

ENGINEERING AND GINNING

Microbial Activity of Stored Cotton Bales with Ambient and Moderate Moisture Levels

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ABSTRACT

Studies on moisture augmentation of cotton bales have shown that excessive amounts of water lead to a reduction in fiber quality. To identify an acceptable range of cotton bale moistures that could be used for moisture restoration without degrading fiber quality during long-term bale storage, cotton was treated with moderate amounts of supplementary moisture and examined after storage for 1 yr (6 mo under ambient warehouse conditions and then 6 mo under processing room conditions). The target moisture contents were 10, 8, and 6%, and a non-treated control bale. The actual initial moisture contents were 9.5, 8.0, 5.4, and 5.0%, respectively. When the bales were opened for sampling, the control and low-level moisture treatment bales gained moisture, while the two high-level moisture treatment bales lost moisture. The bale moisture content tended to equilibrate to between 6 and 7% during storage, so bales treated with higher moisture would most likely be delivered a lower weight bale to the end user. Except for a small color grade change and increase in the short fiber index at the highest moisture treatment, cotton dust potential and fiber quality, especially with regard to color grade, reflectance or yellowness, from the different treatments were not significantly different. Lower viable microbial populations were observed with increasing moisture content, but this did not appear to have any practical significance based on the lack of differences in fiber quality.

To increase profitability, water as a spray, vapor, or steam has been added to ginned cotton to reduce bale packaging forces and increase the bale weight to make up for excessively dry cotton that results from ginning. When excess moisture is added and the bales

are stored at higher than recommended moisture levels, microbial activity can be stimulated so that fiber quality decreases and mold activity increases to the point of being a health risk (Bargerion et al., 1986; Chun and Anthony, 2002; 2004). Microbial activity might not be excessively stimulated if more moderate moisture levels and storage conditions were employed. If adding moisture to reduce excess bale packaging forces, to supply moisture that is lost during ginning (Anthony and Griffin, Jr., 2001), and to improve fiber quality and processing time at the spinning plant by potentially reducing the bale conditioning time (McAlister, 1997) can be obtained simultaneously, the producer and end-user of the cotton could both obtain advantages. This study investigated the fiber quality and microbial activity of cotton bales that were supplemented with moderate levels of water and stored for approximately 1 yr (6 mo under ambient warehouse conditions and 6 mo under spinning room conditions) before opening.

MATERIALS AND METHODS

Cotton and moisture treatment. All cotton used in this study was harvested from the same field on 12 Sept. 2001. The cotton cultivar was PM1218 BG/RR (Delta Pine and Land Co.; Scott, MS). The four bales used in this study were taken from a 16-bale lot ginned on 13 Sept. 2001, from a study being conducted by The Institute of Textile Technology, Inc. (ITT). The cotton was ginned under normal conditions at the Longtown Gin in Longtown, TN, operated by Shelton Wilder with the ginning sequence as follows: module feeder, feed controller, two split-fed tower dryers, two incline cleaners, dryer behind gin stand, incline cleaner, impact cleaner, distributor, gin stand feeder, gin stand, two lint cleaners, battery condenser, Cotton Moisture System (Lewis Electric Company; Memphis, TN), and bale press. The gin drying temperature was set at approximately 176°C.

Treatment moisture was restored by spraying untreated municipal liquid water just before baling at the lint slide using the Cotton Moisture System. This system, automatically, under computer control,

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adjusts the spray level (using up to 5 nozzles that can be activated or deactivated individually) based on the cotton feed rate of each gin feeder and the lint moisture, which is monitored and measured with infrared and radio frequency technology to obtain preset final moisture levels. The moisture treatments consisted of four target moisture levels: a control with no moisture added and 6, 8, and 10% moisture levels. The target and initial moisture content measurements were provided by Institute of Textile Technology (ITT), based on the oven method (ASTM, 1971). Initial high volume instrument (HVI; USDA-AMS Cotton Division Classing Office; Memphis, TN) analysis of the treated cotton was provided by ITT. The bales were protected with a single polypropylene sheet wrap that was bound by six plastic straps. The differences between the initial moisture content and the targeted moisture content of the bales were small. Actual moisture content was 5.4, 8.0, and 9.5%, for the target moisture content of 6, 8, and 10%, respectively (Table 1). The initial moisture content of the control bale was 5.0%, which is considered to be over dried and on the low end of the desirable ginning moisture content range (Anthony et al., 1986). The bales were stored for approximately 1 yr: 6 mo under warehouse conditions before being shipped to the Cotton Quality Research Station (CQRS) in Clemson, SC, where the bales were held for approximately 6 mo in the pilot plant under processing plant conditions ($23.9 \pm 1.1^\circ\text{C}$ and $55 \pm 2\%$ RH) until opened. The warehouse was located approximately 32 km outside of Memphis, TN, and represents a typical commercial warehouse, which protects the cotton bales from the environment, but is not environmentally controlled. The warehouse reflects the ambient weather conditions common to the Memphis area from September to February where the weather can range from an average of 0.5°C and 57.3% R.H. to 27.7°C and 68.2% RH.

