

Evaluation of Cleaning and Washing Processes for Cotton Fiber

Part VI: Histamine Analysis of Cotton Material¹

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ABSTRACT

Chemical and biological assay of dust material obtained from carding washed and unwashed cotton suggests the occurrence of a modest decrease in the histamine content of the dust following the washing process. Histamine-releasing activity is significantly reduced in the washed samples when analyzed by chemical assay. However, biological assay shows only insignificant reduction in histamine releasing after washing.

Experimental

Samples of cotton dust, obtained from the carding process of cotton before and after washing procedures, have been processed for histamine content and histamine release according to standard methods [1]. The analytical determinations consisted of a chemical assay, using the photofluorometric method of Shore *et al.* [5], with the modification introduced by May *et al.* [2], and by the biological method standardized by Nicholls [3] using the ileum segment from the guinea pig.

The release was also assayed, by the chemical as well as by the biological methods just described, after incubation of cotton material extracts, obtained by maceration of the cotton in Tyrode solution, finally adjusted to neutrality, according to the procedure first suggested by Nicholls and Bouhuys [4].

The procedures employed in obtaining the samples of cotton material and the treatment used to wash the raw cotton before carding are described in previous parts of this paper, and the details shall not be repeated here. The number identifying each sample corresponds to the code indicated in Part II. The dust samples were collected on a Pneumafil V-filter frame during carding of the cotton in the N. C. State University model cardroom. The samples were then separated into 20 to 38 μm (coarse) and <20 μm (fine) fractions using the method described by Fornes in Part V.

Results

The results of histamine content in the dust obtained by the chemical analysis and, independently, by the biological method in the control preparations (unwashed cotton, Treatments 1, 2A, and 2B) are reported in Table I. The correlation between the two analytical processes appears to be quite good, with a coefficient of 0.62 (Table II). The average value, read by the chemical method, at 1.01 μg of histamine content per gram of dust, is consistent with the range of mean values reported in recent literature [1]. The biological determination, using the organ preparation from the guinea-pig ileum, yields a smaller value of histamine content, indicating a mean of 0.48 $\mu\text{g/g}$ of dust, approximately one half of the corresponding figure obtained by the chemical determination. Anyway, this finding is certainly consistent with the view that other mediators of smooth-muscle activity, besides histamine, are not likely to be present in this material.

The release phenomenon, studied with the reaction of dust extracts challenging minced porcine lung, indicates a positive release of about 6 μg of histamine per gram of lung, according the chemical assay and a somewhat lower aliquot (1.05 $\mu\text{g/g}$) measured by the biological determination (Table III). Again, the discrepancy between the chemical and the biological assays is consistent with the view that mediators other than histamine are not present or released by the cotton extracts, operating in the specified conditions.

Table I also gives the number of chemical and biological analyses that were run on each sample and the proportion that gave positive reaction to the dusts.

¹ Presented at the second Natural Fibers Textile Conference, Charlotte, North Carolina, September 18-20, 1979.

Table IV gives the same data on the washed (treated) samples. The histamine content as determined by the chemical procedure shows the washed samples (Treatments 3-8) to have half the histamine on the average (0.50 vs. 1.01, Tables II and V) as the unwashed samples. The statistical significance of this reduction in histamine content of the washed samples is slightly under the 0.05 level (Table VI).

TABLE I. Control (histamine expressed in mcg/g).

Bale	Content (dust)	Chemical			Content (dust)	Biological		
		Release (lung)	%	Positive/all		Release (lung)	%	Positive/all
1 (2nd control) (coarse)	1.1 ± 0.2	10.6 ± 3.6	100	12/12	0.2 ± 0.04	1.4 ± 0.4	100	12/12
2A (coarse)	1.2	7.7 ± 0.6	100	2/2	1.0 ± 1.1	0.2 ± 0.1	50	2/2
2A (sonic)	2.1	3.6 ± 0.9	100	2/2	0.8	—	0	0/2
2 (2nd half) (coarse)	0.6 ± 0.2	5.0 ± 2.7	100	4/4	0.2	0.8 ± 0.3	66.6	2/3
2 (2nd half) (fine)	1.0 ± 1.0	1.0	25	1/4	0.4 ± 0.2	1.8 ± 0.3	66.6	2/3
2B (coarse)	0.04 ± 0.03	8.2 ± 3.5	87.5	7/8	0.3 ± 0.01	2.1 ± 1.2	62.5	5/8

TABLE II. Histamine content: chemical vs. biological analysis.

	Control samples	
	Chemical analysis	Biological analysis
Mean	1.01 ± 0.68 mcg/g	0.48 ± 0.34 mcg/g
Variation	68%	70%
Correlation 0.62		

TABLE V. Histamine content: chemical vs. biological analysis.

	Treated samples	
	Chemical analysis	Biological analysis
Mean	0.50 ± 0.38 mcg/g	0.62 ± 0.34 mcg/g
Variation	77.01%	54.7%
Correlation 0.08		

TABLE III. Histamine release: chemical vs. biological analysis.

	Control samples	
	Chemical analysis	Biological analysis
Mean	6.02 ± 3.48 mcg/g	1.05 ± 0.86 mcg/g
Variation	58%	82%
Correlation 0.10		

TABLE VI. Significance of control vs. treated contrast.

Content (chemical assay)	Content (biological assay)
$t = 2.14$	$t = 0.83$
$df = 18$	$df = 18$
$p = < 0.05$	$p = > 0.1$

TABLE IV. Treated (histamine expressed in mcg/g).

