectives:

This project seeks to conserve golf course natural resources while providing quality playing surfaces by improving creeping bentgrass through transformation. We have concentrated on important bentgrass varieties and selections developed for Northeast golf greens.

The goals of this project are to produce improved creeping bentgrass cultivars through a combination of genetic engineering and breeding. Our aim is to provide golf course managers with more effective and selective weed control with herbicides by developing herbicide resistant cultivars. We are also attempting to produce cultivars with improved disease resistance and abiotic stress tolerance that can be maintained in a more environmentatly-sound and cost-effective manner.

The effectiveness of genetically-engineered herbicide resistance in creeping bentgrass has been demonstrated in multiple field tests. This trait is now ready to be incorporated into a commercial cultivar.

We currently have fifty independent transgenic lines of creeping bentgrass expressing one of five potential disease-resistance genes. We have established randomized replicated field trials of these plants that will be evaluated in the summer of 1999.

We also have transgenic plants from bombardments with HVA1, a potential drought and salinity tolerance gene. Plants found to be expressing HVA1 will be screened for effectiveness of the gene.

Cultivar Development and Extreme Temperature Tolerance of Greens-type Poa annua

Pennsylvania State University

David R. Huff

Start Date: 1998
Number of Years: 5
Total Funding: $175,000

Objectives:

1. *Collect, select, breed, and develop genetically stable and phenotypically uniform cultivars of greens-type Poa annua for commercial production.*
2. *Develop techniques to screen large numbers of germplasm accessions for tolerance to extreme temperatures and coverage by sheets of ice.*
3. *Identify genetic markers associated with genetic loci (genes) controlling agronomically important traits and specific stress tolerances in order to aid in the breeding and development of improved cultivar of greens-type Poa annua.*

Annual bluegrass *(Poa annua* L.) makes up a large portion of the putting surfaces in many regions of the United States and Canada. Given its wide-spread occurrence in the golf industry, there is currently a need for high quality, commercially available sources of greens-type *P. annua* for use in constructing, renovating, or maintaining *P. annua* golf greens. Greens-type P. *annua* actually has many characteristics that make them enviable as a putting surface. They typically have high shoot densities (9,000 shoots dm^{-2} or 7 to 24 times higher than that of bentgrass), an upright growth habit, and aggressively inhabit golf greens maintained at extremely close ($\leq$ 0.125 inches) mowing heights. The purpose of this research is not to replace creeping bentgrass as a putting surface, but simply offer an alternative for those golf courses where *P. annua* is simply a better choice.

Germplasm Collection and Evaluation: To date, this project has collected over 2,500 samples of greens-type *Poa annua* from regions including the northeast United States (Pennsylvania, New Jersey, Long Island NY) the mid-Atlantic (Delaware, Maryland, Virginia), and the Pacific northwest (Oregon, Washington). The performance and morphological features of field plot accessions are beginning to demonstrate that there is a tremendous amount of naturally occurring variation between regions, among golf courses within regions, and even among samples within a golf green.

Field resistance to dollar spot disease was observed during summer and fall of 1998 in one particular accession collected from Long Island, NY. This particular accession was completely free of dollar spot disease while all other surrounding plots were moderately to heavily diseased. This disease occurred naturally and was not treated with fungicides.

Currently, our collections of greens-type *Poa annua* exist as a collection of naturally occurring ecotypes and, as such, display a wide range of variation in many, many agronomically important traits. This variation is partitioned among individual plants due to its self-pollinated breeding system and thus, is readily accessible through selection as distinct, uniform, and stable inbred lines. Initially, ecotypic and mass selection of elite germplasm will be used for the development of cultivars. As regional testing and evaluation begins to identify genetically superior strains, these elite strains will begin to serve as parental sources for the cross-hybridization and subsequent single line selection that will eventually result in improved commercial cultivars.

Regional Testing: Based on the 1998-season plot evaluations, a renewed emphasis must be placed on extensive regional testing. In order to enhance and expand regional testing efforts, we have begun to identify cooperators willing to evaluate our experimental strains in golf green plots.

Seed Production and Increase: Seed of the selected accessions were sown into seed production plots (approximately 5 ft. x 20 ft.), in September 1998, for further seed yield evaluation and for generating seed increase for further regional testing. We are expecting a reasonable, though limited, seed harvest for the summer of 1999.

Genetic Identification and Manipulation of Polyhaploids. The evolutionary history of *Poa annua* (allopolyploidy) suggests that observed sexual sterility of particular strains is likely. This may be due to the genetic state of these accessions being sterile polyhaploids (plants derived from an unfertilized, reduced egg). We have begun a set of experiments in an attempt to restore fertility to several sexually sterile accessions.

Transformation of Bermudagrass for Improved Fungal Resistance

Oklahoma State University

Michael P. Anderson

Start Date: 1998
Number of Years: 5
Total Funding: $125,000

Objectives:

1. *Isolate, identify, and characterize chitinases and glucanases and their genes that have high anti-SDS activity.*
2. *Develop an efficient protocol to transform (genetically engineer) bermduagrass.*
3. *Transform and characterize bermdagrass with the antifungal chitinase and/or glucanase genes directed against the spring dead spot casual organism.*

A major disease commonly known as spring dead spot (SDS) causes significant economic damage to bermudagrass in the Southeastern United States. The causal agent for SDS throughout most of the United States is *Ophiosphaerella herpotricha* and *Ophioshpearella korrea.* Both fungal species are very active in the fall and early spring when the temperatures are cool and moisture is plentiful. Infected areas appear as regular circular patches of dead and diseased turf that generally occurs in more mature stands of bermudagrass.

The long-term goal of this project is to increase resistance in bermudagrass turf varieties to SDS through gene transformation technology. This report describes the current progress and results for the development of the transformation system and the isolation and characterization of anti-fungal factors during 1998.

Bermudagrass Transformation. The use of high velocity micro-projectiles (biolistics) to deliver recombinant DNA into intact plant cells has been successfully utilized to transform many grass species, and is considered the method of choice for most grass species. The immature inflorescences of *BRAZOS* bermudagrass, a forage cultivar, were used to induce the formation of embryogenic callus tissue. *BRAZOS* was chosen for this experiment because it had previously demonstrated superior growth and plant regeneration potential in tissue culture. Tissue was transformed with a plasmid containing two chimeric genes of interests, the *bar* and *uidA* genes, under the control of ubiquitin promoters. The *bar* and *uidA* genes serve as a selectable marker and reporter gene, respectively. The GUS enzyme, coded for by *uidA,* can be assayed by accumulation of fluoro-genic products by providing the enzyme substrate. PAT detoxifies bialaphos (the active ingredient in the herbicide Liberty) in the selective media; thereby allowing transgenic cells, and plants to continue to grow. Six hundred and seventy one putative transgenic plants have been recovered from this experiment. We are currently evaluating these putative transformants with PCR to determine if they contain the *bar* gene. PCR positive plants will be characterized by Southern analysis and enzyme assays for phosphinothricin acetyl transferase during 1999.

Anti-SDS Proteins. Living organisms produce many antimicrobial compounds to protect themselves from pathogens, or to give them a competitive advantage for nutrients. They range from the small molecular weight antibiotics and secondary metabolites to the larger macromolecular proteins and assorted polypeptides. Recently we discovered a bacterium that was strongly and persistently inhibitory towards *0. herpotricha.* The bacterium was identified to the genus taxonomic level with confidence by a GC-FAME and BIOLOG technology. This bacterium secreted many proteins into the extra-cellular matrix. Dialysis of the extra-cellular excretions suggested that the anti-fungal factor was a protein. Purification of the anti-fungal proteins on anion exchange, hydroxyapatite, and Mono Q chromatography resulted in the isolation of a 36 kD protein that is most likely expressed as multiple isoforms. Analysis of the purification results suggested that there are at least two distinct anti-fungal factors antagonistic against *0. herpotricha* secreted by the bacterium. Experiments are in preparation to identify, sequence, and characterize the 36 kD protein.

Selecting Seeded Zoysiagrass for Cold Hardiness

University of Missouri-Columbia

Suleiman S. Bughrara

Start Date: 1998
Number of Years: 5
Total Funding: $91,535

Objectives:

1. *Evaluate zoysiagrass germplasm for cold hardiness using the cold chamber techinque.*
2. *Evaluate seed production of selected zoysiagrass strains under Missouri environmental conditions.*
3. *Our long-term objective will be to develop cultivars of zoysiagrass that can be seeded and that have desirable winter hardiness for the transition zone.*

The zoysiagrass breeding program at the University of Missouri was initiated in Summer 1997 by planting over 500 clones in a spaced nursery at Bradford Farm, Columbia, Missouri. The main sources for these clones were the Georgia Plant Introduction Station, Bobbi Murray (Jack Murray's widow) and some clones that collected from golf courses around Columbia, Missouri. Two genotypes with good turf and seed production characteristics are shown in Figure 7.

Figure 7. Clones were planted on 3-foot centers.

Sixty clones that survived the mild winter of 1997-1998 in Missouri and exhibited good seed production were planted in turf plots in spring, 1998, for further evaluation. In addition, 56 clones that were brought from Rutgers University, in cooperation with both Drs. Funk and Meyer were planted in plots adjacent to the Missouri selections. The Rutgers material will be evaluated in both Missouri and New Jersey.

Several laboratory studies were conducted to characterize the germination process and evaluate factors affecting seed dormancy. The floret consists of a caryopsis that is covered by a lemma and palea that adhere tightly. The lemma is very thick, has thickened cell walls and a wax coating on the outer surface that may restrict water penetration. Naked caryopses (lemma and palea removed) germinated up to 80 percent at 22 °C, whereas intact florets germinate less than 10 percent. Florets that were cut transversely either at the base (below embryo) or at the tip (above embryo) germinated almost as well as naked caryopses.

These results suggested that water uptake is a dormancy factor. Water extracts of florets did not affect germination of naked caryopses, but inhibited germination of base and tip cut florets by 25 percent. The nature of the germination is not known. At 35 °C light and 20 °C dark, 90 percent of untreated florets germinated, even in the extract treatment. This indicates the need for high temperature in order for rapid germination to occur. Thirty-percent KOH scarification (15 min.) enhanced germination at 35/20 °C up to 98 percent. Seed treatments can overcome dormancy, which appears to be partially physical, and improve germintation. To understand the genetics of the process we developed 19 half-sib families. These will be evaluated for germination properties in the future.

On-Site Testing Putting Green Variety Trial

National Turfgrass Evaluation Program

Kevin Morris

Start Date: 1996
Number of Years: 5
Total Funding: $71,600

Objectives:

Provide bentgrass and bermudagrass putting green trials on golf course practice putting greens.

The Golf Course Superintendents Association of America (GCSAA), United States Golf Association (USGA) Green Section and National Turfgrass Evaluation Program (NTEP) jointly sponsored on-site trials for bentgrass and bermudagrass cultivars grown on USGA specification putting greens.

Location of Trial Sites. The sixteen sites, course superintendents, and research cooperators are named in Table 10. The bentgrass trial sites are established. The Snoqualmie Ridge Golf Course in Snoqualmie, Washington was the last to be selected and seeded. It is established and has received limited play. It should be in excellent shape for next season.

The bermudagrass sites are constructed and established. Progress on all trial sites is meeting expectations.

Site Visits. As of the date of this report, we have visited trials. All sites visited are doing well. Cooperation is outstanding and enthusiasm among researcher cooperators, host course superintendents, and golfers is extremely positive. Research cooperators will submit their data to NTEP for analysis later this winter. The NTEP will summarize the data and report the first year's results in spring, 1999. A meeting to discuss the on-site trials is scheduled for the GCSAA Conference and Show in Orlando. This meeting will serve as a "feedback mechanism", allowing comment and discussion from the researchers and host superintendents. We will use this information to make appropriate adjustments in data collection and analysis during the 1999-growing season.

Table 10. The GCSAA/USGA/NTEP on-site trials for bentgrass and bermudagrass grown on USGA specification putting greens. Grasses being evaluated at each site are indicated in parenthesis.

