THE PERFORMANCE OF BREEDING LINES WITH PARTIAL RESISTANCE TO *PYTHIUM ULTIMUM* AND *RHIZOCTONIA SOLANI*.

T. A. Wheeler, J. R. Gannaway, S. Fichtner, and S. Yang
Texas Agricultural Experiment Station, Rt. 3, Box 219, Lubbock, TX

Abstract

Nine breeding lines originally selected by a *Pythium* petri plate assay, were tested in a field naturally infested with *P. ultimum*. A susceptible variety, Paymaster (PM) 2326RR was included in the test, without chemical protection against *P. ultimum* and *Rhizoctonia solani* (-) and with chemical protection (+). All other entries, including PM 2326RR-, were treated only with Captan before planting. All seed was inoculated with *R. solani* (grown on oat grains) at planting. Treatments were arranged in a RCBD with four replications. At 28 days after planting, 8 of the 9 breeding lines had significantly ($P = 0.05$) better stands than PM 2326RR (-). All nine breeding lines had stands which were not significantly different than PM 2326RR (+). Five of the breeding lines had a slower rate of emergence at 14 days after planting than PM 2326RR (-), and one line (G52) only had 31 % of the stand it would ultimately achieve. PM 2326RR (-) had significantly poorer yields than PM 2326RR (+) and seven of the breeding lines. However, many of the breeding lines had poorer lint quality, particularly in the area of fiber strength or length than PM 2326RR. The *Pythium* petri plate assay appears to be an efficient method to identify cotton lines with enhaced disease resistance to *P. ultimum* and *R. solani*.

Introduction

Seedling disease accounts for substantial losses across the cotton growing regions of the United States every year. Most varieties grown in the U.S. are highly susceptible to seedling diseases caused by *Rhizoctonia solani* (Figure 1) and *Pythium ultimum* (Figure 2). Exceptions include some Acala varieties like Maxxa which is resistant to *Pythium* spp., but not *R. solani* (Garber et al., 1991). Management of seedling disease for most of U.S. is entirely dependent on fungicide seed treatments and infurrow fungicide applications. When environmental conditions are highly conducive for disease, then chemical methods of control are insufficient. Better resistance to seedling disease in cotton varieties would improve cotton production in the U.S.

Materials and Methods

Nine cotton lines (labeled G46 - G54) were originally selected for seedling disease resistance based on a *Pythium* petri plate assay (Henard, 1997) (Figure 3). These lines were selfed during the 1999 growing season to increase seed for field testing in 2000. The parentage for each line is shown in Table 1. Seed for line G52 was also obtained from open-pollinated plants that had been planted in a dryland study in 1999. All the seed was treated with Captan 75WP (2 oz/100 lb seed). A susceptible check, Paymaster (PM) 2326RR was also included in the test, both with captan treated seed (PM 2326RR-) and with seed treated with Baytan 30 (0.5 oz/100 lb seed) + Allegiance (0.75 oz/100 lb seed) + Thiram 42S (2 oz/100 lb seed), PM 2326RR+. The test field, located at the Lubbock Research and Extension Center, was naturally infested with *P. ultimum*

*(*100 colony-forming units/cm^3 soil). This site also had low levels of *R. solani* and *Thielaviopsis basicola*. An isolate of *R. solani*, (62B, VCG 4) was grown on autoclaved oat grains for 4 weeks and then dried and ground into a fine powder. Plot size was 35.5' long, two rows wide with 40" spacing. The seed for each plot row was placed in a packet (5 seed/ft row,177 seed/packet) with 5 g of *R. solani* inoculum and planted on 28 April. There were 9 breeding lines and a susceptible and fungicide-protected check, arranged in a randomized complete block design with four replications. Soil conditions were warm and dry after planting, so plots were irrigated to saturation 3 days after planting and several times during the seedling phase. Stand counts were taken on both rows weekly until counts did not change. At 30 days after planting, 6 plants were removed from each plot and rated for hypocotyl damage and root necrosis. The hypocotyl rating was based on a scale of 0 to 3 where 0 = no damage; 1 = a superficial lesion; 2 = a sunken lesion; and 3 = a sunken lesion which is killing the plants. Plots were harvested with a two-row plot stripper on 21 November. A sample from each plot was ginned and used to calculate the percentage of lint harvested for each breeding line. A sample of lint was sent to the International Textile Center (Lubbock, TX) to determine micronaire, length, uniformity, strength, elongation, leaf grade, degree of reflectance, yellowness, and color grade. Plant stands, the percent of plants which emerged quickly (stand at 14 days/stand at 28 days), and yield and all the lint properties were compared using analysis of variance. Means of treatments were considered different at $P = 0.05$ using a Waller-Duncan k-ratio t test.

