

**FOURTH
ANNUAL PROGRESS REPORT**

**concerning
DEVELOPING BROWN PATCH AND PYTHIUM DISEASE RESISTANCE
IN BENTGRASS AND ZOYSIAGRASS**

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USGA TURFGRASS PATHOLOGY RESEARCH

FOURTH ANNUAL PROGRESS REPORT 1990

I N D E X

	Page
Executive Summary	i - ii
I. Introduction	1
II. Project Personnel	1
III. <u>Pythium</u> diseases Ongoing Research	
1. Isolate Collection and Storage	2
2. Field Inoculation Technique	2
3. Field Evaluation <u>Pythium</u> blight	3
4. Greenhouse assessment of field evaluation	13
IV. <u>Rhizoctonia</u> Diseases Ongoing Research	
1. Isolate Collection and Storage	18
2. Field Inoculation Technique	18
3. Pathogenicity of <u>Rhizoctonia</u> isolates	19
4. Screening for Multiple Disease Resistance	20
V. Cooperative Research with USGA <u>Rhizoctonia</u> clones	27
VI. Other Diseases Culture Collections Underway	28
VII. Future Investigations	28

Executive Summary

Fourth Annual Progress Report 1 November, 1990

Developing Rhizoctonia Brown Patch and Pythium Disease Resistance in Bentgrass and Zoysiagrass

Principal Investigator: Dr. Phillip F. Colbaugh
Research Period of this report: 1 May 1990 to 1 Nov. 1990

Techniques for handling and inoculating large numbers of bentgrass field samples are being used to identify resistance to Pythium and Rhizoctonia blight diseases. Large culture collections of both pathogens have assisted in the use of highly virulent strains of the fungi for disease screening. Previous inoculation studies with bentgrass genotypes obtained from the GPIN (germplasm introduction nursery), HTS (heat treatment selection nursery), NHTS (not heat treated selection nursery) and the elite bentgrass nursery have examined Pythium foliar blight resistance among members of the collections. Repeated inoculation studies with the standard variety Penncross demonstrated improved Pythium resistance among several members of the experimental bentgrass germplasm collections. Of 1203 germplasm lines screened during 1989, 6% demonstrated resistance to Pythium foliar blight.

Unusually wet weather during the spring allowed research investigations to focus on field evaluations of Pythium blight symptoms in field nurseries. Disease symptoms were observed in June on two field nurseries following lengthy environmental periods favoring maximum disease pressure. Typical visual symptoms of foliar thinning and decline were observed on 15.4% of the NHTS plots and 29.4% on the HTS plots. Disease symptoms appeared to be greater on germplasm lines from Prominent than on other parental lines observed in both nurseries. Greenhouse

studies were used to correlate disease symptoms from the HTS nursery field plots with a seedling bioassay for Pythium blight in McDonald's sundae cups. Blighting of seedling stands in cups produced by soil cores from field plots was greater in cups than on the field plots. Of test experiments showing field resistance (less than 25% disease) 41% were substantiated by seedling blight data. These results indicate the importance of obtaining field plot visual data to identify sources of resistance to Pythium blight. Rhizoctonia spp. inoculation studies on germplasm lines from HTS and NHTS field nursery plots showing field resistance to Pythium blight did not show a high level of resistance to both diseases. Of 105 Pythium resistant germplasm lines examined 11 demonstrated potential resistance to both Pythium and Rhizoctonia blight diseases.

The USGA Turfgrass Rhizoctonia culture collection has also made valuable contributions in the area of developing refined techniques for tissue culture selection of new germplasm lines. Cooperative research is now underway with Dr. Jeff Krans to screen bentgrasses originating from tissue culture lines against Rhizoctonia blight using mass inoculating screening methods. Isolates of fungi causing the Helminthosporium diseases and Sclerotinia dollar spot are being collected and samples will be gratefully received from any sources available during the year. The isolates will be ~~will~~ maintained in long-term storage as with other USGA isolate collections and will be used for a variety of disease screening and pathogen identification studies.

3. Introduction

On 17 February, 1987, the Texas Agricultural Experiment Station and the Texas A&M University Research Foundation accepted research funds as per contract agreement (FPN 5654000) with the United States Golf Association to conduct investigations to develop Rhizoctonia brown patch and Pythium disease resistance in bentgrass and zoysiagrass. This is a cooperative project with the turfgrass breeding and development efforts for both grasses, under the direction of Dr. Milton C. Engelke also at the Texas A&M University Research and Extension Center at Dallas, Texas. This fourth annual research report is for the period 1 May 1990 to 1 November 1990, and represents the past six months of activity on the cooperative disease assessment project.

II. Project Personnel

Mr. Shawn Harvanek served as my assistant on this project for the past summer on field, greenhouse and laboratory research returned to college as a full time student at Tarleton State University. Shawn has contributed measurably to the success of the 1990 USGA pathology research effort. Additional funds have been accepted from the National Turfgrass Evaluation Program to conduct disease screening research on the NTEP Bentgrass Collection. It is anticipated that the additional funds can be used to assign a permanent position to this project and which should overcome some of the recurring problems with employee training and turnover. The position will redefined and filled

during the winter in order to accommodate a busy field research program to be undertaken in the Spring.

III. Pythium diseases Ongoing Research

1. Isolate Collection and Storage:

The USGA isolate collection of Pythium spp. has again been assembled for long-term preservation through encapsulation of the cultures and storage at a temperature known to insure their survival. Storage of the cultures directly in distilled water has largely overcome previous contamination problems and allows for longer preservations of the cultures. Each culture of the collection is routinely plated on water agar in a petri dish and transfers are made on water agar then to vials containing distilled water. The collection can be maintained for long periods at a temperature of 26 C. The unique collection of turfgrass Pythium spp. strains now requires only yearly reexamination for culture viability and contamination. New isolates are being added to the Collection as they become available through cooperation with golf course superintendents. Recent project activities have focused on revitalizing the Pythium culture collection and sending isolates to investigators who requested specific isolates within the Collection. Isolates were sent to Patricia Sanders (Pennsylvania State University) during the summer for use in identifying strains of Pythium aphanidermatum.

2. Developing Field Inoculation Techniques

Previous attempts to inoculate bentgrasses in field plots

with Pythium spp. have generally not been successful, even though some elaborate measures were used to insure their success. Laboratory studies demonstrated that Pythium spp. can survive and infect plants following applications of foliar sprays containing inoculum in water suspended in a light weight oil such as safflower oil. Following inoculations the oil phase separates to form a fine film over applied water droplets which insures moisture retention for longer periods than previously possible. Field research during the summer explored this method of inoculation with Pythium sp (P#16) on var. Penncross growing in field plots. The method was not successful. Failure of the field inoculation technique was attributed to high temperatures developing on the leaf canopy following inoculation. Future research with high temperature Pythium strains may be helpful in overcoming the high temperature effects.

3. Field Plot Evaluations of Pythium blight:

Pythium blight damage to bentgrasses in the South is thought to be both a foliar blight and a root decline condition. Root disease symptoms are usually manifested during the hot summer months. Inoculation studies we have conducted have focused on foliar resistance to Pythium blight and the relationships of these observations to root rot susceptibility of experimental lines is not known. On 4 June, visual observations of Pythium blight damage were made on HTS and NHTS nursery field plots. Each of the seven varieties have 18

different genotype selections with each clone replicated three times. Due to the vast amount of rainfall occurring in the spring, there was an abundance of Pythium blight symptoms for our observations. Disease symptoms included the death and thinning of stands of grass (Fig. 1). Data records were estimates of the percentage disease symptoms occurring in each of the experimental lines of bentgrass. The data was recorded as zero percent being no disease symptoms and 100 percent being completely dead stands of grass (Tables 1 & 2).

Visual evidence of Pythium disease appeared to be most severe in the HTS Plots (Tables 1b, 2b). Experimental bentgrass lines showing greater than 50% disease symptoms (highly susceptible) in HTS plots were 18.8% of the total while similar damage in the NHTS plots was 4% of the total number observed. On the basis of visual symptoms observed, experimental bentgrass lines in the NHTS plots (1) had far greater numbers of experimental lines with no disease, (2) had a lower average mean % disease for the seven varieties and (3) more experimental lines with 25% or less disease symptoms (Table 2b). The variety Prominent had the highest mean % disease rating of the seven bentgrass varieties in both the NHTS and HTS nurseries. The mean % disease among parental variety lines in the HTS nursery were all nearly equal. In contrast, the mean % disease of parental variety lines in the NHTS nursery were different. The greatest resistance was found in seedlings derived from var. Penneagle, ISI6999 and Penncross (Table 2b).

A.



B.



Fig. 1. (A) Field view of HTS bentgrass nursery germplasm lines in June showing Pythium blight symptoms. (B) Close-up photo of disease symptoms.

Table 1. Mean Percentage Pythium blight disease ratings on HTS bentgrass nursery field plots. 4 Jun, 1990

Parent Variety	Seedling Selection	Field Plot Location	Replicate % Disease			Mean % Disease
			1	2	3	
EMERALD	1	01-21, 14-11, 18-05	30	30	90	50.00
EMERALD	2	02-21, 07-11, 10-05	10	10	25	15.00
EMERALD	3	03-21, 08-11, 16-05	0	0	0	0.00
EMERALD	4	04-21, 16-11, 17-05	0	10	20	10.00
EMERALD	5	05-21, 13-11, 02-05	5	50	0	18.33
EMERALD	6	06-21, 10-11, 14-05	40	15	5	20.00
EMERALD	7	07-21, 01-11, 05-05	90	15	45	50.00
EMERALD	8	08-21, 09-11, 04-05	90	5	75	56.67
EMERALD	9	09-21, 15-11, 09-05	30	10	30	23.33
EMERALD	10	10-21, 05-11, 07-05	80	25	25	43.33
EMERALD	11	11-21, 18-11, 13-05	20	5	80	35.00
EMERALD	12	12-21, 12-11, 01-05	25	25	60	36.67
EMERALD	13	13-21, 11-11, 06-05	100	100	80	93.33
EMERALD	14	14-21, 03-11, 03-05	0	0	0	0.00
EMERALD	15	15-21, 17-11, 08-05	5	0	5	3.33
EMERALD	16	16-21, 04-11, 15-05	50	100	80	76.67
EMERALD	17	17-21, 06-11, 11-05	25	75	80	60.00
EMERALD	18	18-21, 02-11, 12-05	0	0	10	3.33
PENNEAGLE	1	01-20, 17-13, 08-07	10	0	10	6.67
PENNEAGLE	2	02-20, 13-13, 15-07	10	0	15	12.50
PENNEAGLE	3	03-20, 18-13, 05-07	0	5	90	31.67
PENNEAGLE	4	04-20, 14-13, 04-07	0	0	30	10.00
PENNEAGLE	5	05-20, 12-13, 17-07	12	40	90	47.33
PENNEAGLE	6	06-20, 03-13, 16-07	0	5	0	1.67
PENNEAGLE	7	07-20, 08-13, 10-07	50	10	15	25.00
PENNEAGLE	8	08-20, 16-13, 06-07	80	5	75	53.33
PENNEAGLE	9	09-20, 06-13, 13-07	10	15	60	28.33
PENNEAGLE	10	10-20, 11-13, 11-07	15	85	75	58.33
PENNEAGLE	11	11-20, 10-13, 14-07	25	40	75	46.67
PENNEAGLE	12	12-20, 02-13, 09-07	50	15	0	21.67
PENNEAGLE	13	13-20, 09-13, 02-07	10	85	-	47.50
PENNEAGLE	14	14-20, 04-13, 18-07	60	25	60	48.33
PENNEAGLE	15	15-20, 15-13, 12-07	10	0	30	13.33
PENNEAGLE	16	16-20, 07-13, 07-07	45	20	15	26.67
PENNEAGLE	17	17-20, 05-13, 01-07	10	0	0	10.00
PENNEAGLE	18	18-20, 01-13, 03-07	0	5	0	2.50
ISI	1	01-19, 18-09, 02-04	0	0	0	0.00
ISI	2	02-19, 12-09, 15-04	0	0	0	0.00
ISI	3	03-19, 17-09, 17-04	0	5	10	5.00
ISI	4	04-19, 04-09, 11-04	3	45	80	42.67
ISI	5	05-19, 01-09, 13-04	5	85	50	46.67
ISI	6	06-19, 03-09, 16-04	40	75	5	40.00
ISI	7	07-19, 07-09, 14-04	5	15	0	6.67
ISI	8	08-19, 10-09, 09-04	70	20	0	45.00
ISI	9	09-19, 15-09, 03-04	5	10	90	35.00
ISI	10	10-19, 02-09, 10-04	10	15	30	18.33
ISI	11	11-19, 14-09, 18-04	15	15	15	15.00

Table 1. Continued:

Parent Variety	Seedling Selection	Field Plot Location			Replicate % Disease			Mean % Disease
					1	2	3	
ISI	12	12-19,	09-09,	05-04	85	90	45	73.33
ISI	13	13-19,	08-09,	12-04	10	10	15	11.67
ISI	14	14-19,	13-09,	07-04	15	20	0	11.67
ISI	15	15-19,	06-09,	01-04	15	45	60	40.00
ISI	16	16-19,	11-09,	08-04	10	50	10	23.33
ISI	17	17-19,	05-09,	04-04	0	75	85	53.33
ISI	18	18-19,	16-09,	06-04	30	5	90	41.67
PSU126	1	01-18,	06-14,	04-02	0	30	80	36.67
PSU126	2	02-18,	17-14,	08-02	0	0	40	13.33
PSU126	3	03-18,	07-14,	15-02	0	0	40	20.00
PSU126	4	04-18,	11-14,	05-02	0	75	85	53.33
PSU126	5	05-18,	15-14,	09-02	20	10	85	38.33
PSU126	6	06-18,	01-14,	18-02	15	5	90	36.67
PSU126	7	07-18,	08-14,	02-02	0	0	0	0.00
PSU126	8	08-18,	14-14,	12-02	10	5	85	33.33
PSU126	9	09-18,	05-14,	13-02	10	25	90	41.67
PSU126	10	10-18,	02-14,	17-02	15	30	40	28.33
PSU126	11	11-18,	09-14,	14-02	50	85	15	50.00
PSU126	12	12-18,	03-14,	07-02	0	15	20	11.67
PSU126	13	13-18,	18-14,	11-02	15	0	20	11.67
PSU126	14	14-18,	10-14,	16-02	0	20	5	8.33
PSU126	15	15-18,	04-14,	03-02	15	25	15	18.33
PSU126	16	16-18,	13-14,	01-02	25	25	95	48.33
PSU126	17	17-18,	16-14,	06-02	15	5	60	26.67
PSU126	18	18-18,	12-14,	10-02	5	10	60	25.00
DUCHESS	1	01-17,	08-08,	06-03	10	10	80	33.33
DUCHESS	2	02-17,	15-08,	16-03	0	15	0	5.00
DUCHESS	3	03-17,	18-08,	18-03	0	30	90	40.00
DUCHESS	4	04-17,	01-08,	14-03	25	40	7	24.00
DUCHESS	5	05-17,	12-08,	17-03	0	0	0	0.00
DUCHESS	6	06-17,	14-08,	08-03	0	90	50	46.67
DUCHESS	7	07-17,	13-08,	01-03	0	95	95	63.33
DUCHESS	8	08-17,	11-08,	12-03	100	50	10	53.33
DUCHESS	9	09-17,	09-08,	03-03	15	25	25	21.67
DUCHESS	10	10-17,	16-08,	02-03	10	0	90	33.33
DUCHESS	11	11-17,	17-08,	13-03	0	20	5	8.33
DUCHESS	12	12-17,	03-08,	15-03	0	10	90	33.33
DUCHESS	13	13-17,	02-08,	09-03	0	60	50	36.67
DUCHESS	14	14-17,	05-08,	05-03	25	-	0	12.50
DUCHESS	15	15-17,	07-08,	07-03	50	25	0	25.00
DUCHESS	16	16-17,	04-08,	11-03	0	0	20	6.67
DUCHESS	17	17-17,	10-08,	10-03	0	20	100	40.00
DUCHESS	18	18-17,	06-08,	04-03	-	-	15	15.00
PROMINENT	1	01-16,	13-12,	02-01	0	85	40	41.67
PROMINENT	2	02-16,	12-12,	07-01	0	90	20	36.67
PROMINENT	3	03-16,	17-12,	18-01	0	75	35	36.67
PROMINENT	4	04-16,	05-12,	06-01	2	10	10	7.33

Table 1. Continued:

Parent Variety	Seedling Selection	Field Plot Location	Replicate % Disease			Mean % Disease
			1	2	3	
PROMINENT	5	05-16, 01-12, 08-01	0	70	50	40.00
PROMINENT	6	06-16, 04-12, 11-01	0	0	40	13.33
PROMINENT	8	07-16, 11-12, 05-01	15	85	60	53.33
PROMINENT	9	08-16, 10-12, 10-01	0	15	50	21.67
PROMINENT	10	09-16, 07-12, 16-01	20	35	5	20.00
PROMINENT	11	10-16, 16-12, 13-01	20	30	75	41.67
PROMINENT	12	11-16, 06-12, 01-01	80	50	80	70.00
PROMINENT	13	12-16, 15-12, 09-01	5	0	10	5.00
PROMINENT	14	13-16, 09-12, 17-01	10	50	25	28.33
PROMINENT	15	14-16, 18-12, 04-01	20	65	45	43.33
PROMINENT	16	15-16, 03-12, 14-01	30	40	85	51.67
PROMINENT	17	16-16, 08-12, 15-01	5	80	5	30.00
PROMINENT	18	17-16, 14-12, 12-01	20	60	75	51.67
PROMINENT	19	18-16, 02-12, 03-01	0	90	0	45.00
PENNCROSS	1	01-15, 15-10, 17-06	5	20	50	25.00
PENNCROSS	3	02-15, 04-10, 05-06	0	25	30	18.33
PENNCROSS	4	03-15, 12-10, 01-06	25	25	90	46.67
PENNCROSS	5	04-15, 14-10, 10-06	15	0	25	13.33
PENNCROSS	6	05-15, 17-10, 16-06	40	5	25	23.33
PENNCROSS	7	06-15, 09-10, 15-06	5	25	15	15.00
PENNCROSS	8	07-15, 06-10, 08-06	20	75	50	48.33
PENNCROSS	9	08-15, 01-10, 13-06	30	75	80	61.67
PENNCROSS	10	09-15, 16-10, 18-06	5	100	50	51.67
PENNCROSS	11	10-15, 02-10, 09-06	20	95	50	55.00
PENNCROSS	12	11-15, 07-10, 02-06	25	90	50	55.00
PENNCROSS	13	12-15, 10-10, 04-06	50	10	30	30.00
PENNCROSS	14	13-15, 13-10, 11-06	40	40	85	55.00
PENNCROSS	15	14-15, 18-10, 14-06	10	10	70	30.00
PENNCROSS	16	15-15, 05-10, 12-06	15	0	50	21.67
PENNCROSS	17	16-15, 11-10, 03-06	0	40	45	28.33
PENNCROSS	18	17-15, 08-10, 06-06	10	25	50	28.33
PENNCROSS	19	18-15, 03-10, 07-06	10	95	35	46.67

Table 1a. Summary by variety of % disease means less than 25% (disease tolerant) and more than 50% (disease susceptible).

Parent Variety	% Disease Mean	Number with 0%	Less than 25%	More than 50%
EMERALD	33.056	2	9	5
PENNEAGLE	27.306	0	10	2
ISI	28.296	2	10	3
PSU126	27.870	1	8	2
DUCHESS	27.676	1	8	2
PROMINENT	35.407	0	5	4
PENNCROSS	36.296	0	6	5

Table 2. Mean Percentage Pythium blight disease ratings on NHTS bentgrass nursery field plots. 4 Jun, 1990

Parent Variety	Seedling Selection	Field Plot Location			Replicate % Disease			Mean % Disease
					1	2	3	
PENNEAGLE	1	-	07-09,	01-06	-	10	10	10.00
PENNEAGLE	2	04-21,	12-09,	02-06	0	0	0	0.00
PENNEAGLE	3	13-21,	11-09,	03-06	0	5	50	18.33
PENNEAGLE	5	05-21,	01-09,	04-06	15	20	0	11.67
PENNEAGLE	6	01-21,	-	05-06	10	-	0	5.00
PENNEAGLE	7	-	13-09,	06-06	-	0	0	0.00
PENNEAGLE	8	-	-	07-06	-	-	5	5.00
PENNEAGLE	9	-	02-09,	08-06	-	0	0	0.00
PENNEAGLE	10	-	18-09,	-	-	80	-	80.00
PENNEAGLE	12	02-21,	-	-	5	-	-	5.00
PENNEAGLE	13	16-21,	-	09-06	0	-	0	0.00
PENNEAGLE	14	18-21,	-	-	10	-	-	10.00
PENNEAGLE	15	20-21,	10-09,	10,06	10	0	25	11.67
PENNEAGLE	16	15-21,	-	-	5	-	-	5.00
PENNEAGLE	17	09-21,	08-09,	11-06	25	15	0	13.33
PENNEAGLE	18	06-21,	-	-	0	-	-	0.00
PENNEAGLE	19	03-21,	-	-	0	-	-	0.00
PENNEAGLE	21	08-21,	09-09,	12-06	0	25	5	10.00
PENNEAGLE	22	10-21,	15-09,	13-06	0	30	20	16.67
PENNEAGLE	24	07-21,	-	-	5	-	-	5.00
PENNEAGLE	25	-	-	19-06	-	-	0	0.00
PENNEAGLE	26	12-21,	-	-	0	-	-	0.00
PENNEAGLE	27	-	17-09,	14-06	-	0	0	0.00
PENNEAGLE	28	-	06-09,	20-06	-	45	5	25.00
PENNEAGLE	29	-	20-09,	-	-	5	-	5.00
PENNEAGLE	30	-	19-09,	15-06	-	40	0	20.00
PENNEAGLE	31	17-21,	03-09,	16-06	0	5	0	1.67
PENNEAGLE	32	11-21,	05-09,	17-06	5	0	0	1.67
PENNEAGLE	33	14-21,	14-09,	-	0	5	-	2.50
PENNEAGLE	34	19-21,	-	-	25	-	-	25.00
PENNEAGLE	35	-	16-09,	-	-	5	-	5.00
PENNEAGLE	36	-	04-09,	18-06	-	5	0	2.50
PSU126	1	07-20,	-	-	10	-	-	10.00
PSU126	2	15-20,	09-08,	-	15	0	-	7.50
PSU126	3	19-20,	-	01-04	80	-	10	45.00
PSU126	4	03-20,	-	02-04	0	-	0	0.00
PSU126	5	-	-	03-04	-	-	0	0.00
PSU126	6	-	17-08,	04-04	-	10	0	5.00
PSU126	7	01-20,	-	05-04	10	-	0	5.00
PSU126	8	16-20,	11-08,	-	0	5	-	2.50
PSU126	9	11-20,	07-08,	06-04	0	0	0	0.00
PSU126	10	09-20,	-	07-04	10	-	0	5.00
PSU126	11	14-20,	18-08,	08-04	0	15	0	5.00
PSU126	12	-	06-08,	09-04	-	0	0	0.00
PSU126	13	-	02-08,	-	-	5	-	5.00
PSU126	14	18-20,	12-08,	10-04	0	5	0	1.67

Table 2. Continued:

Parent Variety	Seedling Selection	Field Plot Location			Replicate % Disease			Mean % Disease
					1	2	3	
PSU126	15	08-20,	19-08,	11-04	0	80	0	26.67
PSU126	16	-	15-08,	-	-	0	-	0.00
PSU126	17	12-20,	04-08,	12-04	5	0	0	1.67
PSU126	18	17-20,	-	-	5	-	-	5.00
PSU126	20	02-20,	03-08,	13-04	5	0	0	1.67
PSU126	21	06-20,	14-08,	-	0	60	-	30.00
PSU126	22	04-20,	-	-	50	-	-	50.00
PSU126	23	-	08-08,	14-04	-	5	50	27.50
PSU126	24	13-20,	-	15-04	0	-	0	0.00
PSU126	25	10-20,	-	-	5	-	-	5.00
PSU126	26	20-20,	13-08,	16-04	0	30	0	10.00
PSU126	28	-	05-08,	17-04	-	0	0	0.00
PSU126	29	05-20,	20-08,	18-04	0	0	5	2.50
PSU126	30	-	10-08,	19-04	-	5	0	2.50
PSU126	31	-	01-08,	-	-	100	-	100.00
PSU126	34	-	16-08,	-	-	5	-	5.00
PSU126	36	-	-	20-04	-	-	0	0.00
ISI	1	05-19,	17-14,	01-05	5	0	0	1.67
ISI	4	03-19,	11-14,	02-05	5	5	0	3.33
ISI	5	-	12-14,	03-05	-	0	0	0.00
ISI	6	20-19,	04-14,	04-05	5	15	5	8.33
ISI	7	06-19,	18-14,	-	0	30	-	15.00
ISI	8	02-19,	10-14,	05-05	10	0	15	8.33
ISI	9	04-19,	-	-	0	-	-	0.00
ISI	10	11-19,	01-14,	06-05	50	0	10	20.00
ISI	11	16-19,	09-14,	07-05	5	0	5	3.33
ISI	12	09-19,	-	08-05	50	-	15	32.50
ISI	13	07-19,	-	09-05	0	-	0	0.00
ISI	14	15-19,	13-14,	10-05	5	0	0	1.67
ISI	15	10-19,	19-14,	11-05	0	0	0	0.00
ISI	17	18-19,	20-14,	-	10	-	-	10.00
ISI	19	-	06-14,	12,05	-	0	0	0.00
ISI	20	08-19,	05-14,	13-05	0	0	0	0.00
ISI	22	14-19,	14-14,	-	0	0	-	0.00
ISI	24	17-19,	15-14,	14-05	70	15	0	28.33
ISI	25	01-19,	07-14,	15-05	0	0	0	0.00
ISI	26	19-19,	08-14,	16-05	0	0	0	0.00
ISI	29	13-19,	03-14,	17-05	15	60	0	37.50
ISI	30	-	16-14,	18-05	-	0	5	2.50
ISI	33	-	-	19-05	-	-	5	5.00
ISI	35	12-19,	02-14,	20-05	0	0	0	0.00
PROMINENT	1	10-18,	20-10,	01-02	95	0	60	51.67
PROMINENT	2	05-18,	11-10,	02-02	0	0	0	0.00
PROMINENT	3	14-18,	19-10,	03-02	70	35	15	40.00
PROMINENT	4	01-18,	09-10,	04-02	5	5	5	5.00
PROMINENT	5	18-18,	16-10,	05-02	90	25	10	41.67

Table 2. Continued:

Parent Variety	Seedling Selection	Field Plot Location			Replicate % Disease			Mean % Disease
					1	2	3	
PROMINENT	6	11-18,	17-10,	06-02	50	75	10	45.00
PROMINENT	7	16-18,	08-10,	07-02	0	15	85	33.33
PROMINENT	8	09-18,	12-10,	08-02	25	0	0	8.33
PROMINENT	9	15-18,	02-10,	09-02	5	0	50	18.33
PROMINENT	10	12-18,	10-10,	10-02	95	0	10	35.00
PROMINENT	13	06-18,	18-10,	11-02	0	0	0	0.00
PROMINENT	15	17-18,	06-10,	12-02	10	20	0	10.00
PROMINENT	17	19-18,	15-10,	13-02	10	5	5	6.67
PROMINENT	19	04-18,	07-10,	20-02	0	10	10	6.67
PROMINENT	20	03-18,	13-10,	14-02	15	0	5	6.67
PROMINENT	24	20-18,	14-10,	15-02	5	0	0	1.67
PROMINENT	29	08-18,	04-10,	16-02	40	0	0	13.33
PROMINENT	30	13-18,	03-10,	17-02	25	0	0	8.33
PROMINENT	32	02-18,	05-10,	19-02	70	20	0	30.00
PROMINENT	36	07-18,	01-10,	18-02	0	5	5	5.00
PENNCROSS	1	05-17,	06-11,	01-01	15	10	20	15.00
PENNCROSS	2	15-17,	13-11,	02-01	0	0	0	0.00
PENNCROSS	3	12-17,	17-11,	03-01	10	0	0	3.33
PENNCROSS	4	08-17,	09-11,	04-01	35	0	5	13.33
PENNCROSS	6	20-17,	03-11,	05-01	-	0	5	2.50
PENNCROSS	8	13-17,	08-11,	07-01	0	5	10	5.00
PENNCROSS	9	09-17,	05-11,	08-01	10	0	5	5.00
PENNCROSS	10	01-17,	04-11,	09-01	0	0	5	1.67
PENNCROSS	12	18-17,	19-11,	10-01	20	10	5	11.67
PENNCROSS	13	04-17,	07-11,	11-01	0	0	20	6.67
PENNCROSS	17	19-17,	02-11,	12-01	20	0	5	12.50
PENNCROSS	19	16-17,	10-11,	13-01	0	0	0	0.00
PENNCROSS	21	10-17,	20-11,	14-01	0	5	10	5.00
PENNCROSS	25	06-17,	11-11,	15-01	5	0	0	1.67
PENNCROSS	26	17-17,	15-11,	17-01	0	0	0	0.00
PENNCROSS	27	11-17,	14-11,	18-01	0	0	50	16.67
PENNCROSS	29	07-17,	16-11,	19-01	0	5	0	1.67
PENNCROSS	30	14-17,	12-11,	20-01	5	0	0	1.67
PENNCROSS	33	03-17,	01-11,	06-01	5	20	10	11.67
PENNCROSS	34	02-17,	18-11,	16-01	25	0	10	11.67
DUCHESS	4	01-16,	17-12,	01-03	-	70	0	35.00
DUCHESS	5	02-16,	18-12,	10-03	10	100	0	36.67
DUCHESS	6	-	01-12,	-	-	0	-	0.00
DUCHESS	7	03-16,	02-12,	02-03	5	0	0	1.67
DUCHESS	10	20-16,	03-12,	03-03	0	50	0	16.67
DUCHESS	11	04-16,	19-12,	11-03	0	10	0	3.33
DUCHESS	13	05-16,	04-12,	12-03	0	0	0	0.00
DUCHESS	15	06-16,	05-12,	04-03	0	0	0	0.00
DUCHESS	16	07-16,	-	13-03	0	-	0	0.00
DUCHESS	17	08-16,	06-12,	14-03	0	0	80	26.67
DUCHESS	19	09-16,	07-12,	05-03	5	10	100	38.33

Table 2. Continued:

Parent Variety	Seedling Selection	Field Plot Location	Replicate % Disease			Mean % Disease
			1	2	3	
DUCHESS	22	10-16, 08-12, 15-03	50	10	5	21.67
DUCHESS	24	- , 09-12, 06-03	-	0	0	0.00
DUCHESS	25	11-16, 10-12, 16-03	0	0	0	0.00
DUCHESS	26	12-16, 20-12, 17-03	0	-	0	0.00
DUCHESS	27	13-16, 11-12, 18-03	0	0	0	0.00
DUCHESS	28	14-16, 12-12, 19-03	25	10	0	11.67
DUCHESS	29	15-16, 13-12, 07-03	50	0	0	16.67
DUCHESS	30	16-16, 14-12, 20-03	20	0	100	40.00
DUCHESS	33	17-16, - , -	-	-	-	-
DUCHESS	35	18-16, 15-12, 09-03	20	0	0	6.67
DUCHESS	36	19-16, 16-12, 08-03	0	0	0	0.00
EMERALD	1	04-15, 08-13, 01-07	0	0	0	0.00
EMERALD	3	14-15, 04-13, 02-07	5	20	0	8.33
EMERALD	5	09-15, 10-13, 03-07	0	0	0	0.00
EMERALD	6	20-15, 17-13, 04-07	0	70	0	23.33
EMERALD	7	11-15, 19-13, 05-07	5	60	20	28.33
EMERALD	10	08-15, 13-13, 06-07	0	5	25	10.00
EMERALD	11	12-15, 16-13, 07-07	15	0	90	35.00
EMERALD	12	15-15, 18-13, 08-07	15	70	10	31.67
EMERALD	13	16-15, 12-13, 09-07	0	10	0	3.33
EMERALD	15	10-15, 15-13, 10-07	25	20	0	15.00
EMERALD	16	01-15, 01-13, 11-07	0	0	0	0.00
EMERALD	17	05-15, 05-13, 12-07	10	25	5	13.33
EMERALD	21	19-15, 14-13, 13-07	0	0	0	0.00
EMERALD	23	13-15, 07-13, 14-07	0	0	0	0.00
EMERALD	24	02-15, 02-13, 15-07	60	10	0	23.33
EMERALD	25	06-15, 09-13, 16-07	5	0	5	3.33
EMERALD	27	07-15, 20-13, 17-07	0	5	0	2.50
EMERALD	31	17-15, 11-13, 18-07	0	10	0	3.33
EMERALD	35	18-15, 06-13, 19-07	0	10	5	5.00
EMERALD	36	03-15, 03-13, 20-07	10	65	50	41.67

Table 2a. Summary by variety of % disease means less than 25% (disease tolerant) and more than 50% (disease susceptible).

Parent Variety	% Disease Mean	No. with 0% Mean	Less than 25%	More than 50%
PENNEAGLE	9.219	9	31	1
PSU126	11.586	8	25	2
ISI	7.396	10	21	0
PROMINENT	18.333	2	13	1
PENCROSS	6.333	3	20	0
DUCHESS	11.591	9	16	0
EMERALD	12.375	5	16	0

4. Greenhouse Assessment of Field Evaluations:

A greenhouse experiment was used to correlate disease symptoms from the field plots with seedling disease activity in McDonalds sundae cups. Grass plugs were collected from all of the HTS nursery plots. Three plugs, each about 1.5 cm in diameter, were taken from each plot and they were then placed in McDonald's sundae cups with sand (Fig 2a). The cups were then placed in the greenhouse and the plugs in sand were overseeded with var. 'Penncross' bentgrass seed. After overseeding, each cup was watered and covered with lids. The lids were kept in place at a 27 C temperature for eight days. The moisture saturated conditions in the cups promoted the growth of both the seedlings and of Pythium spp. from the diseased grass. Percent disease data was recorded for each expervar of the HTS nursery collection. As expected, Pythium blight frequency was greater in the greenhouse cups than on the field plots, however, resistance observed on the field plots was not always correlated with the lack of blight symptoms on test seedlings in the greenhouse (Table 3). Of test expervars showing 25% or less disease in the field, 23 (41%) were substantiated by greenhouse data while 27 expervars had a higher greenhouse disease rating (Table 3). These results may reflect a higher level of disease activity by Pythium spp. in enclosed cups when compared to prevailing environments in the field. The results do indicate the importance of field observations in indentifying Pythium spp. resistant genotypes.

A.



B.



Fig. 2. (A) Recovered field plugs from HTS nursery plots were planted in sand in sundae cups. (B) After eight days % disease on overseeded Penncross seedlings was determined.

Table 3. Comparative results of Pythium blight assessments on HTS field plots and greenhouse seedling disease activity in sundae cups containing soil cores from field plots.

