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14. ABSTRACT

Antimicrobial treatment of clothing and other textile gear is intended to provide advanced protection to the Warfighter in the field by controlling microorganisms that cause problems ranging from odor, skin irritation, athlete's foot, rashes and overall comfort, thereby relieving stressors that can reduce or impair the Warfighter's performance. In addition to providing enhanced protection to the Warfighter, antimicrobially treated textile items may not require laundering as often leading to a reduction in water usage - a benefit highly desired in combat environments.

The objective of this research was to identify, incorporate, and evaluate emerging and promising, commercially available antimicrobial treatments/technologies on military items to provide the Warfighter with advanced protection from unwanted microorganisms in combat operations. The selected antimicrobial treatments were evaluated in both laboratory and field wear tests to demonstrate efficacy to a broad spectrum of microorganisms and durability to wear and multiple launderings without adversely affecting the properties and functionality of the items

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Antimicrobially Treated Products for Military Use

Final Report

Study for U.S. Army Natick Soldier Research, Development and Engineering Center
Contract No.: W911QY-04-C-0079

by

Dr. H. Wayne Swofford, William Hanrahan, Duane Centola, Crystal Isenhour, Joshua Smith, David Ramey, Jeff Chandler

Field Study Conducted by U. S. Army Natick Soldier RD &E Center

Excerpts from
"**Antimicrobial Treated Clothing Items: Field User Evaluation Report**," and
"**Antimicrobial Treated Clothing Items – Part II: Field User Evaluation Report**,"
by Chuck Greene
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Executive Summary

Background

This report is being submitted under Contract No. W911QY-04-C-0079, "Antimicrobially Treated Products for Military Use," performed by Microban Products Company (MPC), Huntersville, NC. The project was awarded to address "Advanced Protection and Integration Technologies and Systems," solicited under the BAA Solicitation Number "03 –5 Natick BAA," Section VI, Part D, "Warrior Systems Technologies," Item 11. Antimicrobial treatment of clothing and other textile gear is intended to provide advanced protection to the Warfighter in the field by controlling microorganisms that cause problems ranging from odor, skin irritation, athlete's foot, rashes and overall comfort, thereby relieving stressors that can reduce or impair the Warfighter's performance. In addition to providing enhanced protection to the Warfighter, antimicrobially treated textile items may not require laundering as often leading to a reduction in water usage - a benefit highly desired in combat environments.

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Experimental

The study was divided into two main portions: the first portion was to antimicrobially treat the Advanced Combat Uniform (ACU) fabric, polyester T-shirt fabric, and boot sock on a pilot scale and then test for durability and efficacy using laboratory microbiological tests against a panel of organisms of Military interest/concern selected by Natick Soldier RD&E Center; the second portion was to conduct a field wear evaluation with Soldiers wearing treated ACUs, T-shirts, and boot socks.

Products were treated by MPC on a pilot scale at Cotton, Inc., Akwatek, and Pickett Hosiery. The samples were brought back to MPC for testing. Microbiological testing was done via AATCC Test Method 100, Test Method 147, and Test Method 30, part III (fungal). Analytical testing was done via HPLC and LC-MS. After the laboratory testing was completed, MPC had fabric antimicrobially treated and items manufactured from the fabric for use in the field evaluation, using current manufacturers of the products and current manufacturing processes.

In August 2005 the Natick Soldier RD&E Center conducted a field wear evaluation (Field Evaluation I) of antimicrobially treated uniforms, T-shirts, and socks with Soldiers from the 1st Battalion, 31st Air Defense Artillery (ADA) Brigade at Fort Bliss, Texas. The treatments evaluated were targeted against common, but harmful, bacteria routinely encountered by Soldiers in the field. The goal of the evaluation was to determine if the protection provided by these treatments would manifest itself in ways that would be perceivable to Soldiers through

occurrence of a wide range of problems and conditions that could be caused by the targeted microbes as well as through other more subjective measures. The items were worn during a seven day field training exercise conducted in the training area at Fort Bliss. While the primary mission of the unit involved is Air Defense, they trained for Infantry duties for future deployment. Data was collected through a series of questionnaires on the day items were issued (background), on day four (midpoint) and on day seven (final). This also allowed for a within-groups dimension to the evaluation to assess performance of the items over time.

Because the data was inconclusive, there were questions regarding the survey methodology, and anecdotal comments indicated benefits that were not captured in the surveys, the field wear evaluation was repeated (Field Trial II) in April of 2006 with Soldiers from the 4th Brigade Combat Team, 1st Cavalry Division, at Fort Bliss, TX. A primary difference in this evaluation from the previous evaluation was that Soldiers were given either all treated or all untreated garments for wear and evaluation, to test the antimicrobial treatments, as opposed to the previous attempt to evaluate each individual type of garment separately. Additionally laboratory testing was preformed on worn items.

