# Fungicide responses of maize hybrids to grey leaf spot

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#### **Abstract**

Grey leaf spot disease of maize (*Cercospora zeae-maydis*) has seriously decreased grain yields in the province of KwaZulu-Natal, South Africa, and has spread to infect maize in neighbouring provinces. No commercial hybrids, resistant to the disease have so far been identified, and fungicides have been shown to reduce disease severity. The response of sixty-four commercial hybrids to grey leaf spot under fungicide treatment were studied over two seasons. Overall, fungicides reduced disease severity and linear regression of gain in yield against disease severity enables the identification of hybrids with optimum responses to fungicides. Under low disease levels hybrids responded less to fungicides than under high disease levels. The most susceptible hybrids had the highest responses in control of leaf-blighting and gain in yield. Hybrids with lower-than-predicted leaf-blighting also had lower-than-predicted yield responses, indicating these to be less susceptible to grey leaf spot. These less susceptible hybrids are likely to require fewer fungicide treatments than more susceptible hybrids and are at lesser risk of serious yield losses.

Abbreviations: GLS - grey leaf spot; AUDPC - area under disease progress curve.

## Introduction

Grey leaf spot (GLS) disease of maize (Zea mays L.), caused by the fungus Cercospora zeae-maydis Tehon and E.Y. Daniels, is a relatively new disease in South Africa. It is capable of reducing maize grain yields by as much as 30 to 60 percent and reduces the yield and quality of maize grown for silage (Ward and Nowell, 1994). Yield losses tend to be more severe with monoculture maize (Beckman and Payne, 1983; Latterell and Rossi, 1983), and conservation tillage practices that retain the previous season's infected maize residue on the soil surface (Rupe, Siegel and Hartman, 1982; Stromberg and Donahue, 1986; Payne, Duncan and Adkins, 1987). GLS may be managed through tillage practices that completely bury infested maize debris. (Payne and Waldron, 1983; Huff, Ayres and Hill, 1988). However, in the United States the disease has

recently been observed to move from reduced tillage situations to become a problem in fields where traditional conventional tillage practices are used (Perkins, Smith, Kinsey and Dowden, 1995).

Rotation with non-host crops is an alternative solution to ploughing as the pathogen does not survive much beyond a year in infected debris (Latterell and Rossi, 1983; Stromberg, 1986; Huff, Ayres and Hill, 1988). In South Africa, maize is grown traditionally under a system of monoculture and few farmers practise any form of rotational cropping (Channon and Farina, 1991). Rotations are unlikely to be used as a means of control, since farmers are reluctant to change cropping practices. Genetic resistance, a highly efficient and cost-effective method of control, is likely to provide the long-term solution to the problem (Lipps and Pratt, 1989). However, no commercially available hybrids resistant to GLS have so far been identified in

South Africa. Chemical control methods offer an interim solution (Ward and Nowell, 1994). At the current maize prices of R450 per ton of grain, the break-even yield for fungicide and its application per treatment is 290 kg of grain ha<sup>-1</sup>. The use of fungicides in commercial maize production in South Africa is therefore economic.

Fungicide sprays protect the upper leaves of the maize plant from disease until the crop is physiologically mature. Research at Cedara has shown that the effective period of control will vary between 29 and 32 days, if fungicides are applied when disease levels are between one and two per cent of leaf area infected (Ward, Laing and Rijkenberg, unpublished results). With early onset of disease more than one fungicide application is necessary to provide protection from disease until the crop is physiologically mature. Ward, Nowell, Hohls and Laing (unpublished results) found that the onset of disease is later with hybrids least susceptible to GLS. Fewer fungicide treatments are required for these hybrids than those that are more susceptible to the pathogen (Ward, Laing and Rijkenberg, unpublished results).

The purpose of this study was to evaluate the response of commercially-grown maize hybrids to fungicide treatment, and to identify those hybrids which have optimum responses to fungicide application.

