# COTTON MODULE STORAGE: MICROBIAL EFFECTS, A PREPARATORY STUDY D. T. Chun and D. D. McAlister USDA, ARS, Cotton Quality Research Station (CQRS) Clemson, SC

# Abstract

A startup study on microbial effects during cotton module storage was initiated as a prelude to looking at storage conditions that may influence microbial deterioration of fiber quality. A simulation module was sampled weekly for 8 weeks and samples were assayed for fiber quality, bacterial density and bacterial profile, and cotton dust potential. The results of this study will be reported.

## **Introduction**

Cotton module usage has gained wide acceptance by cotton producers since its introduction in the early 70's. It's usage has sped up the harvest operation and effectively gets the cotton off the stalk faster because the producer is no longer dependent on trailer availability or on limited ginning resources. This has proven especially significant in an era where the number of operating gins have been on the decline. Without the worry of limiting harvest capacity, ginning can be operated in a more orderly fashion over a longer season for greater efficiency (Metzer).

Detailed methodology has been outlined for proper seed cotton module storage and handling (Metzer; Willcutt and McCarty, 1998) to prevent loss and to maximize the beneficial aspects of more rapid harvest and more orderly ginning on lint and seed quality during storage. Most of these precautions emphasize excess moisture content of the seed cotton, which should not exceed 12 percent. Storage under unfavorable moisture content may result in deterioration of seed quality and a reduction in lint qualities, especially in regard to color and strength. Even when excessive trash is present, the most likely culprit causing lint staining and other lint quality reductions are probably microbial related.

Cotton module storage constitutes the earliest stage of cotton storage that microbial activity can become a concern once the cotton has been produced and harvested. We would like to better understand the role and the conditions that may exacerbate the effect of microbes on fiber quality. While extensive literature has been written on cotton module storage, actually very little has been documented on the role of microbes except to generalize that under excessive moisture conditions, module heating occurs and this fore warns of anticipated seed and fiber quality deterioration. We would like to add to the understanding of the role of microbes. As a first step, a model module will be created and stored for a period of time under ideal storage conditions so that observations of microbial activity can be made and the mechanics of monitoring cotton modules refined. From this preliminary step, future investigations where less than ideal parameters of storage may be incorporated and its effect in relations to microbial effects on reducing fiber quality can be observed. A report of the observations made on a model module storage period where microbial activity is not expected to reduce seed or lint quality will be reported to establish a ground level of microbial activity in module storage.

### **Methods and Materials**

## Simulated Cotton Module

The cotton used was upland seed cotton, 'paymaster 1220', grown in Kingstree, South Carolina, from the 1999 harvest year under ultra narrow row cultivation (7 ½ inch row spacing) on marginal soil. The cotton was

Reprinted from the Proceedings of the Beltwide Cotton Conference Volume 2:1288-1291 (2001) National Cotton Council, Memphis TN stripper picked and transported to the USDA CQRS warehouse in Clemson, SC, in January 2000. Six hundred pounds of this cotton was loosely compressed at 2000 psi into a 5 x 3 x 2 foot bale to simulate a field module. To imitate ideal field storage conditions, the module was stored at ambient conditions under cover at the warehouse for two months. Baling wire was used to hold the module together during that period. To sample the module, the module was uncompressed and two small handful size samples were taken from the middle of the four exposed sides and the top of the module, from approximately 6 inches within the bale; after which the module was recompressed. The samples taken were pooled, ginned with a 6-inch laboratory saw gin and homogenized with a laboratory fibre blender before being assayed for bacterial population and profile, cotton dust potential, and cotton quality. The start of the module storage period began on January 10 as the first module sampling date. From then on, on an approximately weekly basis, the module was sampled. The sampling times are taken as the days in storage starting January 10, 2000.

### **Cotton Quality**

When the last sampling date had been collected, cotton from each of the sampling dates was turned over to the Testing Laboratory at CQRS for HVI testing.

## **Bacterial Profile**

Within 48 hours of each module sampling, cotton from that sampling period was processed and assayed for viable bacteria and its bacterial profile. The methodology has been described previously (Chun and Perkins, 1996 & 1997). Only the frequency of the bacterial genera observed on the lint will be reported for each storage sampling date.