Bale	Content (dust)	Chemical			Content (dust)	Biological		
		Release (lung)	%	Positive/all		Release (lung)	%	Positive/all
3 (coarse)	0.8	2.6	100	4/4	0.5	0.6	50	3/6
3 (fine)	0.1	2.45	100	2/2	1.5	1.2	25	1/4
3 (2nd half) (coarse)	0.6	1.15	50	2/4	0.5	1.2	50	1/2
3 (2nd half) (fine)	0.4	2.6	100	4/4	0.5	1.0	66.6	2/3
4 (2nd half) (coarse)	1.4	2.4	100	4/4	0.5	1.1	100	4/4
4 (2nd half) (fine)	1.0	2.3	100	4/4	0.5	1.6	100	3/3
5 (coarse)	0.5	1.8	100	2/2	0.4	0.0	0	0/2
5 (fine)	0.3	2.2	100	2/2	0.5	0.0	0	0/2
6A ₁ , A ₂ (coarse)	0.15	2.6	100	4/4	0.6	2.4	50	4/8
6A ₁ , A ₂ (fine)	0.14	3.9	100	4/4	0.5	1.1	100	8/8
7A (coarse)	0.7	0	0	0/2	1.0	0	0	0/4
7A (fine)	0.6	2.2	100	2/2	1.1	0.4	25	1/4
8 (coarse)	0.1	2.8	100	12/12	0.3	0.8	66.6	8/12
8 (fine)	0.2	4.2	100	14/14	0.3	1.9	100	14/14

The biological analysis indicates that the histamine content of the washed (treated) samples is a little higher at 0.62 $\mu\text{g/g}$ of dust (Table V) than the unwashed samples, which have 0.48 μg of histamine per gram of dust (Table II). This difference is statistically not significant (Table VI).

The chemical analysis to determine the histamine-releasing effects of this dust shows the unwashed samples releasing about $2\frac{1}{2}$ times more histamine than the washed samples (6.02 $\mu\text{g/g}$ for unwashed *vs.* 2.37 $\mu\text{g/g}$ for washed cotton is highly significant, as seen in Table VIII). However, the biological analysis showed lesser and statistically insignificant reductions in histamine released due to the washed cotton dust. Table VII and Figures 1 and 2 summarize these findings.

Insignificant differences can also be observed between different types of treatments and between coarse and fine dust sizes.

TABLE VII. Histamine release: chemical *vs.* biological analysis

	Treated samples	
	Chemical analysis	Biological analysis
Mean	2.37 \pm 1.02 mcg/g	0.95 \pm 0.72 mcg/g
Variation	43.0%	79.9%
Correlation 0.51		

TABLE VIII. Significance of control *vs.* treated contrast.

Release (chemical assay)	Release (biological assay)
$t = 3.68$	$t = 0.27$
$df = 18$	$df = 18$
$p = <0.01$	$p = >0.1$

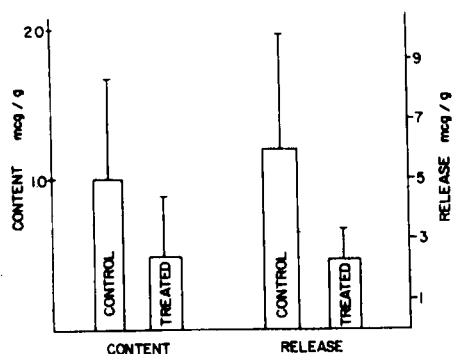


FIG. 1. Histamine chemical assay.

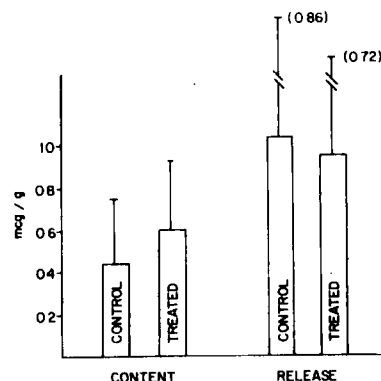


FIG. 2. Histamine biological assay.

Summary

The determination of content of histamine in dust material obtained from an experimental cardroom, before and after washing processes, suggests the occurrence of a modest decrease in this component of the cotton dust, following the washing process. Similarly, the release of histamine, determined by both the chemical analysis and the biological assay, indicates a decrease after washing treatment.

Statistical analysis of these effects indicates that the treatment variable on both content and release is significant when monitored by the chemical procedure but is not when determined by the biological assay. The meaning of this discrepancy remains unclear.

ACKNOWLEDGMENTS. The assistance received from Cotton, Inc. of Raleigh (Dr. Sasser) and from the Southern Region Research Center of the USDA (Dr. Berni) during the conduct of this study is gratefully acknowledged.

Literature Cited

1. Battigelli, M. C. *et al.*, The Role of Histamine in Byssinosis, *J. Environ. Sci. Health* **A12**, 327-339 (1977).
2. May, C. D. *et al.*, Procedures for Immunochemical Study of Histamine Release from Leukocytes with Small Volume of Blood, *J. Allergy* **46**, 12-20 (1970).
3. Nicholls, P. J., Some Pharmacological Actions of Cotton Dust and Other Vegetable Dusts, *Brit. J. Industr. Med.* **19**, 33-41 (1962).
4. Nicholls, P. J., Histamine Release by Compound 48/80 and Textile Dusts from Lung Tissue *in vitro*, in: "Inhaled Particles and Vapours II," C. N. Davies, (Ed.), Pergamon, Oxford, 1967, pp. 69-74.
5. Shore, P. A. *et al.*, A Method for the Fluorometric Assay of Histamine in Tissues, *J. Pharmacol. Exp. Ther.* **127**, 182-186 (1959).