# ASSESSMENT OF YIELD LOSS ATTRIBUTED TO COTTON LEAFROLL DWARF DISEASE IN ALABAMA COTTON

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## Abstract

Cotton leafroll dwarf disease (CLRDD), caused by *Cotton leafroll dwarf virus* (CLRDV), is an emerging disease in U.S. cotton. Impact of CLRDD on the lint yield was assessed in 2019 and 2020 at Alabama Agricultural Experiment Stations (AAES) in Southwest, Central and North East Alabama along with two production fields in Baldwin Co. To establish CLRDD incidence, apex mature leaves on the central leader were collected from symptomatic and asymptomatic plants and tested for CLRDV using PCR. Plant height, along with the numbers of open, unopened, locked and rotted, counts of terminal nodes above the last mature boll, fruiting, and total nodes, as well as stand counts were recorded. Bolls from plants were hand harvested and weighed. At SMREC in 2019, plots were mechanically harvested to determine seed cotton yield. In 2019 at the Northeast AL site, CLRDV+ (positive) infected PhytoGen 440 W3FE plants were shorter, produced fewer bolls and fruiting nodes, and had lower seed yield with an estimated yield loss was 117 lb/A or 10.8% of anticipated yield. At the Central AL site, accentuated verticality was noted on 14.3% plants of Deltapine 1646 B2XF, all of which tested positive for CLRDV. Significantly greater counts for total and additional terminal nodes but with fewer total bolls, fruiting nodes, and a reduced seed yield of 41 lb lint/A or 4.3% of expected yield were recorded for the plants displaying accentuated verticality compared with the CLRDV- (negative) plants with normal terminal growth. For NexGen 5007 B2XF in the production field in Baldwin Co., plants displaying accentuated verticality, comprising 33.1% of the total stand, had greater terminal node counts compared with asymptomatic plants, which produced more bolls and fruiting nodes, along with greater seed yield. Here, estimated yield was 512 lb lint/A or 22.8% of anticipated yield. Finally, all symptomatic and asymptomatic plants sampled were (100%) CLRDV+. While plant height and total node counts were greater, yield losses attributed to CLRDD were not observed in the second producer field in Baldwin Co. For 2020, CLRDD incidence was lower statewide than in the preceding year. At a Central AL site, symptomatic CLRDV+ PhytoGen 480 W3FE plants were taller and had greater total, but not fruiting, and additional terminal node counts. Individual plant yield was also similar for the CLRDV+ and CLRDV- plants. When comparing CLRDV+ and CLRDV- PhytoGen 500 W3FE plants in southwest AL, significantly greater total node but not fruiting and terminal node counts along with individual plant yields were recorded. With a significant PCR test × Sampling date interaction, CLRDV- plant(s) sampled at 2 September produced significantly greater yields compared with CLRDV+ plants collected on that same date, but results are questionable due to low sample numbers. Yield for the 2 September CLRDV+ plants along with both the CLDRV+ and CLRDV- plants sampled on 20 August were similar. Overall, survey results, particularly from 2019, indicate that CLRDD is associated with sizable losses in cotton yield.

## Introduction

*Cotton leafroll dwarf virus* (CLRDV) (genus *Polerovirus*, family *Luteovirdae*) is the causal agent of cotton blue disease (Correa et al. 2005), which is vectored to cotton primarily by the cotton aphid (*Aphis gossypii* Glover) (Cauquil and Vaisayre 1971). First reported in the Central African Republic in 1949, cotton blue disease is widely distributed across sub-Saharan and central Africa (Cauquil 1977) as well as Brazil (Costa and Carvalho 1962; Correa et al. 2005) and Argentina (Campagnac et al. 1986) where two CLRDV strains, typical and atypical occur (Silva et al. 2015;

Agrofoglio et al. 2017). In Brazil over the 2013 to 2015 production seasons, Galbieri et al (2017) reported that the atypical strain accounted for 97.3% of the CLRDV isolated from Fibermax 975WS. Recently, CLRDV has also been detected in symptomatic cotton in India (Mukherjee et al 2012), Thailand, (Sharman et al 2015) and Timor-Leste (Ray et al 2016).

