Diallel Cross Analysis of Resistance to Gray Leaf Spot in Maize

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ABSTRACT

Gevers, H. O., Lake, J. K., and Hohls, T. 1994. Diallel cross analysis of resistance to gray leaf spot in maize. Plant Dis. 78:379-383.

Diallel cross analysis of resistance to gray leaf spot (caused by *Cercospora zeae-maydis*) in an inbred set of diverse heterotic types revealed marked differences in general and specific combining abilities. The former component was of greater relative importance in maize breeding material largely unselected for resistance to this disease, although nonadditive effects were recognized as playing a significant part in the resistance mechanism. Inbreds KO54W and SO507W, representing the F and M heterotic groups, respectively, exhibited the highest resistance and inbred SO713W (P heterotic group) the lowest resistance. Major dominant effects for resistance were associated with KO54W, which makes it particularly suitable for use in backcross recovery programs for gray leaf spot resistance.

Gray leaf spot of maize, caused by the fungus *Cercospora zeae-maydis* Tehon & E. Y. Daniels, has so far not been encountered naturally in the maize breeding program at Ukulinga Research Farm, University of Natal, Pietermaritzburg, Republic of South Africa. However, it has been positively identified in the past two seasons at nearby Cedara College, where a second program is conducted annually. This foliar maize disease has become markedly more prevalent and severe in recent seasons at Cedara, where humid, moist climatic conditions are ideal for its development. Other reports (D. C. Nowell, Pannar (Pty) Ltd., *personal communication*) suggest that the disease has been increasing in recent years in the Natal Midlands and even farther afield. In parts of Natal, gray leaf spot reached epidemic proportions in the 1991-1992 season, and its adverse effects on maize yields may be regarded as serious.

Increased incidence of gray leaf spot has been associated with reduced tillage practices, for instance in the United States (7–9), where yield losses of up to 40% may be expected in maize chemically unprotected against the disease (6). Other deleterious agronomic effects of this disease are increased lodging and reduced grain quality, notably under conditions of prolonged high humidity (2,9), which is characteristic of the Natal Midlands.

There may be several other reasons for the recent dramatic rise in the incidence and spread of gray leaf spot in addition to the effects of reduced tillage. For instance, any genetic vulnerability in locally used breeding material and commercial hybrids would enhance disease severity. This is aggravated in South Africa by the fact that since its inception in 1948, the Natal maize breeding program, which has released numerous widely used hybrids, inbreds, and other germ plasm in recent decades, has undertaken no direct or conscious selection for resistance to gray leaf spot. This same situation also applies to widely used maize breeding material from other countries, notably the United States, where serious attempts toward resistance breeding are of comparatively recent date (2,3,7,12).

In view of the seriousness and potential destructiveness of this "new" disease, it is now urgent to identify sources of resistance in local maize breeding material. We present here the first genetic study on the nature of inheritance of resistance to gray leaf spot in South African maize breeding material.

Accepted for publication 23 July 1993.

Table 1. Characteristics of 12 white modified *opaque-2* maize inbreds used in the diallel cross

Inbred line	Heterotic group	Origin[z]
FO215W	P	Natal Potchefstroom Pearl
SO713W	P	Natal Potchefstroom Pearl
RO452W	M	M37W/21A^2.Jellicorse
RO460W	M	M37W/21A^2.Jellicorse
RO465W	M	M37W/21A^2.Jellicorse
SO181W	M	M37W/21A^2.Jellicorse
SO507W	M	M37W/21A^2.Jellicorse
KO54W	F	Teko Yellow/F2834T
RO504W	F	Teko Yellow/F2834T
RO550W	F	Teko Yellow/F2834T
RO558W	F	Teko Yellow/F2834T
RO594W	F	Teko Yellow/F2834T

[z] After Gevers and Whythe (4).

Table 2. Gray leaf spot ratings[y] of crosses (above diagonal) and parents (on diagonal) and parental array means for a diallel cross of 12 maize inbreds