### **Cotton Dust Potential**

From each sampling date, four 20.0-gram samples were removed for determining cotton dust potential. Each 20.0-gram sample was processed twice (at a setting of 5.5, for a feed rate of a 1.5 minute/run) through a Microdust & Trash Monitor [(MTM), Zellweger Uster, Inc., Technologies, Knoxville, TN] which is an instrument used to measure foreign matter, including microdust and trash, in raw or processed fiber and has been used as a means of ascribing dust releasing potential of cotton (Chun, 1991; Chun & Perkins, 1996; Millner et al., 1988; Sasser, et al., 1986). Six preweighed  $5.0 \,\mu\text{m}$ , 37 mm, Metricel membrane filters (Gelman Sciences, Inc., Ann Arbor, MI) were superimposed equidistant in a clockwise fashion (sequentially labeled as filters 1-6) on the standard MTM filter. The airflow was directed onto the filters by a plastic template that also secured the filters in place. The cotton dust in each 20.0-gram sample is the sum of the dust on the six filters and the average of each sampling date as dust potential (milligrams dust per gram lint) will be reported.

### Statistical Analysis

Data were analyzed using release 6.12 or earlier releases of SAS (SAS, Statistical Analysis System; SAS system for Windows version 4.0950; SAS Institute Inc., Cary, NC, USA) for making mean comparisons. Otherwise additional testing and data manipulation was done with Microsoft EXCEL 2000 or earlier releases of EXCEL (Microsoft Corporation, USA) and plotted using SigmaPlot for Windows version 05 (SPSS, Inc., USA).

#### **Results and Discussion**

The results of the fiber quality HVI measurements showed a greater degree of heterogeneity between the cotton from the different sampling dates (Tables 1 & 2) than would normally be found from baled samples. Microbial deterioration was not suspected since no strong increasing or decreasing trends of any of the fiber qualities could be discerned with storage time and the storage conditions should not have encourage microbial activity – moisture content of the seed cotton averaged 7.8% through the storage period. Most likely, this heterogeneity is variability inherent with seed cotton and will remain an obstacle in future tests.

Over the 58 days storage, cotton dust potential (Figure 1) remained relatively constant over the storage period, averaging around 0.35-mg/g throughout the storage period. This level of dust releasing potential was alarming since these levels were higher than anything we had encountered before and represents more than 5 times the dust releasing potential than found in warehoused baled cottons (Chun and Perkins, 1996). One explanation is that the 1999 harvest year cottons contained hazardously higher levels of cotton dust than in previous years. But a more likely explanation is that in our work, we normally examine ginned cotton or warehoused cottons, and would not be aware of dust potentials in preginned cottons. Our laboratory saw gin used vacuum suction to keep airborne dust levels down, so we are tentatively speculating that commercial ginning operations removes a very much larger fraction of the breathable dust than does our laboratory gin. This is supported by earlier work where seasonal cotton dust production potential increased during the growing season to levels approaching what we observe here (Chun 1991).

Population density shows a tendency to decrease after 5 weeks when the density increases and begins to somewhat level off. Despite this, there is no overall decrease or increase in population density. Rather this probably reflects the static condition of lint storage where the conditions do not favor active bacterial growth or deterioration of fiber quality. The bacterial levels seen here is about 10 to 100 times greater than found in warehoused cottons during its first year of storage. A big increase in microbial population would be indicative of microbial affects on fiber quality and also would be suggestive of a high moisture content in the cotton and possibly even an increase in the module temperature. In future studies, these parameters -- internal module moisture, temperature and carbon dioxide level -- should be monitored.

Approximately 900 bacterial identifications were made to develop the bacterial profile shown in Figure 4. While the identification method permits identification down to the species level and actually in some cases to the specific subgroup level, the identified isolates were grouped into their common genera, and the frequency of the commonly found genera will be presented. At the start of the storage period, a very diverse population of bacterial genera was found, much greater than the bacterial profile than would be expected from cottons grown in this region (Chun and Perkins, 1997). Initially, excluding the artificial category of "No Match", as many as 21 different genera of bacteria was identified (including some Grampositive genera). Following what is to be expected, the diversity of the bacterial population drops during storage so that by the end of the storage period, only about half of the different genera of bacteria remained (Figure 3). When seven of the most commonly found genera of bacteria was followed over the eight weeks of storage, we find that these seven genera not only persisted through storage but made up 50% or more of all of the different genera of bacteria identified. As expected of cottons from this region (Chun and Perkins, 1997) which are characteristically high in endotoxin content, very few Gram-positive genera was observed and the very few that were observed were found when storage was initiated. As expected, the most common genera found in this region of the cotton belt was Pseudomonas, an important contributor to the presence of endotoxin, but what was unexpected was the high occurrence of Salmonella species which besides contributing to the presence of endotoxin, are often associated as a serious health concern. We will need to determine if the high prevalence of Salmonella species here was a fluke or is it really commonly found on seed cotton. But again, since we normally study commercially ginned cotton, very possibly this observation has been missed and may not be a potential health issue if during commercial ginning, the heating used to dry the cotton plays a secondary role of killing viable Salmonella bacteria. This question should be studied later to allay potential health fears

