APPLIED MANAGEMENT STRATEGIES FOR VERTICILLIUM WILT AND ON-FARM COTTON CULTIVAR VARIETY EVALUATIONS

C. Land
K. S. Lawrence
Entomology and Plant Pathology
Auburn, AL
B. Meyer
AGRI-AFC
Decatur, AL
C. H. Burmester
Crops, Soils, and Environmental Sciences
Auburn, AL

Abstract

Verticillium Wilt is caused by *Verticillium dahliae*, which colonizes the vascular cylinder of the plant and causes defoliation, stunting, and yield loss. Eighteen cotton cultivars were planted and evaluated for resistance to *Verticillium dahliae*. The trial was planted on the Tate farm in north Alabama and the field was irrigated with a drip tape irrigation system. Disease incidence and severity ratings were assessed September 20. Ten feet sections of each plot were observed for total number of plants and stems were cut longitudinally to assess disease incidence. Disease severity ratings of foliar symptoms were evaluated on a scale from 1-5 with 1 having no foliar wilting, 3 having interveinal chlorosis and necrosis of the leaves and 5 having completely defoliated plants. Petioles were taken from infected plants of each variety and re-isolated to confirm the presence of *Verticillium dahliae*. In regards to disease severity, the cultivars displaying the least amount of foliar symptoms and appearing most resistant were ST 4946 GLB2 and FM 1944 GLB2. Although none of these cultivars received averaged scores of less than 2 indicating all cultivars displayed some foliar wilt symptoms. In terms of disease incidence, ST 4946 GLB2 had the lowest number of plants with darkened vascular systems. PHY 339 WRF, ST 4747 GLB2 and ST 5032 GLT all displayed vascular staining and the average percent of disease incidence ranged from 50-53%. These percentages were statistically similar and performed better than the resistant check FM 1944 GLB2. In addition to the cultivar test a microplot trial was conducted to determine disease severity with and without irrigation in different soil type commonly found throughout the state of Alabama. The microplot test was arranged in a split plot factorial design with six soil types found throughout the state of Alabama as the main factor and irrigation or dry land condition (natural rainfall) was the secondary factor. Irrigation significantly increased disease incidence and severity compared to the non-irrigated soils. Decatur Silt Loam and the Houston Clay soil types had significantly more disease incidence regardless of the irrigation treatment.

Introduction

This disease was first recorded in Virginia in 1918 by Carpenter who noted its presence on Upland cotton *Gossypium hirsutum*. Currently, Verticillium wilt is present throughout the world affecting major cotton producing countries such as Australia, Brazil, and China. In the United States, it is present in most states in the cotton belt. Approximately, ten million acres of cotton are grown in the U.S, and 97% is planted with Upland cotton (Jefferson). Cotton production is greatly reduced by the presence of fungal pathogens. Cotton losses from Verticillium Wilt for the U.S. exceed 480 billion bales lost between the years of 1990-2013(Disease Database). Verticillium is a serious disease in North Alabama that causes a decline in plant health and a decrease in yields. It is caused by the soil born fungus *Verticillium dahliae,* which colonizes the root and then moves upward in the vascular system of the plant. This colonization of the vascular system prevents water movement giving the plant a wilted appearance. Symptoms in Alabama are not noted until later in the season when boll formation is occurring and the plant prematurely begins to defoliate. Defoliation leads to stunting and yield decrease partially due from the lack of photosynthetic activity. Verticillium wilt causes plant stunting, lack of lateral growth, and decreases in yield, fiber quality, and seed quality. (Bugbee, Xiao)
Verticillium wilt is often a difficult pathogen to control, with limited management options. Chemical and biological controls are not an option because there are none on the market that are recommended for control of Verticillium wilt. Rotation to a non-host such as a monocot will reduce disease but not eliminate the fungal pathogen and is not always an economical route. (McCain) Disease severity increases with the addition of irrigation and fertilization, two very expensive inputs for cotton that should result in yield increases. Producers are selective on the cotton cultivars they plant to combat yield loss; however, there has been little research conducted on cultivar response to *Verticillium*.

Several new cotton cultivars produced by different seed companies are available to be planted in Alabama every year and little is known about the response to a majority of pathogens until they are released and tested in applied field settings. The research performed is important to inform growers of their best cotton cultivar option in heavily infested Verticillium areas.