Inbred	FO215W	SO713W	RO460W	SO507W	RO465W	RO452W	SO181W	RO550W	KO54W	RO558W	RO594W	RO504W
FO215W	2.87[z]	3.20	2.67	2.53	2.68	3.00	2.39	3.33	1.94	3.34	2.81	3.01
SO713W		3.35[z]	2.66	2.73	3.20	2.69	2.92	2.93	1.81	3.27	2.72	3.00
RO460W			2.40[z]	1.73	2.52	3.08	1.93	4.00	1.60	3.00	3.54	2.86
SO507W				0.34[z]	1.28	1.73	1.54	2.46	0.60	2.80	2.86	2.34
RO465W					2.01[z]	2.19	1.47	3.93	1.99	2.91	3.61	2.80
RO452W						1.87[z]	1.53	3.39	0.99	3.00	3.47	2.55
SO181W							1.94[z]	2.73	1.35	2.94	2.99	2.87
RO550W								2.67[z]	1.01	2.52	3.01	3.07
KO54W									0.67[z]	0.46	1.33	1.00
RO558W										2.46[z]	2.98	2.68
RO594W											3.00[z]	3.21
RO504W												2.92[z]
Mean	2.81	2.83	2.62	2.06	2.60	2.51	2.24	2.94	1.28	2.72	2.96	2.67

[y] On a scale from 0 to 5.
[z] Inbreds.

Table 3. Analysis of variance of gray leaf spot ratings for a diallel cross of 12 maize inbreds

Source of variation	df	Sum of squares	Mean square	F value[x]
Replications	2	4.58	2.29	18.37
Blocks	24	18.30	0.76	6.11
Crosses	65	108.86	1.67	13.42
GCA[y]	11	79.62	7.24	57.99
SCA[z]	54	29.24	0.54	4.34
Error	106	13.23	0.12	
Total	197	144.97		

[x] All F values shown are significant at $P < 0.01$.
[y] General combining ability.
[z] Specific combining ability.

MATERIALS AND METHODS

A diallel cross of 12 heterotically divergent, white modified *opaque-2* maize inbred lines, highly selected for yield combining ability but unselected for reaction to gray leaf spot, was used in this study (Table 1). A 9 × 9 triple-lattice design (1) was used to accommodate the 66 single crosses (excluding reciprocals), 12 inbred parents, and three arbitrarily chosen commercial hybrids of unknown gray leaf spot resistance (HL1, PAN6549, and SR52) in a yield trial conducted at Cedara College in the 1991-1992 season.

The plots in this experiment with three replicates consisted of three rows of 20 competitive plants, spaced at a population density of 40,000 plants per hectare. The center row was used for collection of experimental data.

Five plants per plot were rated for gray leaf spot on a widely used scale described by Donahue et al (2): 0 = no symptoms; 1 = trace lesions below the ear; 2 = many lesions below the ear; 3 = large lesions below the ear and lesions on all leaves above the ear; 4 = large lesions on all leaves; and 5 = all leaves dead. Lesions were identified according to descriptions by McGee (9) and Shurtleff (10), augmented by local observations. To accommodate any variations and peculiarities of disease progression relating to stage of plant infection (in this case, natural infection) and prevailing weather conditions, the ratings were done twice. Plot means of the second rating, assumed to be at full disease development, were used for analysis.

The diallel analysis was performed with a standard combining ability model, whereby variation among crosses was partitioned into components for general combining ability (GCA) and specific combining ability (SCA). The model is thus analogous to Griffing's (5) method 4, model 1 analysis, as appropriate for fixed genotype effects in nonrandomly and highly selected parental inbred sets.

RESULTS AND DISCUSSION

The mean gray leaf spot ratings for crosses, parents, and parental arrays are presented in Table 2. The lowest and highest ratings were 0.46 and 4.00 for the crosses KO54W × RO558W and RO460W × RO550W, respectively, and 0.34 and 3.35 for the parents SO507W and SO713W, respectively. This represents a very satisfactory range of disease development within the scale of 0–5.

Inbreds from both the F and M heterotic groups (for instance, KO54W and SO507W, respectively) have relatively low parental and mean cross ratings, while members of these groups also featured prominently among the highest ratings (e.g., RO594W and RO460W, respectively) (Tables 1 and 2). Inbreds from the P heterotic group (SO713W and FO215W) also had relatively high disease ratings, which confirms previous observations at Cedara that this breeding material is generally susceptible to gray leaf spot.

The analysis of variance shows that the single crosses differ significantly in their disease ratings (Table 3). Both the GCA and SCA components of variance were also highly significant. This implies that although the GCA effects of inbreds were accurately reflected in their crosses, certain combinations of lines performed significantly better or worse than was expected on the basis of their GCA effects. The relative magnitude of the GCA and SCA variances confirms that the GCAs of lines accounted for the major portion of variation among crosses. However, as is evident from Table 4, both additive and nonadditive genetic effects are important in resistance to gray leaf spot. For example, in conformity with the observed resistance of inbreds KO54W and SO507W, the greatest GCA effects were associated with these parents, suggesting that they would be very useful sources of disease resistance. The negative sign associated with GCA effects for these two lines arose from the rating scale of 0–5, indicating lowest and highest disease ratings.

