Sources of Resistance to Gray Leaf Spot of Corn

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ABSTRACT

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In 1989, 1,396 corn (*Zea mays*) inbreds were evaluated for resistance to gray leaf spot at Urbana, Illinois, after inoculation with *Cercospora zeae-maydis*. Sixty-two inbreds were selected on the basis of low disease rating, restricted lesion type, and maturity most useful for breeding programs in the midwestern United States. In 1990, selected inbreds and F_1 crosses between selected inbreds and the susceptible inbred FR1141, with the hybrid FR1141 × LH51 as a susceptible check, were evaluated. Crosses of FR1141 with inbreds CI30, CI88A, DS:74:1071, H110, Mo18W, Mo22, and TEX 1 had ratings that were nearly as good as or better than their resistant parent and significantly better than the susceptible hybrid FR1141 × LH51. Many sources of resistance to gray leaf spot are available to improve inbreds used in corn hybrids in the midwestern United States.

Gray leaf spot, a foliar disease caused by *Cercospora zeae-maydis* Tehon & E.Y. Daniels, was first described on corn (*Zea mays* L.) in Illinois in 1925 (17). There were a few reports of the disease in the 1940s (1,8,11,14), 1950s (14), and 1960s (9), but the disease was relatively rare. Beginning in the 1970s the incidence and severity of gray leaf spot were higher because of the increased use of reduced (conservation) tillage (6,10,12,15). *C. zeae-maydis* overwinters in crop debris, and its survival is reduced when crop debris is buried (2,13). Because reduced tillage practices have both economic and environmental benefits that warrant their continued practice, host resistance is the best option for control.

Although sources of resistance have been identified and studied under conditions of natural infection (3–7,16,18,19) and inoculation (7), additional sources of resistance would be useful. Most of the previously identified sources of resistance were found to have genes for resistance that were inherited in an additive manner (3–5,7,18,19). Two studies have identified sources of resistance that may have dominant genes for resistance (4,5). The objective of this research was to identify additional sources of resistance to gray leaf spot, with an emphasis on sources that contribute resistance to the susceptible inbred FR1141, which is a so-called B73-type inbred and is related to many inbreds used widely in the midwestern United States. Sources of resistance that have genes for resistance not present in FR1141 could have value in breeding programs to improve resistance of B73 types.

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MATERIALS AND METHODS

Field plots. In 1989, 1,396 inbreds were evaluated for resistance to gray leaf spot at the Agronomy/Plant Pathology South Farm, Urbana, Illinois. The inbreds included domestic, foreign, and unreleased lines from the Department of Plant Pathology inbred collection. Two replications of inbreds arranged in a randomized complete block treatment design were planted on 17 and 18 May in rows 0.76 m long spaced 0.76 m apart with six plants per row. Sixty-two inbreds were selected, with an emphasis on high levels of disease resistance, resistant lesion types, and maturity that could be useful in breeding programs in the midwestern United States (Table 1). In 1990, selected inbreds and F_1 crosses between selected inbreds and the susceptible inbred FR1141 were evaluated. Inbreds FR1141 and LH51 and the hybrid FR1141 × LH51 were included as susceptible controls. Three replicates of inbreds and hybrids arranged in a randomized complete block treatment design were planted 24 May in rows 4.2 m long spaced 0.76 m apart. Rows were planted with 24 kernels and thinned to 12 plants per row.

Inoculation and rating. Plants were inoculated five times each year between mid-June and early July with a propagule suspension of *C. zeae-maydis*. Inoculum was produced from 7- to 10-day-old cultures grown at room temperature with approximately 12 hr light on V8 agar (350 ml of V8 vegetable juice, 3 gm of $CaCO_3$, 20 gm of agar, and 650 ml of deionized water per liter) amended with 0.25 gm/L of streptomycin sulfate. Cultures from five isolates in 1989 and seven in 1990 were blended together in sterile deionized water, and 2 ml of the blend was spread on the medium surface to produce inoculum. Inoculum was prepared by blending cultures with deionized water, diluting the resulting suspension with water, and adding approximately 0.2 ml/L of Tween 20. Approximately 20 and 46 cultures, from 100-mm petri dishes, were used to make approximately 20 and 53 L of inoculum per 1,000 plants for each inoculation in 1989 and 1990, respectively. In 1989, the spore concentration of inoculum was approximately 6.5×10^3 conidia per milliliter, and a high-boy sprayer was used to apply the inoculum. In 1990, the spore concentration of inoculum was approximately 5.4×10^3 conidia per milliliter, and a hand-held sprayer was used to apply the inoculum.

An overhead mist irrigation system was used, as needed, to maintain a high level of moisture in the leaf canopy to promote disease development. The irrigation system was constructed of 10.2-cm-diameter pipe placed along one edge of the field with valves spaced 12.2 m apart leading to 7.7-cm-diameter pipe extending the length of the field between rows with riser outlets spaced 9.1 m apart. Risers were approximately 2.5 m tall and equipped with low-output sprinklers (Rain Bird model 14VH with 1.6-mm nozzles, Rain Bird Sprinkler Mfg. Corp., Glendora, CA). The output of each nozzle was 3.3 L/min at water pressure of 4.2 kg/cm^2. The output of the total system was about 1.8×10^4 L/hr/ha.

In both years, disease severity was rated on a row basis between mid-August and early September on a 0.5–5 scale in 0.5-increments similar to those used in previous studies (3,5,6,16,18,19), where 0.5 = a few restricted lesions on lower leaves, 1.0 = several scattered lesions on lower leaves, 2.0 = several lesions on lower leaves with a few on middle leaves, 3.0 = several lesions on middle leaves with abundant lesions on lower leaves, 4.0 = several lesions on upper leaves with abundant lesions on middle and lower leaves, and 5.0 = abundant lesions on all leaves. Lesion types on plants in each plot were also recorded as: A = small chlorotic lesion, B = small restricted necrotic lesion with chlorotic halo, C = small rectangular lesion, and D = large rectangular lesion. In 1990, disease severity also was rated as the percent leaf area affected. Severity ratings on the 0.5–5 or the percentage scale were analyzed with Statistical Analysis System software (SAS Institute, Cary, NC) using the analysis of variance procedure (PROC ANOVA). Genotypes were compared by a least significant difference test.

RESULTS AND DISCUSSION

Genotypes differed significantly in both years. In 1989, mean ratings of the 1,396 inbreds ranged from 1.0 to 4.5 (Fig. 1). In 343 inbreds, scores were significantly lower than those of the susceptible inbred FR1141. Sixty-two inbreds were selected for further study on the basis of disease severity, lesion type, and maturity (Table 1). Inbreds Ky128, Mo16W, MP311, and Va3a had mean ratings below 1.49 but were not selected because of late maturity (Table 1). Most of the selected inbreds had a mixture of B and D lesion types. Some inbreds had A-type lesions prior to pollination.

Gray leaf spot was more severe in 1990 than in 1989, possibly because of the increased quantity of inoculum, better coverage of the plants with the inoculum, and frequent natural dews. The mean ratings for the 0.5–5 scale and the percentage scale ranged from an average of 1.0–4.5 and 6.3–57.5, respectively. Many of the selected inbred lines had a mixture of lesion types C and D in 1990, as compared with types B and D in 1989. The mixture of lesion types on the selected inbreds appeared to be due to slow expansion of lesions. Earlier ratings may have been useful in detecting differences between lesion types. In 1990, 33 inbreds had mean scores on either rating scale that were significantly lower than those of the susceptible inbred FR1141. Some inbreds that had much higher ratings in 1990 than in 1989 apparently were escapes in 1989.

Seven hybrids had mean ratings that were significantly lower than LH51 × FR1141 on the 0.5–5 scale and 32 had significantly lower ratings on the percentage scale. Many of the inbreds identified as having resistance did not produce resistant F_1 crosses with FR1141. A similar situation occurred with the F_1 cross between LH51 and FR1141 where the F_1 was as susceptible as the inbred FR1141. On both rating scales, crosses of FR1141 and inbreds CI30, CI88A, DS:74:1071, H110, Mo18W, Mo22, and TEX 1 had ratings that were significantly lower than those of FR1141 × LH51 and similar to those of the resistant parent. We interpret this to indicate that these inbreds could have genes for resistance not present in FR1141 and that they may be of value as sources of resistance. In general, lesion types of the selected inbreds were not expressed in the hybrids that had predominantly type D lesions. In 1989, inbreds CI30, CI88A, DS:74:1071, H110, Mo18W, Mo22, and TEX 1 had lesion types B and D; Mo18W also had lesion type C. In 1990, these inbreds had lesion types C and D; B and C; B; C; B; B and C; and B and C, respectively. The F_1 crosses between these inbreds and FR1141 had lesion types B and D; D; B and D; B; D; D; and D, respectively. Inbreds B68, B68HT, CI64, K054W, Mo18W, NC250, NC290, Pa875,

Table 1. Mean gray leaf spot rating of selected maize inbreds in 1989 and 1990 and F_1 crosses with the susceptible inbred FR1141 in 1990[a]