Key Findings

MPC demonstrated that the ACU, polyester T-shirt, and boot sock could be antimicrobially treated during normal manufacturing operations without affecting performance requirements for the treated articles. The combination of triclosan and PHMB was particularly efficacious, giving a broader spectrum of efficacy on unworn items against three of the four organisms in the laboratory test panel of organisms requested by Natick Soldier RD&E Center and having somewhat enhanced durability.

Additional benefits of antimicrobial treatment with triclosan were reduced wash fading of the ACU and reduced pilling for the polyester T-shirt. The reduced fading may be due to a combination of the MLF 9200-20 and R10800-0 rather than just the use of triclosan, and it is unclear whether the reduced pilling is due to the triclosan or the use of triclosan in the MLF 9200-200 carrier system.

Unfortunately, the results from Field Evaluation I were statistically inconclusive due to a number of possible factors; however, there were trends that suggested that the T-shirt and the boot socks could benefit from the use of antimicrobial technology. Soldiers wearing the antimicrobial T-shirts reported reductions in odor as well as other problems such as general discomfort, heat rash, itching skin, and skin rash, with a reported decrease in intensity over the course of the study and compared with past exercises. Similarly the antimicrobial socks showed an overall reduction in the percentage of Soldiers who reported foot odor, general discomfort, and itching.

There is also some anecdotal evidence that emerges from the data to lend weight to the argument in favor of the antimicrobial treatments. For example, three Soldiers in the treated uniform group reported that they did not experience diarrhea in the field when they normally do. None of the respondents in the treated T-Shirt group reported that they developed a problem that they would not normally have compared to 13% in the untreated group. Fifty percent more Soldiers in the treated sock groups reported a reduction in chronic foot problems when compared to

Soldiers in the untreated groups. There are other examples; these three are cited here in order to be brief.

In addition, three-fourths of our evaluation participants reported that they buy and use products branded as "antimicrobial" for use in the field. More than two-thirds of the survey group urged the adoption of antimicrobial treated clothing items either as standard or optional purchase items and one-fourth felt that the treatments should continue to be researched. Only 6% felt that there was no merit to the technology. It is clear that Soldiers in general are interested in antimicrobial products. They endorse the use of antimicrobial technologies on military items. The fact that Soldiers in the untreated group felt the exact same way as those in the treated group also seems to indicate that they recognize that the benefits of these technologies may not always be apparent. All of this, when taken together, offers a clear basis for further consideration of the antimicrobial technologies evaluated.

Based on the laboratory findings combined with the trends that were observed during the field study, a redesigned field evaluation was performed to obtain significant, differentiating data to make conclusions regarding the benefits of antimicrobial technologies to the Soldier. In the second field evaluation Soldiers were issued either treated T-shirts and treated uniforms together or untreated items together to clearly delineate between antimicrobial technologies and untreated items.

During the second field wear evaluation, numerous significant differences were detected between the treated and untreated groups for uniform and T-shirt performance. Significantly more Soldiers felt that the treated uniform and T-shirt were controlling their body odor than those in the untreated group. Significantly more Soldiers felt that the treated uniform and T-shirt decreased their discomfort, how dirty they felt, and the amount of time spent on personal hygiene. A related significant decrease in the frequency of use of certain hygiene products was also noted in the treated group over the untreated group. Significantly more Soldiers felt that the treated uniform and the T-shirt could be worn longer before needing to be changed; additional days were estimated at three for the uniform and two for the T-shirt compared to one each for the untreated uniforms and T-shirt. Significantly more Soldiers felt that the treated uniform was more comfortable for extended wear than the untreated items.

No significant differences were detected for the rate or intensity of various physical problems for those wearing treated or untreated items; however, we feel that this may have more to do with the format of the question than the properties of the uniform treatment. This is based on the lack of variability apparent in the data. Also, there was some data that seemed to indicate that the opposite may be true and that the treatments did have an impact on these problems.

Findings for the sock were somewhat muted by the low number of respondents in each of the two treated groups (TypB: n=37, Type C: n=24). Also, we did not collect as much data on the sock as we did on the other items – it was considered secondary on the questionnaire to the uniform and T-shirt. However, there seemed to be some impact of the sock treatments on foot odor, hygiene practices, comfort and suitability for extended wear, and safety. A significantly higher percentage of Soldiers with the Type B (triclosan and PHMB combination) felt that it could be worn longer before needing to be changed than those with the untreated sock. A

significantly higher percentage of Soldiers also felt that the Type B sock reduced foot problems more than those in the untreated group. In general we feel that the results of the previous evaluation are more important to assessing treatment performance when used on socks. This data should be viewed as complementary. At some point it might be useful to do a separate dedicated evaluation of antimicrobial socks.

In the second field trial, the application of an antimicrobial treatment, particularly to the T-shirt and uniform, seemed to offer a range of benefits to the user. These included improved odor control, comfort, hygiene, and wear time. Other benefits, to include those related to physical problems and quality of life, are possible but could not be validated based on the available questionnaire data. There is also a great deal of interest amongst the Soldiers in the use of antimicrobial products as a treatment for field uniforms and T-shirts; three-fourths of the Soldiers believe in the effectiveness of these products.

In addition to the results summarized in this work and report, there is a follow-up study in progress, Project W911QY-05-C-0087, "Advanced Antimicrobial & Comfort Technologies for Military Applications." The second study involves the combination of antimicrobial treatment and moisture management finishes on cotton T-shirts as an alternative to polyester T-shirts. An additional part of the study involves the antimicrobial treatment of sleeping bag systems to protect against attack by fungal organisms that damage the fabric as well as reduce bacterial loadings on the fabric.

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Introduction

This report is being submitted under Project No. W911QY-04-C-0079, "Antimicrobially Treated Products for Military Use," performed by Microban Products Company, Huntersville, NC. The project was awarded to address "Advanced Protection and Integration Technologies and Systems," solicited under the BAA Solicitation Number "03 –5 Natick BAA," Section VI, Part D, "Warrior Systems Technologies," Item 11. Antimicrobial treatment of clothing and other textiles intended to provide advanced protection to the Warfighter in the field by controlling microorganisms that cause problems ranging from odor, skin irritation, athlete's foot, rashes and comfort, thereby relieving stressors that can reduce or impair the Warfighter's performance. In addition to providing enhanced protection to the Warfighter, antimicrobially treated items may not require laundering as often leading to a reduction in water usage and increase the life of the items - a benefit highly desired in combat environments.

Objective

The objective of this research was to identify, incorporate, and evaluate emerging and promising, commercially available antimicrobial technologies on military items to provide the Warfighter with advanced protection from unwanted microorganisms. The selected antimicrobial technologies were evaluated in both laboratory and field wear evaluation to demonstrate efficacy to a broad spectrum of microorganisms and durability to wear and multiple launderings without adversely affecting the properties and functionality of the items

Military personnel in the field have varied access to facilities for bathing themselves and laundering their clothes. These may range from the adequate at well-developed support bases, rudimentary near the front lines, to nonexistent in forward areas and combat zones. Troops in combat or undertaking special operations may need to wear the same clothes for days, with limited changes available. Hygiene problems are then exacerbated in hot, humid climates where personnel are perspiring to a greater extent.

The problems arising from these conditions can range from simple comfort problems with odor, itching and rashes as bacteria proliferate on the skin and in clothing and other gear. Odor can also be an operational problem in the field when in close proximity to hostile forces. These problems occur because of bacteria that feed on skin cells and perspiration, proliferate and excrete substances that are odoriferous and irritating to the skin. As an example, the itching and odor that occur when someone has worn a cast for a long period of time is due to bacterial growth between the cast and the skin. Similar problems can occur in clothing worn for long periods of time.

In addition to the hygiene and comfort of personnel, bacteria, mold, and mildew are known to shorten the useful life of clothing and other gear in the field. In this case the microbes feed on the materials damaging the physical properties of the materials, and leading to failure during use. While this may seldom represent a safety problem in the field, assuming proper inspection of equipment, it certainly adds to the overall costs of operations.

What is needed is a method of applying antimicrobial technologies to military items so as to inhibit the growth of bacteria, mold, and mildew in a cost effective manner such that the treatment does not negatively affect the physical properties of the materials. The technology should also be safe for extended skin contact, food contact if applicable, and be durable over the life of the items.

Although such technology already exists and is in use in the commercial/civilian sector in underwear, socks, shirts and general sportswear, product or formulation optimization is required to meet the more stringent requirements for military applications. Field wear evaluations of antimicrobial treatments have not been done previously, particularly under the harsh conditions expected during military operations. Commercially available data on benefits of antimicrobial treatments is limited to small odor studies.

The objectives of this study were:

- to demonstrate that antimicrobial technologies could be applied to military items;
- to evaluate and compare available test methods to use in establishing specifications for antimicrobial technologies;
- to manufacture antimicrobially treated military items for use in a field wear evaluation;
- and to conduct a field wear evaluation to evaluate performance, with emphasis on odor control and comfort. The field evaluation was designed and conducted by the Natick Soldier RD&E Center, Natick, MA.