A new strain of the cotton leafroll dwarf virus, CLRDV-AL, which causes cotton leafroll dwarf disease (CLRDD), was recovered from cotton displaying leaf distortion, curling, rolling along with bluish green discoloration, vein-clearing, and shortened internodes in the fall 2017 in Alabama (Avelar et al. 2019) where disease incidence, based on the occurrence of accentuated verticality symptom pattern, approached 100% in a few southwest Alabama production fields in 2018 (Hagan et al. 2019). Subsequently, CLRDV-AL was identified in symptomatic cotton in Florida (Iriarte et al. 2020), Georgia (Tabassum et al. 2019), Kansas (Ali and Mokhatari 2020), Louisiana (Price et al. 2020), Mississippi (Aboughanem-Sabanadzovic et al. 2020), North Carolina (Thiessen et al. 2020), Oklahoma (Ali et al. 2020), and Texas (Alabi et al. 2020).

For cultivars susceptible to the typical CLRDV strain, yield losses, which are attributed to severe stunting along with reduced bloom set and boll size, shedding of immature squares and bolls, have been estimated at 68% (Santos et al. 2004) to 80% (Silva et al. 2008) compared with a resistant cultivar such as Delta Opel. Yield losses of 13.4 to 21.5% due to the atypical CLRDV strain have been reported for cultivars resistant to the typical strain of the same virus (Galberi et al. 2017). Galberi et al. (2017) also noted that cultivars from the U.S. and Australia are highly susceptible to cotton blue disease. Based on a CLRDD incidence of 3 to 30%, Avelar et al. (2019) estimated yield reduction of 560 kg/ha over 25% of Alabama's 2018 cotton acreage, which translates into a statewide farm-gate income loss of $19 million. Trials were conducted in 2019 and 2020 at AAES research units, as well as in production fields, to assess the impact of CLRDD on the plant growth and yield-related parameters of cotton in Alabama.

## Production Methods

In 2019, Deltapine 1646 B2XF at the Plant Breeding Unit (PBU) in Tallassee, AL, and PhytoGen 440 W3FE at the Sand Mountain Research and Extension Center (SMREC) in Crossville, AL, along with production fields of NexGen 5007 in Malbis and Belforest (Baldwin Co.), AL, were sampled PhytoGen 500 W3FE and Stoneville 5471 GLTP at Brewton Agricultural Research Unit (BARU) and PhytoGen 480 W3FE at the Prattville Agricultural Research Unit (PARU) in Prattville, AL were sampled in 2020. Recommendations of the Alabama Cooperative Extension System for fertility along with insect and weed control, canopy management, and harvest preparation were followed at all locations. The BARU, PBU, PARU, and SMREC study sites were irrigated as needed. The experimental design was a randomized complete block with each plant sampled as an individual experimental unit except for the Sentinel Trial at BARU where a split plot design with planting date as the main plot and cultivar as the split plot treatment. To establish CLRDD incidence, an apex mature leaf on the central terminal was collected from symptomatic and asymptomatic plants at each location and tested for CLRDV using PCR. Plant height, numbers of open, unopened, locked, and rotted bolls, which are presented as total bolls along with counts of fruiting, terminal nodes above the apex mature boll, and total nodes, as well as stand counts were recorded. Bolls from individual plants were hand harvested and weighed. At SMREC in 2019 and BARU in 2020, plots were mechanically harvested to determine seed cotton yield. In the study at BARU and PARU in 2020, significance of PCR result × sampling date and planting date × PCR result interactions, respectively, were determined using PROC GLIMMIX in SAS. Statistical analyses were done on rank transformations for non-normal values. Non-transformed data are reported. Means were separated using Fisher's protected least significant difference (LSD) test ($P \leq 0.05$).

## Results

In 2019 at SMREC in Northeast AL, the CLRDV+ (positive) PhytoGen 440 W3FE plants displaying node stacking in the terminal shoots along with maroon discoloration of the apex mature leaves were significantly shorter in stature, produced fewer bolls and fruiting nodes along with lower individual plant yield compared with the taller and more productive CLRDV– (negative) plants (Table 1). Incidence of symptomatic plants was 15%. Estimated yield loss was 105 kg/ha or 10.8% below estimated yield in the absence of CLRDD of 972 kg/ha.