Methods

Eighteen commonly grown cotton cultivars were planted and evaluated for resistance to *Verticillium dahliae*. The cultivars are listed in Table 1. The trial was planted in a producer's field in Madison County located in northwest Alabama (34N 47' 53.65" 87W 56' 37.93"). The dominate soil types found, were Decatur silt loam (24% sand, 49% silt, 28% clay) and Emory silt loam. Plots were organized to be six rows each, and approximately 500 feet long. The field was irrigated, when needed, with drip tape irrigation system. Disease ratings were taken September 20 when foliar disease symptoms were visible. Ten feet sections of the third row in each plot were evaluated for disease incidence and severity. The total number of plants and stems were cut longitudinally to assess disease incidence, as seen in Figure 1. Disease severity ratings of foliar symptoms were evaluated on a scale from 1-5 with 1 having no foliar wilting, 3 displaying interveinal chlorosis and necrosis of the leaves and 5 being completely defoliated plants (Figure 2). Four replications of each variety were assessed. Petioles were taken from infected plants of each variety and re-isolated to confirm the presence of *Verticillium dahliae.* Yields were taken by hand harvesting ten-foot sections of each cultivar adjacent to the row section where disease ratings were taken.

In addition to the cultivar test a microplot trial was conducted to determine disease severity with and without irrigation in different soil type commonly found throughout the state of Alabama. The microplot test was arranged in a split plot factorial design with six soil types as the main factor and irrigation or dry land condition (natural rainfall) was the secondary factor. This design allowed the interaction of soil type and irrigation effects on Verticillium wilt incidence and severity and the subsequent effect on cotton yield to be determined. The soil types were tested under dryland conditions (natural rainfall events) or irrigated by drip tape applying approximately two liters a day. Each microplot consisted of a 25 liter outdoor plastic tree pot to simulate field conditions. Pots were arranged in a pot in pot design with a brick in between to allow air pruning. Pots were inoculated with 10^7 microsclerotia suspension. Pots were planted with a susceptible cultivar, Croplan 3738 B2RF. Verticillium wilt disease severity ratings were taken near cotton plant maturity with foliar symptoms being evaluated on a scale from 1-5 previously described, and vascular discoloration incidence, plant fresh weights, plant heights, and yields were taken.

Statistical analysis of both the field and microplot trials was done using SAS 9.3 (Copyright © 2013, SAS Institute Inc., Cary, NC, USA). Data was analyzed by Proc Glimmix and Least squares means were compared using Tukey-Kramer HSD (α =0.1). The Pearson-product correlation method was used to examine the relationship among disease incidence, disease severity, and yield.

Figure 1: Vascular discoloration due to *Verticillium dahliae* in the cotton vascular tissues.

Figure 2: Foliar interveinal chlorosis and necrosis.

Results

Disease severity and incidence ratings indicated significant differences between the cultivars tested. The two cultivars with the lowest disease severity rating of the eighteen that were tested were Stoneville (ST) 4747 GLB2 and the resistant check Fiber Max (FM) 1944 GLB2. Both cultivars had average visual rating of less than 2, meaning the plants had slight necrosis and chlorosis of the leaves. The two cultivars that had the highest or most severe ratings were Deltapine Experimental (DPLX) 14R1456 B2R2, DPLX 12R224 B2R2 and Phytogen (PHY) 333 WRF. These cultivars had average scores above 4, indicating the foliage displayed interveinal chlorosis, wilting, and some defoliation. Disease incidence varied between cultivars. There were six cultivars that had less vascular discoloration than the resistant check FM 1944GLB2 which were DPLX 13R352 B2R2, Bayer Experimental (BX) 1534, ST 5032 GLT, Phytogen (PHY) 339 WRF, ST 4747 GLB2, and ST 4946 GLB2. The twelve other cultivars had higher percentages vascular discoloration. Yields indicated significant differences between cultivars. Ranking the cultivars by yield indicates ST 4747 GLB2 and DPLX 14R1455 B2R2 produced the greatest yield. Both cultivars on average increased yield 19% when compared to the resistant check. DeltaPine (DP) 1137 B2RF, ST 4946 GLB2, DPLX 13R352 B2R2 also increased yield on average 5% compared to the check. Comparing the data between disease incidence and severity to find a correlation has shown an interesting result. There is a significant positive correlation (R^2=0.62213; $P \leq 0.0001$) between visual symptoms and the signs of the disease in the vascular system. A correlation between incidence and yield was significant (R^2= -0.49814; $P \leq 0.0001$) indicating a higher disease incidence contributed to a reduced yield by near 50%.

The microplot test indicated significant differences between soil type and irrigation. Irrigation significantly increased disease incidence and severity compared to the non-irrigated soils. Irrigation, as a whole, increased disease across all soil types approximately 121% compared to non-irrigated plots. Visual ratings of the cotton showed a consistence increase in disease among irrigated soils compared to soils which were watered by natural rainfall. Irrigation increased chlorosis and plant height and biomass as seen in Figure 3. Decatur Silt Loam and the Houston Clay soil types had significantly more disease incidence regardless of the irrigation treatment. These soils had the highest clay and silt content and held water thus displayed less moisture stress. These two soil types are our heaviest in the state. The soil type that proved to be the least conducive soil type to Verticillium Wilt was the Hartsells Fine Sandy Loam which had no disease present. There were no significant differences found for the interaction of the soil type and irrigation. Specifically the soil type with the most disease was the irrigated Houston Clay soil. This soil supported 50% more disease than the irrigated Decatur Silt Loam. Soil types that had the least amount of disease present were Hartsells Fine Sandy Loam irrigated, Hartsells Fine Sandy Loam non-irrigated, and Ruston Very Fine Sandy Loam non-irrigated, all these treatments has no disease present. In terms of yields no differences found between soil types or irrigation treatments.