On 20 Aug. 2002, the bales were opened and samples taken for moisture content determined by the oven method (ASTM, 1971), fiber quality, and microbial and dust potential assays. The bales, which had universal density ($53.3 \text{ cm} \times 139.7 \text{ cm} \times 78.7 \text{ cm}$ with an approximate density of 448.5 kg/m^3), were laid on their side with the compression layers parallel to the floor of the pilot plant. Each bale was divided into 10 equal zone layers from the top to the bottom of the bale before the straps were cut. After cutting the bale straps, 10 samples were removed along a diagonal of each zone/layer from the inside surface of each zone.

Cotton dust potential. A Microdust and Trash Monitor (MTM; Schoffner Technologies; Knoxville, TN) was used to determine cotton dust potential (Millner et al., 1988; Sasser et al., 1986) as described by Chun and Perkins (1996) at the CQRS testing laboratory. Twenty-gram samples were assayed, and the dust from each sample was collected on six polyvinyl chloride filters (GLA-5000 Membrane, $5.0\text{-}\mu\text{m}$, 37 mm, PVC membrane filter; Pall Gelman Sciences; Ann Arbor, MI). Cotton dust potential is reported as total cotton dust per 20-gm sample.

Microbial activity and fiber quality. Determining the extent of biological degradation in each moisture treatment consisted of measuring the viable microbial populations (total bacteria, Gram-negative bacteria, and total fungi). Changes to the physical properties of the fiber were measured by the Advanced Fiber Information System (AFIS) and by HVI. AFIS measurements were conducted on site at CQRS. For the HVI analysis, the samples were sent to the USDA-AMS Cotton Division Classing Office in Memphis, TN. The microbial assays used 1g of lint from each sample for total bacterial and Gram-negative bacterial populations, using the method described by Chun and Perkins (1996). The same 1

Table 1. Target moisture treatment, actual initial moisture content, and final moisture content after approximately 1yr of storage

Treatment/target moisture (%)	Actual initial treatment moisture (%) ^y	Final treatment moisture after storage (%) ^z
Normal	5.0	5.7 (0.135)
6	5.4	6.0 (0.121)
8	8.0	6.8 (0.377)
10	9.5	7.2 (0.373)

^y Measurements provided by The Institute of Textile Technology, Inc.

^z Measurements provided by Cotton Quality Research Station. Average of the 10 zones from each bale with standard deviation in parenthesis.

g of lint was used to assay for fungal populations, using the method described by Chun and McDonald (1987). Changes to the protocol included adding 0.05% agar to the dilution blanks to aid suspension and using homemade potato dextrose agar (PDA) instead of commercial PDA. Homemade PDA consists of 500 ml potato broth, 500 ml deionized H₂O, 20.0 g glucose, and 20.0 g agar. The potato broth was made by heating 200 g of sliced potatoes (unpeeled) and 500 ml of deionized H₂O in an autoclave for 40 min. at 121°C and 103.42 kPa (15 psi); then while still hot, the broth was decanted and vacuum filtered through a pad of about 5 to 10 paper filters on a Büchner funnel. Dilution plates were incubated at room temperature (20±2°C) for 3 d for both the total and Gram-negative bacterial assays, and for 7 d for the fungal population assay.

Statistical analysis. One hundred samples were collected from each bale. Of these 100 samples from each bale, 40 were randomly chosen for dust and microbial assays. These 40 sub-samples were chosen as follows: the 10 bale zone layers were grouped into five adjoining zone layer pairs (zones 1 & 2; 3 & 4; 5 & 6; 7 & 8; and 9 & 10) so that there were 20 samples from each zone pair (10 from each zone). Eight samples were then randomly chosen from the 20 samples from each of the five zone pairs. The pooled samples from the four bales were then randomly assigned sample identification numbers from 1 to 160, and the samples assayed sequentially. A completely randomized split block design was used. Microbial data were log transformed for analysis using the equation, $\log_{10}(\text{cfu}+1)$, where cfu = microbial population in colony forming units per gram lint (corrected for dry weight). Data were analyzed using release 8.00 of SAS (SAS Institute Inc.; Cary, NC), and means were separated using Duncan's multiple range test. Microsoft EXCEL 2000 (Microsoft Corporation; Redmond, WA) was used to randomize treatment assignments, to enter, store, and sort data prior to SAS analysis, to correct data for dry weight, to transform data, to summarize and tabulate results, to obtain simple treatment statistics (means, standard deviations, regressions, etc.), and to perform other spreadsheet functions.