	Maize inbreds			F_1 crosses with FR1141	
	0.5–5.0 scale[b]		Percent leaf area affected[c]	0.5–5.0 scale	Percent leaf area affected
Inbred	1989	1990	1990	1990	1990
061	1.75	2.00	23.3	...[d]	...[d]
198	2.25	2.00	13.7	3.00	25.0
25744	2.50	3.33	31.7	3.67	35.0
33-16	2.25	2.17	20.0	3.50	28.3
38-11	2.25	2.67	23.3	3.17	28.3
A622N	3.00	2.50	20.0	3.25	32.5
A669	3.00	3.17	38.3	3.33	36.7
AR214	2.75	2.50	25.0	3.00	23.3
B37HTN	2.50	2.00	14.3	3.00	26.7
B68	3.25	...[e]	...	...	...
B68HT	3.00	...[e]	...	...	...
B85	2.25	3.33	35.0	3.83	46.7
CG12	2.00	3.50	43.3	4.50	57.5
CI7	1.75	2.67	28.3	2.67	20.7
CI30	2.25	2.83	20.0	2.33	15.3
CI38	2.25	2.50	11.7	3.17	25.0
CI41	2.00	2.50	25.0	2.83	20.0
CI43	2.00	2.83	25.0	3.00	26.7
CI64	2.00	2.17	23.3	2.67	26.7
CI66	2.50	2.00	18.3	2.67	21.0
CI88A	2.25	1.83	7.0	2.17	25.0
DS:74:1004	2.00	2.83	30.0	3.33	30.0
DS:74:1071	2.00	2.50	30.0	2.50	26.7
E2558W	2.25	2.17	20.0	3.00	26.7
FR49	1.88	3.00	32.5	3.50	38.3
FR802W	2.75	2.83	21.7	3.33	30.0
H49	3.12	3.50	32.5	3.83	50.0
H50	3.25	2.83	23.3	3.33	30.0
H84	2.75	3.50	38.3	3.67	41.7
H99	2.75	3.83	38.3	3.33	31.7
H110	2.25	2.83	25.0	2.33	23.3
IF192	2.75	3.33	30.0	2.83	28.3
K4KY36-11	2.25	1.83	16.0	3.00	30.0
K64	2.25	2.83	21.7	2.67	21.7
Ky128	1.00	...[f]	...	...	...
LH5	2.75	3.50	36.7	3.67	35.0
LH55	3.00	3.17	30.0	3.33	35.0
LS78	2.75	2.50	15.0	3.00	25.0
M-16	2.50	2.67	28.3	3.50	31.7
MBS 61	3.00	3.17	30.0	3.33	35.0
Mo4	1.25	3.17	31.7	2.67	24.0
Mo10	2.00	2.67	26.7	2.83	26.7
Mo16W	1.00	...[f]	...	...	...
Mo18W	2.00	1.33	10.0	2.33	21.7
Mo21R	1.62	2.17	12.7	3.25	25.0
Mo22	2.00	1.83	14.7	2.50	25.0
MOL3	2.75	3.17	28.3	3.33	35.0
MP305	2.50	2.50	22.5	3.17	26.7
MP311	1.00	...[f]	...	...	...
NC18	1.25	2.00	18.3	2.83	23.3
NC250	1.75	2.50	23.3	3.17	33.3
Oh507	2.75	2.50	20.7	3.00	25.0
(Oh43 × 8brbr)	2.25	1.17	6.3	2.83	23.3
Pa887P	3.00	...[e]	...	...	...

(*continued on next page*)

[a] Means based on six plants per plot in 1989 and 12 plants per plot in 1990 averaged over two and three replicates, respectively. Ratings made between mid-August and early September.
[b] Where 0.5 = a few restricted lesions on lower leaves, 1.0 = several scattered lesions on lower leaves, 2.0 = several lesions on lower leaves with a few on middle leaves, 3.0 = several lesions on middle leaves with abundant lesions on lower leaves, 4.0 = several lesions on upper leaves with abundant lesions on middle and lower leaves, and 5.0 = abundant lesions on all leaves.
[c] Estimate of the percent leaf area affected on a whole plant basis.
[d] Attempt at F_1 cross not successful.
[e] Inbred not evaluated further in our study but found to have resistance in other research.
[f] Inbred highly resistant but not selected for our study because of late maturity.
[g] Plants severely damaged by animals and not rated.
[h] Test mean of 1,396 inbreds.
[i] Mean of inbreds and hybrids evaluated in 1990.
[j] Fisher's least significant difference based on 1,396 inbreds in 1989 and 126 inbreds and hybrids in 1990.

Table 1. *(continued from preceding page)*

	Maize inbreds			F_1 crosses with FR1141	
	0.5–5.0 scale[b]		Percent leaf area affected[c]	0.5–5.0 scale	Percent leaf area affected
Inbred	1989	1990	1990	1990	1990
PB78	2.00	1.83	9.3	3.00	20.0
R193	3.00	2.50	26.7	3.17	31.7
R218A	2.50	3.17	30.0	3.17	35.0
RCI64HT1A	2.25	1.00	...[g]	3.00	26.7
SC301E	2.25	3.33	33.3	3.00	38.3
T101	2.50	3.00	25.0	2.83	28.3
T212	3.25	...[e]	...	...	...
T222	3.00	2.00	23.3	2.67	21.7
T242	2.50	2.00	30.0	2.83	25.0
TEX 1	2.75	2.00	9.0	2.50	21.7
TEX 2	2.00	1.33	16.0	2.83	25.0
Va3a	1.00	...[f]	...	...	...
Va10	2.50	2.25	20.0	3.17	31.7
Va17	4.00	...[e]	...	...	...
Va36	3.00	2.33	21.3	3.00	26.7
Va59	3.25	...[e]	...	...	...
(Va26 × HA494brbr)	2.25	2.33	20.0	3.33	31.7
W153R	2.75	3.17	33.3	3.50	40.0
FR1141	4.00	3.40	35.9		
LH51	3.00	2.83	15.0		
FR1141 × LH51				3.33	38.3
Test mean	3.49[h]	2.82[i]	26.6[i]	2.82	26.6
SD	0.48	0.48	7.09	0.48	7.09
CV	13.79	16.94	26.6	16.94	26.6
FLSD ($P = 0.05$)[j]	0.94	0.77	11.4	0.77	11.4

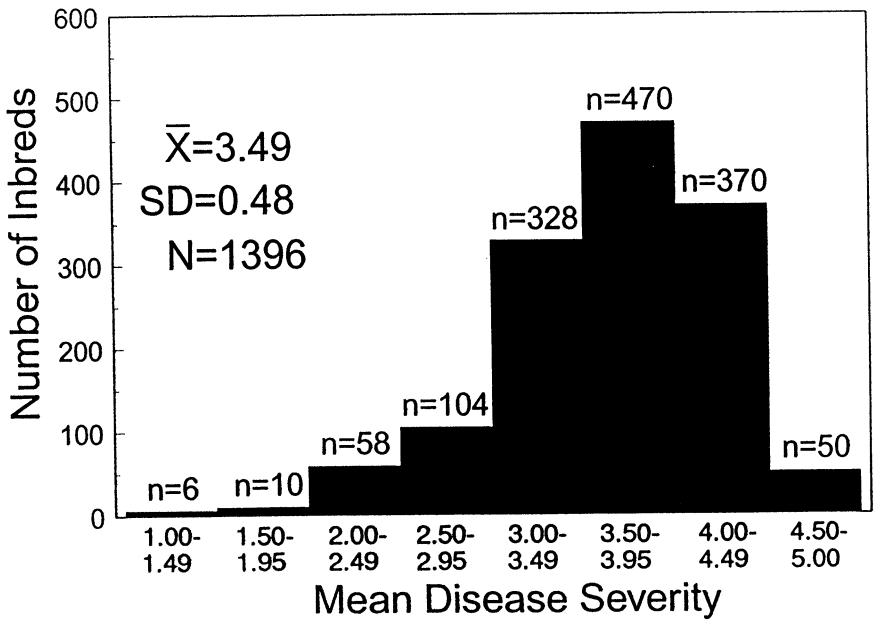

Fig. 1. Mean disease severity of gray leaf spot for 1,396 inbreds in two replications during 1989 rated between mid-August and early September on a 0.5–5.0 scale where 0.5 = a few restricted lesions on lower leaves, 1.0 = several scattered lesions on lower leaves, 2.0 = several lesions on lower leaves with a few on middle leaves, 3.0 = several lesions on middle leaves with abundant lesions on lower leaves, 4.0 = several lesions on upper leaves with abundant lesions on middle and lower leaves, and 5.0 = abundant lesions on all leaves.

Pa887P, S0507W, T212, T222, Va14, Va17, and Va59 have been identified as having resistance in previous studies (3–7,17,18). Inbreds K054W, NC290, Pa875, SO507W, and Va14 were not tested in our study, and inbreds B68, B68HT, T212, Va17, and Va59 were eliminated from further study on the basis of results in 1989. Inbreds 061, 198, B37HTN, DS:74:1004, DS:74:1071, (Oh43 × 8brbr), TEX 1, TEX 2, and (Va26 × HA494brbr) were developed at the University of Illinois. The most resistant inbreds, inbreds that produced the most resistant hybrids, and inbreds that appeared to be unique sources of resistance based on lesion type were selected for further study.

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