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# A Study of Slow Rusting of Southern Rust of Corn: Preliminary Report

Southern rust (*Puccinia polysora*) of corn is potentially devastating because commercially available hybrids are not resistant. More understanding of the nature of resistance and the multiplication and dissemination of the causal agent is needed.

The rate of multiplication and spread of the corn-rust pathogen within a growing season or from one growing season to the next varies greatly. Resistance affects the multiplication of the pathogen, not its spread. The corn-rust fungus has several reproductive cycles each growing season. The development of an epidemic is influenced by the amount of disease present at the start of the epidemic,  $X_0$ , and by its multiplication rate, described by the apparent infection rate  $r$  (13). Resistance may reduce  $X_0$  and/or  $r$  and thereby reduce field severity.

Slow rusting is a kind of resistance that should reduce the severity of the disease by decreasing the reproduction rate of the pathogen. The major components of resistance that might contribute to slow rusting usually include the following items:

Latent period (LP) is defined as the time from infection to spore production. Some examples of cultivar differences in LP have been shown in the wheat "*Puccinia recondita* f. sp. *tritici*" (9), rye "*Puccinia recondita* f. sp. *recondita*" (11), and barley "*P. hordei*" (1, 10) relationships.

Infection frequency (IF) usually is measured as pustule numbers per unit of leaf area. A good example has been reported by Clifford (4). He studied the leaf rust of barley and indicated that significantly fewer (22%) pustules developed on a resistant cultivar than on a sus-

ceptible cultivar. Furthermore, differences in IF or lesion numbers have been reported for most of the host-pathogen systems studied (1,3,4,8,9).

Effect of spore production (SP) on disease spread has been dealt with comprehensively (5,7). However, measuring spore production accurately is extremely difficult because leaf agitation before or during collection may reduce the quantity of spores collected. Thus, most studies of SP have been carried out on seedlings.

In addition, infection type reflects the nature of resistance, and this has been studied by many authors (1,2,14).

The objective of this research was to study the components of slow rusting, both on seedlings (greenhouse) and adult plants (field) of corn hybrids.

## MATERIALS AND METHODS

Two greenhouse tests were carried out. In the first test, seven hybrids were used. Based on previous data, we chose Ab24E x B37R as a resistant check because we knew its level of resistance was high enough to prevent development of pustules. Other hybrids were GT112 x Tx601, Mp305 x Mp496, Mp704 x Tx 601, Mp496 x Mp702, B37 x Va35 and Mp305 x Mp339.

Each hybrid was planted at three kernels/pot in each of twelve 4-in pots on February 26, 1982. Seedlings were thinned to one per pot after emergence. There were three replications and four pots for each replication.

Plants were inoculated on March 15 while in the four- to five-leaf stage. Germinability of uredospores

on water agar immediately before inoculation was 50%. Uredospores were suspended in water amended with Tween 20®. The seeding pots were arranged randomly in a dew chamber and were spray-inoculated as uniformly as possible with a hand sprayer. After 16 hrs incubation in the dew chamber, pots were moved to a greenhouse bench.

Eight days after inoculation (i.e. March 23) symptoms were visible on all hybrids. Plants were evaluated for infection type using the nine grades as follow:

- 1 = No symptoms
- 2 = Flecking indistinct
- 3 = Flecking distinct
- 4 = Necrotic or chlorotic spots, but no pustules

- 5 = Necrotic or chlorotic spots, with a few small unruptured pustules
- 6 = Numerous small pustules, with only a few ruptured
- 7 = Small pustules, most of which had ruptured
- 8 = Moderate-sized pustules and almost all ruptured
- 9 = Large-sized pustules and almost all ruptured

Additional evaluations were made on March 24, 26 and 29.

In the second greenhouse test, Ab24E x B37R again was used as the resistant check. In addition, two single crosses, Ab24E x SC376 and B37 x GT106, were selected as a susceptible check. Other hybrids tested were Mp496 x Mp702, Mp704

x Tx601, B37 x Va35, Mp305 x Mp496, Va35 x Mp78:518, Mp410 x SC212M and Mp490 x Tx601. Five were repeated from the first test.

The hybrids were planted on April 30 in the greenhouse. Each hybrid was planted at three kernels/pot in each of eight 4-in pots. Seedlings were thinned to one seedling per pot after emergence. The six pots with the best plants were used and divided into three replications with two seedlings (pots) in each replication. Inoculation was carried out on May 18 when the seedlings were at the four- to five-leaf stage. The germinability of spores tested before inoculation was 45%. After 15 hr of incubation (i.e., on May 19, 9:30 a.m.) the pots were moved to the laboratory to avoid the high temperature of the greenhouse. Plants were grown under artificial light using a 14 hr day/10 hr night regime. Eight days after inoculation plants were evaluated for infection type using the 1-9 rating scale previously given. Additional evaluations were made at 10, 13 and 17 days after inoculation. All the other operations were the same as in the first greenhouse test.

### Greenhouse Studies

The latent period has been defined as the time from infection to spore production (13). We feel that a practical definition of latent period for southern rust would be the time from inoculation to pustule rupture. Even though a few pustules ruptured with a rating of 6 on the rating scale we used, we feel that, when plants rate 6-7 or 7, the latent period has ended. In our tests, the latent period for most hybrids was 10 days (Table 1). Two exceptions were 9 days for GT112 x Tx601 in the first test and 13 days for Va35 x Mp78:518 in the second test. The only disagreement between the tests was with the hybrid Mp305 x Mp496, and the reason for this incon-

Within five days after the appearance of symptoms, most pustules had ruptured. Spores were collected from a previously marked area on leaves 13 days after inoculation, using a mini-cyclone spore collector, and were weighed. The marked areas of leaves then were excised and measured by an automatic leaf-area meter. Finally, a count was made of pustules---both ruptured and unruptured---within the marked area.

Using seed of the same hybrids grown in the second greenhouse test, a field experiment was planted on June 7. The experiment was located on the Plant Science Research Farm at Mississippi State University. Single-row, 20-plant plots, 5 m long with 1 m between rows, were planted, with a single row of a susceptible hybrid grown in alternate rows to serve as spreader rows. The test was replicated three times. On the evening of July 23 (before tasseling) 2 ml of a spore suspension were injected into the whorl of each plant in the spreader rows. The purpose was to inoculate plants in the spreader rows from which inoculum would

disseminate to test rows.

Twenty-five days after inoculation (approximately two disease cycles) on August 17, notes on rust severity were taken. An 11-point (to 10) severity scale based on percentage of leaf area covered by pustules (0, 0.1%, 0.5%, 1%, 2.5%, 5%, 10%, 15%, 20%, 25%, 30% or more) was used. Leaves with a 10 rating were considered 100% infected because there was little or no further increase of uredinia on them before they became completely desiccated. This visual scale was an interpolation of a key produced by James for rating leaf rust of cereals based on percentage of leaf area covered by uredinia (6).

Visual severity ratings were taken at two positions on the plants---(1) the ear leaf and two leaves above the ear leaf and (2) the top three leaves. Ratings for the two leaf positions were averaged. Severity was estimated independently by two people, and the average was determined. Additional recordings were made on August 20 and 27 and on September 3.

## RESULTS

sistency is not readily apparent.

Only limited differences were noted for maximum disease ratings among the hybrids tested. In the first test, B37 x Va35 rated one grade higher than the other hybrids, and, in the second test, the two susceptible checks had the largest pustules (Table 1).

Other resistance components, such as infection frequency (pustule number) and the amount of spores produced, were significantly different among hybrids (Table 2). One of the susceptible checks, B37 x GT106, had the highest spore production and ranked third for pustule numbers, but the other susceptible check Ab24E x SC376

was less susceptible. Data on the resistant check, Ab24E x B37 were not used in the analysis but are presented for comparison with the other hybrids in the test. Even if Mp305 x Mp496 is not considered, three-fold differences in numbers of spores produced were noted among hybrids (Table 2). Mp410 x SC212 had more than twice the number of pustules found on Mp496 x Mp70

### Field Test

Data obtained in the field indicated that the two hybrids selected as susceptible checks were indeed susceptible and that another hybrid

Table 1. Infection type and latent period of southern rust on seedlings of corn hybrids grown in the greenhouse, by pedigree of hybrids.

Pedigree	First Test					Second Test				
	Days after inoculation					Days after inoculation				
	8	9	10	14	L.P. (days)	8	10	13	17	L.P. (days)
Ab24E x B37R (Res. ck)	2	2	3	3	-	1	1	2	2	-
Mp496 x Mp702	6	6	7	7-8	10	5	6-7	7	7	10
Mp704 x Tx601	6	6	7	7-8	10	5	6-7	7	7	10
B37 x Va35	6	6	7	8-9	10	6	6-7	7	7	10
Mp305 x Mp496	6	6	7	7-8	10	4	4-5	5-6	6	-
Va35 x Mp78:518	-	-	-	-	-	5	5-6	6-7	7	13
Mp410 x SC212M	-	-	-	-	-	6	6-7	7	7	10
Mp490 x Tx601	-	-	-	-	-	6	6-7	7	7	10
Ab24E x SC376 (Sus. ck)	-	-	-	-	-	6	6-7	7-8	8	10
B37 x GT106 (Sus. ck)	-	-	-	-	-	6	7	7-8	8	10
Mp305 x Mp339	6	6	7	7-8	10	-	-	-	-	-
GT112 x Tx601	6	7	7-8	7-8	9	-	-	-	-	-

Table 2. Pustule numbers and spore amounts of southern rust on seedlings of hybrid corn grown in the greenhouse, by pedigree of hybrids.

Pedigree	Spore Amounts <sup>(x)</sup> (mg/cm <sup>2</sup> )	Pustule Numbers (Per cm <sup>2</sup> )
B37 x GT106 (Susceptible ck)	0.68 a	38.92 bc
Mp410 x SC212M	0.55 ab	53.76 a
Ab24E x SC276 (Susceptible ck)	0.44 abc	27.97 cd
B37 x Va35	0.43 abc	44.73 b
Mp490 x Tx601	0.30 bc	30.92 cd
Mp704 x Tx601	0.22 c	33.90 cd
Va35 x Mp78:518	0.21 c	30.90 cd
Mp496 x Mp702	0.21 c	23.29 d
Mp305 x Mp496	0.004 d	13.78 e <sup>(y)</sup>
Ab24E x B37R (Resistant ck)	None	None <sup>z</sup>

<sup>x</sup>: Means not followed by the same letter differ significantly (5% level) from each other according to Student-Newman-Keul's Test.

<sup>y</sup>: Some of the numbers are flecks.

<sup>z</sup>: There are some flecks; the density is 0.94/cm<sup>2</sup>.

B37 x Va35, was equally susceptible (Table 3). The other hybrids all had some characteristics of slow rusting. The differences appeared to be primarily in pustule numbers (coverage of leaf) because differ-

ences of one grade or less were observed for pustule size (infection type). The differences in disease severity were striking.

The degree of correlation between components of slow rusting is given

in Table 4. Both infection type and pustule numbers were highly related with spore production in greenhouse test. Spore production in the greenhouse was correlated with disease severity in the field.

Table 3. Response to infection with Puccinia polysora of corn hybrids in field tests, by pedigree of hybrids.

Pedigree	Severity of the End of Epidemic (%) <sup>(x)</sup>	Field Infection Type
B37 x GT106 (Susceptible ck)	30.00 a	8
Ab24E x SC376 (Susceptible ck)	28.33 a	7-8
B37 x Va35	28.33 a	7-8
Va35 x Mp78:518	10.83 b	8
Mp305 x Mp496	9.43 bc	7
Mp410 x SC212M	8.20 b-d	7
Mp496 x Mp702	6.83 c-e	7-8
Mp490 x Tx601	5.30 de	7
Mp704 x Tx601	3.50 e	7-8
Ab24E x B37R (Resistant ck)	0.00 <sup>(y)</sup>	2-3

<sup>x</sup>: Means not followed by the same letter differ significantly (5% level) from each other according to Student-Newman-Keul's Test.

<sup>y</sup>: Hybrid Ab24E x B37R was not used in analysis of variance because it did not develop pustules.

Table 4. Correlation coefficients of resistant components of slow rusting of corn hybrids grown in the field.

	Spore Amount (SA)	Pustule Number (PN)	Severity of the end of Epidemic (SE)
Infection type (IT)	0.7762**	0.4873	0.5819 <sup>(x)</sup>
Spore Amount	-----	0.7753**	0.6479*
Pustule Number	-----	-----	0.2350

\*\* : Significant at 1% level (1% significant level = 0.765).

\* : Significant at 5% level (5% significant level = 0.632).

<sup>x</sup> : This is the correlation coefficient between the field severity and field infection type.

## DISCUSSION

The resistant components of slow rusting generally consist of longer latent period, fewer spores produced per unit leaf area, lower infection frequency in terms of pustule number and lower infection type. In the greenhouse test, one hybrid had a longer latent period than did the other hybrids. Some studies indicate that genotypes differ for latent period, and other studies do not. For instance, barley cultivars tested by Clifford (1) and Parlevliet (10) differed in latent period for barley leaf rust. A similar pattern was observed in rye for *P. recondita* f. sp. *recondita* (11). But, a test carried out on oats with *Puccinia graminis* f. sp. *avenae* showed that the host genotypes did not differ in latent period (15). Therefore, the differences in latent period among cultivars apparently vary with different crops and different diseases. In some crops, the latent

period is an important component of resistance.

There was a highly significant correlation ( $P = 0.01$ ) between infection type and spore production and between pustule numbers and spore production (Table 4). It appears that these three major components of slow rusting (resistance) are closely associated. Different components of resistance, lower infection type, reduced spore production and lower infection frequency are correlated. The variation in spore production was always closely associated with infection type. (Infection type classification is based on size of pustules.) This conclusion had been drawn by Dehne (2) who made a comprehensive study of the yellow rust disease of wheat caused by *Puccinia striiformis* and is also shown clearly in the barley leaf-rust relationship (8, 10, 12).

The susceptible checks, B37 x GT105 and Ab24E x SC376, were the most susceptible hybrids in both adult and seedling tests. In the field Mp410 x SC212M did not show as much susceptible reaction as in the greenhouse. Compared with others, B37 x Va35 was the most susceptible hybrid in both seedling and adult stages. Several other hybrids showed somewhat less disease than the susceptible check; however, this difference was not always significant in the greenhouse tests.

The disease on the susceptible hybrid in spreader rows was uniform and severe. This shows that the inoculation method of injecting spore suspension into the whorl was successful. However, the inoculating rows did not play an important role in the field test because there was an abundance of natural inoculum in 1982.

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