#### Materials and methods

The trials were conducted at the Cedara Agricultural Development Institute, Cedara (29°31'S, 30°17'E and alt. 1 070 m), South Africa. Maize has been continuously grown at Cedara since the National Maize Hybrid Cultivar Trials commenced in 1982. Hybrids were evaluated for response to fungicide treatment during the 1992/93 and 1993/94 growing seasons. The experiments comprised 49 hybrids laid out in a  $7 \times 7$  triple lattice design under stubble tillage. The experiment was replicated for fungicide sprayed and unsprayed treatments. The site was gently sloping and soils were well-drained, sandy-clay loams of the Hutton Form and Doveton Series (MacVicar, 1991). The trial was chiselploughed to a depth of 120 mm in the winter and again in October, prior to planting. The chisel-plough tines were spaced 310 mm apart and fitted with sweeps. The stubble residue on the soil surface prior to planting was calculated using a sitting frame described by Lang and Mallett (1982). The residue cover was 31%. Planting lines were drawn immediately prior to planting, when fertilizer sufficient for an eight-ton grain crop was band applied. A topdressing of 100 kg N ha<sup>-1</sup> was broadcast when the maize was knee-high. Normal weed and pest control practices were followed. Hybrids were planted in plots of two 6.6 m rows, spaced 0.75 m apart, with in-row plant spacings of 0.30 m. The trials were jabplanted by hand in early November each year and two seeds per plant station were planted. Approximately 30 days after planting (DAP) the seedlings were thinned to 44 000 plants ha<sup>-1</sup>. The harvested area of two, 6.0 m rows, were hand-harvested.

The 1992/93 season was characterised by low rainfall and hot days. The weather conditions until anthesis and during early grainfill were hot and dry and it was only in mid- to late-grainfill from mid-February that rainfall normalised. In contrast, rainfall during the 1993/94 season was above average and well-distributed throughout the growing season. Mists were abundant, especially in January and February, 1994. Temperatures were slightly lower than average (Table 1).

#### Cultivars

Sixty-four commercially-available hybrids in the National Cultivar Phase II series of trials were studied in the 1992/93 and 1993/94 seasons.

#### Fungicide treatment

In both seasons fungicide treatments commenced at first signs of disease: 76 days after planting (DAP) in 1992/93 and 74 DAP in the 1993/94 seasons. Three applications were made each season at approximately 17-day intervals. Benomyl fungicide was applied in the first season at a rate of 375 g of active ingredient (ai)  $ha^{-1}$  and in 1993/94 a combination of 187.50 g carbendazim and 93.75 g flusilazole ai  $ha^{-1}$  was applied (Punch Xtra, Du Pont de Nemoirs and Coy). Spray solutions were applied with a  $CO_2$  – pressurised backpack sprayer fitted with a vertically mounted sprayboom having three Whirlrain  $\frac{1}{4}$ " WRW2-20° nozzles, spaced one meter apart. Full-cover sprays of 450 L  $ha^{-1}$  were applied to each maize row.

## Disease and grain yield assessments

Whole-plant standard area diagrams described by Ward, Laing and Rijkenberg (submitted for publication) were used to in estimate percent disease severity. Assessments were made regularly on plants in the centre of each plot, commencing at first signs of disease until physiological maturity. These data were

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	Oct	Nov	Dec	Jan	Feb	Mar	Apr	Total
Rainfall (mm)	-							
1992/93	34	83	69	69	108	115	25	503
1993/94	133	63	162	206	127	113	37	846
Mean monthly	125	116	126	158	130	103	32	789
Mean Temperature	1							
1992/93	17.7	18.5	20.3	21.0	20.0	19.4	17.3	19.2
1993/94	17.0	18.3	19.4	19.9	19.4	19.0	17.1	18.6
Mean monthly	17.1	18.4	19.9	20.6	20.4	19.2	17.6	19.0

Table 1. Rainfall and temperature at Cedara for the 1992/93 and 1993/94 growing seasons

used in calculating the area under disease progress curve (AUDPC), which is an integrated summary of the disease epidemic. The AUDPC, calculated by the trapezoidal integration program (Berger, 1981), was standardised by dividing the AUDPC value by the duration of the epidemic. This was compared to the critical (single) point model of disease severity. As the correlations were highly significant, it was decided to use the critical point model as the disease index in the linear regression analysis. Grain yields were expressed in kg ha<sup>-1</sup> at 12.5% moisture. Relative yield and disease data were obtained by dividing by the trial mean, and are expressed as a percentage. The relative disease is expressed similarly. The disease severities and yields have been presented on a relative basis to remove seasonal effects.

## Regression analysis

A linear regression model was used to determine the yield responses and disease severity of GLS, and these were estimated by the linear regression model:

$$Y = Bo + BiXl + Ei$$

where Y is the response variable (yield), Bo is the intercept (response when disease is zero), Bi is the slope of the regression line, XI is the regressor variable (disease severity at a particular stage) and Ei is the unexplained variation (error or residual). The regression analysis was conducted on Genstat 5.2.