For the Central AL PBU site, 14.3% of Deltapine 1646 B2XF planted in early June displayed the accentuated verticality symptom pattern associated with CLRDD, all of which were CLRDV+ in mid-November. In addition, 2.4% of plants, which were stunted with limited or no bolls set, also tested positive for this virus. While plant height

along with counts of nodes above the apex boll and total nodes were greater for the CLRDV+ plants, counts of total bolls and fruiting nodes were significantly reduced (Table 1). Estimated yield, which was calculated from the mean of four stand counts taken in each of the four sampled blocks, was 883 kg/ha for the CLRDV- compared with 846 kg/ha for the symptomatic and CLRDV+ cotton for an estimated yield loss of 37 kg/ha or 4.3%.

At the Belforest production field in Baldwin Co in Southwest AL, accentuated verticality was recorded in mid-November 2019 on 33.1% for early-June planted NexGen 5007 B2XF plants, which were significantly taller with greater counts of nodes above the apex mature boll but fewer total bolls and fruiting nodes (Table 1). Individual plant yield for the symptomatic and asymptomatic plants was 33.6 and 107.6 g, respectively, which translates into an estimated yield loss of 461 kg/ha or 22.8%. All symptomatic and asymptomatic plants tested positive for CLRDV.

For the production field located in Malbis, AL in Baldwin Co., accentuated verticality was the diagnostic CLRDD symptom observed. Symptomatic plants were significantly taller with greater total node counts compared with plants with a 'normal' growth pattern (Table 1). Similar individual plant yield was recorded for both the symptomatic compared with asymptomatic plants.

Table 1. Impact of CLRDD as indicated by CLRDV virus presence or diagnostic symptom patterns on plant dimensions, growth, boll set, and productivity of individual cotton plants at four survey sites across Alabama in 2019.

| Visual symptoms | Plants Sampled | CLRDV PCR | Plant height | Total bolls | Fruiting Nodes | Nodes above mature apex boll | Total nodes | Individual plant yield |
|---|---|---|---|---|---|---|---|---|
| Survey site | # | +/- | M | # | # | # | # | g |
| SMREC | | | | | | | | |
| Stunting | 9 | + | 1.08 b [z] | 17.7 b | 11.9 b | --- | --- | 33.6 b |
| Normal | 15 | - | 1.24 a | 34.6 a | 15.2 a | --- | --- | 107.6 a |
| | | | | | | | | |
| PBU | | | | | | | | |
| Accentuated Verticality | 24 | + | 1.32 a | 10.3 b | 8.7 b | 5.6 a | 24.6 a | 29.4 b |
| Normal | 36 | - | 1.26 b | 15.6 a | 11.0 a | 1.8 b | 20.5 b | 41.8 a |
| | | | | | | | | |
| Belforest | | | | | | | | |
| Accentuated Verticality | 21 | + | 1.03 a | 8.8 b | 6.6 b | 8.3 a | 21.5 a | 33.6 b |
| Normal | 15 | + | 0.90 b | 27.6 a | 13.3 a | 2.1 b | 20.6 a | 107.6 a |
| | | | | | | | | |
| Malbis | | | | | | | | |
| Accentuated Verticality | 15 | --- | 1.33 a | 31.3 a | 15.9 a | --- | 25.0 a | 122 a |
| Normal | 26 | --- | 1.23 b | 37.9 a | 17.2 a | --- | 21.3 b | 145 a |

[z] Means followed by the same letter are not significantly different according to Fisher's protected least significant difference (LSD) test ($P \leq 0.05$).

At the PARU in 2020, PhytoGen 480 W3FE was significantly taller with greater numbers of fruiting and total nodes as well as individual plant yield when sown at the May1 than 1 June planting date (Table 2). Nodes above the apex boll and open boll counts (data not shown) recorded at both planting dates were similar. In addition, CLRDV+ plants were taller with greater numbers of total nodes, which is an indication of accentuated verticality, but fruiting node and nodes above the apex boll along with individual plant yield did not significantly differ by PCR reaction. For PhytoGen 480 W3FE % gin out and lint yield values were 42.5% and 207 kg lint/ha.

At BARU, two tropical storms, which occurred after cut out, separated the lint and seed from the bolls, thereby delaying harvest by nearly a month and causing loss of 35 to 40% below historical yields. Total nodes but not fruiting node or nodes above the apex boll counts were greater for the CLRDV+ compared with CLRDV- PhytoGen 500 W3FE (Table 3). Also, these same variables did not differ by the date mature apex leaves were collected for PCR analysis for CLRDV. Individual plant yield did differ significantly by CLRDV status and sampling date. Greatest yield was noted for CLRDV- plants sampled at 2 September as compared with 20 August as well as the CLRDV+ plants sampled at both the above dates. Yield for PhytoGen 500 W3FE was 877 kg lint/ha.

Table 2. Impact of CLRDV on plant height, growth, and individual plant yield of PhytoGen 480 W3FE at the Prattville Agricultural Research Unit in 2020.

| Source of Variation | Plants sampled | Plant height | Nodes | | | Yield /plant |
|---|---|---|---|---|---|---|
| | | | Fruiting | Above apex boll | Total | |
| | # | m | # | # | # | g |
| Planting date | --- | <0.0001*** | 0.0053** | 0.0034** | 0.3625 | 0.0015** |
| CLRDV | --- | 0.0119* | 0.0644^ | 0.3315 | 0.4038 | 0.7299 |
| Planting date × CLRDV | --- | 0.8971 | 0.2499 | 0.7037 | 0.8753 | 0.6459 |
| | | | | | | |
| Planting date | | | | | | |
| May 1 | 15 | 1.22 a | 10.0 a | 4.9 a | 23.6 a | 34.3 a |
| June 1 | 41 | 1.05 b | 7.5 b | 5.5 a | 21.4 b | 21.7 b |
| | | | | | | |
| CLRDV Status | | | | | | |
| + | 32 | 1.17 a | 9.1 a | 5.5 a | 23.2 a | 28.7 a |
| - | 24 | 1.09 b | 8.4 a | 4.9 a | 21.8 b | 27.4 a |

[z] Significance of *F* values at the 0.10, 0.05, 0.01, and 0.001 levels is indicated by ^, *, **, or ***, respectively.
[y] Means in each column followed by the same letter are not significantly different according to Fisher's protected least significant difference (LSD) test ($P \leq 0.05$) unless otherwise indicated.

Table 3. Impact of CLRDV on plant height, growth, and individual plant yield for PhytoGen 500 W3FE at the Brewton Agricultural Research Center in 2020.

| Source of Variation | Plants sampled | Plant height | Nodes | | | Yield /plant |
|---|---|---|---|---|---|---|
| | | | Fruiting | Above apex boll | Total | |
| | # | m | # | # | # | g |
| CLRDV | --- | 0.3819 | 0.0794^ | 0.3866 | 0.1657 | 0.1522 |
| Sampling date | --- | 0.2274 | 0.9172 | 0.9743 | 0.9230 | 0.1068 |
| Sampling date × CLRDV | --- | 0.4518 | 0.1480 | 0.1919 | 0.3994 | 0.0581^ |
| | | | | | | |
| CLRDV Status | | | | | | |
| + | 35 | 1.10 | 10.5 a | 4.4 a | 21.0 a | --- |
| - | 12 | 1.16 | 11.9 a | 2.5 a | 16.8 b | --- |
| | | | | | | |
| Sampling date | | | | | | |
| 20 August | 41 | 1.08 a | 11.2 a | 3.4 a | 19.0 a | --- |
| 2 September | 6 | 1.17 a | 11.2 a | 3.5 a | 18.8 a | --- |
| | | | | | | |
| CLRDV × Sampling date | | | | | | |
| CLRDV- 20 August | 11 | --- | --- | --- | --- | 37.1 b |
| CLRDV+ 20 August | 30 | --- | --- | --- | --- | 42.7 b |
| CLRDV- 2 September | 1 | --- | --- | --- | --- | 77.8 a |
| CLRDV+ 2 September | 5 | --- | --- | --- | --- | 39.3 b |

[z] Significance of *F* values at the 0.10, 0.05, 0.01, and 0.001 levels is indicated by ^, *, **, or ***, respectively.
[y] Means in each column followed by the same letter are not significantly different according to Fisher's protected least significant difference (LSD) test ($P \leq 0.05$) unless otherwise indicated.

