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Proceedings of the 27th Annual Meeting, Southern Soybean Disease Workers (April 6-8, 2000, Fort Walton Beach Florida)

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PROCEEDINGS OF THE SOUTHERN SOYBEAN DISEASE WORKERS



27TH ANNUAL MEETING

April 6–8, 2000 | Fort Walton Beach, Florida

PROCEEDINGS OF THE

SOUTHERN SOYBEAN DISEASE WORKERS

27th ANNUAL MEETING

APRIL 6-8, 2000

FORT WALTON BEACH, FLORIDA

Thursday, April 6

Friday, April 7

Saturday, April 8

Steering Committee Meeting

Invited Paper Contributed Papers State Reports Business Meeting Steering Committee Exit Meeting



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SOUTHERN SOYBEAN DISEASE WORKERS

1999 - 2000 OFFICERS

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TABLE OF CONTENTS

BUSINESS SECTION

Pa	ige
Southern United States Soybean Disease Loss Estimate for 1999S. R. KoenningNorth Carolina State University1	l
Treasury ReportP. S. KingAuburn University.6	5
INVITED PRESENTATION	
Bean Pod Mottle: A Soybean Disease on the Rise in the New Millennium S. A. Ghabrial	
University of Kentucky	
CONTRIBUTED PAPERS	
Response of Selected Mid-South Soybean Varieties to the Reniform Nematod G. W. Lawrence, K. S. McLean, and S. M. Baird Mississippi State University and Auburn University	le
Field Response of Soybean Cultivars to the Reniform Nematode Rotylenchulus reniformisP. S. King, D. B. Weaver, and R. Rodriguez-Kabana Auburn University.9	
Approaches to Race Determination in Soybean Cyst NematodeA. J. Palmateer, M. E. Schmidt, S. R. Stetina, and J. S. RussinSouthern Illinois University10	
Nematological Survey of Selected Soybean and Cotton Fields in Alabama D. G. Robertson and R. Rodriguez-Kabana Auburn University	
Soybean Meal-Based Compositions as Organic Amendments for Control of Plant-Parasitic NematodesC. F. Weaver and R. Rodriguez-Kabana Auburn University.12	
Cell Selection Approach for Generating Soybean with Resistance to Macrophomina phaseolinaN. A. Reichert, G. L. Sciumbato, SH. Lin, L. Chen, B. L. Keeling, and A. L. Woods Mississippi State University.13	

 Identification of Molecular Markers Linked to a New Gene Conferring Resistance to Frogeye Leaf Spot in 'Peking' Soybean W. Yang, D. B. Weaver, J. Qiu, and B. Nielsen Auburn University. 	14
 Soil Physical, Chemical, and Biological Properties Associated with Sudden Death Syndrome in Southern Illinois J. P. Bond, A. J. Hoskins, C. M. Vick, S. K. Chong, and J. S. Russin Southern Illinois University 	15
 Response of Soybean Sudden Death Syndrome to Amelioration of the Soil Physical Environment C. M. Vick, S. K. Chong, J. P. Bond, and J. S. Russin Southern Illinois University 	16
 Differential Enzyme Activity in Two Soybean Cultivars Resistant and Susceptible to Sudden Death Syndrome S. A. Bates, D. A. Lightfoot, O. Myers, Y. Luo, and J. S. Russin Southern Illinois University	17
Evaluation Of Azoxystrobin On Soybean Disease, Yield, and Seed Quality K. S. McLean, G. W. Lawrence, L. Carter, and L. Campbell Auburn University and Mississippi State University	18
Soybean Disease Loss Estimates for the United States from 1996-1998 J. A. Wrather and W. C. Stienstra University of Missouri and University of Minnesota	19

L.

SOUTHERN UNITED STATES SOYBEAN DISEASE LOSS ESTIMATE FOR 1999

Compiled by Stephen R. Koenning Extension Specialist, Department of Plant Pathology, Campus Box 7616, North Carolina State University, Raleigh, NC 27695-7616

Since 1974, soybean disease loss estimates for the Southern United States have been published in the Southern Soybean Disease Workers Proceedings. Summaries of the results from 1977 (7), 1985 and 1986 (1), 1987 (2), 1988 to 1991 (6), 1992 to 1993 (9), 1994 (12), 1994 to 1996 (4) have been published. A summary of the results from 1974 to 1994 has also been published (8).

The loss estimates for 1999 published here were solicited from: Bill Gazaway in Alabama, Clifford Coker in Arkansas, Robert Mulrooney in Delaware, Tom Kucharek in Florida, Richard Davis in Georgia, Don Hershman in Kentucky, Ken Whitam in Louisiana, Gabe Sciumbato in Mississippi, Allen Wrather in Missouri, Steve Koenning in North Carolina, Phil Pratt in Oklahoma, Charles Drye in South Carolina, Melvin Newman in Tennessee, Joseph Krausz in Texas, and Patrick Phipps in Virginia. As recommended by Dr. Arvydas Grybauskas, University of Maryland, the Maryland soybean disease loss estimate was provided by Robert Mulrooney of Delaware. Various methods were used to obtain the disease losses, and most individuals used more than one. The methods used were: field surveys, plant disease diagnostic clinic samples, variety trials, questionnaires to Cooperative Extension staff, research plots, grower demonstrations, private crop consultant reports, and foliar fungicide trials. The actual production figures for each state were supplied by the state crop reporting services. Production losses were based on estimates of yield in the absence of disease. The formula was: potential production without disease loss = actual production + 1-percent loss (decimal fraction).