Although the inbreds KO54W, SO507W, and SO181W all had superior GCA effects for resistance to gray leaf spot (Table 4), least significant differences derived for the genetic variance components (*not shown*) indicated that only KO54W had a significantly greater GCA variance than the remaining lines. The GCA variances of SO507W and SO181W were not significantly different at the 0.01 significance level. The lines SO713W, FO215W, RO550W, and RO594W, which were susceptible, had relatively high positive GCA effects, indicating a low frequency of genes for resistance.

Although the GCA component of variance in this relatively unselected material was more important, the SCA component was numerically greater for inbreds RO465W, RO452W, SO181W, and RO558W, indicating greater deviations from additivity in some lines. However, the overriding importance of the additive component of variance suggests that active selection for resistance to gray leaf spot in this material, as well as use of the most resistant lines in crosses or in backcross programs, should be very successful. Similar conclusions were reached by Donahue et al (2) in a 14-line diallel cross analysis and by several other workers (3,7,12) in inheritance studies with inbred lines from U.S. maize breeding programs. Further, numerous corn belt inbreds, including widely used lines such as B73, were found to be susceptible to gray leaf spot (2,12).

Results obtained in this study are generally confirmed in Figures 1 and 2, which summarize the associations between parental and combining ability effects. The inbreds KO54W and SO507W, on the one hand, and SO713W and FO215W, on the other, represent the extremes in resistant and susceptible genotypes (Fig. 1). Figure 2 indicates that the F and M heterotic groups were represented in both the most resistant and the most susceptible crosses; the greatest specific resistance was recorded for the F × F group.

The breeding merit of this material is perhaps best illustrated in Table 5, which

Table 4. Specific combining ability (SCA) effects (above diagonal), general combining ability (GCA) effects, and GCA and SCA components of variance for a diallel cross of 12 maize inbreds[z]

Inbred	FO215W	SO713W	RO460W	SO507W	RO465W	RO452W	SO181W	RO550W	KO54W	RO558W	RO594W	RO504W
FO215W		0.04	−0.30	0.17	−0.15	0.12	−0.23	−0.01	0.47*	0.39*	−0.43*	−0.07
SO713W			−0.45*	0.28	0.28	0.05	0.24	−0.40*	0.38*	0.23	−0.65**	−0.01
RO460W				−0.56**	−0.34*	0.52**	−0.45*	0.84**	0.32	0.06	0.46*	−0.11
SO507W					−0.71**	−0.22	−0.09	0.01	−0.03	0.57**	0.33	0.26
RO465W						−0.53**	−0.72**	0.81**	0.72**	0.00	0.63**	0.01
RO452W							−0.72**	0.35*	−0.32	0.34*	0.48*	−0.06
SO181W								0.08	0.64**	0.55**	0.24	0.45*
RO550W									−0.54**	−0.70**	−0.34*	−0.10
KO54W										−0.93**	−0.39*	−0.33
RO558W											−0.40*	−0.11
RO594W												0.08
RO504W												
GCA	0.33**	0.34**	0.17*	−0.53**	0.04	0.01	−0.30**	0.45*	−1.36**	0.16*	0.47**	0.22**
σ^2_{GCA}	0.10	0.11	0.02	0.27	0.00	0.00	0.08	0.19	1.84	0.02	0.21	0.04
σ^2_{SCA}	0.00	0.01	0.11	0.04	0.20	0.06	0.13	0.15	0.18	0.14	0.10	0.00

[z] One and two asterisks indicate that the effect is significantly different from zero at $P < 0.05$ and $P < 0.01$, respectively.

Table 5. Parents, crosses, and controls in a diallel analysis of 12 maize inbreds ranked by gray leaf spot rating

