COLLECTION OF UNIFORM, CARD GENERATED, VERTICALLY ELUTRIATED DUST FOR INTERLABORATORY COMPARISON OF DUST ENDOTOXIN ASSAYS H.H. Perkins, Jr., Research Chemist, USDA, ARS, Cotton Quality Research Station Clemson, SC S.A. Olenchock, Captain US Public Health Service, NIOSH Morgantown, WV R.E. Harrison, Agricultural Engineer USDA, ARS, Cotton Quality Research Station Clemson, SC

#### Abstract

For a number of years, researchers and others concerned with endotoxin levels in cotton lint or cotton dust have known that when identical samples are assayed for endotoxin content, level differences, often in orders of magnitude, may exist in results between laboratories. An experiment was designed in which about 10 laboratories with established endotoxin assay protocols would participate in an interlaboratory test to assay "uniform", vertically elutriated dust to gain insight into the problem. An elaborate cotton blending and dust blending and collection protocol was developed and implemented to produce this dust. The methodology is described and results on dust uniformity are reported.

## Introduction

When identical samples of cotton lint or cotton dust are assayed for endotoxin content, level differences, often in orders of magnitude, may exist in results between laboratories. Thus, results reported in the literature must be interpreted with caution and in consideration of extraction methods and the laboratory conducting the analysis. On a number of occasions, dating back as much as 4 or 5 years, the need for and possibility for conducting an interlaboratory test on uniform dust was discussed among interested scientists, most notably -- Henry H. Perkins, Jr., USDA, ARS, Clemson, SC; Stephen A. Olenchock, NIOSH, ALOSH, Morgantown, WV; Ragnar Rylander, University of Gothenburg, Sweden; and Robert R. Jacobs, University of Alabama, Birmingham, AL. Things moved rather slowly, but after discussions with Stephen Olenchock in the Summer of 1995, a statistical protocol for sampling was designed to generate samples for testing by up to 10 laboratories. The statistical aspect of the project will not be discussed here. We then took the initiative to collect the dust at the USDA, Cotton Quality Research Station, Clemson, SC. The elaborate cotton blending procedure and the air handling techniques used to generate and collect uniform, vertically elutricated dust are described. Results detailing dust uniformity are reported.

### **Preparation of Cotton and Dust Generation**

As mentioned, the statistical protocol will not be discussed except as it concerns the number and locations of vertical elutriators required. To supply the required number and distribution of dust laden filters to 10 laboratories, 12 vertical elutriators - four each at three locations - were used. Fifteen individual collection runs were scheduled over about a 2-week period to collect 180 filters, 18 for each laboratory. The process to prepare the cotton for carding and the carding conditions utilized to ensure that the dust generated would be consistent for each dust collection run are described in general in the following paragraphs.

An elaborate blending procedure was used to prepare the cotton for carding to ensure that the dust generated would be uniform for each of the dust collection runs. Cotton was purchased commercially from the Midsouth Area as a 25bale lot of uniform quality, predominantly Grade Code 52, cotton (MQ-202). The 25 bales were placed systematically behind 12 blender hoppers and cotton from each of the 25 bales was fed simultaneously to the hoppers. The production from each hopper fell onto an endless belt in a manner that formed a sandwich of cotton comprised of cotton from each of the 25 bales in the lot. The sandwich blend was picked up pneumatically at the belt delivery and was transported about 100 feet through a duct to the feed mechanism of a bale press. Twenty four blended bales were formed. Theoretically, each of the blended bales is a proportioned composite of the cotton from each of the 25 original bales. The delivered bales were numbered 1-24 and bales #7, 12, and 15 were used for the dust generation. Thus, these bales were produced well after a "steady state" of blending had been reached and well before any "tailing" that might have occurred in the last one or two bales.

In the textile opening line the cotton from each bale, #7, 12, 15, was fed into a separate blending hopper. The delivery from each hopper fell onto an endless belt to form a sandwich blend of the cotton from the three bales. The sandwich blend, theoretically containing equal portions of cotton from each of the three bales, was picked up pneumatically and transported through a duct to a single opening - cleaning point consisting of a bladed beater to open the tufts of cotton and remove trash. The cotton was picked up at this point in large wheel boxes and was transported manually to the blending picker. The cotton was placed on a large apron behind the picker. The picker consists of a spiked beater and a lap forming section. As cotton passes through the beater, small tufts are produced and these subsequently are formed into a "lap" that is a thin batt (13 oz/sq.yd.) of cotton rolled up as a cylinder

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weighing about 40-42 pounds. All of the blended cotton was processed into laps (36 laps). To continue the blending process, 4 laps of the 36 were chosen systematically and were refed as a sandwich blend through the picker again to form 4 new blended laps. Additional groups of 4 were processed until all of the original 36 laps had been processed. This latter process was repeated by again systematically selecting 4 blended laps and processing them as a sandwich blend into 4 new blended laps. This process continued until all laps had been processed into blended laps. These 36 blended laps comprised the stock fed to the card to generate dust for collection by vertical elutriators. This is an ultimate blending scheme that ensures that any one pound of cotton fed to the card is essentially identical to any other pound of cotton fed.