Host Club	Course Superintendent	Research Cooperator
1. Bent Tree Country Club Dallas, Texas (Bentgrass & Bermudagrass)	Keith Ihms	Dr. Milt Engelke
2. Lassing Pointe Golf Course Florence, Kentucky (Bentgrass)	Jerry Coldiron	Dr. A. J. Powell
3. Country Club of Mobile Mobile, Alabama (Bermudagrass)	Ron Wright	Dr. Bryan Unruh
4. Crystal Springs Golf Course Burlingame, California (Bentgrass)	Ray Davies	Dr. Ali Harivandi
5. Fox Hollow at Lakewood Lakewood, Colorado (Bentgrass)	Don Tolson	Dr. Tony Koski
6. Snoqualmie Ridge Golf Course Snoqualmie, Washington (Bentgrass)	Mark Cupit	Dr. Gwen Stahnke
7. Lakeside Country Club Houston, Texas (Bermudagrass)	Mike Sandburg	Dr. Richard White
8. North Shore Country Club Glenview, Illinois (Bentgrass)	Dan Dinelli	Dr. Tom Voigt
9. Purdue University North Course West Lafeyette, Indiana (Bentgrass)	Jim Scott	Dr. Clark Throssell
10. SCGA Members Club Murrieta, California (Bentgrass & Bermudagrass)	John Martinez	Dr. Robert Green
11. Country Club of Birmingham Birmingham, Alabama (Bentgrass & Bermudagrass)	Lee Mc Lemore	Dr. Elizabeth Guertal
12. Country Club of Green Valley Green Valley, Arizona (Bentgrass & Bermudagrass)	Mark Clark	Dr. David Kopec
13. Jupiter Island Club Hobe Sound, Florida (Bermudagrass)	Rob Kloska	Dr. John Cisar
14. The Missouri Bluffs St. Charles, Missouri (Bentgrass and Bermudagrass)	Robert Deardeuff	Dr. John Dunn
15. Westchester Country C8lub Rye, New York (Bentgrass)	Joe Alonzi	Dr. Jim Murphy
16. Westwood Golf Course Vienna, Virginia (Bentgrass)	Walter Montross	Dr. David Chalmers

NTEP Statistical Analysis Grants

National Turfgrass Evaluation Program

Kevin Morris

Start Date: 1999
8Number of Years: 1
Total Funding: $23,000

Objectives:

1. *Evaluation of ANOVA diagnostics and the validity of assumptions about turf quality ratings.*
2. *Assessment of spatial variation.*
3. *Evaluation of factors affecting the success or failure of varietal separation.*
4. *Quantification of the value of test locations - varietal separation and uniqueness of ranking.*
5. *Assessment of plot size and experimental design efficiency.*

In an interest to improve the acquisition and analysis of National Turfgrass Evaluation Program variety trial data, the Policy Committee discussed the current trial setup, experimental design, and statistical analysis procedures. Five one-year statistical analysis projects were selected for funding in 1999. NTEP data available for evaluation includes the Bentgrass Putting Green and Fairway Trial, Kentucky Bluegrass Trial, and Perennial Ryegrass Trial. Research proposals were considered for the following five areas.

Evaluation of ANOVA diagnostics and the validity of assumptions about turf quality ratings. Is the current 1-to-9 rating system the best system for accurately assessing quality. The rating scale assumes a quantitative measurement when in reality it is qualitative in nature. Much of the rating scale is not used by some (or many) cooperators, therefore a normal distribution (bell-shaped curve) is not produced. If cooperators used more of the rating scale would better data be produced?

Assessment of spatial variation. How effective are cooperators at establishing uniform sites and collecting uniform data? For instance, disease data is often not very significant statistically. Is this because the disease did not develop uniformly throughout the plot area? What procedures might we use to determine if plots are uniform?

Evaluation of factors affecting the success or failure of varietal separation. Why do some locations achieve more varietal separation than other locations? When using more of the rating scale do we see more varietal separation or less?

Quantification of the value of test locations - varietal separation and uniqueness of ranking. Can the use of cluster analysis and correlation among locations, years and seasons within years be used to group or separate locations? Research in this area could lead to logical geographic groupings of locations and specific regional analysis.

Assessment of plot size and experimental design efficiency. Are there any changes that can be made to the way tests are designed (the efficiency of the randomized complete block design), proper plot size, number of replications, etc. that can make for better tests and data?

Short summaries of the five funded projects will be discussed in next year's annual summary.

On-Site Fairway Overseeding Trials

National Turfgrass Evaluation Program

Kevin Morris

Start Date: 1999
Number of Years: 2
Total Funding: $ 20,674

Objectives:

Evaluate new cultivars on bermudagrass fairways at golf courses in the Southern and Western United States that will provide scientific information of a more applied nature about cultivars for overseeding.

With the initiation of on-site testing of bentgrass and bermudagrass on putting greens, interest is now increasing for the evaluation of other grasses used on golf courses. Grasses are needed that provide exceptional playing surfaces with less pesticides, fertilizer and water. Therefore, grasses that have superior drought, cold, heat, disease and insect resistance need to be identified.

Overseeding bermudagrass fairways is a common practice throughout the southern half of the United States. Millions of pounds of seed are bought and sown each autumn on golf courses in this region. Golf course owners, managers and superintendents seek grasses that establish quickly, exhibit exceptional playability, are aesthetically pleasing and require less input. This project will evaluate new cultivars on bermudagrass fairways at golf courses in the Southern and Western United States. This on-site testing program will provide scientific information of a more applied nature about cultivars for overseeding.

Information from this project will be valuable to the golfing industry because it will determine the adaptation of grasses for golf course use. Information obtained from on-site testing will be of particular value to plant breeders, researchers, extension educators, USGA agronomists, golf course architects, and superintendents who need to select the best adapted cultivars for overseeding in a particular regional climate.

Location and Number of Trial Sites. The evaluation trials will be jointly sponsored by the Golf Course Superintendents Association of America (GCSAA), the United States Golf Association (USGA) Green Section and the National Turfgrass Evaluation Program (NTEP). Trial sites will be located on golf courses near a land grant university with a turfgrass research program or in a major metropolitan area that is readily accessible to a university turfgrass scientist. Ten evaluation trial sites are proposed. Trials will be positioned strategically in the following areas: southern California, Arizona, south Texas

(Houston), Gulf Coast (LA, MS, FL), central or south Florida, Myrtle Beach, SC, North Carolina/Virginia, Georgia/Alabama, Oklahoma/southern Kansas.

Trials will be located on active play sites where golfers hit fairway golf shots and/or drive golf carts. Host clubs will provide daily maintenance of the fairway site. It is preferred that host clubs have a history of supporting the USGA and receiving visits from USGA agronomists. The superintendent should have excellent skills and a strong record of supporting GCSAA and the USGA. The superintendent should have good relationships with the university scientist, who will have ultimate responsibility for the trial.

The Executive Director and Special Projects Coordinator of NTEP, USGA Construction Education Coordinator and Green Section Director of Research and the GCSAA Research Director will determine the location of trial sites.

Trial Specifics. The NTEP will function as the coordinating agent for this two-year cultivar trial. Because overseeded grasses provide a temporary playing surface mainly in fall and winter and are reseeded each year, cultivars will be seeded in two consecutive years. Trials will be conducted under mutually agreed upon guidelines, procedures and funding outlined in a research agreement to be drafted and signed by appropriate representatives of GCSAA, USGA and NTEP. Trials will be conducted under the leadership of a university turfgrass research scientist (i.e., research cooperator), who has a faculty appointment. This person will sign a research agreement and will be responsible for establishment of the trial, coordination of the maintenance regime, collection and submission of the data to NTEP.

The NTEP will solicit entries for the trial from sponsoring companies. Trials will be conducted with named cultivars and commercially available blends or mixtures. Various species used in overseeding, such as perennial ryegrass and *Poa trivialis* will be allowed. Experimental lines that will be released in the immediate future (i.e. before the end of the testing cycle) may also be included in this trial at the sponsor's discretion.

Trials will be maintained according to agreed upon procedures. Establishment and maintenance procedures will be based on recommendations set by an advisory committee consisting of representatives from GCSAA, USGA, NTEP, universities and the turfgrass seed industry. Daily maintenance will be conducted by the golf course superintendent at the expense of the host club.

The NTEP will administer the program and its funding, set the advisory committee and gather their input and recommendations for the trial. The NTEP will organize and distribute the seed that will constitute entries for each trial location. The NTEP will provide maintenance and data collection protocols to each site, collect, analyze and disseminate the performance data in annual and final reports, and conduct an annual site visit of each trial site.

Data Collection. The research cooperator will be responsible for data collection. The following data will be collected from each trial:

1. Establishment rate, seedling vigor, percent ground cover (4-6 weeks after seeding).
2. Turfgrass quality (monthly).
3. Plot color (twice - late fall/early winter and spring).
4. Texture (once per season).
5. Rate or speed of transition from overseeded grass to bermudagrass.
6. Environmental stress, traffic and divoting damage, disease and insect damage and other data deemed appropriate and feasible by the research cooperator.

The research cooperator will be responsible for submission of data to NTEP by August 1 of each year.

Special Report on Turfgrass Diversity

Diversity, GRCS, Inc

Deborah Strauss

Start Date: 1998
Number of Years: 2
Total Funding: $36,000

Objectives:

1. *The results and analysis of the USGA turfgrass study, and overviews of the relevant USGA research grants.*
2. *A history of major turfgrass germplasm collections and their evaluation for use in developing golf courses.*
3. *A report on the National Turfgrass Evaluation Program (NTEP).*
4. *Articles from experts within both the golfing industry and the larger research and breeding community on innovative turfgrass breeding research including how biotechnology is or could be used in turfgrass breeding regimes.*
5. *A perspective on how US turfgrass breeding and use relates to international efforts.*
6. *Interviews with key leaders in forage and turfgrass research and breeding, including the chairs of the US Forage and Turfgrass Crop Germplasm Committee (CFC), and the Guelph Turfgrass Institute, NTED, and others.*
7. *Interviews with representative industry (seed and golf) leaders.*

The rapidly developing interest in golf around the world has put increasing demands on the golfing industry to develop and maintain improved and more environmentally sound golf courses. These demands come not just from the leisure and sporting community, but also from the communities in which the growing number of golf courses are located and where they fill significant needs for green belts and park spaces, particularly in the drier areas of the world.

These circumstances underscore the need for the dissemination of scientific information and expert views about the often-overlooked agricultural commodity of turfgrass. Golf courses require the genetic materials that will keep them

green—hardy turfgrass that is of high quality and resistant to the scourges of pests, heat, and drought. The success of golf courses—environmentally and economically—depends upon the availability of a wide range of turfgrass germplasm for plant breeders as they continually search for ways to breed new varieties that exhibit these qualities.

The independent, quarterly news journal DIVERSITY is a most appropriate vehicle through which to bring together global information and viewpoints on the agricultural and environmental aspects of turfgrass research. DIVERSITY is the flagship publication of the non-profit Genetic Resources Communication Systems, Inc. (GRCS). GRCS has had a long, successful record of accomplishment in addressing information needs of the plant genetic resources community. During the next year, GRCS will prepare and publish a special report on turfgrass—a subject area that has received very little attention—in order to bring together and engage both agricultural and environmental interests.

Environmental Impact of Golf

The public is concerned about the effects of golf courses on the environment. In response to this concern, the USGA has conducted research examining the fate of turfgrass pesticides and fertilizers since 1991. The USGA continues to support scientifically based investigations on the environmental impact of golf courses. The focus remains on research to understand the effects of turfgrass pest management and fertilization on water quality and the environment.

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

Pesticide and Nutrient Fate Models are used to predict the environmental impact of turfgrass pesticides and fertilizers. Prediction models are only as good as the information upon which they are developed and calibrated. From 1991 through 1997, research sponsored by the USGA demonstrated:

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

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

Development of a Layered Model to Predict Pesticide Transport in Turfgrass Thatch

University of Maryland

Mark J. Carroll
Robert L. Hill

Start Date: 1998
Number of Years: 2
Total Funding: $49,880

Objectives:

1. *To develop a two phase layered pesticide transport model which considers equilibrium or non-equilibrium transport with in each layer and the use of appropriate pesticide adsorption coefficients for each layer.*
2. *To evaluate the use of the model for two of the pesticides used in the previously funded USGA study.*
3. *To evaluate the effectiveness of the model to predict pesticide transport in comparison to commonly used pesticide transport models such as PRZM2 or GLEAMS.*

Pesticides applied to mature turf move into the soil only after being washed off foliage and move through turfgrass thatch. Any attempt to predict the movement of pesticides applied to turf requires that the retention characteristics of the pesticide to foliage and thatch be known.

In 1997 and 1998, a series of sorption and transport studies were conducted to characterize the movement of carbaryl in soils containing a surface layer of turfgrass thatch. The sorption studies were conducted using a device called a mechanical vacuum extractor. This device precisely controls the rate at which a solution moves through a column of porous media.

The adsorption and desorption properties of a 3½ year old, 2.3 cm thick *SOUTHSHORE* creeping bentgrass thatch, and a 6 year old, 3.4 cm thick *MEYER* zoysiagrass thatch were compared with the soil residing below each thatch layer.

The adsorption kinetics of carbaryl to thatch and soil were similar. Sorption equilibrium was achieved within 4 hours in all media. The thatch from both turfgrass species had much higher carbaryl adsorption capacities than the underlying soil. There was, however, no difference in the adsorptive capacities of the two turfgrass species thatch. The normalized sorption coefficients of the four media were similar suggesting that differences in the carbaryl sorptive capacities of thatch and soil were solely due to differences in the organic carbon content of the media.

Desorption losses were evaluated by subjecting columns of thatch or soil to three successive leaching events. The leaching events took place after allowing carbaryl to adsorb to the thatch or soil for 24 hours. The amount of carbaryl detected in the leachate was used to determine the proportion of carbaryl that was desorbed from the sample. Carbaryl retention in soil was much lower than in thatch during the first leaching event. By the end of the third leaching event, there was little difference in the proportion of carbaryl retained in the bentgrass thatch and soil. In contrast, zoysiagrass thatch always retained a greater proportion of carbaryl than the underlying soil. This suggests that carbaryl is more tightly bound to zoysiagrass thatch than to the underlying soil (Figure 8).

Undisturbed columns of soil, or soil plus a surface layer of thatch, were used to determine the effect of thatch on the carbaryl transport in soil. Columns having a surface layer of zoysiagrass thatch were more effective in reducing carbaryl transport than columns having a surface layer of creeping bentgrass thatch. Visual examination of the bentgrass site columns revealed extensive earthworm burrowing. The channels present in these columns likely reduced the effectiveness of bentgrass thatch to inhibit carbaryl transport.

Bromide and carbaryl breakthrough curves obtained from the transport study were used to evaluate the performance of the linear equilibrium (LEM) and the two-site non-equilibrium (2SNE) models to predict carbaryl transport. The latter model uses a non-equilibrium form of the convective-dispersion equation to predict solute movement in porous media while the former uses a linear equilibrium form of the equation to predict solute movement. The carbaryl breakthrough curve (BTC) data were also used to compare the use of column

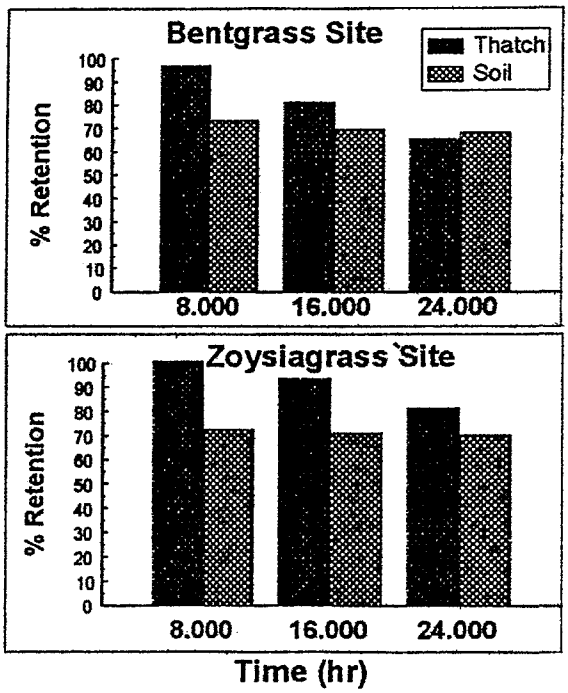


Figure 8. Cumulative proportion of carbaryl retained to thatch and soil following three successive 8 hour leaching events.

retardation factors (R) based on our laboratory measured thatch and soil sorption coefficients with model fitted R's to predict carbaryl transport.

Modeling of bromide transport presented strong evidence of significant two-domain flow in all columns except the zoysiagrass soil columns. In columns exhibiting two-domain flow, use of retardation factors based on laboratory measured adsorption coefficients accounted for 74 to 94 percent of the variability in carbaryl transport. Slightly improved estimates of carbaryl transport were obtained when R was kept as a fitting parameter. In columns where two-domain flow was not apparent, the LEM model satisfactorily described carbaryl transport only when R was curve-fitted. Use of R's based on laboratory derived adsorption coefficients resulted in poor LEM estimates of carbaryl transport. The 2SNE model gave reasonable estimates of carbaryl transport when R was calculated using the adsorption coefficients determined in our sorption studies.

Nitrogen and Phosphorus Leaching and Runoff from Golf Greens and Fairways

University of Georgia

Larry M. Shuman

Start Date: 1998
Number of Years: 3
Total Funding: $75,000

Objectives:

1. *Quantify the amounts of nitrogen and phosphorous that leach from USGA greens under various management practices.*
2. *Determine the amounts of nitrogen and phosphorous that runoff from a Southeastern piedmont soil under various management practices including the effect of buffer zone width and irrigation scheduling with respect to fertilizer application.*
3. *Determine the effects of forms of phosphorous, dissolved organic carbon (DOC), soil compaction and crusting, and climatic variables on phosphorous leaching and runoff. This information will be incorporated existing fate prediction models.*
4. *Develop best management practices to limit leaching and runoff on nitrogen and phosphorous form golf course greens and fairways.*

A project was initiated to determine the potential transport of nitrogen and phosphorus by runoff of surface water from fairways and by leaching through golf greens. The research especially emphasizes studies on phosphorus transport. Experiments are being carried out at four research venues developed by Dr. Albert Smith to study pesticide fate. The results from three of these areas are reported here summarizing preliminary data, since this is the first year of the project.

Runoff of phosphorus was greatest at the first simulated rainfall event from bermudagrass plots with a 5 percent slope and receiving three rates of a 10-10-10 fertilizer (Figure 9). The runoff decreased dramatically during subsequent rainfall events. Step-wise increases in phosphorous concentrations in the runoff were found for the 5 and 11 kg ha^{-1} rates for the first runoff event. The total mass of phosphorous transported for all four events was 10.6 and 11.5 percent of that added for the 5 and 11 kg ha^{-1} rates, respectively. Nitrate runoff followed a different pattern resulting in a higher mass of nitrate during the second rainfall event, when the runoff water volume was highest. Since the ammonium form of nitrogen was applied, the amounts of nitrate in the runoff would depend on rates of nitrification as well as transport parameters.

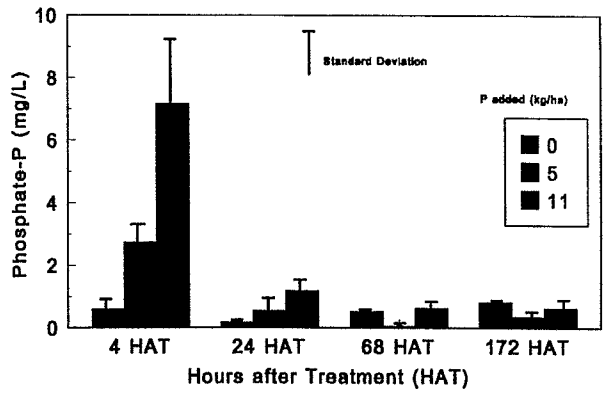


Figure 9. Phosphate concentration in runoff for three rates of 10-10-10 fertilizer. Simulated rainfall at two inches for 4 hours after treatment (HAT), two inches at 24 HAT, and one inch at 68 HAT, and one inch at 172 HAT.

A greenhouse experiment was carried out with columns made to USGA specifications for greens and sodded with bermudagrass. Two sources of balanced fertilizers were applied at four rates to determine potential leaching. The sources applied were a water-soluble fertilizer and a sulfur and poly-coated micro-granular fertilizer to study both fast and slow-release types. These rates were added every other week for a total of six weeks with the last treatment being made at week eleven. Phosphorus concentrations in the leachate were much higher for the soluble source at the end of the eleventh week of the experiment (Figure 10). The difference is especially great at the lowest phosphorous rate (5 kg ha^{-1}). In fact, phosphorous concentrations in the leachate were not different from the control at the granular source rate.

Leaching of nitrogen and phosphorus has been monitored for two working putting greens at an Atlanta Country Club since January 1995. The bentgrass greens were constructed in

the fall of 1994 and were fitted with three lysimeters each. The nitrate concentrations in the leachate did not exceed the 10 mg L^{-1} drinking water standard for the first three years of monitoring. For 1997 the nitrogen concentrations increased in the leachate about 20 to 30 days after application. For all the years, increases in the mass of nitrogen tended to correspond with rainfall events. The concentration of nitrate and the total mass in the leachate is increasing over time. Phosphorus concentrations in the leachate were highest the first year and decreased dramatically thereafter. These initial higher concentrations were probably caused by higher phosphorous applications in 1994 during grow-in.

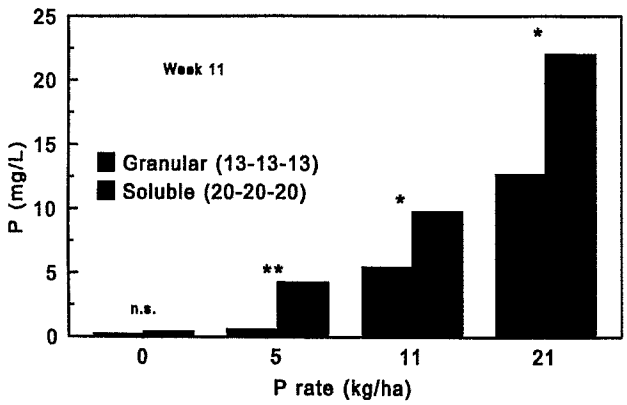


Figure 10. Concentrations of phosphorous through simulated green columns. Rates of applied were applied bi-weekly for a total of six times ceasing on week 11. *, ** indicate significance for sources at each rate (P = 0.05 or 0.01, respectively).

Innovative Water Quality Management Utilizing Wetlands Construction on a Golf Course

Purdue University

Ronald F. Turco

Start Date: 1998
Number of Years: 5
Total Funding: $125,000

Objectives:

Our objective is to evaluate the use of a "closed-loop" water in situ treatment system in terms of:

1. *Use of a golf course wetland to improve residential runoff.*
2. *Protection and improvement of a sensitive wetland environment.*
3. *Regeneration of water supplies for golf course use.*

Previous USGA-funded studies have documented the chemical makeup of water formed during golf course runoff and leaching events. Our work goes beyond monitoring to assess how innovative golf course water quality management, based around a constructed wetland, can reduce pollution from the golf course and from adjacent non-golf course sources. Not only do the wetlands accept water originating from the golf courses but also runoff from a watershed that includes a gas station, retail businesses and parking lots, over 500 residences, and two major city highways. The quality of water will be monitored throughout the system for nutrients, pesticides, salt, automobile fluids, and other possible contaminants.

Earlier results from across the country demonstrated that the quality of water originating from the golf course is expected to be good. We have established an innovative management scheme in which golf course runoff and urban runoff are passed through created wetlands and then used as the primary water source for golf course irrigation. This arrangement is designed to both reduce impacts from the golf course and commercial / residential runoff on an important wetland adjacent to the golf course and to provide a reliable source of water for golf course irrigation. This approach will provide a blueprint that allows for a reduction in golf course nutrient applications and groundwater withdrawals for irrigation.

This project is a model for any location where a golf course interfaces with natural areas or other high value property. The ability of the constructed wetland to remove contamination is being evaluated and documented. The use of the wetland system to clean and remediate roadway water and water from commercial and residential areas is also being followed. For locations where water is expensive or not available, this approach may prove to be an extremely useful way to improve water supply. This approach will add environmental value to the golf course. Roadway water that would have been directly discharged, untreated, to surface water now will be treated in the golf course wetland system before release.

Work to date has concentrated on the integration of the constructed wetland and the golf course. Purdue University's athletic department constructed the Kampen Golf Course that opened in the summer, 1998 adjacent to the new Turfgrass Research and Diagnostic Center. The Kampen Golf Course is a Pete Dye designed facility intended to display state-of-the-art environmentally sensitive golf course management as well as providing an excellent playing surface. In order to meet real and anticipated environmental problems, the Kampen golf course design includes a series of constructed wetlands between the course and the adjacent natural wetland. Moreover, the course is constructed to capture water from the adjacent city highway and residential area. The constructed wetlands will intercept and process runoff, tile water directly

from the golf course, and the water captured from the adjacent urban road, commercial, and residential areas.

We have completed installation of sampling equipment (flow meters, samplers, and infield chemical detection systems) at five of the six sites established as part of this project on the constructed wetland. The samplers began running in September of 1998. However, we have not had a significant enough storm to allow for water collection. It is fully anticipated that most of our run off collections will be starting within the next five weeks.

Description of sample sites:

Site 1. Evaluation of the *typical* water flowing from a mature residential and light industrial setting to the golf course.

Site 2. By subtraction (site 1 from 2), water quality and quantity from a greens-fairway complex.

Site 3. The treatment ability of a single wetland cell for municipal water as well as golf course materials (site 2 from 3).

Site 4. Water quantity and quality as affected by treatment in cell series (site 1 from 4).

Site 5. Water volume and quantity for untreated conditions. By subtraction (site 4 from 5), the impact of a wetland series on the quality of discharge waters.

We are presently employing a technician on the project. To date, the funds for the technician have come from matching monies on the project. We are presently search for a student to be employed on the project. We have several excellent applications and hope to have someone in place by January of 1999.

The Effects of Turfgrass Root Architecture on Nitrate Leaching and Nitrogen Use Efficiency

North Carolina State University

Daniel C. Bowman

Start Date: 1998
Number of Years: 5
Total Funding: $97,830

Objectives:

1. *Extend our current column lysimeter study comparing six different warm-season turfgrasses for NO3 leaching and nitrogen efficiency.*
2. *Measure root architecture (depth, density, dynamics) and other root characteristics (cation exchange capacity, carbohydrate release, microbial association, viability) for the six species.*
3. *Measure the kinetic parameters of nitrogenuptake for each species.*
4. *Determine whether root architecture or uptake kinetics explains the differences between the species.*
5. *Use a state-of-the-art-flow-through nutrient solution culture system to screen germplasm for nitrogen uptake efficiency and to simultaneously determine rooting depth of the genotypes.*
6. *Use genotypes identified in objective five to validate the conclusions regarding rooting architecture vs.uptake kinetics as a primary determinant of nitrogen efficiency.*

As part of the initial phase of this study, several model systems/methodologies have been developed and tested. Large column lysimeters were constructed and installed at the NCSU Phytotron. Each is equipped with sampling hardware to permit recovery of all leachate. A preliminary study to evaluate lysimeter performance was conducted using hybrid bermudagrass sod. We hypothesized that supplementing the fertilizer with soluble carbohydrate could reduce nitrate leaching during turf establishment. This would stimulate microbial immobilization of the fertilizer, and tie the nitrogen up in the rootzone rather than having it leach.

Ammonium nitrate was applied approximately monthly for four months at a rate of 50 kg N ha^{-1}, with sucrose added at rates of 0, 50, 150 and 250 kg C ha^{-1}. Irrigation was applied to maintain a high leaching fraction and maximize leaching potential. Mass emission of nitrogen from the controls amounted to 23, 28, 9 and 7 percent of the applied nitrogen for months one through four, respectively. The reduction in loss with time corresponds to root development. Sucrose addition reduced both $N0_3$ concentration and mass emission 40 to 65 percent compared to controls (Figure 11), suggesting significant increases in microbial immobilization. Sucrose addition did not affect root distribution, which also supports the role of microbial activity in reducing leaching. These data indicate the need to better understand turfgrass soil microbiology, especially regarding nitrogen nutrition. The experiment also validated the performance of the lysimeters, which will be used to monitor root development during year two of the project.

A second objective of the research project is to compare the nitrogen uptake kinetics of several warm-season turfgrass species. We have previously characterized uptake by cool-season turfgrasses using the Classen-Barber depletion technique to quantify the kinetic parameters V_{max} and K_m. The method requires a flowing solution culture system to minimize diffusion limitations. The most common system design uses hydraulic pumps, which are expensive and often troublesome. We have designed a simplified flow-through system using the air-lift principle, which reduces both cost and complexity while maintaining rapid solution flow. Sod will be grown in culture rings until a healthy root system has developed.

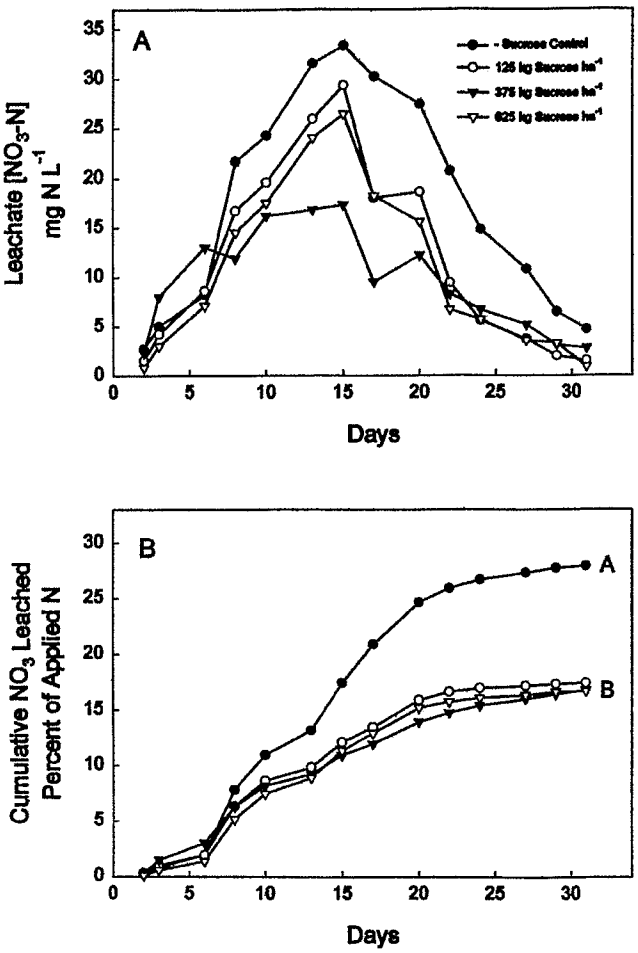


Figure 11. Nitrate-N concentration in leachate (A) and mass emission of nitrogen (B) with time.

Kinetic parameters will then be determined. This work is scheduled for summer of 1999, once sod becomes available.

We are also evaluating the possibility of screening germplasm for nitrogen uptake efficiency using ^{15}N. Seventeen genotypes of Kentucky bluegrass were grown in large flow-through solution culture systems. Nitrogen was maintained in the solution at either constant low-nitrogen concentration with continuous addition via a peristaltic pump, or at high (1 mM) concentration with periodic addition. Screening at low concentration should select for differences in uptake affinity (K_m) while screening at the high concentration selects for uptake capacity (V_{max}). ^{15}N-labeled fertilizer was added transiently to label the plant material. Plants were harvested, separated into roots and shoots, dried, weighed, and ground. The tissue is currently being analyzed by commercial mass spectrometry. Uptake will be expressed on a root weight basis. We will be looking for genotypes that vary significantly in uptake affinity and capacity. Uptake kinetics of selected genotypes will then be verified using the flow-through system.

Comparing Nutrient Losses Via Runoff from a New Golf Course and the Golf Course Site's Previous Native Condition.

Kansas State University

Steve Starrett

Start Date: 1998
Number of Years: 5
Total Funding: $118,155

Objectives:

1. *Compare the nutrient loading, by way of surface water runoff, from a new golf course, and the site's previous native prairie condition.*
2. *Investigate the new golf course's impact on surface water quality during construction and during golf course operation.*

The objective of this research is to compare the nutrient loading, by way of surface water runoff from a new golf course (Colbert Hills Golf Course), and the site's previous native prairie condition. The nutrient loading from the golf course site into the main surface water stream (Little Kitten Creek) will be determined during construction and during operation. Surface water samples are being collected during runoff events from at three locations on Little Kitten Creek. Currently, automated samplers are installed where Little Kitten enters the golf course property, where a small tributary enters the property and where Little Kitten Creek exits the property. About 300 water samples have been collected since February. Water samples will be tested for nutrient concentrations and other physical and chemical parameters. Surface water runoff amounts will be determined so those mass amounts of nutrients contained in the runoff can be calculated.

Kansas State University in cooperation with Jim Colbert, PGA TOUR, GCSAA, and various alumni are building Colbert Hills Golf Course, a 27-hole championship course, near Manhattan, Kansas. Colbert Hills is being built on land that has a prairie-woodland mix that is typical of the Flint Hills Region. The only previous land use was occasional grazing for beef cattle. Data on water quality from the nearby Konza Prairie research area (NSF Long-Term Ecological Site and USGS Benchmark site) has been collected for close to 20 years and comparisons in water quality from Colbert Hills and the Konza Prairie will be made.

Development and Testing of Indices and Models of Pesticide Volatilization from Turfgrass

Cornell University

Douglas A. Haith

Start Date: 1998
Number of Years: 3
Total Funding: $27,724

Objectives:

1. *Develop and test concise indicators of volatilization hazard that can be used by turf managers to determine the likely degree of health hazards associated with pesticide applications.*
2. *Develop and test alternative models of turfgrass pesticide volatilization.*

Mathematical models can potentially be used to estimate volatilization of chemicals applied to turf. However, the complexity and limited testing of volatilization models restrict their general applicability. An alternative procedure estimates concentrations using simple *volatilization indicators* which are determined from basic chemical properties and the temperature and wind speed at the application site. Using data from field studies for eight different turf pesticides, three different indicators were evaluated for their ability to predict vapor concentrations. Chemical vapor pressure was the simplest indicator considered, and it was 70 percent effective in predicting variations in vapor concentrations. The effectiveness increases to 90 percent when factors related to solubility, adsorption and wind speed are added to produce the G/V indicator.

We further tested the use of volatilization indicators by using them to classify the inhalation hazards associated with 37 different applications of the eight pesticides to grass. Health hazards were determined by comparing inhaled dose to the EPA's reference doses. Inhaled doses were computed using both measured vapor concentrations and concentrations determined from the indicators. As shown in Table 11, the volatilization indicators produced the same rankings of health hazards that were obtained from the measured concentrations. Although further testing is necessary, the research suggests that with a chemical properties table and a weather forecast, it may be relatively easy to identify whether or not application of a pesticide to turf on a particular day may be hazardous to golfers or lawn users.

Table 11. Inhalation Hazard Quotients (HQ) Determined from Measured and Calculated Concentrations.

Chemical	HQ from Measured Concentration	HQ from Concentration Regressions on Vapor Pressure	HQ from Concentration Regressions on G/V
Bendiocarb	0.01	0.03	0.02
Carbaryl	0.00	0.01	0.00
Chlorpyrifos	0.09	0.04	0.11
Diazinon	2.80	2.70	1.50
Ethoprop	70.20	61.90	64.40
Isazophos	5.20	13.10	11.20
Isofenphos	0.12	0.17	0.15
Trichlorfon	0.04	0.04	0.03

Pesticide Runoff Model for Turfgrass: Development, Testing and Application

Cornell University

Douglas A. Haith

Start Date: 1998
Number of Years: 3
Total Funding: $30,798

Objectives:

1. *Adapt a previously developed pesticide runoff model to turfgrass conditions and test the accuracy of model predictions by comparisons with data from field experiments.*
2. *Use the model to estimate pesticide runoff probabilities (return periods) for a range of chemicals and locations in the eastern United States.*

We are currently testing two pesticide runoff models, PRZM, and PESTRUN, to see if they are capable of predicting the quantities of water that may run off from turfgrass. Following testing of the water portion of these models, we will evaluate their abilities to predict the pesticide content of runoff water. Both models base runoff estimates on adaptations of the U.S. Soil Conservation Service Curve Number equation.

Comparisons of observed runoff data and results from calibrated runs of the two models are shown in Figure 12 for *classic* plots that were seeded to a mixture 50 percent Kentucky bluegrass, 20 percent fescue, and 30 percent ryegrass. The calibration process was subjective, i.e., curve numbers were adjusted until predicted runoff seemed to reasonably follow the observed values. The selected curve numbers (CN) are indicated on the figure legends. Best results were obtained for PESTRUN when different curve numbers were used for each year (39, 20, and 25 for 1986, 1987 and

1988, respectively. In most months, the models' results were a reasonable match to observations. A although the tests are not conclusive, it appears that these models are capable of describing turf runoff.

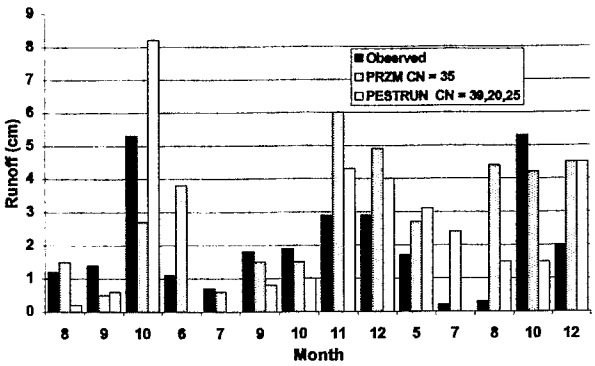


Figure 12. Comparison of calibrated model estimates with observed runoff - Pennsylvania State University Classic Seed Mixture Turf Plots 1986-88.

Characterization of Leaching at the Coeur d'Alene Golf Course Floating Green

Washington State University

William J. Johnston

Start Date: 1998
Number of Years: 3
Total Funding: $32,000

Objectives:

1. *Quantify water flow and movement of NO_3 and NH_4 through a large-scale sand-based putting green under actual golf course conditions.*
2. *Demonstrate the effect of nitrogen fertilization and application methods on sand-based putting greens to promote environmental safety and support the highest level of turfgrass quality.*
3. *Explore monitoring root growth and water movement utilizing computerized mesorhizotron technology newly developed at Washington State.*
4. *Monitor movement of nutrients (other than N) and pesticides.*
5. *Develop water movement models and calibration in sand-based greens.*

The objective of this project is to determine whether modern management practices provide adequate prevention of nitrogen leaching while maintaining the high level of playability required on modern golf facilities. The unique research site at the floating 14th green at the Coeur d'Alene Resort Golf Course allows for complete leachate collection from the green surface due to its self-contained design. This permits an accurate determination of concentration and quantity of leachate.

An automated flow meter with a sampler line is located in a storage tank beneath the green surface. As water is leached through the soil profile and collected by the drain lines located within the green, the flow meter monitors flow rate and total flow. Every 24 hours, a leachate sample is automatically taken from the drainage water prior to its entering the storage tank. The leachate sample is then stored within the refrigerated sampler until removal for analysis.

Currently a soluble 20-0-20 fertilizer is being applied at 0.1 lb N/1000 ft^2 every 7 to 10 days throughout the growing season. For research purposes, nitrogen rate was briefly increased to 0.3 and 0.6 lb N/1000 ft^2 to observe leaching at higher fertilizer rates. Future objectives include the use of a granular fertilizer to observe the effect of form on nitrate and ammonia leaching.

To date, leachate data indicate very low concentrations of ammonia and nitrate. Nitrate levels range from 0.11 to 1.79 ppm, well below the EPA limit of 10 ppm (Figure 13). Ammonia levels ranged from 0.18 to 0.47 ppm. Following closure of the green to play in November 1998, six micro-lysimeters were placed in the rootzone to allow for additional sampling.

To develop a nitrogen balance, soil and clipping samples are being taken to determine the concentration and form of nitrogen present. Clipping samples are taken from daily mowing, weighed, sub-sampled, then frozen for later analysis. Currently clipping samples range from 3.3 and 6.1 percent

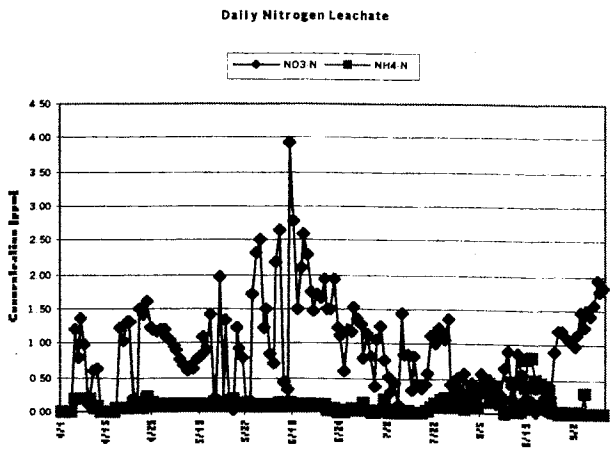


Figure 13. Nitrogen concentrations (NO_3 and NH_4) found in leachate recovered from the floating green located at Coeur d'Alene Resort.

nitrogen on an oven dry-weight basis.

Future research includes the formation of a nitrogen balance. In addition, construction of a water balance can be made leading to a better understanding of water use efficiency in the turfgrass environment. The findings of the project will provide a clearer understanding of how golf course management practices affect the environment. It will allow the public to directly view how management practices on sand-based greens affects groundwater and the environment. It will demonstrate to the public the high level of effort being placed on insuring that the environment is preserved through current golf course practices.

Gaseous Losses and Long-Term Fate of Nitrogen Applied to Kentucky Bluegrass Turf

University of Illinois

Bruce Branham

Start Date: 1998
Number of Years: 5
Total Funding: $124,270

Objectives:

1. *Detemine the quantity and form of gaseous nitrogen losses from turf.*
2. *Develop long-term (20+ years) field plots examining the fate of nitrogen applied to a mature turf.*

The objectives of this project is to determine how nitrogen is lost from fertilized turf stands, which is being conducted at the University of Illinois; and to determine the long-term fate of nitrogen at Michigan State University (MSU). The project at MSU includes the development of long-term turfgrass plots that will be treated the same every year for an indefinite period. We envision these plots as a kind of "Morrow" plots for turf. The Morrow plots are an experimental field at the University of Illinois that has continually produced corn since 1868. The plots at MSU will be continually under turf management for the indefinite future at the same, known level of management inputs. These plots will be an invaluable resource for future researchers long after this study is over. The project at MSU consists of establishing the long-term plots on a site that contains four large lysimeters that will be monitored for nitrogen leaching under two fertility regimes. The high maintenance plots receive 5 lbs nitrogen per 1000 square feet per year while the low maintenance plots receive 2 lbs. nitrogen per 1000 square feet per year. The lysimeters will be monitored continuously for nitrate leaching under these two fertility programs. Beginning in 2000, a study will be initiated to monitor the fate of labeled fertilizer nitrogen in these plots.

The results from monitoring the leachate of the two nitrogen regimes in 1998 indicate slight increases in nitrogen leaching compared to the baseline levels established in these same plots during a study conducted from 1991 through 1993. Average nitrogen losses ranged from 1.83 to 2.85 mg N L^{-1} for the 2 lbs. N/1000 ft^2/Yr treatment and 2.37 to 4.61 mg N L^{-1} for the 5 lbs. N/1000 ft^2/Yr treatment. While these values are well below the drinking water standard of 10 mg N L^{-1}, they are above the levels detected by Miltner et al. (1996) in a similar study. Values detected by Miltner et al. were generally below 1 mg N L^{-1} with a maximum detection of 3.8 mg N L^{-1}

Research at the University of Illinois is focused on gaseous losses of nitrogen from fertilized turf stands. Turfgrasses are fertilized yearly but lose insignificant amounts of nitrogen to leaching. Yet, turfgrass systems must be losing nitrogen or fertilization could be stopped without any loss in turfgrass growth or quality. Since nitrogen leaching appears negligible, the only other avenue for loss of nitrogen from the turf is through nitrogen volatilization. Nitrogen volatilization losses can occur by two different pathways. Ammonia volatilization and denitrification will both be studied in these experiments.

Most of our time in 1998 was devoted to developing the systems needed to study nitrogen volatilization under field conditions. In particular, denitrification is very difficult to study under field conditions. One of the most vexing problems encountered was a method to measure the volume of the lysimeters to be used in the field measurements. The lysimeters, plastic tubes inserted into the turf, must protrude above the turf surface so that a cover can be placed over the lysimeter and a sample of the air inside the lysimeter taken to determine gaseous nitrogen loss. Determining the head space volume inside the lysimeter is critical to getting an accurate estimation of the total nitrogen gasses volatilized. While many easy approaches were considered, none of them provided acceptable results. The method developed introduces a known volume of neon gas into the lysimeter prior to sampling, after equilibration, a sample is withdrawn and the concentration of neon gas is determined by mass spectroscopy. The dilution of the neon gas can be used to accurately determine the head space volume of the lysimeters.

Our field system has been tested and is ready for our first field studies which will begin in May of 1999. These trials should be sufficiently sensitive to determine nitrogen volatilization losses from fertilized turf stands.

An Assessment of the Risks Associated with Pesticides Volatilized and Dislodged from Golf Turf

University of Florida

George H. Snyder
John L. Cisar

Start Date: 1998
Number of Years: 3
Total Funding: $75,000

Objectives:

1. *Identify by survey golfer practices and habits which lead to exposure from dislodged pesticides.*
2. *Quantify the amount of various pesticides dislodged, volatilized, and transferred during play.*
3. *Characterize the risk incurred by pesticide exposure while playing golf.*

A series of pesticide dislodgeability studies were conducted to evaluate the risks associated with golfer exposure to pesticides. The work was performed by W. Raymond H. Snyder, as part of a Master of Science degree program, at the University of Florida. The pesticides 2,4-D and dicamba were applied as liquids to a *TIFGREEN* bermudagrass putting green with and without *Poa trivialis* overseeding, and to a *TIFDWARF* green. Isazofos, chlorpyrifos, and fenamiphos were spray-applied to the *TIFDWARF* green. Pesticides were dislodged with damp cheesecloth rubbed on the turf surface, damp cotton cloth or leather pressed on the surface, by putting a golf ball over the surface, by rolling golf grips on the surface, and in a short rough off to the side of the green by swinging a golf club through the grass and wiping the club surface with damp cheesecloth.

Generally, the amount of pesticide dislodged decreased with time after application, and was greatly reduced following irrigation. By combining the data, risk assessment calculations could be made for various scenarios. For example, exposure to chlorpyrifos on 18 greens one hour after application every day for a lifetime was calculated to provide a Hazard Quotient of 0.31. Hazard quotients (HQ) less than one indicate that the residues present are at concentrations below those that would cause effects in humans. A HQ greater than one does not necessarily infer the residue levels will cause adverse effects, but rather that the absence of adverse effects is less certain. A similar calculation for exposure after irrigation was 0.02. Chlorpyrifos has a rather high Reference Dose (i.e., acceptable amount of exposure) that reduces the HQ value. Calculations for the other pesticides, some of which have higher Reference Doses, will be reported next year.

A stabilized organic polymer (SOP) coated on sand for reducing pesticide leaching was field evaluated twice. Inclusion of the SOP-sand at the rate of 20 percent by volume in the lower 10 cm of the 30-cm USGA rootzone profile was very effective. Fenamiphos leaching was reduced up to 100 percent and fenamiphos metabolites up to 76 percent.

Recreational Exposure of Golfers to Pesticides Applied to Golf Courses

University of Massachusetts

John Marshall Clark

Start Date: 1998
Number of Years: 1
Total Funding: $25,000

Objectives:

1. *The role of vapor pressure and temperature will be evaluated in terms of developing a screening system for turfgrass pesticides.*
2. *Pesticides with possible safety concerns will be further evaluated in the context of best management practices, including the role of spray volume and adjuvants.*
3. *The role of thatch accumulation on the dissipation of volatile and dislodgeable residues will be assessed.*

Volatilization and dislodgeable foliar residues of turfgrass pesticides following application are major routes of exposure to golfers via inhalation and dermal penetration. Our past research has determined that pesticides with high vapor pressures and inherent high toxicities result in Inhalation Hazard Quotients (IHQs) greater than 1.0. From this determination, our research has progressed to examine methods of suppressing volatilization and dislodgeable residues in order to reduce potential golfer exposure. The adjuvant, Silwet L-77, was selected for its superior wetting, thatch penetrating, and volatilization suppressing properties (Policello et al., 1995). Three pesticides with relatively high vapor pressures were applied to small circular turfgrass plots with or without the adjuvant. Volatilization was measured with high volume air samplers and using the Theoretical Profile Shape (TPS) method. Dislodgeable residues samples were collected by wiping treated turf plots with dampened cheese cloth. The potential hazard associated with exposure to the volatile and dislodgeable residues was determined by IHQs and Dermal Hazard Quotient (DHQs) determination using the USEPA Hazard Quotient method (Murphy et al., 1996ab).

Wildlife Links Program

Golf courses offer excellent opportunities to provide important wildlife habitat in urban areas. With more than 15,000 golf course in the United States comprising in excess of 1.5 million acres, great potential exists for golf courses to become an important part of the conservation landscape.

Wildlife Links is a cooperative program with the National Fish and Wildlife Foundation that funds innovative research, management, and education projects that will help golf courses become an important part of the conservation landscape. The United States Golf Association is providing $200,000 annually to fund the grants through the Wildlife Links program. Since the program began in 1996, eight projects committing over $430,000, were funded to enhance wildlife conservation on golf courses. The objectives of Wildlife Links are to:

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

Wildlife Links Highlights

National Meetings

The National Audubon Society invited Peter Stangel, chairman of *Wildlife Links* and director, Neotropical Migratory Bird Conservation Initiative, to host a workshop on golf and the environment at their annual meeting in Estes Park, Colorado, July 6 through11. The annual meeting typically attracts 600 to 800 chapter members from around the country. The workshop included speakers from the USGA and NFWF, as well as Ron Pulaski, Superintendent at Seabrook Island, who provided details on the Cooperative Sanctuary Program. Jens Tripson, of Pelican Island Audubon, spoke on chapter participation in the Cooperative Sanctuary Program. This was an important event, even a bit of a breakthrough, and should go a long way toward making National Audubon a more involved partner in our conservation programs.

NFWF recently funded, through our *Restore Our Southern Rivers* Program, a stream restoration project on the Hutton Creek in Abingdon, Virginia. About 1,400 feet of this one mile stretch of creek runs through the Greenway Creek Golf Course. Agricultural impacts have resulted in severe degradation of this creek and the adjacent riparian habitat. When golf course construction began in March of 1998, the owners agreed to cooperate on a restoration project that will meet criteria for successful use of the golf course and provide for enhancement of the stream. A variety of bank stabilization practices will be used to create in-stream and riparian diversity, including bank sloping.

Wildlife Links in the News

- Special show on conservation on golf courses, highlighting Jim Howard's work with amphibian conservation on golf courses, Audubon's Cooperative Sanctuary Program, and the Wildlife Links Program. Maryland Public Television, February 1998.
- "Looking After Amphibians." Idaho State Journal, November 1997.
- "Teeing off from a frog's handicap." Dishneau, Daivd, The Patriot News, November 1997
- "Birdies, Woodpeckers, and Eagles." Bird Conservation Magazine, Fall 1997.

1999 Wildlife Links Request for Proposals and Funding. The Request for 1999 Proposals brought in over 40 preproposals. Funding decisions were made in late October 1998, with projects beginning in February of 1999. Approximately $75,000 was available from the USGA and the National Fish and Wildlife Foundation. The RFP appeared in the following publications:

- Entomology Society of America newsletter
- Ecological Society of America newsletter
- Society for Conservation Biology newsletter
- Ornithological newsletter
- The Wildlifer (The Wildlife Society newsletter)
- Copeia
- Froglog
- Herp Review
- Journal of Field Ornithology

Wildlife Links Advisory Committee. The *Wildlife Links* Advisory Committee is responsible for the selection and monitoring of grants funded under the Wildlife Links Program. Committee members are experts in fields of wildlife biology and management, pesticide management, habitat management and conservation, education, and the game of golf.

Peter Stangel, Director; *Katie Distler*, Coordinator, Neotropical Migratory Bird Conservation Initiative, National Fish and Wildlife Foundation, Washington, DC
Mike Lennartz, U.S. Forest Service, Forest Environmental Research, Washington, DC
Ron Dodson, President Audubon International, Inc. Selkirk, NY
Dan Petit, U.S. Fish and Wildlife Service, Office of Migratory Bird Management, Arlington, VA
Jim Snow, National Director; *Kimberly Erusha*, Director of Education, Green Section, United States Golf Association, Far Hills, NJ
Paul Engman, Fairfax County Park Authority, Fairfax, VA
Tom Franklin, The Wildlife Society, Bethesda, Maryland

Wildlife Links Technical Committee. The *Wildlife Links* Technical Committee gives input on technical issues that arise during the project selection process. They review proposals and comment on materials produced by the grantees when needed.

Dr. John Atwood, Manomet Observatory, Inc.
Melody Allen, The Xerces Society
Dr. John Fitzpatrick, Cornell Lab of Ornithology
Dr. Gregory Butcher, American Birding Association
Dr. Steve Sherrod, G.M. Sutton Avian Research Center
David Stubbs, European Golf Association, Ecology Unit

For additional information about Wildlife Links and other NFWF programs, please contact:

Katie Distler, Coordinator
Neotropical Migratory Bird Conservation Initiative
National Fish and Wildlife Foundation
1120 Connecticut Avenue NW, Suite 900
Washington, DC 20036
phone: (202) 857-0166
email: distler@nfwf.org

Golf Courses and Bird Conservation: A Management Manual

Colorado Bird Observatory

Scott Gillihan

Start Date: 1996
Number of Years: 3
Total Funding: $48,760

The Colorado Bird Observatory is creating a practical guide for golf course architects and superintendents to improve golf course habitat for bird species. *Bird Conservation on Golf Courses* will be available for purchase through USGA and Sleeping Bear Press in 1999. It is the first book that brings together the latest information on bird and bird habitat management as it applies to golf courses and similar settings.

The hands-on manual covers everything from general concepts, to specific techniques, and vital information on:

- Design and management for habitat conservation
- Management techniques for specific bird species
- Artificial nest structures
- Plants beneficial to bird populations
- Birds and golf course maintenance
- Dealing with *problem* birds

Wetlands Management Manual for Golf Courses

MACED

Don Harker
Gary Libby

Start Date: 1996
Number of Years: 3
Total Funding: $35,000

Objectives:

Develop an illustrated wetlands management manual for golf courses that uses a general narrative overview, drawings, case studies, key restoration techniques and indicator species to walk managers through a process of understanding wetlands.

This project will create an illustrated booklet of key wetlands restoration techniques and case studies for golf course superintendents. A new revision of *Wetlands Management Manual for Golf Courses* is expected during the summer of 1999. The booklet should be available for purchase through USGA in early 2000.

The approach is to use a general narrative overview, drawings, case studies, key restoration techniques and indicator species to walk managers through a process of understanding wetlands, leading to the ability to conserve, create/restore, and manage wetlands. The booklet will be as brief as possible and still cover the necessary material.

A reference method for the golf course manager to follow when working to conserve, restore, or construct a wetland was designed for the booklet. That method combines drawings with a plant species matrix. The drawing depicts (in aerial cross-section) different wetland conditions for the wetland types. A matrix contains the key species for that type and gives information about where in the wetland to plant a particular species, what restoration techniques to use, some wildlife value, flower color and size, and bloom period information. This easy reference approach is new and should prove to be a useful approach for land managers.

Data Management System for Information on Wildlife Habitat on Golf Courses

Audubon International

Ron Dodson

Start Date: 1996
Number of Years: 3
Total Funding: $77,500

Develop a computer-based system that accesses present and future wildlife and habitat information gathered from participants in the Audubon Cooperative Sanctuary Program.

Audubon International is computerizing its database of information gathered through its Cooperative Sanctuary System, a voluntary program for golf courses interested in creating and enhancing wildlife habitats and conserving and sustaining natural resources. The database will be open to golf course managers and others in the near future. Accessing data in this manner will help Audubon International staff to better direct members of the program in regard to conservation activities. In addition, it will establish a foundation from which wildlife research can be generated and give a clear picture of the resources presently under management by program members.

The creation of the *Managed Lands Database System* began in late August 1995. A review of all the *Resource Inventory Information* contained in hard copy at Audubon International headquarters was first completed. All member golf courses of the Audubon Cooperative Sanctuary Program System that completed a resource inventory form since 1991 were broken down into quantifiable information. This information was then transferred onto a standard form from which the data could easily be placed into a database. There are close to 950 bird species, 600 different species of reptiles and amphibians, and 100 species of mammals contained within the database.

A model was designed to help make entries into the database. This model included a very limited and general species list for birds, reptiles, amphibians, mammals, trees, and

even insects. It contained a breakdown of all habitat types such as desert, prairie, or woodland, and if the woodland was deciduous or coniferous. It also broke down the water features of the site, by number of ponds and pond acreage, number of lakes and lake acreage, and the amount of wetland area. The model also incorporated the address, state, and zip code and contact name fore each site.

A series of reports can now be generated based on the *Resource Inventory Information* logged into the *Managed Land Database Information.* For example reports dealing with geographic regions, address information, land and water acreage, and habitats were developed. This kind of information is very useful and beneficial to the Audubon Cooperative Sanctuary Program and its members.

Developing Methods to Enhance Amphibian Diversity on Golf Courses: Effects of Golf Course Construction on Amphibian Movements and Population Size

University of Rhode Island

Peter Paton

Start Date: 1998
Number of Years: 3
Total Funding: $72,000

Objectives:

1. *Determine the pre-construction population size for amphibians breeding at ponds within the boundaries of a proposed golf course.*
2. *Determine pre-construction travel corridors and movement patterns for amphibians at this same site.*
3. *Quantify population size and movement patterns following construction on the golf course.*

Amphibian movement chronology and community structure was monitored in three ponds in the middle of the proposed golf course construction site starting mid-February 1998. A total of 7,911 amphibian captures representing 11 species were recorded since project initiation. In addition, two species of snakes and seven species of mammals were detected.

Experimental evidence showed that frogs prefer to move through wooded habitats rather than turf areas ($G = 3.6$, $P = 0.058$) or barren areas ($G = 9.2$, $P = 0.002$). This preliminary finding suggests that dispersal corridors from ponds to upland wintering areas will be more effective if designed to include woodlands. However, other research showed that amphibians would readily cross turf.

Experiments with various grass heights (0.25, 0.5, 1.0, and 2.5 inches) found no evidence that grass height affected frog movement patterns ($G=3.7$, $P = 0.29$). This suggests that varying grass height is not a management option to increase frog use of a potential movement corridor.

Frogs readily crossed a 68 m (225 ft.) wide, mowed grass field, but there was little evidence of amphibian movement across a 175 m (575 ft.) wide grass field. This preliminary evidence suggests that the vast majority of fairways do not represent a dispersal barrier for most species of frogs in New England.

One of the most important scientific findings of this summer's research was that we documented non-random migration of metamorph frogs (e.g., newly transformed young) away from our monitored ponds. We established two 200-m long drift-fence arrays, 100 m to the east (habitat = woodlands) and to the west (habitat = woods and turf fields) of monitored ponds. Several species (Green Frog, Pickerel Frog, and Spotted Salamander) radiate out at random directions from breeding ponds. On the other hand, American Toads, Gray Tree Frogs, Spring Peeper, Wood Frogs, and Red-spotted Newts exhibited habitat preferences, most species were more likely to move through wooded habitats. This suggests that among some species of frogs, metamorphs have an innate genetic predisposition to migrate in specific directions. This has very important implications for management strategies.

Proposed research for 1999. Future funding for this research project during the 1999 field season will be used support three types of investigations: 1) we will continue monitoring natural movement patterns amphibians in the North Woods study site (this research will focus on adult movements to/from breeding sites, which was missed during the 1998 field season); 2) a series of experiments will be conducted in the North Woods area to further refine our knowledge of habitat characteristics of amphibian movement corridors, and 3) we propose to initiate a large-scale quantitative survey of the habitat characteristics of breeding sites used by amphibians on golf courses on southern New England, including habitat characteristics of potential movement corridors.

Pesticides and Nutrients in Surface Waters Associated with Golf Courses and Their Effects on on Benthic Macroinvertebrates

University of Maryland

William Lamp

Start Date: 1998
Number of Years: 2
Total Funding: $54,896

Objectives:

1. *Measure the concentration of pesticides and nutrients residing in the water column of streams associated with golf courses.*

2. *Measure the concentration of pesticides reding in the sediments and sediment porewater of streams associated with golf courses.*
3. *Assess the impact of golf courses on stream macroinvertebrate communities.*
4. *Determine the sublethal impacts of selected pesticides on benthic macroinvertebrates.*

Golf courses provide citizens with a convenient recreational opportunity while preserving green space and natural settings. Yet, their intensive management necessitates the use of pesticides and fertilizers, thus provoking concerns of environmental damage. One of the overall goals of this project is to determine if surface waters, and their sediments, associated with golf courses are contaminated by pesticides and/or fertilizers. Potential contamination can occur especially in association with high runoff events such as storms. However, because contamination varies with time, a second overall goal was to develop the use of stream macroinvertebrates and their communities as long-term indicators of water quality. This will allow us to determine if pesticides and/or fertilizers are impacting stream macroinvertebrate communities.

Water samples for nutrient level measurement have been collected and analyzed once or twice every month since March 1998. In addition, we have collected water from five run-off events and have analyzed this water for nutrients. Water and sediment samples for pesticide analysis have also been collected five times following run-off events. The water samples have been filtered and processed using solid phase extraction. The sediment samples are being stored using methods required to maintain the integrity of any pesticides. We are now in the process of analyzing the samples using gas chromatography and mass spectrometry. These samples are being analyzed using protocols developed at USDA.

Macroinvertebrates associated with natural leaf packs are collected using artificial leaf pack samplers. Five leaf packs, each consisting of dried leaves (standardized by leaf taxa and dry weight) and connected to a brick with a strap, are placed in the stream 21 days prior to the sampling date to allow for colonization by benthic macroinvertebrates. On the sampling date, the leaf packs are collected and water quality parameters measured. In the laboratory, invertebrates in each sample are sorted, preserved, and identified to family level. Community comparisons, using taxonomic diversity and invertebrate density, are being performed by calculating various community statistics for each golf course and site.

During 1997 and 1998, invertebrates were collected five times. During 1997, these samples yielded 24,555 individuals representing 46 families of invertebrates. The most abundant types of invertebrates collected were members of the families Chironomidae (midge flies), Simuliidae (black flies), Hydropsychidae (net-spinning caddisflies), Elmidae (riffle beetles), and Capniidae (winter stoneflies). No significant differences were seen in either taxonomic richness (P=0.59) or invertebrate density (P=0.65) when comparing upstream with downstream sites, illustrated in Figures 14 and 15. Of the physical and chemical parameters measured, only turbidity

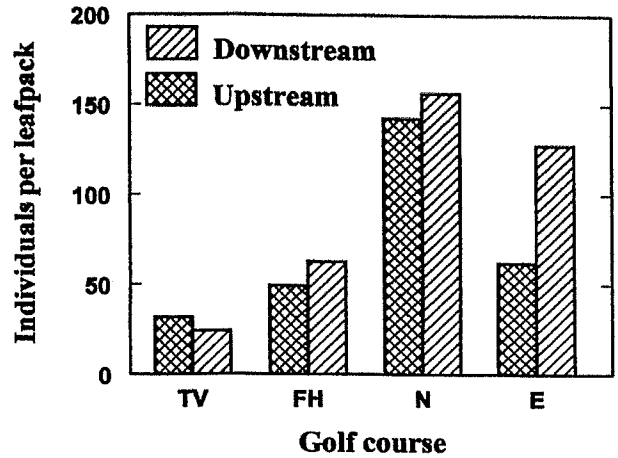


Figure 14. Average invertebrate diversity for all sample dates (1997) at upstream and downstream locations on four golf courses, TV, FH, N, and E.

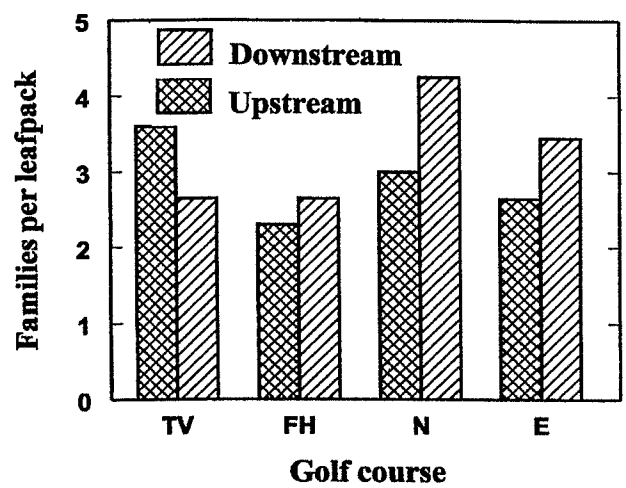


Figure 15. Average taxonomic diversity for all samples (1997) for upstream and downstream locations on four golf courses,TV, FH, N, and E.

showed a trend across all golf courses; water from sites upstream from the courses were more turbid than water collected downstream from the courses.

Based on this preliminary analysis, golf course management practices are not significantly impacting the invertebrate community. However, when one analyzes the trends seen in the invertebrate density and taxonomic diversity data, there is an increase in these two population indices at the downstream sites at three of the four courses, albeit, they are not significant increases. We will collect additional data and perform further analyses over the next year.

Area golf courses routinely use five fungicides: Daconil 2787, Bayleton, Aliette, Banol, and Subdue. Furthermore, the application of nitrogen and phosphorus is commonplace on area golf courses. Therefore, laboratory and field studies are being used to determine if pesticides and/or fertilizers influence consumption and decomposition of coarse particulate organic matter (CPOM).

Specifically, field studies are being conducted to measure the decomposition and consumption of organic matter in our streams associated with golf courses. Mesh bags containing predetermined amounts of leaf material are left in the field for various amounts of time during which the leaves are allowed to decompose or subjected to consumption by benthic macroinvertebrates. At the end of the study period, the bags are removed from the stream and the remaining leaf matter is weighed. Using this information, we can determine if golf courses are influencing the organic matter processing via alterations in decomposition of the leaf matter by periphyton or consumption of the matter by benthic macroinvertebrates.

Laboratory studies are being conducted to measure the decomposition of maple leaf discs in the presence of the five fungicides listed above. We will try to determine if the presence of these fungicides inhibits decomposition of organic matter by fungi and bacteria. In addition, laboratory studies will look at the effect of the presence of these fungicides on the consumption of maple leaf discs by stoneflies. It has been shown that consumers of organic matter are really using the periphyton growing on the organic matter as an energy source. Therefore, we are trying to determine if the presence of these pesticides has a sublethal affect on invertebrates through altering their consumption of organic matter, possibly due to altering periphyton growth on organic matter.

Golf Course Maintenance and Amphibian Conservation

Frostburg State University

Dr. James Howard

Start Date: 1997
Number of Years: 3
Total Funding: $105,036

Objectives:

Laboratory Studies:

1. *To test the relative toxicity of the most commonly used pesticides (insecticides, fungicides and herbicides) with three diverse taxa of amphibians.*
2. *To develop a more complete and biologically realistic testing protocol including: a) multiple species; b) short term acute and long term chronic tests; c) multiple life history stages; d) multiple indicators of biological impact; and e) an environment that provides the opportunity to detoxify or potentiate chemicals with more biological realism.*

Field Studies:

1. *To access the feasibility of "stocking" wetlands in order to establish breeding populations of desired amphibian species.*
2. *To evaluate the relative success of small temporary wetlands versus a larger permanent body of water stocked with the same amphibians.*

Laboratory study. The toxicity of three pesticides (carbaryl, chlorpyrifos, and imidacloprid) was investigated using American toad tadpoles (*Bufo americanus*). These trials were completed by August and the data analyzed by October. Effects on survival, growth, and time to metamorphosis analysis were consistent with previous results obtained using chorus frogs (*Pseudacris triseriata*). Concentrations of pesticide had a significant effect on survival. Prior to initiation of the *Bufo* trial, the LC_{50} (concentration of pesticide needed to kill 50% of test organisms) was determined for each pesticide. The estimated LC_{50}s in parts per billion were 468,000 for imidacloprid, 63,167 for carbaryl, and 1,316 for chlorpyrifos. All tadpoles placed in high (0.5 x LC_{50}) concentrations died during the trial whereas survival of tadpoles at all other concentrations was above 95 percent. Subsequent analyses were performed only on medium (0.1 x LC_{50}), low (0.01 x LC_{50}) concentrations and controls. Growth of tadpoles was significantly ($P < 0.05$) decreased by chronic exposure to 0.1 x LC_{50} concentrations of carbaryl and chlorpyrifos (Figure 16). Significant differences between control tadpoles and those raised in medium concentrations were observed in time to metamorphosis (measured as day front limbs emerge). Tadpoles in medium concentrations of all pesticides took an average of three days longer to reach metamorphosis when compared with controls. Sublethal effects on time to metamorphosis and growth would be expected to have negative effects on population persistence.

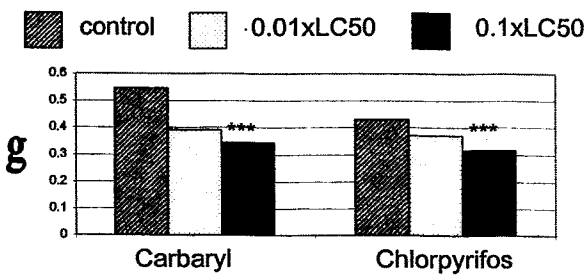


Figure 16. The effects of carbaryl and chlorpyrifos on average growth per tadpole (g) after three weeks. Treatments marked * are significantly different from controls.**

Water and sediment samples collected during the *Pseudacris* larval trials were analyzed for chlorpyrifos by The Institute of Wildlife and Environmental Toxicology. Results indicate that pesticide is rapidly absorbed by the sediment. For example, chlorpyrifos was added to a tank at a concentration of 112 ppb, and 24 hours later the concentration in water was 23 ppb. At termination of the experiment (after 4 repeated applications of pesticide), the concentration of chlorpyrifos in the sediment was 443 ppb. Because many frog tadpoles feed in the substrate and are detritivores, this pathway may be a more important contributor than pesticide residues in the water column.

In summary, Larval trials on *Bufo* were completed and data indicates that pesticide concentrations have effects on survival, growth, and time to metamorphosis of tadpoles which are significant and similar to previous trials on the genus *Pseudacris*. Sediment analysis indicates that pesticide added to the water column becomes concentrated in the sediment. Larval trials using *Rana* are in progress.

Field Studies. In March of 1998, egg masses of *Ambystoma jeffersonianum* (Jefferson salamander) and *Pseudacris triseriata* (chorus frog) were translocated into each of the six experimental ponds. Estimates indicate that 90.7 percent of *A. jeffersonianum* and 64.5 percent of *P. triseriata* larvae had successfully hatched. After larvae metamorphosed, individuals were captured by pitfall traps, funnel traps, or time constrained searches. Captured individuals were marked via toe clipping and/or freeze branding for identification in subsequent seasons. Experimental ponds, as well as ponds located on the Rocky Gap Golf Course, have been monitored for natural colonization by local amphibian species. Egg masses found have been identified to species, total number of eggs was estimated, and location of deposition within experimental ponds has been mapped. Larvae that have been dipnetted have also been identified to species.

In the spring and summer of 1998, the following species (in addition to the introduced species) had colonized experimental ponds: *Rana clamitans* (green frog), *Rana sylvatica* (wood frog), *Bufo americanus* (American toad), *Pseudacris crucifer* (spring peeper), *Hyla veriscolor* (gray treefrog), and *Notophthalmus viridescens* (red-spotted newt). Egg masses and/or larvae of the following species were found on ponds associated with the golf course at Rocky Gap: *Rana catesbeiana* (bullfrog), *R. clamitans*, *B. americanus*, and *P. crucifer*.

Although the species composition of experimental and golf course ponds seem similar, several important distinctions should be clarified. The absence of *R. catesbeiana* colonization in experimental ponds should aid in the colonization of smaller species of frogs because *R. catesbeiana* has been implicated in the local extirpation of smaller species due to predation. In addition, ponds associated with the golf course are not colonized as ubiquitously by smaller frog species as experimental ponds are. For instance, not only have we failed to find evidence of *H. versicolor* breeding in golf course ponds, but larvae of *P. crucifer* only were found in one golf course pond while all experimental ponds have contained them. The design of our experimental ponds may promote colonization success of some species. Nearly 60 percent of egg masses were deposited on narrow pond shelves designed to support vegetation that, in part, provides structure for the oviposition of amphibian egg masses. Similarly, the one golf course pond constructed with a shallow shelf on its perimeter is the only course pond that has shown evidence of *B. americanus* and *P. crucifer* colonization.

In summary, egg masses of two species of amphibians have been translocated into experimental ponds at Rocky Gap State Park. Hatching success has been monitored in the egg masses and metamorphosed individuals of both species have been captured and marked for future identification. Experimental ponds, as well as golf course ponds, have been monitored for natural colonization of amphibian species. We have detected six amphibian species that use experimental ponds for breeding and we have detected four species that use golf course ponds for breeding. More importantly, the species composition of our ponds suggests that golf course ponds lack the colonization of smaller species of frogs. However, they support the colonization of a large species (i.e., *R. catesbeiana*) that prey on (and could extirpate) smaller species.

Avian Community Response to a Golf Course Landscape Unit Gradient

Clemson University

David H. Gordon

Start Date: 1998
Number of Years: 3
Total Funding: $60,188

Objectives:

1. *Determine the composition and species richness of avian communities occupying a gradient of golf course landscape units located along the South Atlantic Coast during the breeding season.*
2. *Determine the composition and species richness of avian communities for habitat types found on golf course landscape units located along the South Atlantic Coast during breeding season.*
3. *Examine the influence of landscape context and characteristics of golf course landscape units on the composition and species richness of avian communities.*
4. *Produce a set of outreach products including a technical publication with management and design recommendations, a brochure, and color poster targeted at golf course stakeholder groups.*

David Gordon is assessing the value of golf course landscapes to avian communities. The results of the assessment will be used to produce a technical manual with management and design recommendations, as well as a brochure and color poster targeted at golf course stakeholder groups. The study will

be conducted on several golf courses along the *Grand Strand* area near Myrtle Beach, South Carolina.

Conservation of Native Pollinators on Golf Courses

Xerces Society
Melody Allen

Start Date: 1997
Number of Years: 3
Total Funding: $136,500

Objectives:

The aim of the project is to foster and increase insect pollinator populations including native bees, wasps, moths, flies, and butterflies to offset the effects of habitat fragmentation, and to augment the species composition of native plants in the out-of-play areas to produce continuous flowering throughout the growing season.

In the late summer of 1997, consulting scientists and Xerces Society staff visited several golf courses in eastern Washington and Oregon to rate their appropriateness for study. The interest of the golf course superintendent in participating in the plant and pollinator enrichment program also was determined. Soon after conducting the site visits and interviews, the project team selected four golf courses for inclusion: Wildhorse Resort in Mission, OR; Veterans Memorial Golf Course in Walla Walla, WA; Horn Rapids Golf Course in Richland, WA; and Coeur d'Alene Resort in Coeur d'Alene, ID.

Insect Surveys. The research scientists then initiated surveys on Wildhorse, Veterans Memorial, and Horn Rapids to obtain a background estimate of the number of individuals and species of flower-visiting *Hymenoptera* (bees and wasps) present in late summer and fall. The survey was implemented by a Master of Science student at Washington State University. Coeur d'Alene Resort was declared a reference site because of its abundance of insects and native plant communities.

In late spring of 1998 through fall of 1998, the same estimation process was implemented under Dr. Heidi Dobson at Whitman College in Walla Walla, Washington. In addition to collecting specimens, nesting block stations using blocks of wood drilled with varying sized holes to attract a diversity of hole nesting bee and wasp species were set up. The blocks were attached to 3-foot posts and placed in the ground at each golf course and in a reference site approximately one mile away. The reference sites were chosen for quality of native vegetation.

Preliminary analysis of these nesting blocks is in Table 12. Entomologists at the USDA Bee Biology and Systematics Laboratory in Logan UT have opened the blocks, extracted, and dissected each nest into its component cells. After recording this data, the cells were placed in gel capsules and stored at 3 to 5 °C for the winter. Next spring they will be returned to the golf course where the bees and wasps will be allowed to emerge normally.

Collection Technique. The baseline data at Wild Horse Horn Rapids and Veterans Memorial has been obtained using a passive trapping technique with colored plastic cereal bowls. The bowls were filled with a dilute solution of detergent and water that attracts bees, wasps and flies. The insects land on the surface of the solution and drown. The trap bowls were laid out once per week in transects in the out-of-play areas of the three courses. The survey design called for alternation of the 45 three colored bowls (15 each, white, yellow, and blue), at 10 to 15 feet apart.

The surveying activities are ongoing; however, the project scientists have identified the specimens from 1997. Thus far, 79 species of bees (all native except for the introduced honeybee) and 51 species of wasps from weekly collections in August, September, and October 1997 were trapped at the three golf courses. Specimens from the 1998 season are being prepared for identification at the USDA laboratory. Data will be available next year. The bees include a wide range of species from genera whose members tend to be somewhat specialized in their flower-foraging habits (*Anthidium, Dianthidium, Megachile, Andrena, Nomadopsis*), to those that are quite generalized (all genera in the bee family *Halictidae*).

The wasps also represent a surprising diversity for such a brief sampling period. All are predaceous on arthropods, many of which are considered pests. For example, all *Eumenidae* capture caterpillars, many *Larridae* (*Liris, Larropsis, Tachytes,* and *Tachysphex*) prey on *Orthoperans* (grasshoppers and crickets). *Oxybelus* and *Bembix* are valuable because they hunt and kill flies, and *Podalonia* is a cutworm predator. Although it is early in the analysis, a pattern of abundance seems to be present among the three courses. For a number of *Hymenoptera* individuals captured per bowl trap, Wildhorse had significantly more than the other two golf courses. Walla Walla had significantly more than Horn Rapids (Wildhorse averaged from 8.9 to 19.1 insects per bowl; Walla Walla averaged from 1.4 to 6.8 insects per bowl; and Horn Rapids averaged only 1.0 to 3.7 insects per bowl). This result was somewhat surprising in that Horn Rapids, on cursory inspection, appeared to have the highest representation of native plant species. At the same time, the total vegetation cover at Horn Rapids appeared rather low.

Project scientists have also analyzed the 1997 data for the influence of bowl color but have found no consistent differences among colors.

Native Plant Surveys. During the 1997 field season, the project team contracted with a local botanist and soil scientist to conduct plant and soil analyses of Wildhorse and Veterans Memorial Golf Courses. The goal was to develop a list of native plants occurring historically in the two geographic areas. From these lists, the researchers created a list of target plants for augmenting the existing plantings in the out-of-play areas of the two golf courses. The plant lists were annotated to provide fuller information on each plant species to make it easier for course superintendents to choose and purchase plants. A botanist employed by the Horn Rapids Golf Course produced a plant list

Table 12. Nesting blocks were set out on each golf course (onsite) and in three additional reference sites (off-site). Each nesting block was drilled with rows of varying sized holes (50 total) to attract different genera and species of bees.8

Site Description	No. of Blocks	Hole Size	No. of Nests	Cells per Nest	Bees	(Bee Type)	Wasps
Wildhorse Onsite	17	4	2	0			
		5	57	7.7			
		5	80	8.3			
		7	25	5.4			
Totals			164		142	(leafcutter)	24
Horn Rapids Off-site	20		7	9	5	(leafcutter)	2

Note: More nesting block data will be tabulated by next year. The nesting blocks were still being actively used at the end of the 1998 reporting period and were not shipped to the USDA laboratory in Logan, UT for analysis.

(also included in this report) for that area, but did not do a soil analysis or an historic comparison.

Future Objectives. Having established baseline data for 1997, the next step is to identify the insects collected during the full field season of 1998. That information will be furnished by the USDA Bee Biology and Systematics Laboratory in Logan, UT during 1999. A major 1998 through 1999-project goal is to increase the numbers of native plants used in the out-of-play landscaping of the three golf courses. The annotated plant lists are being used by superintendents to purchase landscaping materials. The Xerces Society is also supplying plants for this initial enrichment. The pre-enrichment data on native bees, wasps, plant, and nest-sites will then be used as a baseline to compare insect data gathered in subsequent, post-enrichment years. The plant enrichment program is beginning in late fall 1998 and will continue throughout the project. The project scientists also plan to expand the use of nesting sites on the golf courses to bolster the local populations of native bees and wasps. Sand nests, nesting blocks, trap nests, and log nests will be added in appropriate locations. The nests will be analyzed for species and number of cells.

During 1999, the Xerces Society will produce a *Pocket Guide to Insect Pollinators*. This guide is for the lay audience and will include color illustrations and basic life history information for the common groups of North American pollinating bees, wasps and flies. The guide will allow a lay person to identify the groups of bees, wasps, and flies that are the ubiquitous pollinators. It also will provide sufficient information on the appearance, habits, and life histories of most native bees and wasps. As material for the publication is produced, Xerces will produce educational sheets for the golf courses. The aim of this project is to educate people about the major groups of pollinating insects, to encourage them to use pollinator-attracting plants in their own backyards, and to notice and appreciate beneficial insects while spending time outdoors. As people gain knowledge about the diversity of insects responsible for pollination and the habitat requirements for sustaining pollinators, native plant restoration efforts will gain momentum and insect pollinators will thrive. The pocket guide will be produced under rigorous scientific standards, emphasize the beauty and fascinating biology of pollinators, and encourage the reader to appreciate these beneficial insects.

The Audubon Cooperative Sanctuary System Program for Golf Courses

Audubon International

Ron Dodson

With support from the United States Golf Association, the Audubon Cooperative Sanctuary Program (ACSP) for Golf Courses was created in 1991. The ACSP promotes ecologically responsible land management and natural resource conservation. Participation in the program assists golf land managers to plan, organize, implement, and document comprehensive environmental management programs on golf courses, while preserving the natural heritage of the game of golf. The goals of the program are to:

1. Enhance wildlife habitats on existing golf courses by working with the golf course superintendent and providing advice for ecologically sound course management;

2. Encourage active participation in conservation programs by golfers, golf course superintendents, golf officials, and the general public;

3. Recognize the people who are actively participating in environmentally responsible projects; and

4. Educate the public and golfing community about the benefits of golf courses and the role they play relative to the environment and wildlife.

ACSS accomplishments during the past year can be broken down into three categories: A) Seminars And Presentations, B) ACSS Golf Member Awards And Recognition, and C) Program Accomplishments.

A. Seminars & Presentations
- Golf & the Arizona Environment Conference, Phoenix AZ
- Wildlife Management & Habitat Conservation (Developing an Environmental Plan), Rutgers University
- Creating an Environmental Plan, Mid-Atlantic Association of Golf Course Superintendents
- Golf & the Environment Conference, Orlando, FL
- South Dakota Superintendents Association Annual Conference, Sioux Falls, SD
- Wildlife Management & Habitat Conservation Seminars
  Northeast Turfgrass Association Conference, Syracuse, NY
  GCSAA Conference & Show, Orlando, FL
  South Dakota Superintendents Association, Sioux Falls, SD
- USGA Green Section Regional Conference, Creating an Environmental Plan, Orlando & Palm Beach Gardens, FL

B. ACSS Golf Member Awards & Recognition
- GCSAA Environmental Steward Awards: 33 out of 37 winners were ACSP members
- 3M Tartan Park, MN: 3M's Environmental Leadership Award
- Panama Country Club, FL: National Society of the Daughters of the American Revolution award for Distinguished Conservation Record
- Superior National at Lutsen, MN: Department of Natural Resources grant recipient to enhance wildlife habitat

C. Program Accomplishments
- Golf & the Environment Status Report & Recommendations
- Project Flight Plan: 208 golf courses participated pledging to complete 1487 projects to enhance and protect habitats for migratory birds.
- ACSS Case Studies: Completed case studies of nine certified golf courses
- ACSS Case Study Master List: Master list of exemplary golf course conservation projects including contact people.
- ACSS Case Study Project Survey: Survey of 160 certified golf members to elicit more detailed case studies of conservation projects.
- Review and revise goals, objectives, and certification process to increase self-evaluation, increase project options, and increase educational opportunities in environmental planning.
- Review, revise, and update environmental fact sheets.

There are currently 2,212 golf course members (including 200 Canadian members) of which 174 are certified Audubon Cooperative Sanctuary golf courses. These include 14 Canadian, 1 Southeast Asian, and 2 European golf courses.