**Full Length Research Paper**

**Influence of boll sampling method and water stress on fiber quality of irrigated cotton** (*Gossypium hirsutum* L.)

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The quality of cotton fibers in Brazil has been studied. This was done by studying the fiber samples obtained from bolls removed from the middle third of the plants. These fiber samples are referred to as “standard sample”. This way to collect fiber data requires lots of labor, and may disguise the results obtained in experimental appraisals, due to human errors in gathering boll. Besides, cotton yield and quality is influenced by water availability, especially during abiotic tests with water deficit, in which, fiber quality samples may be affected by boll position. Thus, the objective of this study was to evaluate the influence of sampling method on the technological characteristics of cotton fibers, in irrigated and water stress tests at different stages of the crop cycle. Two methods were made to collect cotton fiber. The first method was the standard sample and the second way was gathering sample of bolls in randomized position, through all experimental plot (called randomized sample). The results show that the analysis performed by standard sample tend to overestimate the values of the fiber quality parameters, differing from the results obtained with the randomized sample that is representative of all plot. It was observed that the variability of cotton fiber quality affected by water stress treatments were best represented using bolls obtained by randomized method. Consequently, in the case of experiments with water stress, the most representative method to collect cotton fiber, is through a sampling of all the plant, and not only of the middle third.

**Key words:** Boll position, cotton fiber, HVI, standard sample, water stress.

**INTRODUCTION**

Cotton profitability relies on both yield and quality of cotton fiber, and also depends on the interaction of several factors, such as crop management, environment factors and genetics.

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Beltrão and Azevedo (2008) affirmed that cotton fiber is mainly conditioned by hereditary factors, although some technological characteristics are decisively influenced by environmental factors and crop management. Thus, the cultivars selection in breeding programs is an important activity, in which crop behavior, conditioned by environmental features must be taken into account during evaluation.

Whilst cotton fiber yield is easily quantified, fiber quality is a complex parameter (Bradow et al., 1997). Measurements of fiber quality are complicated due to natural and environmental variations in fiber structure and maturity. These variations may occur both in bale, plant, boll or seed. Thus, this is essential for breeding programs to make progress in quality improvements, and the results obtained from the research are reliable. Uniformity of the sample collected for analysis represents the real condition of the fiber quality of any experimental area or plot (Bradow et al., 1997).

Fiber quality analysis in Brazil are done using a “standard sample” from each plot, in which bolls are harvested from the middle third of the plants, and, in this case not representative of all experimental plot, considering that plants may suffer any stress, after or during boll formation. Therefore, this methodology can generate erroneous estimates of the cotton fiber yield or characteristics of the quality fiber measure by High Volume Instruments (HVI) (Belot and Dutra, 2015; Kelly et al., 2015).

The standard sample consists of harvesting 20 bolls of the middle third of the plants, which may mask research results. According to Belot and Dutra (2015), comparing the boll of top and lower position with the middle third of the plants, great discrepancies in some characteristics such as micronaire, maturity and percentage of fibers were found. These differences occur probably due to complexes interactions among soil properties, soil water and nutrients availability and plant populations (Bradow et al., 2000). So, the use of standard methodologies in research, harvesting only in specific positions, do not allow safety evaluation of the cotton fiber quality produced in field (Belot and Dutra, 2015).

This problem can be further aggravated in the case of tests with abiotic stresses, such as water stress, since this may occur at different stages of the crop cycle, disturbing the cotton fiber formation with consequent changes in quality. These changes depend on the fruit positions at the time of the water deficit. Thus, ideally, samples should be taken to represent all the fruiting points of the plant, and not only those of the middle third.

The objective of the present study was to evaluate the influence of the sampling methodology on the characteristics of cotton fiber quality to cultivars under irrigation system, with and without water stress, in the Brazil's semi-arid region.

### MATERIALS AND METHODS

The experiment was carried out from June to November 2015, at the Experimental Farm of the Agricultural Research Company of Rio Grande do Norte (EMPARN), located in Apodi, RN. Experimental area have central geographical coordinates of 5°37'19"S and 37°49'06"W, with altitude range of 128 to 132 m.

The climate of the region is characterized as hot and semi-arid tropical, with predominance of BSwh't type, according to Köppen's climatic classification. The soil of the experimental area was classified as eutrophic Cambisol (Santos et al., 2006), clay-sandy texture, with 49% sand, 45% clay and 6% silt. It was used no-tillage system with a 3 row mechanized sowing machine, and no thinning was required. Fertilization was performed according to the technical recommendations for the crop, based on soil fertility analysis (Table 1).