Results and Discussion

The weather during the seedling period of development was unusually hot and dry during the 2000 growing season.. There was 0.45" of rain that occurred one day after planting, and then 0.5" of rain 27 days after planting. However, the addition of *R. solani* in the seed and the heavy irrigation after planting caused significant seed rot. The susceptible check, PM 2326RR(-), had the poorest plant stands for all evaluation periods and the lowest yields (Table 2). With the addition of a fungicide package with activity against *R. solani* and *P. ultimum*, plant stand for PM 2326RR increased from 0.5 plants/ft row to 2.3 plants/ft row and yield increased from 167 lbs of lint/a to 666 lbs of lint/a (Table 2). At 14 days after planting, two of the breeding lines (G50 and G53) had significantly higher stands than PM 2326RR(-), and 8 of the 9 breeding lines had stands not significantly different than PM 2326RR(+) (Table 2). At 21 days after planting, all of the breeding lines had better stands than PM 2326RR(-) and the breeding lines all had stands not different than PM 2326RR(+) (Table 2). At 28 days after planting, 8 of the breeding lines with significantly better stands than PM 2326RR(-) and not significantly different than PM 2326RR(+) (Table 2). Only G52 had stands which were poorer than PM 2326RR(+). This line was the only one to use seed which came from a dryland study, as opposed to purely selfed seed. That may have impacted the germination of the seed. Unfortunately there was not sufficient seed to run germination studies.

Improved resistance to seedling disease was an objective of the MAR breeding program developed by Dr.Luther Bird. In the MAR program, cotton lines with slow germination for 8 days (at 56° F) were associated with resistance to seedling disease (El-Zik, 1989). To examine whether the breeding lines in this study had slow germination, the number of plants which had emerged at the first evaluation period (at 14 days after planting) was compared with the number

of plants which ultimately emerged for each plot. The lines with the fastest germination rate included PM 2326RR(-) (79 % of final stand)), G53 (78 % of final stand), and G50 (72 % of final stand). However, G50 and G53 had a final stand of 2 plants/ft, which was not significantly different than lines with slower initial emergence. The lines with the slowest emergence included G52 (31 % of final stand), G54 (44 % of final stand), and G47 (46 % of final stand). G47 had the highest final stand (2.4 plants/ft) and G54 also had one of the best final stands (2.2 plants/ft). G52 never did achieve an acceptable final stand (1.4 plants/ft), but that may have been a function of poorer quality seed.

The was no root necrosis measured on the samples dug at 30 days after planting. There was some hypocotyl damage, though none of the breeding lines had significantly lesshypocotyl damage than the susceptible and chemically protected check (Table 2). G53 had significantly more hypocotyl damage (1.48) than G47 (0.67), G54 (0.79), and PM 2326RR (+) (0.83).

Yield of PM 2326RR(-) was substantially affected by the poor stands, resulting in a 500 lb lint/a decrease compared to PM 2326RR(+) (Table 2). Yield of PM 2326RR(-) was significantly lower than 7 of the 9 breeding lines (Table 2). Only G46 and G47 had yields which did not differ from PM 2326RR(-), and both of those lines had much better plant stands than PM 2326RR(-). All the other breeding lines had yields not significantly different than PM 2326RR(+) (Table 2).

Micronaire was best (premium) for PM 2326RR(+), G49, and G47 (Table 3). Micronaire was at a base level for G54, G50, G51, G53, PM 2326RR(-), and G46 (Table 3). Micronaire was discounted for G48 and G52. Fibers were longest for G47 (staple 36) and PM 2326RR(+) (staple 35) (Table 3). Breeding lines G49, G48, G50, and PM 2326RR(-) all had a staple length of 34. Breeding lines G46, G54, G52, and G51 had a staple length of 32, and G53 had a staple length of 31 (Table 3). Lint uniformity was high for G48, G47, PM 2326RR(+), and PM 2326RR(-) (Table 3). Lint uniformity was intermediate for all other breeding lines. Breeding lines with very strong lint were G47, G48, and G46 (Table 3). Breeding lines with strong lint were PM 2326RR(+), G49, and PM 2326RR(-). Breeding lines with intermediate lint strength were G50, G51, G54, and G53. The lint for G52 was rated as weak. Lint elongation which is the amount that a fiber will stretch prior to breakage, was better only for G54 than for PM 2326RR(-) (Table 3). However, none of the breeding lines had significantly worse lint elongation than PM 2326RR(-) or PM 2326RR(+) (Table 3). Leaf grade was better for PM 2326RR(-) than for G50 and G47 (Table 3). Leaf grade was better for G49 and G54 than PM 2326RR(+). The degree of reflectance of the lint indicated that G49, G46, and G47 all had whiter lint than PM 2326RR(-) (Table 3).