In the southern states, the 1999 average soybean yield and acreage decreased from that reported in 1998 (6). In 1999, 447.7 million bushels were harvested from 17.6 million acres in 16 southern states. The overall average for the 16 reporting states was 24.1 bushels/acre. The overall average reported in 1998 was 27.5 bushels/acre. The 1999 total acres harvested, average yield in bushels per acre, and total production in each state are presented in Table 1.

Percentage loss estimates from each state are specific as to causal organism or the common name of the disease (Table 2). The total average percent disease loss for 1999 was 11.3 %, a slight decrease from the 11.53 % loss for 1998 (6). In 1999, Mississippi reported the greatest percent loss at 18.36%, followed by Louisiana at 17.30 %. Virginia reported the least at 4.20 % (Table 2).

The estimated reduction of soybean yields is specific as to the causal organism or the common name of the disease (Table 3). The estimated reduction in soybean yield due to diseases during 1998 was greatest in Missouri with 18.36 million bushels and least in Florida with 0.04 million bushels. The total reduction in soybean yield due to diseases in the 16 southern states was 57.27 million bushels in 1999, down from 78.282 million bushels in 1998; partially due to lower production. The estimated dollar value of this loss was \$286,350,000 (based on \$5.00/bu).

In 1999, the highest estimated percent loss was caused charcoal rot 3.39% (22.39 million bushels) (Table 2 & 3). The soybean cyst nematode was the second most damaging pathogen effecting an estimated 2.19% yield loss (10.48 million bushels). The least reported disease was brown stem rot. Brown stem rot was not reported as occurring in any of the 16 southern states.

In 1999, diseases continued to cause significant loss in soybean production throughout the 16 southern states that participated in this disease loss estimate. It is essential that Extension and University research continue their efforts to discover methods to control these diseases and to educate soybean producers concerning the best methods to prevent yield loss due to soybean diseases.

State	Acres harvested	Yield/acre (bu) Tota	al production (bu)
Alabama	200,000	16	3,200,000
Arkansas	3,400,000	27	91,800,000
Delaware	201,000	. 26	5,200,000
Florida	19,000	30	600,000
Georgia	180,000	18	3,200,000
Kentucky	1,100,000	19	20,900,000
Louisiana	1,010,000	25	20,900,000
Maryland	480,000	30	14,400,000
Mississippi	1,950,000	26	50,700,000
Missouri	5,300,000	27	143,100,000
North Carolina	1,300,000	23	29,900,000
Oklahoma	460,000	23	10,500,000
South Carolina	600,000	21	12,600,000
Tennessee	1,040,000	19	19,800,000
Texas	320,000	31	9,900,000
Virginia	460,000	24	11,000,000
Total	18,020,000	Avg. =24.1	447,700,000

Table 1. Soybean production for 16 southern states in 1999.

Table 2. Estimated percentage loss of soybean yields for 16 southern states during 1999 a.