The experiment was carried out in a randomized complete block design with split plot arrangement, and with water deficit periods in main plot. Cotton cultivars were in subplots, and sampling methods in sub-subplots with four replications. In the plots, treatments consisted of 6 periods of water deficit, named:

1. Initial (IN)
2. Floral bud (FB)
3. Early bloom (EB)
4. Peak bloom (PB)
5. Open bolls (OB) and
7. Control without water deficit (IR).

Cotton cultivars were:

1. BRS 286
2. BRS 335
3. BRS 336
4. BRS 372
5. BRS 368RF
6. BRS369RF
7. BRS370RF and
8. BRS 371RF

Sub-subplots sampling methods: the standard sample (SS) and plot sample (PS). The standard sample consisted of 20 bolls harvested in the middle third of plants, while the plot sample consisted of 100 g of cotton fiber, randomly collected from the all experimental plot, harvested in different sample positions.

Each experimental unit consisted of 4 spaced rows of 0.8 and 6.0

### Table 1. Soil chemical characteristics at depth of 0-40 cm, in experimental area of Apodi, RN.

<table>
<thead>
<tr>
<th>Year</th>
<th>pH water</th>
<th>OM (g kg⁻¹)</th>
<th>P (mg kg⁻¹)</th>
<th>Na⁺</th>
<th>K⁺</th>
<th>Ca²⁺</th>
<th>Mg²⁺</th>
<th>H + Al</th>
<th>CEC</th>
<th>BS</th>
</tr>
</thead>
<tbody>
<tr>
<td>2014</td>
<td>6.20</td>
<td>16.4</td>
<td>10.7</td>
<td>0.4</td>
<td>1.6</td>
<td>34.8</td>
<td>10.0</td>
<td>23.1</td>
<td>69.9</td>
<td>46.8</td>
</tr>
</tbody>
</table>

(OM) - organic matter; CEC - cation exchange capacity, BS – Base Sum.
Figure 2. Water deficit period in each treatment.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Start of water suppression</th>
<th>Net irrigation depth (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial (IN)</td>
<td>After stand establishment</td>
<td>650</td>
</tr>
<tr>
<td>Floral Bud (FB)</td>
<td>Beginning with the first flower bud at least in 10% of the plants</td>
<td>634</td>
</tr>
<tr>
<td>Early Bloom (EB)</td>
<td>Opening of first flower at least in 10% of the plants</td>
<td>577</td>
</tr>
<tr>
<td>Peak Bloom (PB)</td>
<td>Boll loading. At least 10% of plants heavily fruited where first bolls were completely full</td>
<td>584</td>
</tr>
<tr>
<td>Open Boll (OB)*</td>
<td>Opening of the first bolls in 10% of the plants</td>
<td>621</td>
</tr>
<tr>
<td>Without Water Stress (IR)</td>
<td>Without deficit irrigation during all crop cycle.</td>
<td>700</td>
</tr>
</tbody>
</table>

*After treatment, cotton plants did not receive water anymore, since crop cycle was in conclusion.

Table 2.

Table 3. Agronomic data and irrigation parameters during cotton crop cycle.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Period</th>
</tr>
</thead>
<tbody>
<tr>
<td>Planting date</td>
<td>30/06/2015</td>
</tr>
<tr>
<td>Line space</td>
<td>0.8 m</td>
</tr>
<tr>
<td>Planting density</td>
<td>8-12 plants m⁻¹</td>
</tr>
<tr>
<td>Fertilization at planting</td>
<td>150 kg ha⁻¹ of P₂O₅ and 30 kg of N (MAP* form)</td>
</tr>
<tr>
<td>Topdressing</td>
<td>150 kg of N ha⁻¹ (Urea)</td>
</tr>
<tr>
<td>Last irrigation</td>
<td>21/10/2015 (106 DAE)</td>
</tr>
<tr>
<td>Harvest date</td>
<td>17/11/2015</td>
</tr>
<tr>
<td>Crop cycle duration</td>
<td>131 days</td>
</tr>
<tr>
<td>Total rainfall in season</td>
<td>0.0 mm</td>
</tr>
</tbody>
</table>

*MAP – Monoammonium phosphate.