All nine of these breeding lines demonstrated better resistance to seedling disease than PM 2326RR. Breeding line G49, was similar or better then PM 2326RR in all measured traits. Most of the breeding lines had a lower staple length and weaker strength than PM 2326RR. These lines do offer the potential to substantially reduce losses to seedling disease without compromising yield, though lint quality still needs some improvement. This field test also offers evidence that the rapid laboratory screen using *P. ultimum* on a petri plate is a valid method of identifying cotton lines with better resistance to seedling disease.

References

El-Zik, K. M., and P. M. Thaxton. 1989. Genetic improvement for resistance to pests and stresses in cotton. p. 191-224. *In* R. E. Frisbie, K. M. El-Zik, and L. T. Wilson (eds) Integrated Pest Management Systems and Cotton Production. John Wiley & Sons, NY.

Garber, R. H., J. E. DeVay, and R. J. Wakeman. 1991. The role of cultivar tolerance in cotton seedling disease control. p. 163-164. In D. J. Herber and D. A. Richter (eds) Proceedings of the Beltwide Cotton Conference. National Cotton Council, Memphis TN.

Henard, R. S., J. R. Gannaway, and T. A. Wheeler. 1997. Screening for resistance in upland cotton (*Gossypium hirsutum*) to *Pythium ultimum*. p. 118. *In* P. Dugger and D. A. Richter (eds) Proceedings of the Beltwide Cotton Conference. National Cotton Council, Memphis, TN.

Table 1. The designation of nine breeding lines identified with improved resistance to *Pythium ultimum*

Breeding line	Designation
G46	Acala 442-58
G47	Acala 442-64
G48	CA1012 = CA 491 x CA 788 (K810)
G49	92'93 G-33
G50	CA 2253 = CA 1012 x G1802 (NM)
G51	CA 2266 (2) = EP60-611 x CA 1056 (F. Q. 109)
G52	CA 2266 (2) = “ ”
G53	CA 2266 (2) = “ ”
G54	CA 1056 = CA 803 (K904) x 6024-11-1 (V667)

Table 2. The effect of seedling disease on plant stand and yield.

Breeding line	Plants/ft (days after planting) 14 DAP	28 DAP	Early[a] germ	Hypo.[b]	lbs of lint/a
PM 2326RR-[c]	0.3	0.5	0.79	0.96	167
PM 2326RR+	1.6	2.3	0.66	0.83	666
G46	1.1	2.0	0.54	1.13	340
G47	1.0	2.4	0.46	0.67	382
G48	1.1	1.9	0.52	1.04	441
G49	1.2	1.8	0.62	1.00	492
G50	1.4	2.0	0.72	1.17	542
G51	1.0	1.5	0.59	1.08	475
G52	0.5	1.4	0.31	0.75	443
G53	1.6	2.0	0.78	1.46	581
G54	1.0	2.2	0.44	0.79	542
MSD[d]	1.0	1.0	0.23	0.53	250

[a]Early germination is determined by dividing the stand at 14 days after planting by the stand at 28 days after planting.

[b]The hypocotyl rating scale is 0 = no damage; 1 = a superficial lesion; 2 = a sunken lesion; and 3 = a sunken lesion which is killing the plant.

[c]The susceptible variety Paymaster (PM) 2326 was either treated with the fungicide Captan, (-)which has no activity of *Pythium ultimum* and *Rhizoctonia solani*, or was treated with Vitavax-PCNB and Allegiance (+), which does have activity against *P. ultimum* and *R. solani*.

[d]MSD is the minimal significant difference between treatments, based on the Waller Duncan k-ratio t test ($P = 0.05$).

Table 3. Influence of breeding lines with *Pythium* resistance and Paymaster (PM) 2326RR on lint quality, as defined by the USDA in 1995

Breeding lines	Micr[b]	Length[c]	Uniform[d]	Strength[e]	Elong[f]	Leaf grade[g]	Degree of reflec[h]
PM 2326RR-[a]	4.35	1.05	82.75	28.70	6.35	2.5	64.5
PM 2326RR+	4.15	1.08	82.85	29.05	6.55	3.5	65.6
G46	3.45	1.01	79.50	31.35	6.15	2.5	68.1
G47	3.65	1.12	82.95	35.30	6.20	4.0	66.7
G48	5.20	1.06	83.95	32.05	6.55	3.5	64.4
G49	3.80	1.06	81.50	29.00	6.80	2.0	68.2
G50	4.60	1.06	80.45	28.10	6.00	4.5	64.5
G51	4.55	0.99	80.35	27.40	6.45	2.5	66.0
G52	5.05	0.99	80.30	24.75	6.75	3.0	64.2
G53	4.50	0.98	80.40	27.10	6.20	2.5	66.2
G54	4.60	1.00	81.65	27.35	7.05	2.0	65.3
MSD[i]	0.48	0.05	1.38	1.82	0.63	1.4	1.9