#### Results

## Hybrids

Data from 64 hybrids, evaluated between one and two seasons were used in the regression analyses. Only 34 of the more commonly grown hybrids are listed in Table 2. Seven of these, evaluated for two seasons and representative of the main disease susceptibility groups, were selected for ease of presentation. PAN 6479 and PAN 6480 were least susceptible, SNK 2888, RS 5206 and RS 5232 were most susceptible, whilst NS 9100 and PAN 6549 were of intermediate susceptibility to GLS (Table 2).

# Disease severity

Disease severity varied between seasons. There was a lower mean disease severity of 31% in the 1991/92 season, compared to 82% in the 1993/94 season (Table 3).

The regression of disease severity of fungicidesprayed hybrids against disease severity of unsprayed hybrids accounted for 64.3% of the variance (P < 0.001). The slope of 0.08484 was highly significant (P < 0.001) (Figure 1). At low disease levels experienced in 1992/93 the predicted disease severity of the sprayed maize was generally over-estimated (Table 3). Under the high disease levels experienced in 1993/94, the sprayed maize disease severities were much closer to those predicted. With the less susceptible hybrids, the actual disease severity of PAN 6479 was higher than predicted, and was lower than predicted for PAN 6480 (Figure 1). Of the susceptible hybrids, RS 5232 had nearly double the disease severity than predicted, while RS 5206 and SNK 2888 also had higher than predicted levels of disease. The model predicted slightly higher disease severity for the intermediate susceptible PAN 6549, while NS 9100 had lower than the predicted disease level.

#### Grain yield and response to fungicide treatment

The overall grain yields and gain in yield due to fungicide treatment were lower in 1992/93 than in the 1993/94 season. In both seasons the hybrids with the lowest GLS levels, PAN 6479 and PAN 6480,

Table 2. The mean relative disease severity and relative yields of 34 maize bybrids in response to fungicide treatment across the 1992/93 and 1993/94 season

		Relative	Relative grain yield			
Hybrid	i	Disease severity	Unsprayed	Sprayed		
PAN	6480	75	124	104		
SNK	2200	95	122	94		
CRN	3584	55	121	101		
SNK	2266	123	115	100		
SNK	2154	77	113	100		
PAN	6479	54	113	98		
PAN	6034	110	113	117		
SNK	2954	122	113	108		
PAN	6363	66	112	100		
TX	24	82	112	101		
PAN	6496	97	109	107		
SNK	2888	123	109	101		
Α	1598	91	106	100		
PAN	6578	75	105	117		
SNK	2042	88	105	94		
SNK	2151	86	105	94		
Α	1556	89	104	95		
SNK	2255	114	104	105		
CRN	3816	89	104	95		
SNK	2665	112	103	98		
SNK	2246	103	101	91		
PAN	6364	112	101	107		
PHB	3412	83	101	103		
SNK	2021	87	101	93		
PAN	6146	106	99	120		
NS	9100	77	98	100		
SNK	2950	122	98	100		
PAN	6549	87	98	100		
CRN	4502	105	98	88		
PAN	6552	136	93	108		
RO	413	108	92	91		
CRN	3414	118	89	_		
CRN	4523	140	89	87		
RO	410	160	88	104		
A	1849	131	87	115		
PHB	3442	87	87	102		
CRN	4605	150	86	87		
PAN	6140	112	86	121		
RS	5206	142	85	109		
RO	430	120	83	94		
PAN	6528	126	82	106		
CRN	4403	112	82	91		
PHB	3427	106	78	94		
SNK	2265	108	78	99		
RS	5232	149	76	95		

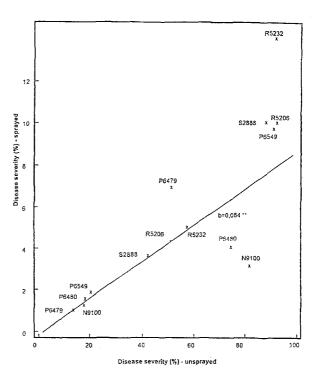


Figure 1. Regression analysis of fungicide-sprayed grey leaf spot disease severity against unsprayed disease severity at Cedara. Disease severity (expressed as a percent) of whole plants in control plot areas, was estimated at the milk stage of growth (R3 stage), 120 days after planting using standard whole plant diagrams. These regression analyses represent all hybrids evaluated over the 1992/93 and 1993/94 seasons. The means of only seven of the hybrids regressed are shown. P6479 and P6480 are least susceptible, R5206 and R5232 are most susceptible, S2888 is tolerant and P6549 and N9100 are intermediate in their reactions to grey leaf spot.

had the highest unsprayed grain yields and the lowest responses to fungicide. In contrast, the most susceptible hybrids, RS 5206 and RS 5232 had the lowest unsprayed yields and the highest response to fungicides. SNK 2888, also with relatively high disease, had among the highest unsprayed yield. In 1992/93, the response of SNK 2888 to fungicides was relatively low (2301 kg ha<sup>-1</sup>), whereas in 1993/94 the response was intermediate (4543 kg ha<sup>-1</sup>) (Table 3).