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Disease	A	AR	DE	Н	В	کح	P	QW	WS	QM	NC	ð	sc	N	Ĕ	٨	Avg.
			Ĺ	Ę	070	020	ŝ	C F	010		02.0		5		Ş	F	2
Anthrachose	3.00	0.40	Ľ	B .	0.0	200	8	2	040		8	200	3	NZ.N	30.0	4	0.03
Bacterial diseases	TR	0.01	000	000	0.00	0.01	TR	0.0	TR		0:00	TR	0.03	0.00	0.00	0.0	0.12
Brown leaf spot	0.00		0.00	0:00	0.00	0.10	0.00	0.00	0.10		0.00	0.00	0.10	0.20	0.20	TR	0.04
Brown stem rot	0.00	0.00	0.00	0.00	000	0.00	000	0.00	0.00		00.0	00:0	0.00	0.00	0.00	0.00	0.00
Charcoal rot	0:50	4.00	5.00	1.00	0.20	10.00	4.00	5.00	14.00	2.50	0.40	4.00	0.10	1.50	2.00	0.00	3.39
Diaporthe/Phomopsis	0.10	0.01	ТR	2.00	0.40	0.20	1.00	ТR	TR	0.25	0.10	0.30	0.01	0.00	1.00	1.00	0.40
Downy mildew	0:50	0.01	0.00	0.00	0.10	0.01	TR	0.00		•	0.00	TR	0.05	00.0	0.00	0.00	0.40
Frogeye	0:50	0.10	0.00	0.50	0:50	0.01	0.20	0.00	0.05		0.00	0.00	0.05	1.00	0.20	TR	0.19
Fusarium wilt and rot	TR	0.01	0:00	0.00	0:00	0.01	ТR	0.00		0.50	0.00	0.00	0.01	00.0	0.20	TR	0.05
Other diseases ^b	0.00		0:00	0:00	0.40	0.00	0.20	0.00			5.05	0.00	0.00	0.00	0.10	0.00	0.36
Phytophthora rot	0:00	0.04	0.00	0:00	0.00	0.01	0.20	0:00	0.01	1.50	0.20	0.50	0.01	0.10	0.10	TR	0.17
Pod & stern blight	3.00	0:60	TR		0.10	0.20	2.00	ТR	2.00		0.80	1.00	0.80	0.00	0.50	TR	0.69
Purple seed stain	0.40	0.25	0.00	0.10	0.10	0.01	3.00	0:00	0.70		0.01	0.40	0.25	0.00	1.00	0.00	0.39
Aerial blight	000	0.10	0:00	0.00	0.00	0.00	2.00	0:00	•		0.00	0.00	0.00	0.00	0.10	0.00	0.14
Sclerotinia	0:00		0.00	0.00	0.00	0.00	0.00	00:0	TR		0.00	0.00	0.00	0.00	0.00	0.00	0.00
Seedling diseases	0.20	1.00	0:50	1.00	0.60	0.10	TR	0:50	0:50	0:50	0.01	0.75	0.10	1.50	0.40	0:50	0.51
Southern blight	0.10	0.01	0.00	0.10	0.20	0.01	TR	0.00	ТR	•	0.05	ТR	1.25	0.10	0.20	TR	0.13
Soybean cyst nematode	2.00	1.25	3.00	1.00	3.00	2.00	1.00	3.00	0.10	2.50	5.70	1.50	3.00	4.00	0.00	2.00	2.19
 Root-knot nematode 	1.00	1.45	TR	0.10	3.00	0.00	200	0.00	TR	0.10	0.90	0.10	3.00	0.10	0.20	0.10	0.75
Other nematodes ^c	2.00	0.10	0.00	•	0.50	0.00	0.50	0.00	00:0	•	0:30	0.00	2.00	0.20	0.00	0:50	0.38
Stern Canker	0:00	0.25	TR	0.00	0:50	0.01	TR	Ч	TR		0.00	0.00	0.00	0.10	0.10	0.00	0.06
Sudden death syndrome	1.00	0.10	0.00	0.00	00:0	0.01	0.00	0.00	TR	0.25	0.00	0.00	0.00	0:30	0.10	0.00	0.11
Virus ^d	TR	0.04	000	0.00	0.10	0.10	0.20	00:0	0.50	•	0.20	0.10	1.50	0.00	0.10	0.10	0.18
Total disease %	14.30	9.81	8.50	6.80	9.80	13.29	17.30	8.50	18.36	8.10	13.80	8.95	13.26	9.30	9:50	4.20	11.34

^a Rounding errors present. TR indicates trace.

^b Other diseases listed were: ozone in NC, red crown rot caused by Cylindrocladium parasiticum in NC and VA; target spot in LA, and Phymatotrichum root rot

in TX. ^c Other nematodes listed were: Stubby root in VA; Columbia lance in NC and Georgia; Lance in LA; and Reniform in AL, AR, and

NC. ⁴ Viruses were identified as: soybean mosaic in AL, AR, GA, KY, MS, NC, and VA; bean pod mottle in AR, KY, LA, MS,NC, and VA; Tobacco ringspot

in AR; and peanut mottle in VA.

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soybean	
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reduction	
Estimated	
Table 3.	

southern states during 1999	,													
Disease	AL	AR	Ш		GA	Υ	LA	QM	MS	MO	NC	Я	sc	TN
Anthracnose	0.11	0.49	.	0.01	0.00	0.12	0.25	0.00	0.25	0.00	0.03	0.03	0.15	0.04
Bacterial diseases		0.01	00.00	0.00	0.00	0.00		0.00	•	0.00	0.00		0.00	0.00
Brown leaf spot	0.00	0.00	00.0	0,00	0.00	0.02	00.0	0.00	0.06	0.00	0.00	0.00	0.01	0.04
Brown stem rot	0.00		0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Charcoal rot	0.02	4.07	0.29	0.01	0.01	2.41	1.01	0.79	8.69	3.89	0.14	0.46	0.01	0.33
Diaporthe/Phomopsis	0.00	0.01	•	0.01	0.01	0.05	0.25	0.00		0.39	0.03	0.03	0.00	0.00
Downy mildew	0.02	0.01	00.00	0.00	0.00	0.00	•	00.00		0.00	0.00	•	0.01	0.00
•												1	•	

Tot.