RESULTS AND DISCUSSION

Results of the variance analysis to lint percentage (%Lint), and to fiber quality characteristics as length (UHM), uniformity (UNF), short fiber index (SFI), strength (STR), elongation (ELG), micronaire index (MIC) maturity (MAT), reflectance (Rd) and yellowing degree (+b) are shown in Table 4.

To accomplish the influence of the sampling methodology on cotton fiber quality, the discussion focused on the sample factor and its interaction with water deficit and cultivars. Observing the results presented in Table 4, it can be noted that the sampling method had no influence on the data of UNF and SFI. For the interaction between the factors, considering the water deficit versus sampling, the interaction was not significant for the STR and ELG parameters, while the cultivar versus sampling interaction was significant only for the UHM data. These results proved that there is variation in cotton fiber quality within the same plant, depending on the boll position, as discussed by several authors, such as Bradow and Davidonis (2000), Bauer et al. (2009) and Feng et al. (2011).

Thus, if the bolls are harvested bolls just in the middle third of the plants, the result of fiber quality analysis can be masked, not representing the real condition of the plot, agreeing with the study of Belot and Dutra (2015). Additionally, for experiments or field appraisal, in which cotton plants endured abiotic stresses, water deficit is the best way to estimate cotton fiber quality in harvesting the whole plant.
Table 4. Mean squares for lint percentage and fiber quality characteristics evaluated as a function of water deficit, cultivars and sampling method, Apodi, 2015.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>GL</th>
<th>% Lint</th>
<th>UHM</th>
<th>UNF</th>
<th>SFI</th>
<th>STR</th>
<th>ELG</th>
<th>MIC</th>
<th>MAT</th>
<th>Rd</th>
<th>+b</th>
</tr>
</thead>
<tbody>
<tr>
<td>Block</td>
<td>3</td>
<td>3.37*</td>
<td>1.41</td>
<td>0.39**</td>
<td>0.24</td>
<td>2.28</td>
<td>0.32</td>
<td>0.36</td>
<td>0.0002</td>
<td>0.96</td>
<td>0.32</td>
</tr>
<tr>
<td>Déficit (D)</td>
<td>5</td>
<td>10.86**</td>
<td>30.2**</td>
<td>54.83</td>
<td>18.7**</td>
<td>50.17**</td>
<td>0.52*</td>
<td>7.03**</td>
<td>0.04*</td>
<td>27.44**</td>
<td>9.57**</td>
</tr>
<tr>
<td>Residue a</td>
<td>15</td>
<td>0.91</td>
<td>1.03</td>
<td>0.66**</td>
<td>0.29</td>
<td>2.90</td>
<td>0.17</td>
<td>0.35</td>
<td>0.0002</td>
<td>3.64</td>
<td>0.34</td>
</tr>
<tr>
<td>Cultivar (C)</td>
<td>7</td>
<td>146.6**</td>
<td>113.91**</td>
<td>19.03</td>
<td>6.75**</td>
<td>108.41**</td>
<td>0.91**</td>
<td>7.03**</td>
<td>0.04*</td>
<td>27.44**</td>
<td>9.57**</td>
</tr>
<tr>
<td>C × D</td>
<td>35</td>
<td>3.29**</td>
<td>1.4</td>
<td>1.64</td>
<td>0.79**</td>
<td>4.68</td>
<td>0.33*</td>
<td>0.68**</td>
<td>0.0004**</td>
<td>3.16</td>
<td>0.55*</td>
</tr>
<tr>
<td>Residue b</td>
<td>126</td>
<td>1.57</td>
<td>1.42</td>
<td>1.29</td>
<td>0.28</td>
<td>4.02</td>
<td>0.18</td>
<td>0.19</td>
<td>0.0001</td>
<td>2.58</td>
<td>0.34</td>
</tr>
<tr>
<td>Sampling (S)</td>
<td>1</td>
<td>4.49*</td>
<td>41.81**</td>
<td>1.26</td>
<td>0.58</td>
<td>56.51**</td>
<td>0.82*</td>
<td>8.28**</td>
<td>0.006**</td>
<td>86.29**</td>
<td>24.9**</td>
</tr>
<tr>
<td>D × S</td>
<td>5</td>
<td>4.14**</td>
<td>7.87**</td>
<td>2.98*</td>
<td>0.91*</td>
<td>2.91</td>
<td>0.24</td>
<td>0.78**</td>
<td>0.0005**</td>
<td>9.02**</td>
<td>1.42**</td>
</tr>
<tr>
<td>C × S</td>
<td>7</td>
<td>1.31</td>
<td>2.13*</td>
<td>0.94</td>
<td>0.50</td>
<td>2.73</td>
<td>0.11</td>
<td>0.16</td>
<td>0.0001</td>
<td>2.75</td>
<td>0.19</td>
</tr>
<tr>
<td>D × C × S</td>
<td>35</td>
<td>1.28*</td>
<td>0.86</td>
<td>1.77*</td>
<td>0.47</td>
<td>3.76</td>
<td>0.28</td>
<td>0.13</td>
<td>0.00008</td>
<td>2.13</td>
<td>0.26</td>
</tr>
<tr>
<td>Residue c</td>
<td>144</td>
<td>0.79</td>
<td>0.79</td>
<td>1.22</td>
<td>0.32</td>
<td>3.02</td>
<td>0.20</td>
<td>0.09</td>
<td>0.0001</td>
<td>2.67</td>
<td>0.28</td>
</tr>
</tbody>
</table>