## Summary

While CLRDD is distributed throughout all U.S. cotton producing states except for Arizona and California, the impact of this disease along with Cotton Blue Disease in Brazil and Argentina on lint yield and quality factors is not well documented. Since first documented in U.S. cotton, southwestern Alabama is the epicenter of CLRDD with

accentuated verticality symptoms displayed by 100% of plants observed in multiple cotton fields in several central Baldwin Co communities (Hagan, personal observation). In that area of Alabama, disease incidence, as indicated by positive PCR tests, was nearly 100% in 2019 compared with approximately 50% at multiple south and central AL sites in 2020; thus, a sharp decline in CLRDD occurred in the latter than the former production year. In Brazil, incidence of cotton blue disease with the atypical CLRDV strain is positively correlated with populations of the aphid vector, while a negative correlation was noted between yield and aphid populations (Galbieri et al. 2017).

In Brazil and Argentina, Santo et al. (2004) and Silva et al. (2008) reported yield losses of 68 and 80%, respectively, to Cotton Blue Disease incited by the typical strain of CLRDV. While losses of that magnitude have not been reported in the U.S., Avelar et al. (2019) estimated a yield reduction of 560 kg/ha over 25% of Alabama's 2018 cotton acreage, which translates into a statewide farm-gate income loss of $19 million. Estimated yield loss to CLRDD in 2019 and particularly 2020 was much lower compared with 2018 (Hagan, personal observation).

Here, an attempt was made to define the potential for yield loss due to CLRDD in cotton. In 2019, disease related yield loss as indicated by CLRDV status was 4.3% for Deltapine 1646 B2XF in Central AL and 10.8% for PhytoGen 440 W3FE in Northeast AL. Unfortunately, the cotton was not ginned and lint quality traits were not recorded. In 2020 at BARU, a yield loss was noted for CLRDV+ compared with CLRDV- plants at the 2 September sampling dates. However, the number of plants sampled (5 CLRDV+ and 1 CLRDV-) was small resulting in questionable results. Finally, a sizable 33.1% yield loss for plants displaying accentuated verticality compared with a normal terminal growth pattern in Belforest, AL was recorded at a location near those where 100% accentuated verticality symptoms were noted the previous year. Notably, all plants sampled, regardless of symptom pattern, were CLRDV positive. Such a result is not surprising given that the cotton aphid (*A. gossypii*) vector was widely distributed in Alabama cotton into mid-November and virus transmission will likely continue until the cotton is defoliated. Infection timing often has a sizable impact on the impact of CLRDV-incited diseases on plant growth and ultimately yield (Agrofoglio et al. 2017). Prior to pin head square, cotton infected with the typical strain of CLRDV is severely stunted with severely deformed leaves and no boll set (Sharman. 2015), while plants infected during flowering and early boll set are symptomatic only on leaves in the shoot terminals (Agrofoglio et al. 2017). For mature plants infected during boll maturation, such as those at production field in Belforest, impacts on yield are likely minimal.

Over the past three years, CLRDD has emerged as a widespread and potentially damaging disease in Alabama cotton. While disease incidence declines from southwest to north Alabama, yield losses were recorded at a northeast and central Alabama sites in 2019. To date, however, CLRDD incidence has been negligible in sentinel trials in the Tennessee Valley, a major cotton production region in Alabama. The risk of significant yield loss is greatest in late May- or early June- planted cotton in Baldwin, Escambia, Mobile, and Monroe counties in southwest Alabama. In the absence of resistant cultivars, recommended control strategies should focus on cotton stalk destruction, and winter weed control in and around fields slated to be cropped to cotton, and early planting (Hagan et al. 2019).

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