[a]The susceptible variety Paymaster (PM) 2326 was either treated with the fungicide Captan, (-)which has no activity of *Pythium ultimum* and *Rhizoctonia solani*, or was treated with Vitavax-PCNB and Allegiance (+), which does have activity against *P. ultimum* and *R. solani*.
[b]Micronaire is a relative measure of fiber linear density determined by air permeability. Micronaire is considered premium in the range between 3.7 and 4.2. Micronaire is in a base range from 3.5 to 3.6 and 4.3 to 4.9. Micronaire is in a discount range when it is 5 or above or 3.4 and below.
[c]Length is expressed in hundredths of an inch which approximates the classer's staple length, which is31 for a HVI length of 0.96 to 0.98, 32 for a HVI length of 0.99 to 1.01, 34 for a HVI length of 1.05 to 1.07, 35 for a HVI length of 1.08 to 1.10, and 36 for a HVI length of 1.11 to 1.13.
[d]Uniformity is a measure of the uniformity of fiber length expressed as a percentage, where 83 to 85 is high, 80 to 82 is intermediate, and 77 to 79 is low.
[e]Strength is the force required to rupture a fiber sample in grams per tex. A rating of 31 or higher is very strong, 29 to 30 is strong, 26 to 28 is intermediate, and 24-25 is weak.
[f]Elongation is the amount that a fiber sample will stretch prior to breakeage.
[g]Leaf grade is the percentage of the fiber sample area covered by non-fiber materials, as determined by a video scanner. The percentage is converted to units from one through eight and

the higher values indicate more foreign material.
[h]Degree of reflectance is a measure of how light or dark the fiber sample is, expresesed as a percentage. The lower values indicate a grayer sample.
[i]MSD is the minimal significant difference between treatments, based on the Waller Duncan k-ratio t test (P = 0.05).

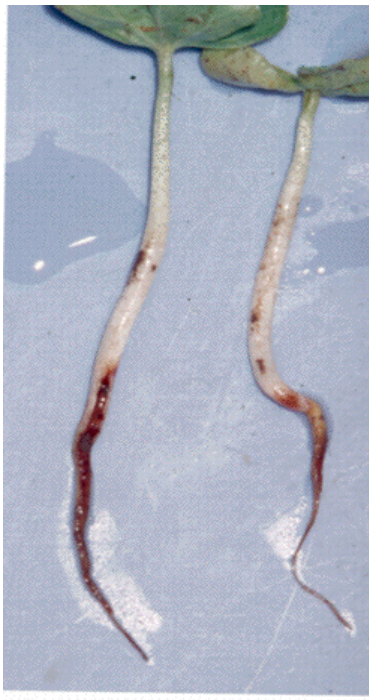

Figure 1.
Rhizoctonia attacking cotton seedlings

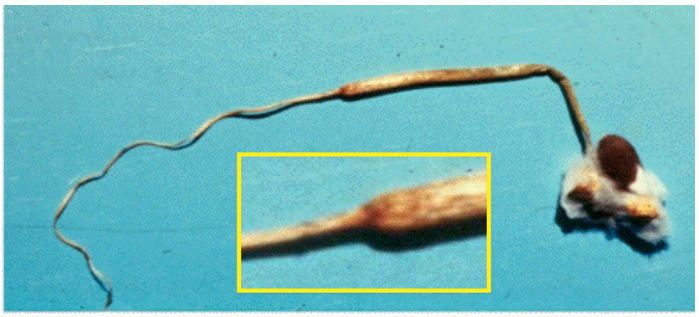

Figure 2.
Pythium attacking cotton seedling

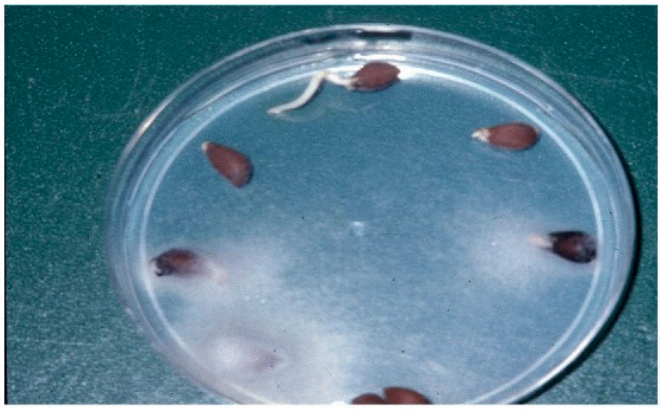

Figure 3.
Fungus pythium attacking cotton seed