When the gain in yield was regressed against disease severity, the linear regression accounted for 76% of the variation. Since the quadratic model only resulted in a 3.3% improvement, the linear model was retained. The intercept of the linear regression was -22.0 and the slope was 53.64, which was highly significant (Figure 2). This model implies that with each percent increase in disease severity, there was a corresponding increase of 53.64 kg ha<sup>-1</sup> response to fungicide treatment. Under the low disease levels in

Table 3. The actual and predicted disease severity, grain yield and gain in yield (response) of unsprayed and fungicid	е
sprayed maize hybrids in 1992/93 and 1993/94	

	Hybrid	Unsprayed	l	Sprayed			Gain in yields	
		Yield	Disease	Yield	Disease (%)		kg ha -1	
		kg ha <sup>-1</sup> %		kg ha <sup>-1</sup>	Actual	Predicted	Actual	Predicted
1992/93	PAN 6479	5931	14	6711	0.7	1.2	780	740
	PAN 6480	6704	18	7119	0.3	1.6	416	960
	NS 9100	5327	17	7294	1.0	1.4	1966	874
	PAN 6549	5722	20	7115	0.3	1.7	1393	1050
	SNK 2888	6020	43	6689	0.7	3.7	669	2301
	RS 5206	5113	53	7504	0.7	4.5	2292	2837
	RS 5232	4678	58	6865	0.7	4.9	2186	3105
	Trial Mean	5571	31	7075			1504	1641
1993/94	PAN 6479	5563	52	9298	7.0	4.4	3735	2751
	PAN 6480	5924	75	9770	4.2	6.4	3846	3855
	NS 9100	4644	82	8802	3.3	7.0	4157	4233
	PAN 6549	4286	90	9321	9.7	7.6	5035	4631
	SNK 2888	5112	88	9839	10.0	7.5	4728	4543
	RS 5206	3526	92	10259	10.0	7.8	6693	4750
	RS 5232	3161	92	8458	14.2	7.8	5297	4719
	Trial Mean	4629	82	9166			4536	4376

the drought of 1992/93 PAN 6480, SNK 2888, RS 5206 and RS 5232 responded less to fungicides than predicted. The response of PAN 6479 was within the 95% confidence limits, while NS 9100 and PAN 6549 responded better than predicted by the model. In the more humid 1993/94 season, highly conductive to GLS disease, the yield response of most of the seven hybrids was close to that predicted by the model (Table 3 and Figure 2), except for RS 5206 which had the highest response to fungicides, of nearly 50% higher than predicted.

#### Discussion

The regression analysis of the effect of disease severity on the response of maize hybrids to fungicide treatment may serve as a basis for selecting the most suitable hybrids in areas where GLS is problematic.

Fungicide treatment reduced overall disease severity, with the most susceptible hybrids having the highest responses in the control of disease (leaf-blighting) and gain in yield (Table 3). Under the low disease levels in 1992/93, these responses were not as apparent as under high disease levels in 1993/94. The

hybrids, RS5206 and SNK 2888 were among the most susceptible to leaf-blighting. RS 5206 showed greater susceptibility to GLS by its yield response after treatment which was nearly 50% higher than predicted (Figure 2). This high-yielding hybrid was widely grown by farmers, has significantly reduced yields in the presence of GLS. The yield response of SNK 2888, was close to that predicted, substantiating previous findings that this hybrid is more tolerant of disease, in spite of high levels of leaf blighting. In contrast, hybrids less susceptible to GLS had lower than predicted yield responses as shown by the response of PAN 6480. This hybrid, with lower than predicted leaf-blighting also had a lower than predicted yield-response, indicating that it is less-susceptible to GLS.