Table 1. Cotton cultivar results for disease severity, disease incidence, and yield

Cotton cultivars		Disease severity**	Disease incidence percentages***	Yield (lb/A)
1	CROPLAN 3738 B2RF	2.9 abc*	87.8 ab	2733 bcde
2	DP 1321 B2RF	2.8 abc	73.3 abc	3122 abcde
3	DP 1133 B2RF	2.9 abc	74.3 abc	2993 abcde
4	DP 1137 B2RF	3.3 abc	83.5 abc	3870 abc
5	DPLX 12R224	4.1 a	89.8 a	2059 e
6	DPLX 13R310	3.1 abc	83.5 abc	2263 ed
7	DPLX 13R352	2.5 abc	66.3 abcd	3672 abcd
8	DPLX 14R1455	2.4 abc	67.0 abc	4048 ab
9	DPLX 14R1456	4.3 a	90.0 a	2047 e
10	PHY 333 WRF	4.0 a	89.3 a	2823 abcde
11	PHY 339 WRF	2.8 abc	50.3 cd	3542 abcde
12	PHY 499 WRF	3.8 ab	73.0 abc	3297 abcde
13	FM 1944 GLB2	1.8 bc	66.5 abcd	3542 abcde
14	ST 4747 GLB2	1.6 c	51.5 cd	4360 a
15	ST 4946 GLB2	2.4 abc	44.3 d	3753 abcd
16	ST 5032 GLB2	2.6 abc	53.0 bcd	2846 abcde
17	ST 6448 GLB2	2.9 abc	73.3 abc	3508 abcde
18	BX 1534	2.9 abc	59.5 abcd	2289 cde
Mean (P < 0.10)		2.9	71.0	3155.47

* Means followed by the same letter do not differ significantly at the 0.1 level of probability.

** Disease severity ratings of foliar symptoms were evaluated on a scale from 1-5 with 1 having no foliar wilting, 3 displaying interveinal chlorosis and necrosis of the leaves and 5 being completely defoliated plants.

*** Disease incidence is the number of stems cut with vascular discoloration.

Figure 3. An aerial view of the microplot test. Plots with a red line beneath indicate natural irrigation and plot with a blue line below indicate irrigation.

Table 2. Cotton Microplot Irrigation Trial Results

Irrigation Type	% Disease Incidence *
1 Irrigated	5.28 a
2 Non- Irrigated	2.38 b

* Means followed by the same letter do not differ significantly at the 0.1 level of probability

Table 3. Cotton Microplot Irrigation Trial Results

Soil Type	% Sand	% Silt	% Clay	% Disease Incidence *
1 Dothan Sandy Loam	57	28	15	12.33 ab
2 Decatur Silt Loam	24	49	28	19.83 a
3 Houston Clay	6.5	48	45	19.50 a
4 Ruston Very Fine Sandy Loam	59	33	8	2.50 ab
5 Hartsells Fine Sandy Loam	56	33	12	0.00 b
6 Lloyd	52	23	25	7.83 ab

* Means followed by the same letter do not differ significantly at the 0.1 level of probability

Summary

The results from the 2014 field trial indicate ST 4747 GLB2 supported the lowest disease severity indicated by foliar wilting and defoliation, and ST 4946 GLB2 had the lowest disease incidence or number of plants with vascular discoloration. The cultivars with low disease severity scores and high disease incidence would be considered tolerant to the disease if they also supported high yields. Two cultivars had the highest yields compared to the other sixteen, ST 4747 GLB2 and DPLX 14R1455 B2R2. The correlation shows that visual symptoms only account for over half the disease present in the field. Cotton cultivars are often taken off the market to make way for new and improved cultivars or to avoid resistance build up; continuing cultivar testing is needed to inform producers of responses to pathogens, particularly such as yield limiting pathogen such as Verticillium wilt.

Understanding environmental and cultural influences that enhance Verticillium Wilt pathogenicity are important when calculating control measures. The addition of water enhanced the pathogenicity of disease. Irrigation is an expensive input that could potentially enhance disease causing greater yield losses. Heavier soils, or soils with higher silt and clay content, were also found to have greater disease infection compared to lighter sandier soils. This is consistent with where infection occurs in the state. Understand what growers may be at higher risk will help provide better control strategies for the prevention of the difficult disease.

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