In carding, the dust generated in the card room was forced to an outlet and was transported pneumatically through a small duct about 85 feet to the remote dust sampling facility. This process of channeling the dust into the confined space of the duct for transportation to the remote facility is an effective forced blending of the dust. The air for the card room came directly from outside using a specific filtering and conditioning system for this room. The card room was operated under positive pressure to ensure that no contamination entered the card room when the door was opened. The air conditioning system for the remote facility is a completely independent unit. The fan on the unit was operated in the "on" position to prevent any surges of air that might occur if the fan was in the "automatic" position. All filters in the air conditioning systems in the card room and in the remote room were changed daily in the morning before starting dust collection runs.

In the carding runs to collect dust, the card room always started from a clean condition. Air was transported to the remote sampling facility continuously when no cotton was being processed. The absence of or extremely low background level of dust in this control configuration was verified by a Continuous Aerosol Monitor (CAM) system in the remote facility that gives a continuous, instantaneous reading of the dust level. When the cotton was started into the card, the time was noted and the card was operated for about 30 minutes (minimum of 20 minutes) before the vertical elutriators were started. We know by observing the CAM readings and from past experience that the dust level in the remote facility will stabilize during this time.

The card room where dust was generated and the remote room where dust was collected were specially designed to generate, measure, and collect dust and have been used effectively for this purpose for more than 15 years. In the remote facility, 12 vertical elutriators were utilized to collect dust. The elutriators were arranged in 3 locations with 4 positions at each location. Elutriator intakes were 5 feet above the floor. The locations were designated A, B, and C ;and the positions within a location were designated 1, 2, 3, and 4. The distances between locations and between positions within a location were 2.5 feet and 9 inches, respectively. Thus, the 12 elutriators were located in a relatively small cluster. Several preliminary runs were conducted to verify that each of the elutriators was collecting relatively equivalent amounts of dust. The carding rate (30 lbs/hr) and elutriator run time (90 minutes) were established to provide about 0.5 mg of dust on a filter with an acceptable range of 0.3 - 0.7 mg. Seventeen dust collection runs were conducted and 204 filters were collected. Several "clean room" runs were conducted that verified that background dust levels were negligible.

# **Dust Uniformity**

Once the conditions for conducting the dust collection were established, a controlled, trial run was conducted to collect dust for endotoxin analysis at the NIOSH laboratories in Morgantown, WV. The results of endotoxin measurements on these filters are shown in Tables 1 and 2. The endotoxin levels have been adjusted by a factor to provide for a blind test of the actual level. Thus, no units are shown. The results in Table 1 show that there are no significant differences in endotoxin levels between elutriator locations and the results in Table 2 show that there are no differences between positions at the various locations. The statistical protocol was designed to be effective even if differences between locations or positions were present. Since there are no significant differences, the potential statistical power of the experiment is very good.

The average dust weights of the four filters at each location for each of the 17 dust runs are shown in Figure 1. All of the weights are within the target range (0.3-0.7 mg). There are a few minor differences in levels between runs that can be attributed to factors such as slight difference in weight of laps fed to the card, slight differences in airflow from card room to remote room, and in at least 2 runs, premature cutoff of the timer controlling elutriator run time. These weight level differences will have no effect on the interlaboratory test of endotoxin levels.

#### Summary

Uniform, card generated, vertically elutriated cotton dust has been collected for use in an interlaboratory test of endotoxin extraction and analysis methods. Endotoxin analysis of dust samples collected under standard test conditions showed that the endotoxin levels did not vary significantly either between elutriator locations or between positions within locations. The process of selecting laboratories for participation in the interlaboratory test is in progress.

Table 1. Endotoxin Levels VE Filters		
Location	Endotoxin	Average $+$ S.D. <sup>1</sup>
	Level	
А	242	
А	245	
А	228	
А	211	232 <u>+</u> 16 A
В	197	
В	228	
В	201	
В	198	206 + 15 A
С	214	—
С	184	
С	235	
С	233	217 + 24 A
ALL		218 + 20

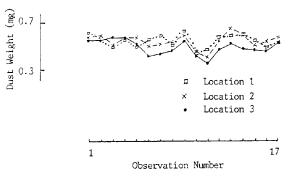


Figure 1. Average weight of dust on vertical elutriator filters from each sampling location during dust collection runs

 $^{1}$ Means having a letter in common are not significantly different (alpha = 0.05)

Table 2. Endotoxin Levels VE Filters		
Position	Endotoxin Level	Average $+$ S.D. <sup>1</sup>
1	242	
1	197	
1	214	218 <u>+</u> 23 A
2	245	
2	228	
2	184	219 <u>+</u> 31 A
3	228	_
3	201	
3	235	221 + 18 A
4	211	—
4	198	
4	233	214 + 18 A
ALL		$218 \pm 20$

<sup>1</sup>Means having a letter in common are not significantly different (alpha = 0.05)