GLS disease at Cedara usually infects maize at or near anthesis. The loss of photosynthetic leaf-area from leaf-blighting translates into loss in grain-yield. Fungicides delay the development of disease and treatment in areas prone to GLS produce yield gains (Ward et al., 1995). This was true across seasons, although the yield responses to fungicides were lower in the dry season of 1992/93, which resulted in lower levels of disease. The ability to predict yield responses of hybrids to

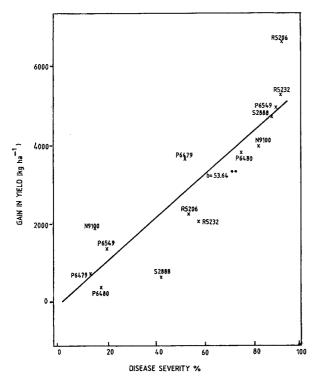


Figure 2. Regression analysis of gain in yield due to fungicide treatment against disease severity at Cedara. Disease severity (expressed as a percent) of whole plants in control plot areas, was estimated at the milk stage of growth (R3 stage), 120 days after planting using standard whole plant diagrams. The regression analysis represent all hybrids evaluated over the 1992/93 and 1993/94 seasons. The means of only seven of the hybrids regressed are shown. P6479 and P6480 are least susceptible, R5206 and R5232 are most susceptible, S2888 is tolerant and P6549 and N9100 are intermediate in their reactions to grey leaf spot.

fungicide treatment under varying levels of disease can assist in deciding whether to apply fungicides. For instance, is fungicide treatment of maize warranted when disease-levels are relatively low in dry-seasons which are infavourable for GLS? The model predicted a response of 1641 kg ha<sup>-1</sup> to fungicides when the mean disease level near physiological maturity was only 31%. With lower disease levels of 18%, the predicted yield response of PAN 6480 was 960 kg ha<sup>-1</sup>. Being a less susceptible hybrid the actual response was lower, 416 kg ha<sup>-1</sup>. This response was the result of three fungicide treatments, and, at current maize prices, the break-even yield for fungicide treatment is 290 kg ha<sup>-1</sup> per treatment. In this instance, spraying would not be economical as the cost of three fungicide treatments would have exceeded the actual yield response. In contrast, the higher than average diseaselevel of 53% of RS 5206 in 1992/93, resulted in a predicted yield response of 2,837 kg ha<sup>-1</sup>. With an actual response of 2,292 kg ha<sup>-1</sup>, the economics of treatment is justified. However, under low-disease levels, it would be more economical to produce grain from unsprayed PAN 6480 than RS 5206 with three spray treatments. The situation under high disease pressure is different. PAN 6480 with 75% leaf-blighting, the yield response predicted by the model was 3,855 kg ha<sup>-1</sup>. Three fungicide applications, with a break-even yield response of 870 kg ha<sup>-1</sup>, is therefore justified. There is however, less risk in selecting hybrids less susceptible to GLS than more susceptible hybrids like RS 5206, which may have a higher yield response to fungicide sprays. Ward et al. (1995), showed that disease develops earlier and more rapidly in more susceptible hybrids. These are likely to require more spray treatments than less susceptible hybrids in which disease is later and slower in developing. The additional sprays require additional costs, and if the timing of the sprays is delayed, there would be greater risk of yield losses which may negate the higher yield potential of a susceptible hybrid such as RS 5206.

Disease severity is usually less severe in early maize plantings, as infection is largely due to primary inoculum on debris of the previously infected maize crop. Later planted maize is often more severely diseased as infection is due to both primary inoculum produced on infected stubble from the previous crop, as well as secondary inoculum produced on lesions of infected earlier planted adjacent maize. This, however, does not always follow, as conditions more favourable for disease may occur with earlier plantings and become less favourable in later planted maize. In these circumstances, earlier planted maize is more diseased and may suffer more severe grain yield losses than later planted maize. This was the situation in 1993/94 when earlier planted maize in other trials planted at Cedara (results not shown) were more diseased and grain yields lower than the same hybrid planted later in the trial under discussion.

The timing of the initial fungicide spray relative to disease severity is to be important to the effectiveness of the treatment (Ward et al., unpublished results). The timing of the initial spray application at first signs of disease in the trial under discussion, however, was unlikely to influence final grain yields of the sprayed treatments. The initial spray was applied when disease severities, estimated by the logistic model, (Vanderplank, 1963), varied between 0.5 and 1.5 percent (results not shown). These disease severity levels were within the threshold limits prescribed for initial spray

treatment (Ward et al., unpublished results), and the frequency and intervals between spray applications were sufficient to provide optimum control of GLS.

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