## Disclaimer

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## Table 1. HVI Properties,<sup>1</sup> Part 1.

|      |     | Color |     |       | Trash  |       |
|------|-----|-------|-----|-------|--------|-------|
| Days | Mic | Rd    | +b  | Grade | Leaf,% | Count |
| 0    | 3.1 | 60.9  | 9.7 | 63-1  | 17     | 123   |
| 9    | 3.4 | 57.0  | 9.2 | 62-2  | 35     | 215   |
| 16   | 3.2 | 60.6  | 9.1 | 62-1  | 24     | 162   |
| 23   | 3.5 | 58.0  | 8.1 | 62-2  | 42     | 193   |
| 30   | 3.2 | 59.7  | 8.6 | 62-1  | 30     | 173   |
| 37   | 3.3 | 60.0  | 9.0 | 62-2  | 25     | 170   |
| 44   | 3.3 | 61.9  | 9.8 | 53-2  | 14     | 149   |
| 51   | 3.3 | 61.0  | 9.1 | 62-1  | 17     | 120   |
| 58   | 3.5 | 57.8  | 8.6 | 62-2  | 42     | 187   |

<sup>1</sup> Days = Days from module initiation, January 10, 2000; Mic = micronaire reading, microgram/inch; Color: Rd = color reflectance, +b = color + b, Grade = color grade; Trash: Leaf = leaf area, percent, Count = actual count; UHM= upper half mean; St = staple length, 1/32 of an inch; Unif. = uniformity index, mean/upper half mean; Str = strength, grams/tex; El g. = elongation, percent.

Table 2. HVI Properties,<sup>1</sup> Part 2.

| Days | UHM  | St | Unif. | Str. | El g., % |
|------|------|----|-------|------|----------|
| 0    | 1.17 | 37 | 83.5  | 28.6 | 6.9      |
| 9    | 1.18 | 37 | 84.5  | 29.6 | 6.8      |
| 16   | 1.15 | 37 | 82.0  | 31.2 | 6.7      |
| 23   | 1.16 | 37 | 84.6  | 29.0 | 6.9      |
| 30   | 1.20 | 37 | 83.4  | 30.9 | 7.0      |
| 37   | 1.15 | 37 | 83.7  | 31.8 | 6.8      |
| 44   | 1.15 | 37 | 84.3  | 29.7 | 6.8      |
| 51   | 1.15 | 37 | 83.1  | 28.4 | 7.0      |
| 58   | 1.16 | 37 | 83.8  | 27.4 | 7.0      |

<sup>1</sup> Days = Days from module initiation, January 10, 2000; Mic = micronaire reading, microgram/inch; Color: Rd = color reflectance, +b = color + b, Grade = color grade; Trash: Leaf = leaf area, percent, Count = actual count; UHM= upper half mean; St = staple length, 1/32 of an inch; Unif. = uniformity index, mean/upper half mean; Str = strength, grams/tex; El g. = elongation, percent.

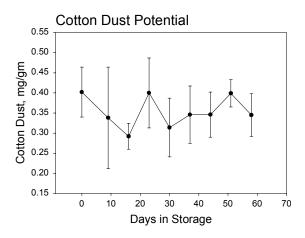


Figure 1. Average cotton dust potential during storage, days from module initiation, January 10, 2000 (each half bar represents 2 s.e.).

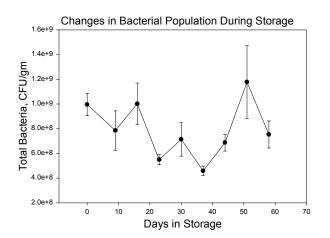


Figure 2. Population density changes during storage, days from module initiation, January 10, 2000 (each half bar represents 2 s.e.).

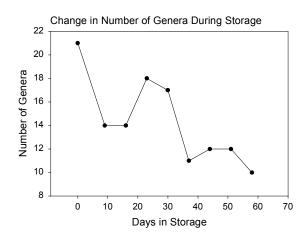


Figure 3. Change in the number of genera observed during storage, days from module initiation, January 10, 2000.

Seven of the Most Frequent Found Genera

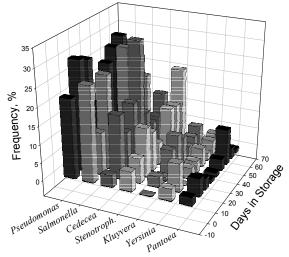


Figure 4. Seven of the most frequently found bacterial genera observed during storage, days from module initiation, January 10, 2000, frequency of observation (%).