Rank	Genotype	Mean rating[z]	Rank	Genotype	Mean rating[z]
1	SO507W	0.34 a	42	SO713W × RO452W	2.69 ef
2	KO54W × RO558W	0.46 a	43	SO713W × RO594W	2.72 ef
3	SO507W × KO54W	0.60 ab	44	SO713W × SO507W	2.73 ef
4	KO54W	0.67 abc	45	SO181W × RO550W	2.73 ef
5	RO452W × KO54W	0.99 abcd	46	SO507W × RO558W	2.80 ef
6	KO54W × RO504W	1.00 abcd	47	RO465W × RO504W	2.80 ef
7	RO550W × KO54W	1.00 abcd	48	FO215W × RO594W	2.81 ef
8	SO507W × RO465W	1.28 abcde	49	SO507W × RO594W	2.86 ef
9	KO54W × RO594W	1.33 abcdef	50	RO460W × RO504W	2.86 ef
10	SO181W × KO54W	1.35 abcdef	51	FO215W	2.87 ef
11	RO465W × SO181W	1.47 abcdef	52	SO181W × RO504W	2.87 ef
12	RO452W × SO181W	1.53 abcdef	53	RO465W × RO558W	2.91 ef
13	SO507W × SO181W	1.54 abcdef	54	RO504W	2.92 ef
14	RO460W × KO54W	1.60 abcdef	55	SO713W × SO181W	2.92 ef
15	HL1	1.67 abcdef	56	SO713W × RO550W	2.93 ef
16	RO460W × SO507W	1.73 abcdef	57	SO181W × RO558W	2.94 ef
17	SO507W × RO452W	1.73 abcdef	58	RO558W × RO594W	2.98 ef
18	SO713W × KO54W	1.81 abcdef	59	SO181W × RO594W	2.99 ef
19	RO452W	1.87 abcdef	60	RO594W	3.00 f
20	RO460W × SO181W	1.93 abcdef	61	RO460W × RO558W	3.00 f
21	SO181W	1.94 abcdef	62	FO215W × RO452W	3.00 f
22	FO215W × KO54W	1.94 abcdef	63	SO713W × RO504W	3.00 f
23	RO465W × KO54W	1.99 abcdef	64	RO452W × RO558W	3.00 f
24	RO465W	2.01 bcdef	65	FO215W × RO504W	3.01 g
25	RO465W × RO452W	2.19 cdef	66	RO550W × RO594W	3.01 g
26	PAN6549	2.20 cdef	67	RO550W × RO504W	3.07 g
27	SO507W × RO504W	2.34 cdef	68	RO460W × RO452W	3.08 g
28	FO215W × SO181W	2.39 def	69	SO713W × RO465W	3.20 g
29	RO460W	2.40 def	70	FO215W × SO713W	3.20 g
30	RO558W	2.46 def	71	RO594W × RO504W	3.21 g
31	SO507W × RO550W	2.46 def	72	SO713W × RO558W	3.27 g
32	RO550W × RO558W	2.52 def	73	FO215W × RO550W	3.33 g
33	RO460W × RO465W	2.52 def	74	FO215W × RO558W	3.34 g
34	FO215W × SO507W	2.53 def	75	SO713W	3.35 g
35	RO452W × RO504W	2.55 def	76	RO452W × RO550W	3.89 g
36	SO713W × RO460W	2.66 def	77	RO452W × RO594W	3.47 g
37	RO550W	2.67 def	78	RO460W × RO594W	3.54 g
38	FO215W × RO460W	2.67 def	79	RO465W × RO594W	3.61 g
39	SR52	2.68 def	80	RO465W × RO550W	3.93 g
40	FO215W × SO507W	2.68 def	81	RO460W × RO550W	4.00 g
41	RO558W × RO504W	2.68 ef			

[z] Ratings followed by the same letter do not differ significantly according to Snedecor and Cochran's (11) Q test.

shows the genotypes ranked in order of resistance, with fiducial limits. The high resistance to gray leaf spot associated with inbreds KO54W, SO507W, and SO181W is clearly reflected in the high rankings of their crosses. This is particularly evident for line KO54W, all of whose crosses appeared within the first 23 rankings and within the same fiducial limits. The fact that the cross between this line and the most susceptible inbred, SO713W (ranked 75th), is placed 18th implies that within the predominantly additive nature of gene action, some genes exhibit major dominant effects. Similarly, the cross KO54W × RO558W, with mean disease rating 0.46, is as resistant as the parent KO54W (0.67), and its rating is markedly lower than both the rating of the susceptible parent (2.46) and the midparent value (1.56), which suggests major gene action (Table 5). This inbred would therefore be particularly useful in backcross recovery programs for disease resistance. In contrast, SO507W (which had the lowest disease rating) and SO181W were both represented much less frequently among this top-ranking group.