RESULTS AND DISCUSSION

Since the report by Chun and Anthony (2002; 2004) indicated degradation of fiber quality from storing cotton at high moisture contents, this study

was conducted using more moderate target moisture levels. When the bales were opened at the end of storage, no moisture was found on the bale wrappings and the cottons did not feel 'wet'. The control bale and 6% moisture treatment bales gained 0.7 and 0.6% moisture, respectively (Table 1). The two high moisture treatments, 8 and 10%, lost 1.2 and 2.3% moisture, respectively. The moisture content and weight of bales may increase or decrease during storage as the cotton equilibrates with the storage environment (Anthony and Herber, 1991), so these changes were not unexpected and mirror the changes observed by Chun and Anthony (2002; 2004). These bales were wrapped in a single polypropylene sheet wrap and started with lower levels of added moisture, while the bales studied by Chun and Anthony (2002; 2004) were covered with three 0.15 mm thick plastic bale bags to maintain the moisture in the bales to investigate the response to higher moisture levels. The bales in this study were stored for a longer period and probably equilibrated to 'safe' storage moisture levels (Hall and Elting, 1951) earlier during the storage period than the bales studied by Chun and Anthony (2002; 2004). There were some significant differences between treatments in selected AFIS measurements of the card mat (chute stock), card sliver, and finisher sliver of the treated bales; however, these differences are very small and do not appear to reflect practical differences or reduction in fiber quality (Table 2).

Before storage, the initial reflectance and yellowness readings of the cottons were not affected by the addition of moisture, as shown by the identical color grade measurement that combines the reflectance and yellowness values of the bales in the HVI measurements taken in 2001 (Table 3). Except for the 10% moisture treatment, fiber qualities of the bales stored for 1 yr were not adversely affected by treatment with moisture at the levels used in this study. With the 10% added moisture treatment, reflectance decreased and yellowness increased that resulted in a change in color grade from white to light spotted (31 to 32), which might reduce the value of the cotton (Table 3). Any beneficial effect of moisture on fiber quality and processing that might be expected as a result of the added moisture to potentially reduce the bale conditioning time, as described by McAlister (1997), would not be expected to be realized. Micronaire, short fiber index, and strength were not improved with added moisture (Table 3).

Table 2. Advanced fiber information system fiber quality measurements of water treated bales

Treatment/target moisture (%)	Fiber quality ^y				
	Uqlw (cm)	Sfcw (%)	Maturity ratio (%)	Nep (counts/gram)	VFM (gm)
Card mat/chute stock					
Normal	2.69 b ^z	11.4 a	0.92 bc	325 a	0.76 b
6	2.67 b	10.8 ba	0.93 ba	302 a	1.04 a
8	2.74 a	10.6 b	0.94 a	311 a	0.88 ba
10	2.69 b	9.8 c	0.91 c	308 a	1.07 a
Card sliver					
Normal	2.72 b	10.3 a	0.96 a	45 a	0.22 a
6	2.74 a	9.8 ba	0.95 b	46 a	0.22 a
8	2.77 a	9.2 b	0.95 b	50 a	0.20 a
10	2.77 a	9.0 b	0.94 b	47 a	0.24 a
Finisher sliver					
Normal	2.82 b	10.6 a	1.00 a	38 a	0.23 a
6	2.79 b	10.9 a	1.00 a	35 a	0.32 a
8	2.84 a	8.9 a	1.00 a	36 a	0.30 a
10	2.82 b	9.7 a	0.99 a	44 a	0.25 a

^y Uqlw=upper quartile length by weight in cm; Sfcw=short fiber content by weight; Maturity ratio=percentage maturity ratio; VFM=visible foreign matter

^z Means within column followed by the same letter are not significantly ($P \leq 0.05$) different according to Duncan's multiple range test.

Table 3. High volume instrumentation fiber quality data for treated bales before and after storage

September 2001 ^z				September 2002 ^y					
Treatment/target moisture (%)	Color grade	Reflectance (%)	Yellowness (+b)	Color grade	Reflectance Rd (%)	Yellowness (+b)	Micro-naire	Strength (cN/tex)	Short fiber index
Normal	31	76.0	9.1	31	76.0	9.0	5.28	27.66	10.15
6	31	76.0	9.0	31	76.0	9.3	5.30	27.42	10.20
8	31	76.0	9.0	31	75.8	9.2	5.30	27.66	10.33
10	31	77.0	9.2	32	74.8	9.6	5.33	27.66	10.48

^zData provided by The Institute of Textile Technology, Inc. HVI data of fiber properties before bales were placed in storage.