** and * Significant at 1 and 5% of probability, respectively.

Figure 1. Lint percentage (A) and fiber length (B) as a function of water stress and sampling methods for cotton fiber quality.

Environmental variation that occurs within the plant canopy, between plants and between plots, causes great variability in fiber quality characteristics, not only at boll level, but also among plants and plots (Bauer et al., 2009; Bradow and Davidonis, 2000; Feng et al., 2011). In this way, the more uniform and representative the conditions of the plant and the plot as a whole is with the sampling, the more representative the results of the fiber quality analysis will be.

The cotton fruits generally develop rapidly up to 16 days after the anthesis (DAA), reaching their maximum size approximately 24 DAA, being mature and open between 40 and 60 days after the anthesis (Kim, 2015). When water stress is applied in diverse phases of the crop phenological cycle, the stress will affect the bolls differently, depending on the stage of the fiber formation. Thus, for the determination of the fiber quality in tests of water stress, the most indicated is to gather boll samples representing all plant positions, in order to avoid results with mistaken estimates. Figures 1 to 5 show the values of the cotton quality parameters of the evaluated fibers. It is observed that the lint percentage (Figure 1A), determined from the SS, was underestimated just when water deficit occurred at the early flowering, being overestimated in the other treatments when the analysis was performed based the SS gathering. This is due to the fact that in the standard sampling method, only the middle third of the plants are selected, excluding the bolls of top and bottom, and so, inadequate for assessments in water stress treatments, on which lint percentage may be affected depending on the phenological phase of the harmful stress.
Belot and Dutra (2015) found that the lint percentage is higher in the middle third of the plants, when compared with the bottom position, and lower in relation to those in the upper position. Thus, for treatment with early flowering period of water stress application, the bolls of the middle third were the most affected, and the values of lint percentage were underestimated when compared to the whole plant data in this phase.

Several authors such as Wen et al. (2013), Brito et al. (2011), De Tar (2008) and Pettigrew (2004) have shown that cotton is influenced both in yield and lint percentage, as fiber quality when submitted to irrigation with water deficit. Beltrão and Azevêdo (2008) stated that cotton fiber is mainly conditioned by hereditary factors, although some technological characteristics are decisively influenced by environmental factors as temperature, luminosity or water availability, and also depend on the crop management. Hence, the sampling performed in an unrepresentative manner of the applied treatments can lead to errors in the interpretation of the results.

Similar overestimation behavior can be observed for the fiber length parameter (Figure 1B). Results showed that, except to data collected through SS in the treatment with water deficit during early flowering, the remaining data were overestimated for this sampling method. Possibly, in this phase where the water stress was applied, bolls in the middle third were being formed and, therefore, they had a greater influence of the applied treatments, which demonstrates the importance of the data collection of the whole plant for fiber analysis, especially when crop was submitted to water stress. The period of fiber formation, according to Abidi et al. (2010), occurs within 3 weeks after the anthesis, thus periods of water stress in this phase can compromise the length of the fibers formed in these bolls.

Considering the fiber length overestimation in the other treatments to data acquired by SS method, the result is due to the fact that, according to Kelly et al. (2015), the fiber length values vary according to boll position in the plant, being larger in the middle and bottom thirds, and lower in the upper positions. Consequently, when merely collecting the samples of the middle third, the values tend to display overestimation in relation to samples of the whole plant that are more representative.