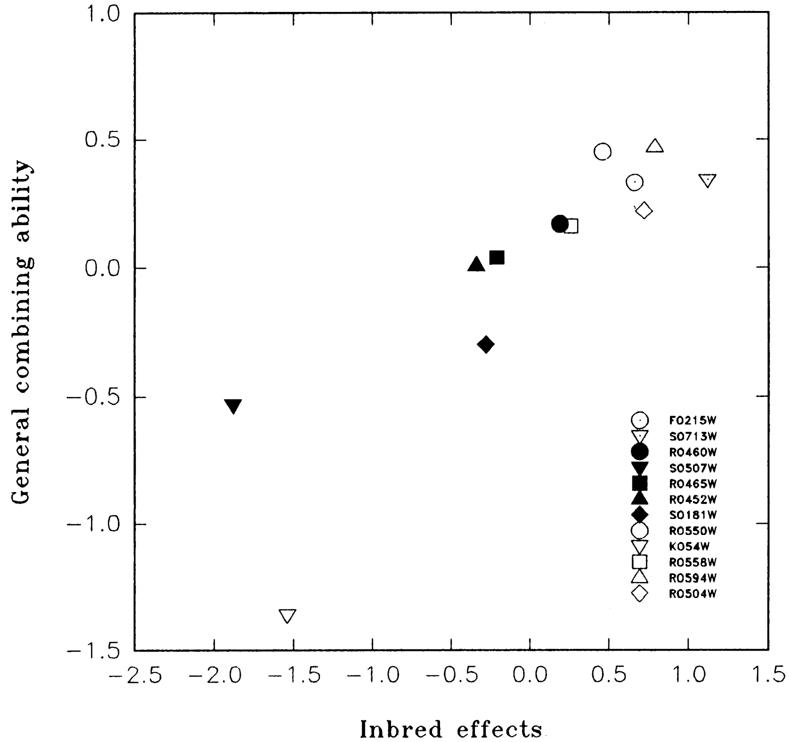

Fig. 1. Relationship between inbred effects and general combining ability for a diallel cross analysis of 12 maize inbreds.

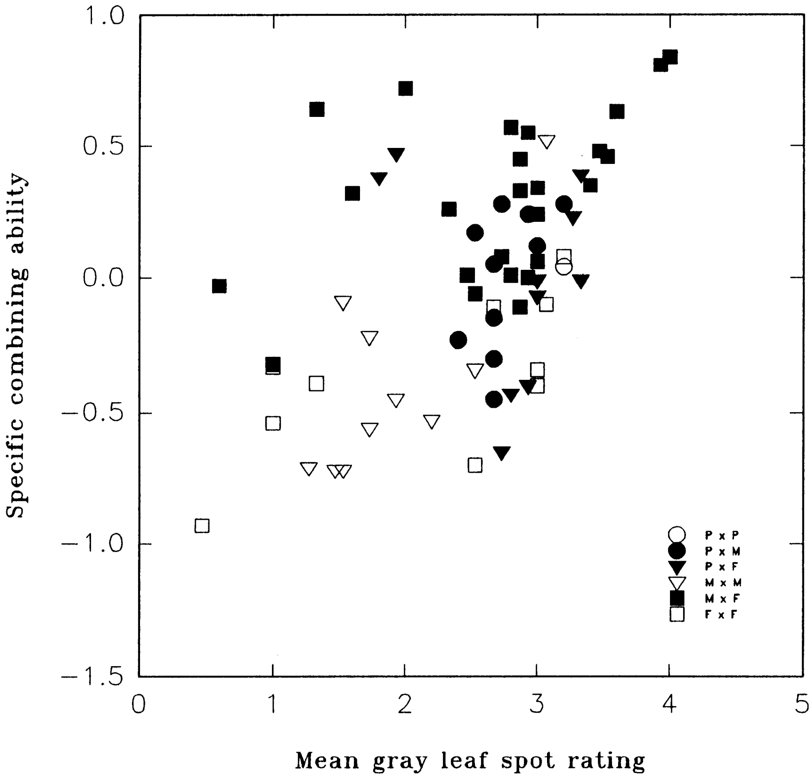

Fig. 2. Relationship between mean gray leaf spot ratings and specific combining ability effects of crosses in a diallel analysis of 12 maize inbreds. P, M, and F refer to the heterotic groups from which the lines were derived.

Other inbreds showed variably less dominance and usefulness. The disease ratings of commercial hybrids HL1, PAN6549, and SR52 (1.67, 2.20, and 2.68, respectively) represent a range of values that may be expected in local breeding material relatively unselected for this character.

Although our results demonstrate the importance of additive gene action in the inheritance of gray leaf spot resistance and identify elite sources of resistance, further evaluation of this and other breeding material at numerous locations is advisable to confirm the stability of available resistance. This is important because gray leaf spot is a relatively "new" disease and may not have been fully evaluated and established locally. However, local maize breeders may now incorporate into recurrent and backcross programs sources of resistance such as that found in the inbred KO54W, which in this first study has shown not only high levels of resistance and high general combining ability, but also evidence of major gene action for resistance to gray leaf spot.

ACKNOWLEDGMENT

Thanks are due to K. A. O'Reilly for technical assistance during the recording of disease data.

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