TECHNICAL REPORT

For

John Gildersleeve
International Distribution Alliance

AATCC Test Method 100 - 2004

Assessment of Antibacterial Finishes on Textile Materials

White Material treated with AM 7200

Analysis performed by
John Limbach
Microbiologist

Approved by
Andy Konopacki
Laboratory Director

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1. Materials Submitted for Testing:

One square foot of medical tent black and white laminate was submitted for AATCC 100 testing.

2. Significance and Use:

The AATCC 100 test method is designed to quantitatively test the ability of fabrics and textiles to inhibit the growth of microorganisms or kill them, over a 24 hour period of contact, at ambient room temperature. Among the various tests for antimicrobial activity of fabrics, this has emerged as the textile industry's standard

3. Scope:

This test method provides a quantitative procedure for the evaluation of the degree of antibacterial activity. Assessment of antibacterial finishes on textile materials is determined by the degree of antibacterial activity intended in the use of such materials. If only bacteriostatic activity (inhibition of multiplication) is intended, a qualitative procedure which clearly demonstrates antibacterial activity as contrasted with lack of such activity by an untreated specimen may be acceptable. However, if bactericidal activity is intended or implied, quantitative evaluation also provides a clearer picture for possible uses of such treated textile materials.

4. Terminology:

4.1 Activity of an antibacterial agent – a measure of effectiveness of the agent.
4.2 Antibacterial agent – in textiles – any chemical which kills bacteria (bactericide) or interferes with the multiplication, growth or activity of bacteria (bacteriostat).

5. Test Specimens:

The black/white laminate is cut into round pieces, each with a diameter of 1.9 inches with a surface area of 2.84 square inches. Latex gloves are worn while handling the fabric to prevent any oil contamination from the skin from altering test results.

6. Test Procedure:

Organism. *Staphylococcus aureus* (ATCC #6532)
Culture medium. The Staphylococcus aureus is grown on a Brain/Heart Infusion Agar slant for 24 hours at 35° C.
Inoculum. The bacteria is collected in a 99 mL potassium phosphate buffer blank containing magnesium chloride.

Inoculation. One mL of the bacterial suspension is added to a second 99 mL phosphate buffer blank. One mL of this bacterial dilution is placed in the center of a petri dish and the round fabric sample is placed upon the bacterial dilution. The petri dish is covered and placed into a Ziploc bag and sealed to prevent any moisture evaporation.

Analysis. The bacterial samples are collected at 4 hours and at 24 hours. The round, non absorbent, sample is rinsed off with a 4 mL aliquot from a 9mL phosphate buffer dilution blank. The bacterial dilution is placed into the 9mL dilution blank and pour plate counts are prepared with Standard Methods Agar. The pour plates are incubated at 35° C for 48 hours. The bacterial counts are obtained and recorded.

7. Results:

Sample ID	Staphylococcus aureus count - cfu/mL	
Initial Control	7,400,000	
Control at 4 hours	5,600,000	
Black surface at 4 hours	750,000	86.6% reduction
White surface at 4 hours	650	99.988% reduction
Control at 24 hours	3,700,000	
Black surface at 24 hours	210,000	94.32% reduction
White surface at 24 hours	<10	99.999% reduction

Calculation of the "Antibacterial Activity": This is the difference in the logarithm of the viable cell count found on an antimicrobial-treated product and an untreated product after inoculation with, and incubation of, the bacteria. The following equation is used: $R = (U_t-U_o) – (A_t-U_o) = U_t-A_t$

Where: R = the "Antibacterial Activity"

U_o = the average of the common logarithm of the number of viable bacteria (bacteria/mL) recovered from the untreated test specimens immediately after inoculation.

U_t = the average of the common logarithm of the number of viable bacteria (bacteria/mL) recovered from the untreated test specimens after 4 hours o contact and 24 hours of contact.

A_t = the average of the common logarithm of the number of viable bacteria (bacteria/mL) recovered from the treated test specimens after 4 hours of contact and 24 hours of contact.

4.0 Conclusions:

Antibacterial Activity of the Black surface at 4 hours.	**0.873**
Antibacterial Activity of the White surface at 4 hours.	**3.935**

Antibacterial Activity of the Black surface at 24 hours.	**1.246**
Antibacterial Activity of the White surface at 24 hours.	**5.568**

Antibacterial Activity	%Kill compared to control	Comment
<1.5	<96.8	poor
1.5 to 2.0	96.8 to 99.0	borderline
2.0 to 3.0	99.0 to 99.9	good
>3.0	>99.9	excellent

REDUCTION/INHIBITION PERCENTAGE RESULTS FOR APPLICATION OF Vex-Protex-All **ON THE FOLLOWING ARMY UNIFORMS SAMPLES AGAINST *STAPHYLOCOCCUS AUREUS*.**

REFERENCED METHODS:
AATCC Test Method 100 - 1993

Test article: Army Uniforms-US Dept. of Defense

Samples:
1) 50/50 Nylon/Cotton Ripstop Control Sample.
2) 50/50 Nylon/Cotton LW Twill Marpat Woodland Control
3) 50/50 Nylon/Cotton Midweight Twill Marpat Woodland Control
4) 100% Nylon 500 Denier Cordura Control
5) 100% Nylon 70 Denier Control

Sample Size: 48mm circle
Number of Layer (s): 2
Time of Contact: 24 hours
Contact Temperature: 23° C
Date Contact Initiated: 27April 2009
Incubation Temperature: 37° C
Incubation Time: 24 hours
Date of Plating: 28April 2009

Test Organism: Initial Inoculum
Staphylococcus aureus 1.84×10^6 CFU/ml

RESULTS:

Staphylococcus aureus

Sample	CFU/ml after 24 hr Contact Time	% Reduction	% Inhibition
Sample "1" Control	**2.09×10^9**	**N/R**	**NI**
Sample "1" Treated	**2.50×10^2**	**99.98%**	**99.98%**
Sample "2" Control	**3.40×10^9**	**NR**	**NI**

Sample "2" Treated	**<10**	**99.99%**	**99.99%**
Sample "3" Control	**$2.70x10^8$**	**NR**	**NI**
Sample "3" Treated	**<10**	**99.99%**	**99.99%**
Sample "4" Control	**$2.20x10^8$**	**NR**	**NI**
Sample"4" Treated	**$2.10x10^2$**	**99.98%**	**99.98%**
Sample "5" Control	**$1.60x10^9$**	**N/R**	**N/I**
Sample "5" Treated	**<10**	**99.99%**	**99.99%**

The overnight culture contained a concentration of $1.84x10^8$ CFU/ml and was diluted 1:100 for application to the substrate of 1ml at an estimated $1.84x10^6$ CFU/ml.

The following formulas were used to calculate the percent reduction:

Percent Reduction (R) = [(A-B) / A] x 100

Where:

A = Population of bacteria/fungi recovered from untreated samples after 24 hrs of contact
B = Population of bacteria/fungi recovered from treated samples after 24 hrs of contact

Note:
NR= No Reduction
NI = No Inhibition