It is also observed in Figure 1B more accentuated overestimation to irrigated treatments or for treatments with water deficit in the initial phase (IN) and floral bud (FB) stages, because water stress did not occur or was less severe in the fruit formation phase. Less accentuated overestimation was observed to treatments where stress occurred in the filling and opening boll periods, since stress happened in the phase of fruits formation, and probably affecting fruits that were being formed in others parts of the plant, like the top positions (Figure 2).

Fiber uniformity is presented in Figure 2A. This characteristic was less influenced by standard sampling method, with changes observed only when water stress was applied in the peak bloom (boll filling stage). Another characteristic that affected SS method was the short fiber index (Figure 2B), this was influenced by the sampling method that is well irrigated in the control treatment, and to application of water stress in the peak bloom (boll filling phase), showing values overestimated and underestimated, respectively. The fiber analysis results were affected by sampling method, since it is completely irrigated cotton, that is the best condition, the SS sampling method conferred an overestimation of values, due to the collection being performed in the middle third of the plants.

In contrast, the most sensitive phase to water stress is during cotton boll filling stage (peak bloom), in which water deficit was imposed (Cock et al., 1993; Gwathmey et al., 2011; Snowden et al., 2014). So, the sample
collection by SS method in this phase resulted in an underestimation of the results, probably because the sampling may occurred in the area of the plant with the most affected bolls by water stress (Figure 3). Independent of the water stress treatment was applied, SS method affected characteristics as strength (Figure 3A) and elongation (Figure 3B), with values overestimated and underestimated, respectively.

One of the most important cotton fiber parameter of quality to be evaluated is micronaire index (Figure 4A). Results showed overestimation for all evaluated treatments when used with SS method to sampling fiber. But, there were no statistically different results with water stress beginning from early flowering, if compared with the two methods of sampling.

Several authors such as Cordão Sobrinho et al. (2015) and Zonta et al. (2015) have reported micronaire index values above 5.0 in experiments with irrigated cotton in the semi-arid region, considered coarse fiber, and above the tolerable value by the textile industry (Fonseca and Santana, 2002). According to the results founded in this study, this high value of micronaire index can be associated with the SS method to collect fiber samples, in which mainly first-rank bolls are collected, where fiber bring the highest values of micronaire (Belot and Dutra, 2015).

For the control treatment and using the method of sampling to whole plant, it was observed that the average of micronaire values was 4.5. This value is considered, acceptable by the textile industry, that range from 3.8 to 4.5, and so, not resulting in discount in the fiber price.

Maturity (Figure 4B) also followed the same general behavior of the other characteristics, being overestimated when samples were determined by SS method. According
to Kelly et al. (2015), the fiber maturity decrease towards from bottom to top of the plants, this explains the overestimation of this parameter when bolls were obtained for the middle third of the plant (SS) in relation to the whole plant sampling. Fiber maturity is an important parameter for the textile industry, because its variability has negative impact on the final product, especially during dyeing, since the immature fibers have a lower ink absorption capacity, making the fabric not uniform (Kelly et al., 2015; Kim, 2015). This statement demonstrates the importance of the correct determination of this parameter.

For the characteristics related to fiber color, reflectance (Figure 5A) and yellowing degree (Figure 5B), the values also followed the tendency to be overestimated when determined from the SS method, and when compared to the values determined by sampling the whole plant. Discoloration of a cotton sample may be an indication of problems such as exposure of the fiber to conditions that lead to reduced strength of fiber such as long exposure to the climate under field conditions, hence the importance of its precise determination (El Mogahzy and Chewning, 2001).

Conclusions

Results showed that fiber analysis performed from samples collected from the middle third of the plants (Standard Sample) tend to overestimate fiber quality parameters when compared to the results of data obtained from fiber samples of the whole cotton plant. These result are worsen with the occurrence of water stress in different phases of the cotton phenological cycle, because standard sample harvested bolls just to the middle third positions of the plant, and so, can dissemble the influence of water stress on cotton quality properties. At the time of water stress, bolls being formed may be affected and further, when in the middle third position of plants, their fibers can be measured, influencing the results obtained. On the other hand, if bolls in formation are in the middle third position during the water stress, the effect of water stress will not be observed afterwards using standard sampling, which also changes the results. Thus, for the determination of the cotton fiber quality in water stress tests, the most indicated method to collect samples is to gather boll samples representing the whole plant, using all positions, not only the fruitful positions of the middle third of the plants, in order to avoid results with mistaken estimates.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES