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3.10 0.82 0.31 0.61 57.27 10.48 0.75 1.810.020.000.000.0470.0470.801.031.031.030.0470.0470.0470.020.000.0000.48 0.23 0.01 0.00 0.00 0.00 0.01 0.00 0.06 0.06 0.00 0.00 0.00 0.12 0.00 1.04 0.02 0.00 0.02 0.01 0.01 0.01 0.02 0.00 0.04 0.00 0.02 0.00 0.22 0.00 0.02 0.01 0.33 0.11 0.01 5 $\begin{array}{c} 0.00\\ 0.00\\ 0.00\\ 0.00\\ 0.02\\ 0.02\\ 0.02\\ 0.02\\ 0.02\\ 0.02\\ 0.00\\ 0.00\\ 0.00\\ 0.00\\ \end{array}$ 2.03 0.22 0.00 0.00 0.02 .93 0.44 0.29 0.00 0.00 0.22 0.44 0.00 0.00 0.18 0.04 0.01 0.00 0.00 0.00).12 0.01 0.01 0.00 0.00 0.01 1.03 0.05 0.00 0.00 0.09 0.00 0.00 0.06 0.12 0.17 0.02 1.98 0.31 0.10 0.00 0.00 4.79 0.00 0.00 00.0 0.00 0.00 1.75 0.07 0.28 3.89 0.16 0.00 0.39 0.00 12.61 $\begin{array}{c} 0.00\\ 0.78\\ 0.00\\ 0.00\\ 0.00\\ 0.00\\ 0.00\\ 0.00\\ 0.00\\ 0.00\\ 0.00\\ 0.00\\ 0.00 \end{array}$ 11.40 0.31 . 0.06 0.01 1.24 0.43 0.03 . 0.31 • 0.47 0.00 0.00 0.00 0.00 0.00 1.34 $\begin{array}{c} 0.00\\$. 0.00 0.05 0.25 0.51 0.13 4.37 . 0.05 0.05 0.51 0.76 0.51 0.00 0.05 0.48 0.00 0.00 0.00 0.00 0.02 3.20 0.02 0.00 00.0 0.00 0.00 0.05 00.0 0.11 0.11 0.02 0.00 0.00 0.35 0.020.000.010.000.000.000.020.020.020.00 0.00 0.04 0.00 0.00 0.00 0.00 0.00 00.0 0.00 0.00 0.01 0.00 0.01 0.00 0.49 0.00 0.00 0.00 0.00 0.00 0.03 0.03 0.17 66.6 0.25 0.10 0.04 0.10 0.10 I.48 0.10 1.02 0.01 0.04 0.25 0.61 1.27 0.53 $\begin{array}{c} 0.01\\ 0.00\\ 0.00\\ 0.01\\ 0.07\\ 0.07\\ 0.07\\ 0.00\\ 0.00\\ 0.04\end{array}$ 0.00 0.02 0.11 Sudden death syndrome Soybean cyst nematode Fusarium wilt and rot Root-knot nematode Seedling diseases Pod & stem blight Other nematodes Purple seed stain Phytophthora rot Other diseases Southern blight Stern Canker Aerial blight Sclerotinia Frogeye /irus Total

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SOUTHERN SOYBEAN DISEASE WORKERS 1999 TREASURY REPORT

Operational

Planters Bank, Hawkinsville, GA

Receipt Summary	
Interest on Operational Account	\$ 93.74
1999 Meeting Registration Receipts	\$ 7 10.00
1999 Soybean Disease Atlas Sales	\$ 0.00
Total Receipts	\$ 803.74

Disbursement Summary	
Printing Fees	\$ 732.69
Postage Fees	\$ 112.10
1999 Annual Meeting Costs	\$ 227.74
SSDW Association Awards	\$ 262.46
Bank Account Fees(12/31/98 - 12/31/99)	\$ 60.00
Total Disbursements	\$ 1394.99

SSDW Assets - December 31	, 1999
Beginning Balance - 12/31/98	\$ 4778.94
Receipts	\$ 803.74 +
Disbursements	\$ 1394.99 -
Net Assets - 12/31/99	\$ 4778.94
Balance of Operational Account	\$ 4187.69



BEAN POD MOTTLE: A SOYBEAN DISEASE ON THE RISE IN THE NEW MILLENNIUM

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Bean pod mottle virus (BPMV) is widespread in many of the soybean growing areas in the southeastern United States. Increased incidence of BPMV has recently been observed in several major soybean growing regions in some central and southeastern states. Soybean yield losses of 10 - 55% have been reported as a consequence of BPMV infection. The primary sources of BPMV inoculum in soybean fields early in the season have not been critically studied. We examined the roles of overwintering beetles and seeds from infected soybean plants as possible primary sources of BPMV inoculum. None of the virus-containing naturally overwintered beetles transmitted the virus to healthy soybeans. Likewise, beetles exposed to artificial overwintering conditions showed little or no transmission of BPMV. Overwintered beetles regained the ability to transmit virus following acquisition feeding on infected plants. Regurgitants from overwintered beetles were infectious by mechanical inoculation and viral RNAs extracted from such regurgitants showed no apparent changes in their integrity. Limited proteolysis of the large capsid protein, however, was detected and is believed to be related to the loss of beetle transmissibility.

Evidence to date from seedling grow-out and ELISA tests of seeds collected from fields with high BPMV incidence failed to demonstrate seed transmission of BPMV. Northern hybridization analysis of RNA from several BPMV isolates collected from soybean fields in three states indicated the occurrence of at least two distinct subgroups of BPMV strains.

Disease management through genetic resistance is not possible at present because no soybean cultivars with resistance to BPMV are commercially available. A limited number of transgenic soybean lines that express the precursor to the two coat proteins (CP-P) have been generated. Two of these lines showed delayed onset of symptoms and produced milder symptoms than those induced on non-transformed soybeans. We have previously shown that the expressed CP-P did not assemble into virus-like particles (VLPs) since assembly requires the presence of the individual CP subunits. It is believed that aggregation of CP subunits and assembly into VLPs is a prerequisite for displaying efficient CP-mediated virus resistance. Plans are underway to generate transgenic plants that express the individual CP subunits that can be assembled into VLPs.

RESPONSE OF SELECTED MID-SOUTH SOYBEAN VARIETIES TO THE RENIFORM NEMATODE

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Twenty-seven mid-south soybean varieties in maturity groups (MG) IV through VI were evaluated for resistance and yield response to the reniform nematode (*Rotylenchulus reniformis*). Included in the study were eleven MG IV, thirteen MG V and three MG VI varieties.

Reniform resistance reactions were evaluated in the greenhouse. Each soybean variety was inoculated with 2,000 juvenile and vermiform adult reniform nematodes and allowed to grow for 60 days in the greenhouse. Resistance for each cultivar was calculated by dividing the number of eggs and vermiform stages of reniform nematodes at harvest by the number of nematodes used as inoculum. A product less than one indicated that cultivar was resistant to the reniform population used in this test. Field evaluations were conducted in a field located at Inverness, Mississippi which was naturally infested with the reniform nematode. Each variety was planted with and without the nematicide Temik 15G at 5.0 lb formulated material per acre.

Resistance to the reniform nematode was identified in five MG V varieties. These included Asgrow 5602, Delta and Pine Land 5354, 5644 RR, 5806 R and Hornbeck 5770. One variety Asgrow 6101, was determined to be resistant in MG VI. None of the MG IV varieties included in these tests showed resistance to this isolate of the reniform nematode.

In the field evaluations, each soybean variety varied in their response to the Temik 15G application. Nine varieties did not result in an increase in yields with the use of the nematicide. Soybean yields in MG IV ranged from 39.3 to 52.3 bu/acre from Asgrow 4601 and Delta and Pine Land 4909, respectively. Temik 15G improved yields in 10 varieties ranging from 2.9 to 12.2 bu/acre over the untreated plants. In MG V, yields ranged from 41.2 to 52.8 bu/acre from Terral-Norris TV5666 RR and Pioneer Brand 95B33, respectively. Yields were improved in six of the varieties ranging from as little as 0.6 to 8.0 bu/acre over the untreated MG V plants. Maturity group VI yields were 38.7, 43.2, and 51.1 bu/acre for Hornbeck 6600, Pioneer Brand 96B01, and Asgrow 6101, respectively. Pioneer Brand 96B01 did not respond to the application of Temik 15G, however, soybean yields were improved 3.8 and 6.3 bu/acre from Asgrow 6101 and Hornbeck 6600, respectively.

FIELD RESPONSE OF SOYBEAN CULTIVARS TO THE RENIFORM NEMATODE ROTYLENCHULUS RENIFORMIS

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Twenty-eight soybean cultivars (*Glycine max*) were evaluated for resistance to *Rotylenchulus* reniformis in a field in central Alabama heavily infested with the nematode (>400/100cm³ soil). This field had been under continuous cotton production for the preceding ten years. Each cultivar was planted in plots that were 2-rows (30 inch spacing) wide and 25 feet long. There were four replications per cultivar and plots were arranged in a randomized complete block design. The selected cultivars covered maturity groups ranging from 5 to 8, and a wide range of known resistance and susceptibility levels to root-knot (*Meloidogyne* spp.) nematodes and SCN (*Heterodera glycines*). No cultivar tested showed high resistance to *R. reniformis*. Three cultivars (Motte, Stonewall, and Boggs) showed moderate resistance (70-100 nematodes/100cm³ soil). All other cultivars tested showed little or no resistance (250-800 nematodes/100cm³). Very little correlation could be determined between nematode populations and yield response.

APPROACHES TO RACE DETERMINATION IN SOYBEAN CYST NEMATODE

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The current method for race determination in soybean cyst nematode is based on the development of females and cysts on soybean roots. With this approach, the female count is more of a developmental index than a reproductive index unless one assumes equal reproductive rates of all females across all differentials. The objectives of this study were to compare the present race identification scheme based on female and cyst counts with the same scheme based on egg and juvenile counts and to examine the effect of temperature on soybean cyst nematode race determination.

Race designations for soybean cyst nematode race 3 and 4 populations were examined at 20, 27, and 30 °C in a water bath. For each temperature and race, the four standard soybean differential varieties (Peking, Pickett, PI 88788, PI 90763) and the susceptible control Hutcheson were planted in a completely randomized design with four replications. Trials were harvested 26 (30 °C) or 30 (20 and 27 °C) days after inoculating soybean seedlings with a suspension of 2,000 soybean cyst nematode eggs in 5 ml tap water. The number of females, eggs, and juveniles (hatched at 19 days) were recorded and an index based on each life stage was calculated.

At the lower temperature of 20 °C, results based on the three life stages evaluated were inconsistent suggesting that temperature influenced soybean cyst nematode race designations for these populations. Race determinations based on female, egg, and juvenile indices for the soybean cyst nematode race 3 and 4 populations were consistent and matched the original race designations at 27 and 30 °C. This shows that developmental and reproductive indices are consistent at these temperatures, therefore the current method of counting females is not a limitation of the race scheme. However, one could use a more "foolproof" egg counting method to determine race for soybean cyst nematode.

NEMATOLOGICAL SURVEY OF SELECTED SOYBEAN AND COTTON FIELDS IN ALABAMA

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A field survey was conducted in Baldwin County in southwest Alabama to assess the change in nematode population dynamics from a soybean dominant agro-economic setting to a cotton dominant agro-economic setting. The last field survey was done over 15 years ago in the south half of Baldwin County. When this first survey was conducted in the early 1980's the principal economic crop grown was soybean, followed by corn and potatoes. Today, the primary economic crop grown is cotton, followed by peanut, soybean, corn, potatoes and vegetables. This nematode survey was conducted, in part, to determine if there have been changes in nematode populations as the field crops have changed.

Results indicate that root-knot nematode (*Meloidogyne* spp.) is still dominant, while soybean cyst nematode (*Heterodera glycines*) populations have declined. The reniform nematode (*Rotylenchulus reniformis*) is now identified in south Baldwin county. With the shift to cotton as the primary money crop in Baldwin county, surveys will need to be conducted regularly to monitor possible increases in reniform nematode populations. Yield loss estimates can only be done after surveys determine the various nematode populations.

SOYBEAN MEAL-BASED COMPOSITIONS AS ORGANIC AMENDMENTS FOR CONTROL OF PLANT-PARASITIC NEMATODES

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A greenhouse experiment was conducted to assess the nematicidal activity of soybean meal [SBM] and sorghum meal [SGM] when used alone and in combination. Soil for the experiment was collected from a soybean field in southeast Alabama which was heavily infested with a mixture of root-knot nematodes [RKN] (*Meloidogyne incognita* + *M. arenaria*). The [SBM] was applied at rates of 0, 2.5 and 5.0 g/kg soil, and to each of these were added [SGM] at rates of 0, 2.0, 4.0, and 8.0 g/kg soil. Nematode populations were assessed in soil 10 days after treatment. 'Young' soybean was then planted and allowed to grow for 8 weeks, after which, nematode populations were assessed in soil and roots. Results from the 10-day post-treatment soil sampling showed that [SBM] when used alone at rates of 2.5 and 5.0 g/kg soil resulted in significant suppression of [RKN] as did [SGM] when used alone at rates of 4.0 and 8.0 g/kg soil. Combining [SBM] and [SGM] did little to further enhance the suppressive effect on [RKN] over the 10-day interval. This was in contrast to the experiment termination sampling in both soil and roots which did show a more synergistic effect in the combination formulas over the long-term. The experiment termination sampling showed in most instances that [SBM] + [SGM] was more effective in suppressing [RKN] than when either one was used alone.

CELL SELECTION APPROACH FOR GENERATING SOYBEAN WITH RESISTANCE TO MACROPHOMINA PHASEOLINA

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The soybean disease charcoal rot (CR), caused by the fungal pathogen *Macrophomina phaseolina*, is found in many soil types throughout Mississippi. As such, 100% of soybean seeds/seedlings can be inoculated/infected within 3-4 days after planting and *Macrophomina* can also persist on, and within, the seed coat after harvest. To date, no known genetic resistance to CR exists in soybeans, nor is fungicide control effective. Development of genetic resistance via a tissue culture cell selection procedure is being attempted.

Previously, researchers determined that at least one toxin, phaseolinone, was excreted by *Macrophomina*. Our initial soybean seed germination tests in water containing varying concentrations of cell-free, fungal culture filtrate (CF; fungal growth medium plus excreted substances) determined that CF could induce disease symptoms without the presence of the live fungus. Abnormal seedlings emerged displaying both stunted root and shoot growth. No seedling differences were noted when comparing CF that had been filter-sterilized (0.2 μ m filter; CF-F) or autoclaved (CF-A).

Tissue culture cell selection protocols were developed for the inclusion of CF-A (50%; v/v) into soybean shoot regeneration media. A previously-developed regeneration procedure utilizing seedling hypocotyl section explants was used. This approach relies on CF-A acting as a selection agent and mutagen to develop/create resistance with the mutated cells capable of growth and regeneration. Therefore, regenerants may contain resistance/tolerance to CR. They, and subsequent progeny, will be grown in inoculated greenhouse and field trials to determine actual levels of resistance. Initial varieties being screened are Tracey and Williams.

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IDENTIFICATION OF MOLECULAR MARKERS LINKED TO A NEW GENE CONFERRING RESISTANCE TO FROGEYE LEAF SPOT IN 'PEKING' SOYBEAN

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Frogeye leaf spot (FLS), caused by *Cercospora sojina* Hara, is a worldwide foliar disease of soybean (*Glycine max* L. Merr.) in tropical and subtropical soybean-growing areas, where warm and humid conditions often occur. It is capable of causing severe yield losses in the southeastern USA. The use of resistant cultivars is an environmentally friendly, economical and effective way to control the disease.

Screening soybean for resistance to FLS using artificial inoculation techniques is laborious and subject to environmental variability. Identification and characterization of resistance genes in soybean by molecular markers may play an important role in tagging and incorporating these desirable genes into improved cultivars. A recent study indicated that 'Peking' is resistant to most isolates of *C. sojina*. Further study indicated that the gene in 'Peking' is non-allelic to Rcs_3 , which is resistant to all known races. The objective of this study was to identify DNA markers associated with this gene.

A total of 116 F2 individuals derived from the cross Peking (resistant) Ψ Lee (susceptible) were tested using amplified fragment length polymorphism (AFLP) and simple sequence repeat (SSR) markers. Sixty-four AFLP primer combinations were used to screen the two parents, 30 were found polymorphic between the two parents. One hundred eleven AFLP markers were found by screening the 30 AFLP primer combinations within the F2 population. Among these markers, one was found highly correlated with the resistance gene in Peking. In order to locate this gene in the soybean genome, several SSR markers were chosen based on disease resistance gene clusters of soybean linkage groups to screen the F2 population. One SSR marker was found linked to the gene. This will provide soybean breeders an opportunity to use these markers for marker assisted selection and pyramiding the resistance gene for FLS resistance in soybean.

SOIL PHYSICAL, CHEMICAL, AND BIOLOGICAL PROPERTIES ASSOCIATED WITH SUDDEN DEATH SYNDROME IN SOUTHERN ILLINOIS

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The interrelationship between soil chemical, physical, and biological factors and sudden death syndrome (SDS) was evaluated in a 3.5-ha field plot and in transects at multiple sites in southern Illinois. Sites were selected based on a history of prior SDS outbreaks. The field plot was established in DeSoto, IL and consisted of an area measuring 120 m x 130 m. The plot was subdivided into 130 subplots each 9 m x 10 m. The entire plot was managed according to traditional no-till management recommendations. In each of the subplots, soil samples were collected to determine bulk density, porosity, available soil moisture, and nutrient status. Disease expression was measured by rating SDS incidence and severity at R5, and yield was collected at maturity. In addition, *Fusarium solani* f. sp. glycines (FSG) levels in the soil and roots as well as the initial and final population densities of *Heterodera glycines* (SCN) were determined.

Two of the three transects were established in an irrigated field in Carmi, IL. A third transect was established in a non-irrigated rain fed field. Each of the three transects originated in an area of low disease expression and continued through areas of more severe disease. Every 1.5 meters, soil and plant samples were collected to determine the previously mentioned soil physical, chemical, and biological properties along the transect.

In the field plot, root infection by FSG increased and final SCN population densities decreased as bulk density of the soil increased. Initial and final SCN population densities correlated positively with higher SDS severity. Soybean yield correlated inversely with SDS severity and with initial and final SCN population densities.

For transects in the irrigated field, as bulk density increased, soil conductivity, disease severity, and CFU / g of root increased resulting in yield loss. Soybean cyst nematode and spiral nematode (*Helicotylenchus* spp.) population densities were lower as bulk density increased and did not correlate with yield loss.

RESPONSE OF SOYBEAN SUDDEN DEATH SYNDROME TO AMELIORATION OF THE SOIL PHYSICAL ENVIRONMENT

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Sudden death syndrome (SDS) is caused by the soilborne pathogen *Fusarium solani* f. sp. *glycines* (FSG). Previous research conducted at Southern Illinois University indicated that soil compaction was directly related to disease incidence and severity. Therefore the purpose of this project was to determine whether SDS could be suppressed by decreasing soil compaction. Anecdotal observations indicate that disease symptoms are more severe when cool, wet conditions prevail in the early part of the growing season. Our hypothesis was that increasing porosity would allow soils to drain more rapidly thus providing a dryer root zone, which would hinder root infection by FSG and subsequently reduce development of foliar symptoms.

A field was selected in Jackson County IL where previous research focused on soil physical properties and their relationships to SDS. Within this field, a 120 m x 120 m plot was established to evaluate the relationship between soil variables and SDS. Across the field, strips (9m wide) tilled to a depth of 41 to 46 cm were alternated with no-till strips. Each strip was divided into 10 subplots, each 9m x 10m. Within these plots, we measured soil physical and biological properties as well as soil moisture at field capacity. Physical properties included bulk density and porosity, whereas biological properties included initial and final populations of soybean cyst nematode (*Heterodera glycines*) as well as soybean root colonization by FSG. Plots were rated at R5 for SDS incidence and severity and yields were measured at maturity.

In contrast to no-till plots, tilled plots showed reduced bulk density and porosity as well as lower soil moisture at field capacity. Final populations of SCN were greater in tilled than in no-tilled plots. This likely is due to lower soil moisture, which can enhance nematode reproduction. Across tillage treatments, soil bulk density correlated directly with root colonization by FSG, which resulted in greater SDS disease levels. Soil moisture was correlated directly with bulk density, root colonization by FSG, and SDS disease level, and correlated inversely with soil porosity.

DIFFERENTIAL ENZYME ACTIVITY IN TWO SOYBEAN CULTIVARS RESISTANT AND SUSCEPTIBLE TO SUDDEN DEATH SYNDROME

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Soybean sudden death syndrome (SDS), caused by the fungus *Fusarium solani* f. sp. *glycines*, has become an increasing problem in Illinois and most soybean growing states. No soybean cultivars have been found to be completely resistant although partial resistance has been observed in some cultivars. Mechanisms of resistance are not fully understood. Studying enzymes related to disease resistance will add to better understanding of resistance mechanisms. The objective of this study was to identify differences in activities of selected enzymes which may be involved in SDS resistance.

Soybean varieties Essex (susceptible) and Forrest (resistant) were planted in a peat-lite potting medium and placed on greenhouse benches. After 17 days, plants were transplanted to a 1:1 (v:v) sand:soil potting medium lacking or containing $4x10^4$ macroconidia/gram soil of the fungus. Plants were sampled 1, 3 and 7 after inoculation. Roots were excised from plants, crushed in liquid nitrogen and stored at -30 C until assayed. Enzymes were extracted and their activities were measured spectrophotometrically using standard protocols. Enzymes tested were phenylalanine ammonia lyase, polyphenol oxidase, hydroperoxidase and glucose-6-phosphate dehydrogenase.

Phenylalanine ammonia lyase levels increased over time and variety differences were observed. Phenylalanine ammonia lyase levels in Forrest were consistently higher than those in Essex. Polyphenol oxidase levels one day after inoculation were higher in Essex compared to Forrest. However, polyphenol oxidase levels in Forrest were higher than in Essex at 3 and 7 days after inoculation. Glucose-6-phosphate dehydrogenase levels were consistently higher in Essex compared to Forrest.

EVALUATION OF AZOXYSTROBIN ON SOYBEAN DISEASE, YIELD, AND SEED QUALITY

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Two fungicide tests were conducted on soybeans at the AAES Monroeville Experiment Fields in Alabama. Tests were planted May 17, 1999 at a rate of 10 seeds per ft of row in a RCBD with five replications. Plots consisted of four rows 25 ft long with a 36 in. row spacing. In test 1, fungicides Azoxystrobin and Iprodione were applied as in-furrow sprays, Metalaxyl plus LS022 and Carboxin plus Thiram plus Metalaxyl were applied as seed treatments, and Azoxystrobin was also applied as a foliar spray at the V2. In test 2, fungicides Azoxystrobin and Benomyl were applied as foliar broadcast sprays at R3 and R5. All in-furrow fungicide and foliar broadcast spray treatments were applied using a CO_2 charged system with flat tip 8001 nozzles calibrated to 10 GPA at 30 PSI. Foliar broadcast sprays were applied at 23, 77 and 94 days after planting for the V2, R3 and R5. Stand counts and skip index were determined 23 days after planting. Stem canker and foliar disease ratings were made at 77, 94, and 107 days after planting. The yield was measured and recorded. Harvested soybean seeds were aseptically plated on APDA to identify the seed borne pathogens.

In Test 1, the seedling stands at 23 days after planting ranged from 114 to 182 plants for the LS 275 and the Iprodione treatments, respectively. Emergence was reduced by Azoxystrobin applied in-furrow as compared to the control. The control, Iprodione and Azoxystrobin applied at V2 produced a more uniform stand than all other treatments. No stem canker was observed. Yields ranged from 25.44 to 20.33 bushels per acre for the Azoxystrobin (V2 application) and control, respectively. Fifty three percent to 61% of the soybean seed harvested were colonized by fungi. *Phomopsis longicolla* was isolated significantly more frequently from the LS 022 plus Metalaxyl seed treatment than the control and LS 275 seed treatment. LS 275 seed treatment significantly reduced the number of *Phomopsis longicolla* and *Macrophomina phaseolina* isolated recovered from soybean seed. Azoxystrobin applied only as a foliar spray significantly reduced the presence of *Alternaria alternata* on soybean seed.

In Test 2, foliar diseases. Cercospera blight, Frogeye leaf spot and Rhizoctonia foliar blight were not observed at any date sampled. Yields ranged from 17.39 to 21.95 bushels per acre for the Azoxystrobin 0.15 lbai/a R5 application and Azoxystrobin 0.15 lbai/a R3 application respectively. The total number of fungi isolated from the harvested soybean seed was reduced by the Azoxystrobin foliar sprays applied at R5 compared to Azoxystrobin 0.15 lbai/a applied at R3. *Phomopsis longicolla* was isolated most frequently from Azoxystrobin 0.15 lbai/a applied at R3 compared with all the Azoxystrobin foliar sprays applied at R5 and to the Benomyl applied at R3 was increased by Benomyl applied at R3.

SOYBEAN DISEASE LOSS ESTIMATES FOR THE UNITED STATES FROM 1996-1998

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Soybean disease loss estimates were compiled for the 1996-1998 harvested crop from all soybean producing states in the United States. Scientists from each state provided estimates of yield losses due to diseases. Methods used to estimate soybean disease losses were field surveys, information from field workers and university extension staff, and research plot data. Total yield losses caused by *Heterodera glycines*, soybean cyst nematode, in the United States were greater than those caused by any other disease (Table 1). The reduction of U.S. soybean yields due to SCN was 5,819,300 metric tons (t) in 1996, 5,958,800 t in 1997, and 7,593,300 t in 1998. Next in order of yield reduction caused over 3 years were Phytophthora root and stem rot, seedling diseases, Sclerotinia stem rot, brown stem rot and charcoal rot. Yield loss estimates due to particular diseases to diseases in the United States were 10.9 million t in 1996, 11.9 million t in 1997, and 14.0 million t in 1998.

1996, 1997 and 1998 ^a .	·			
Diseases	1996	1997	1998	Total
Anthracnose	105,500	244900	188800	539200
Brown spot	88,300	. 124100	167000	379400
Brown stem rot	837,500	653300	369500	1860300
Charcoal rot	336,000	452400	1036700	1825100
Phomopsis seed rot	93,800	43200	243900	380900
Phytophthora rot	1,101,600	1459100	1148600	3709300
Pod and stem blight	139,500	346700	143400	629600
Purple stain	217,300	100600	112400	430300
Root-knot nematodes & Other nematodes	161,100	134300	128000	423400
Sclerotinia stem rot	668,800	957300	508900	2135000
Seedling diseases	852,900	666900	776600	2296400
Soybean cyst nematode	5,819,300	5958800	7593300	19371400
Sudden death syndrome	100,800	303700	900500	1305000
Virus	65,200	58600	250600	374400

Table 1. Estimated reduction of U. S. soybean yields (metric tons) due to major diseases during 1996, 1997 and 1998^a.

a Rounding errors may occur.