^yData provided by Cotton Quality Research Station. HVI data of fiber properties after bales were in storage.

Generally, higher dust potential has been associated with higher levels of microbial activity. In this study, the dust potential did not significantly change with added moisture, but the trend in average dust levels was weakly correlated with the bacterial and fungal populations associated with the moisture treatments (Table 4). When the average level of dust for each moisture treatment is regressed against the total bacterial, Gram-negative bacterial, and fungal populations, the coefficients of determination (R^2) were 0.27, 0.27 and 0.25, respectively. When the individual dust values

and corresponding microbial population are looked at, the significance of the relationship was less ($R^2 = 0.01$, 0.00 and 0.01, respectively). The two higher moisture levels tended to have significantly lower viable total bacterial, Gram-negative bacterial, and fungal population densities. There was a poor correlation between both the total or Gram-negative bacterial population densities and fungal population densities ($R^2 = 0.001$ and 0.002, respectively). While higher moisture levels may be generally associated with higher microbial levels, the lower populations were probably the result

of microbial death. The initial higher moisture content probably broke the dormancy survival stage causing short-term high microbial activity so that later, when the cotton equilibrated with the storage environment, the moisture content dropped below levels that would sustain growth and caused death, leaving a lower number of viable bacteria and fungi to be assayed (Table 4). This is in contrast to cottons that are normally warehoused without the benefit of moisture restoration. In this situation, represented by the untreated bale in this study, the viable microbial population density can remain dormant and unchanged for a year of storage. Only after longer storage does the population density begin to slowly decrease with storage time (Chun and Perkins, 1996). Also, this situation is not comparable to the studies involving washed cotton (Jacobs et al., 1993). In the washed cotton studies, microbial agents were removed by extraction into the wash water by the washing process, and this wash water was separated from the lint before storage. With moisture restoration, the amount of water added is much smaller and is not removed, so microbial agents would not be removed by extraction or 'being washed off'. This decrease in microbial population densities is in contrast to the observation made earlier by Chun and Anthony (2002; 2004) in which the moisture content remained high enough through storage to sustain fungal growth and resulted in a significantly higher fungal density (Chun and Anthony, 2002; 2004; Hall and Elting, 1951).

In summary, the cottons equilibrated with storage conditions to reach moistures of about 6 to 7%. The added moisture did not appear to have caused the cottons to suffer from microbial activity, which was supported by the failure to observe significant fiber quality deterioration, increased dust potential, or increased viable microbial populations, associated with the increased moisture. On the other hand, the fiber quality did not improve with increased moisture either, which indicates that added moisture before baling would not replace laydown conditioning to

improve fiber quality and processing as suggested by McAlister (1997). This work and the work by Chun and Anthony (2002; 2004), indicates that the fiber quality might be reduced near the 8% target moisture, as evidenced by the color grade loss in the 10% moisture treatment. Reduction of bale packaging forces and increased bale weight might be the main advantage of adding water; but the level of water treatment, which would prove the most advantageous, is perilously close to the levels where fiber quality deterioration and increased microbial activity (Chun and Anthony, 2002; 2004) might occur, which might offset the small profit from adding additional water to increase bale weight. With the careful application of the moderate levels of moisture as applied in this study, reduced bale packaging forces and modest recovery of weight lost from drying should be achievable. Additional studies regarding the restoration of bale moisture and its effect on fiber quality and microbial populations are being conducted or are near completion.

DISCLAIMER

Mention of a trademark, warranty, proprietary product or vendor does not constitute a guarantee by the U.S. Department of Agriculture and does not imply approval or recommendations of the product to the exclusion of others that may also be suitable.

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Table 4. Cotton dust potential and average microbial population in treated bales

Treatment/target moisture (%)	Average dust (mg/20 g lint) ^z	Total bacteria [$\log_{10}(\text{cfu}+1)$] ^z	G(-) bacteria [$\log_{10}(\text{cfu}+1)$] ^z	Fungi [$\log_{10}(\text{cfu}+1)$] ^z
Normal	2.82 a	6.474 a	6.071 a	4.13 a
6	2.73 a	6.456 a	6.125 a	3.93 a
8	2.75 a	6.011 b	5.575 b	3.24 c
10	2.70 a	6.099 b	5.556 b	3.62 b

^zMeans within column followed by the same letter are not significantly different ($P \leq 0.05$) according to Duncan's multiple range test.

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