JULY 2, 1990

Parent Variety	Selection	Mean % Disease on field plots	Mean % Disease greenhouse cups	Deviation
EMERALD	1	50.00	21.67	28.33
EMERALD	2	15.00	28.33	13.33
EMERALD	3	0.00	30.00	30.00
EMERALD	4	10.00	95.00	85.00
EMERALD	5	18.33	33.33	15.00
EMERALD	6	20.00	56.67	36.67
EMERALD	7	50.00	63.33	13.33
EMERALD	8	56.67	66.67	10.00
EMERALD	9	23.33	38.33	15.00
EMERALD	10	43.33	33.33	10.00
EMERALD	11	35.00	0.00	35.00
EMERALD	12	36.67	33.33	3.34
EMERALD	13	93.33	50.00	43.33
EMERALD	14	0.00	33.33	33.33
EMERALD	15	3.33	50.00	46.67
EMERALD	16	76.67	51.67	25.00
EMERALD	17	60.00	71.67	11.67
EMERALD	18	3.33	55.00	51.67
PENNEAGLE	1	6.67	26.67	20.00
PENNEAGLE	2	12.50	3.33	9.17
PENNEAGLE	3	31.67	96.67	65.00
PENNEAGLE	4	10.00	38.33	28.33
PENNEAGLE	5	47.33	66.67	19.34
PENNEAGLE	6	1.67	60.00	58.33
PENNEAGLE	7	25.00	0.00	25.00
PENNEAGLE	8	53.33	66.67	13.34
PENNEAGLE	9	28.33	63.33	35.00
PENNEAGLE	10	58.33	26.67	31.66
PENNEAGLE	11	46.67	0.00	46.67
PENNEAGLE	12	21.67	66.67	45.00
PENNEAGLE	13	47.50	35.00	12.50
PENNEAGLE	14	48.33	58.33	10.00
PENNEAGLE	15	13.33	38.33	25.00
PENNEAGLE	16	26.67	75.00	48.33
PENNEAGLE	17	10.00	78.33	68.33
PENNEAGLE	18	2.50	16.67	14.17
ISI	1	0.00	66.67	66.67
ISI	2	0.00	0.00	0.00
ISI	3	5.00	33.33	28.33
ISI	4	42.67	68.33	25.66
ISI	5	46.67	75.00	28.33
ISI	6	40.00	83.33	43.33
ISI	7	6.67	93.33	86.66
ISI	8	45.00	100.00	55.00
ISI	9	35.00	63.33	28.33
ISI	10	18.33	26.67	8.34

Table 3. Continued:

Parent Variety	Selection	Mean % Disease on field plots	Mean % Disease greenhouse cups	Deviation
ISI	11	15.00	41.67	26.67
ISI	12	73.33	35.00	38.33
ISI	13	11.67	33.33	21.66
ISI	14	11.67	26.67	15.00
ISI	15	40.00	33.33	6.67
ISI	16	23.33	50.00	26.67
ISI	17	53.33	63.33	10.00
ISI	18	41.67	63.33	21.66
PSU126	1	36.67	51.67	15.00
PSU126	2	13.33	58.33	45.00
PSU126	3	20.00	0.00	20.00
PSU126	4	53.33	41.67	11.66
PSU126	5	38.33	90.00	51.67
PSU126	6	36.67	83.33	46.66
PSU126	7	0.00	30.00	30.00
PSU126	8	33.33	10.00	23.33
PSU126	9	41.67	66.67	25.00
PSU126	10	28.33	30.00	1.67
PSU126	11	50.00	63.33	13.33
PSU126	12	11.67	90.00	78.33
PSU126	13	11.67	63.33	51.66
PSU126	14	8.33	66.67	58.34
PSU126	15	18.33	60.00	41.67
PSU126	16	48.33	33.33	15.00
PSU126	17	26.67	8.33	18.34
PSU126	18	25.00	1.67	23.33
DUCHESS	1	33.33	41.67	8.34
DUCHESS	2	5.00	63.33	58.33
DUCHESS	3	40.00	33.33	6.67
DUCHESS	4	24.00	48.33	24.33
DUCHESS	5	0.00	100.00	100.00
DUCHESS	6	46.67	46.67	0.00
DUCHESS	7	63.33	80.00	16.67
DUCHESS	8	53.33	41.67	11.66
DUCHESS	9	21.67	86.67	65.00
DUCHESS	10	33.33	58.33	25.00
DUCHESS	11	8.33	50.00	41.67
DUCHESS	12	33.33	40.00	6.67
DUCHESS	13	36.67	50.00	13.33
DUCHESS	14	12.50	100.00	87.50
DUCHESS	15	25.00	100.00	75.00
DUCHESS	16	6.67	66.67	60.00
DUCHESS	17	40.00	75.00	35.00
DUCHESS	XX	15.00	12.50	2.50
PROMINENT	1	41.67	83.33	41.66
PROMINENT	2	36.67	0.67	36.00
PROMINENT	3	36.67	93.33	56.66
PROMINENT	4	7.33	63.33	56.00
PROMINENT	5	40.00	35.00	5.00
PROMINENT	6	13.33	28.33	15.00

Table 3. Continued:

Parent Variety	Selection	Mean % Disease on field plots	Mean % Disease greenhouse cups	Deviation
PROMINENT	8	53.33	51.67	1.66
PROMINENT	9	21.67	0.00	21.67
PROMINENT	10	20.00	16.67	3.33
PROMINENT	11	41.67	36.67	5.00
PROMINENT	12	70.00	0.00	70.00
PROMINENT	13	5.00	16.67	11.67
PROMINENT	14	28.33	16.67	11.66
PROMINENT	15	43.33	1.67	41.66
PROMINENT	16	51.67	0.00	51.67
PROMINENT	17	30.00	8.33	21.67
PROMINENT	18	51.67	50.00	1.67
PROMINENT	19	45.00	100.00	55.00
PENNCROSS	1	25.00	66.67	41.67
PENNCROSS	3	18.33	63.33	45.00
PENNCROSS	4	46.67	60.00	13.33
PENNCROSS	5	13.33	31.67	18.34
PENNCROSS	6	23.33	55.00	31.67
PENNCROSS	7	15.00	66.67	51.67
PENNCROSS	8	48.33	60.00	11.67
PENNCROSS	9	61.67	60.00	1.67
PENNCROSS	10	51.67	36.67	15.00
PENNCROSS	11	55.00	63.33	8.33
PENNCROSS	12	55.00	56.67	1.67
PENNCROSS	13	30.00	63.33	33.33
PENNCROSS	14	55.00	33.33	21.67
PENNCROSS	15	30.00	36.67	6.67
PENNCROSS	16	21.67	41.67	20.00
PENNCROSS	17	28.33	58.33	30.00
PENNCROSS	18	28.33	63.33	35.00
PENNCROSS	19	46.67	50.00	3.33

IV. Rhizoctonia Diseases Ongoing Research

1. Rhizoctonia Isolate Collection and Storage:

Turfgrass Rhizoctonia isolates are being maintained in long-term storage in replicate prescription vials (3 drams) on PDA at a temperature of 20 C. The collection is the larger of the two fungal culture groups being maintained for the research program. Some of the Rhizoctonia spp. isolates appear to develop a leathery crust on the surface of the culture which is very difficult to obtain subcultures for use in inoculation studies. Many of the cultures also have only short-term viability in storage vials on agar. For this reason, transfers from the isolate collection will be made at 6 month intervals to insure maximum survival of isolates in the Rhizoctonia collection.

2. Development of Field Inoculation Technique

Previous investigations have shown that inoculation of turfgrasses in the field is advantageous because various forms of field resistance can be identified through inoculation studies. Inoculations of the same bentgrass germplasm lines taken directly from the field or from a greenhouse bench indicated 35% more resistance among the population of field-grown plants. Mass inoculation procedures presently in use also often require extensive wounding of the leaf canopy and need long-term exposure to be effective.

Field studies were undertaken during the fall to determine if field inoculations with Rhizoctonia solani could be

achieved on field plots of Raleigh St. Augustinegrass and Penncross bentgrass. Both of the field plots had been established for more than one year and received heavy fertilization and irrigation prior to applying inoculum. Rhizoctonia solani isolate (R-31) was used as a source of inoculum in the study as the isolate was shown to be highly pathogenic in greenhouse pathogenicity tests (fig 3). The fungus was cultured for one week on a mixture of sterile grain seed including equal portions of millet, fescuegrass and barley in canning jars. The colonized seed was passed through a 0.5 inch wire mesh screen and dried on a greenhouse bench 48 hrs before being used as inoculum. The colonized seed was scattered by hand over the field plot (one seed/ft²) before applying water by hand. Typical disease symptoms were noted on the Raleigh St. Augustinegrass plot within 7 days but no symptoms were observed on the bentgrass plots. Additional field inoculations on bentgrass will utilize higher fertility levels to stimulate Rhizoctonia blight activity.

3. Indexing Pathogenicity of Rhizoctonia Isolates:

Study emphasis during the summer months sought to identify sources of bentgrass resistance to both Pythium and Rhizoctonia foliar blights. Research during the past two years has primarily concentrated on the identification of Pythium resistance within bentgrass germplasm lines. Additional research using the same mass disease screening procedures examined Pythium disease resistant lines for

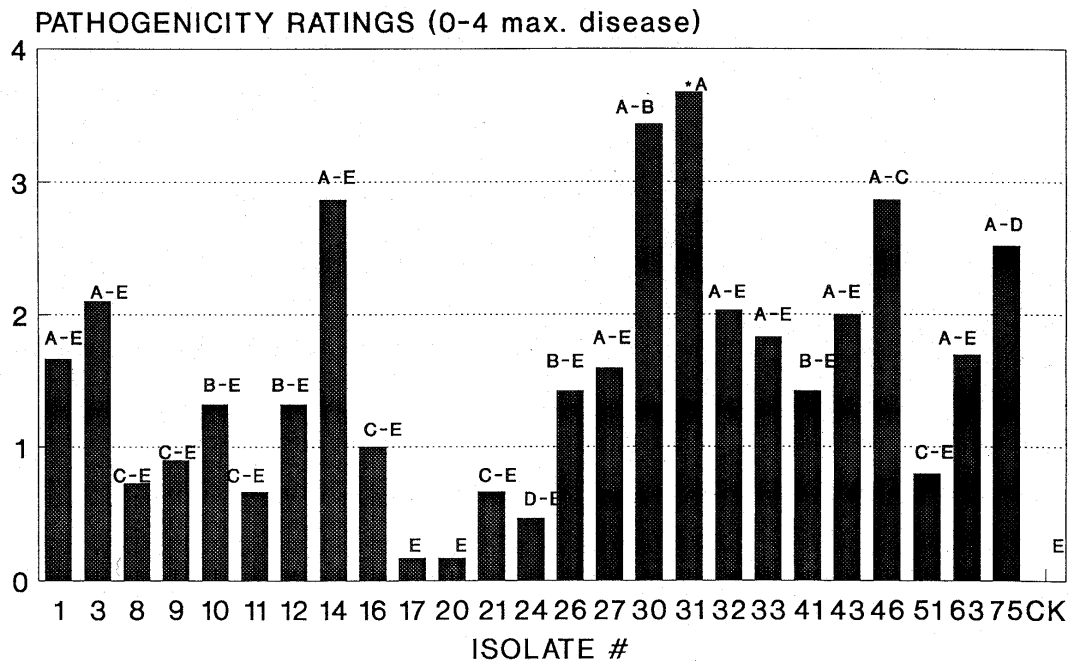
tolerance or resistance to Rhizoctonia blight. As with previous studies with Pythium spp., the inoculation studies employed a virulent strain of Rhizoctonia applied to field collected plants which were maintained in the walk in environmental chamber in plastic McDonald's cups.

The pathogenicity of 25 Rhizoctonia solani isolates resembling AG2-2 was determined in greenhouse inoculation studies. Agar cultures of the fungal isolates were prepared on PDA agar in petri dishes where the fungi were allowed to colonize for four days prior to inoculation of a standard Penncross turf. Greenhouse benches with Penncross bentgrass were used to uniformly inoculate the 25 strains in a completely randomized block with three replica inoculations for each fungal strain. Plastic cups were placed over areas of the turf where a small agar disc containing fungal mycelium was placed on the leaf canopy. After misting the inoculated turf with sterile water the inoculum was left to colonize the turf for a period of five days. A disease index of 0-4 where 4 = maximum diameter of the diseased grass (5 cm dia) was used. Results of the inoculation study are shown in Fig. 3. Five of the fungal isolates; nos. 14, 30, 31, 45 and 75 were more pathogenic than other isolates tested.

4. Screening Study for Multiple Disease Resistance

On 5 August three replicate field cores of 26 expervars (fig. 4) were removed from the HTS nursery and 79 expervars from the NHTS nursery to screen the germplasm lines for Rhizoctonia blight

Fig. 3. PATHOGENICITY RATINGS FOR
RHIZOCTONIA
ON PENNCROSS BENTGRASS



* DNMRT 0.05 %



A.



B.

Fig. 4. (A) *Rhizoctonia solani* AG2-2 isolate R-31 on experimental bentgrass. (B) covered sundae cups used for experimental disease resistance study.

resistance. Field cores (8 cm dia) were placed in McDonald's sundae cups and taken immediately to the walk-in environmental chamber for inoculation. Agar discs from Rhizoctonia solani isolates R-31, R-30 and R-75 were removed from the periphery of four day old colonies growing on potato-dextrose-agar in petri dishes and used for inoculation of the germplasm lines. Six days after inoculation and incubation at 27 C the severity of disease on replicate cores was determined using a disease index (0-5 max).

The selections used in this study were based on visual selections from the field evaluations earlier in the year (Tables 1 & 2). Results of the study do not indicate a strong correlation of expervars showing field resistance to Pythium blight and those showing resistance to Rhizoctonia blight under conditions of this experiment (Table 4,5). The mean overall disease ratings for progeny of six parental varieties in the HTS nursery were very similar (Table 4) and ranged from 3.4 to 4.9. Disease ratings for progeny of the same parental varieties in the NHTS nursery were somewhat different and ranged from 2.7 to 5.0. The mean % disease for parental varieties in the HTS group (4.3) were generally higher than those in the NHTS group (3.6) where the same varieties are compared. The lowest index of disease was observed in the NHTS germplasm group for the parental varieties penneagle (2.7) and Prominent (2.9) (Table 5). Rhizoctonia blight resistance was generally concentrated among individual selections from Penneagle, Prominent, PSU6999 and Penncross.

Table 4. Rhizoctonia solani (R-31) foliar blighting on HTS Bent grass germplasm lines showing field resistance to Pythium blight

Parent Variety	Field Plot Location	<u>Rhizoctonia</u> blight Disease (0-5 max) *	Mean Var. Rating
Emerald	03-11	5	
Emerald	03-21	4.5	
Emerald	08-11	4	
Emerald	16-05	4.5	
Emerald	18-21	5	4.6
Penneagle	03-20	4.3	
Penneagle	10-13	3	
Penneagle	16-07	4	
Penneagle	17-13	5	
Penneagle	18-13	5	4.3
ISI 2999	01-19	4.5	
ISI 2999	02-19	5	
ISI 2999	03-19	5	
ISI 2999	05-19	5	
ISI 2999	15-19	5	4.9
PSU 126	01-18	2	
PSU 126	02-18	5	
PSU 126	10-14	5	
PSU 126	12-18	4	
PSU 126	14-18	5	
PSU 126	18-14	4.5	4.3
Duchess	13-17	4.5	4.5
Penncross	02-15	4.5	
Penncross	05-10	4	
Penncross	14-10	2.5	
Penncross	16-15	2.5	3.4

*/ Disease data was taken using a disease index (0-5) where 0=no disease and 5= 100% disease.

Table 5. Rhizoctonia solani (R-31) foliar blighting on NHTS Bent grass germplasm lines showing field resistance to Pythium blight

Parent Variety	Field Plot Location	Rhizoctonia blight Disease (0-5) *	Mean Var. Rating
Emerald	04-15	5	
Emerald	09-07	4.7	
Emerald	09-13	4.7	
Emerald	14-13	5	
Emerald	16-07	4.5	
Emerald	16-15	2.8	
Emerald	18-15	2	
Emerald	19-15	5	4.22
Penneagle	03-21	3.5	
Penneagle	04-21	4.5	
Penneagle	05-09	2	
Penneagle	06-06	3	
Penneagle	06-21	3	
Penneagle	09-06	3.5	
Penneagle	11-06	4	
Penneagle	13-09	2.5	
Penneagle	14-06	1	
Penneagle	14-21	2	
Penneagle	15-06	0.8	
Penneagle	16-21	4	
Penneagle	17-09	1.8	
Penneagle	17-21	3.5	
Penneagle	19.06	1	2.74
ISI 2999	01-05	5	
ISI 2999	01-14	4	
ISI 2999	05-05	3.5	
ISI 2999	05-14	4	
ISI 2999	06-19	2.5	
ISI 2999	07-14	4.7	
ISI 2999	09-05	3	
ISI 2999	11-05	4	
ISI 2999	12-05	5	
ISI 2999	13-05	4.5	
ISI 2999	14-05	5	
ISI 2999	15-05	4.5	
ISI 2999	16-05	2	
ISI 2999	16-14	3.5	
ISI 2999	19-05	5	
ISI 2999	19-14	4.7	
ISI 2999	20-19	2	3.92
PSU 126	02-04	3	
PSU 126	03-04	4.5	
PSU 126	05-04	3	
PSU 126	05-08	4	
PSU 126	06-04	4	

Table 5. Continued:

Parent Variety	Field Plot Location	Rhizoctonia blight Disease (0-5)	*
PSU 126	08-04	5	
PSU 126	09-04	5	
PSU 126	10-08	1	
PSU 126	11-04	4.8	
PSU 126	12-04	4.5	
PSU 126	12-20	3.5	
PSU 126	13-08	5	
PSU 126	14-08	3	
PSU 126	14-20	5	
PSU 126	16-08	2.5	
PSU 126	16-20	3	
PSU 126	18-20	5	
PSU 126	20-08	1	3.70
Duchess	15-12	5	5.00
Penncross	03-17	4.7	
Penncross	04-17	2	
Penncross	09-17	3	
Penncross	10-11	2.5	
Penncross	11-11	5	
Penncross	13-01	4.5	
Penncross	15-01	5	
Penncross	15-11	2.5	
Penncross	15-17	5	
Penncross	16-17	3	
Penncross	17-01	5	
Penncross	17-17	3	
Penncross	19-01	5	3.96
Prominent	03-10	3	
Prominent	10-10	4	
Prominent	12-02	4	
Prominent	13-10	1	
Prominent	15-02	4	
Prominent	16-18	2	
Prominent	17-02	2.5	2.93

*/ Disease data was taken using a disease index (0-5) where 0=no disease and 5= 100% disease.

V. Cooperative Research with USGA Rhizoctonia clones

1. Fatty Acid Analysis of Strains:

The technology for separation and identification of the strains of fungi causing the Rhizoctonia diseases has advanced rapidly during the past decade. Dr. Roger Jones who is a plant pathologist at University of Minnesota is using fatty acid analysis as a tool to determine the identity of Rhizoctonia strains. Rhizoctonia is a complex of several species and subspecific groups. Certain species within the genus have been well studied and are consistently associated with symptoms that can be identified. Other species are less well understood. The separation of Rhizoctonia species within the four groups causing turfgrass diseases is difficult and now relies on anastomosis testing to determine the affiliation of strains. Anastomosis is the ability of growing cells of one isolate to fuse (recognize) cells of another isolate. Two isolates that are capable of this level of recognition are said to belong to the same anastomosis group. Dr. Jones has characterized Rhizoctonia strains from turfgrasses using the fatty analysis method of strain identification. Most of the USGA Rhizoctonia culture collection for turfgrass was sent to Jones during the past winter for this research effort. Dr. Jones has characterized Rhizoctonia strains from turfgrasses using the fatty analysis method of strain identification. This method of strain identification may prove to be very useful for use in breeding programs and fungicide screening programs for

controlling the Rhizoctonia diseases.

2. Tissue Culture Experiments with Rhizoctonia Isolates:

Rhizoctonia cultures from the USGA Collection were also sent to Mississippi State University in a cooperative research effort with Dr. Jeff Krans who has done extensive tissue culture screening with Penncross bentgrass explants. The tissue culture screening procedure to determine resistance to Rhizoctonia solani uses one isolate that produces a toxic factor that diffuses through the agar to the callus tissue. Dr. Krans is examining several isolates from the Rhizoctonia USGA collection that are highly pathogenic and others that are only weakly pathogenic to determine the relationships of toxic factor presence among isolates and their known pathogenicity on Penncross bentgrass.

VI. Other Diseases

In response to requests to add additional pathogens to the USGA fungal collection, cultures of fungi causing the Helminthosporium leafspot diseases and the dollar spot disease have been started. Most of these cultures will be obtained during the upcoming spring and fall season during periods of highest activity by these fungi. Since we have not determined appropriate methods for long term preservation of these fungi, they are now being held as agar cultures in test tubes at temperatures favoring their preservation. Additional isolates will be solicited from colleague scientists during the upcoming summer months.

VII. Areas of Future Investigation:

Inoculation Studies with Rhizoctonias on bentgrasses will be continued in the GPIN bentgrass nursery at Dallas in order to identify potential germplasm lines of resistance to the foliar blighting stage of both diseases. Variations of the field inoculation procedures that were initiated during the past year will also attempt to develop a suitable method for use in field inoculations.