Experimental

Antimicrobial Treatments

The antimicrobials used for treating the fabrics in this study were triclosan (2,4,4'-trichloro-2'-hydroxydiphenyl ether), quat silane (3-(Trimethoxysilyl)propyl dimethyl octadecyl ammonium chloride), and PHMB (poly(hexamethylene biguanide) hydrochloride or poly(iminoimido-carbonyliminoimidocarbonyliminohexamethylene) hydrochloride). Microban Products Company supplied the triclosan as MLF 9200-200 and the quat silane as MLF SiS 7200 AM. The PHMB was supplied by Arch Chemical Company as Reputex 20. In pad application on cotton and poly/cotton fabrics the MLF 9200-200 was applied with a fixing agent, MLF R10800-0.

Fabrics and Treatment Procedures

ACU: Mil-C-44436-Class 8

The fabric for the Advanced Combat Uniform (ACU), a sixty inch inside width, 6.5 oz per square yard, 50% / 50% nylon / cotton rip stop fabric, was dyed and printed in Universal pattern. The application of the antimicrobial technology was achieved utilizing a pad and stenter frame.

The antimicrobial chemicals were added to the standard wrinkle free pad bath. The wrinkle free pad bath consisted of a resin finish and softeners. The pick-up rate of the fabric was 50%. The fabric was dried and cured in a four zone dryer with temperatures of 410F, 420F, 430F and 430F respectively. The speed of the frame was set at 80 yards per minute resulting in a fabric temperature of 330°F +/- 5°F.

For the initial screening trials, the triclosan was added as Microban MLF 9200-200 at 2% with 4% MLF R10800-0, the quat silane as 3% Microban MLF SiS 7200 AM, and the PHMB as 2% Reputex 20, based on the dry weight of the fabric. For the field studies the antimicrobial treatment combined the triclosan and PHMB with 1.5% Microban MLF 9200-0, 3% MLF R10800-0, and 2% Arch Reputex 20. The actual letdown in the bath would depend on the percent pickup so that at 50% pickup, the letdown in the bath for the field studies for MLF 9200-200 would have been 3%, i.e. double the target rate on the dry fabric.

T-Shirt: CR/PD 04-13 Sand

The fabric, a tubular knit 100% polyester 22/1 MJS in various diameters (to accommodate different sizes), was treated with Akwatek, the moisture management chemical. The application of the antimicrobial treatment was achieved utilizing two processes; a pressurized jet dyeing machine and a pad.

The antimicrobial Microban MLF 9200-200 was applied via the jet dye machine. The various diameter knit fabrics were sewn together to achieve a 1000 pound dye lot. The fabric was loaded into the jet and a volume of water was added to attain a 10:1 liquor ratio. The dye stuff and auxiliaries were added and the bath was buffered to a pH of 5.5. Microban MLF 9200-200 was then added. The temperature of the bath was raised at a rate of 3° C per minute to 130°C. Once the bath came to temperature a 45 minute dye cycle was run. Upon completing the dye cycle the bath was cooled at a rate of 2°C per minute to 55°C. The machine was then drained and unloaded.

The antimicrobials PHMB (Arch Reputex 20) and quat silane (MLF SiS 7200 AM) were applied via a two place pad, co-applied with the standard silicone softener. The first pad reduced excess water from the jet to a 60% pre-wet condition. The second pad added the softener and antimicrobial at a rate to attain an additional 15% wet pick-up. The fabric was the dried in a conventional dryer at 245° F.

For the initial laboratory screening triclosan was applied as MLF 9200-200 at 2%, the quat silane as MLF SiS 7200 AM at 3%, and the PHMB as Arch Reputex 20 at 2%, based on weight of the dry fabric. For the field evaluation, triclosan and PHMB were combined, with the triclosan being applied as MLF 9200-200 at 1.5% and the PHMB being applied as Arch Reputex 20 at 2%, based on weight of dry fabric. For the field evaluation the triclosan was applied first in the jet dye machine and then the PHMB was applied in the pad bath with the softener.

Boot Sock: CR/PD 03-18

The boot sock was an over the calf style double welt top having a double covered elastic yarn. The fiber content was 85% cotton, 10% nylon and 5% spandex/nylon. The standard antimicrobial military boot sock calls for a 2.5% silver coated nylon fiber. For this evaluation the silver coated nylon fiber was replaced with a standard 100% nylon fiber as to not cause a false positive result in testing.

The socks were knit, scoured and dyed in the normal fashion. The antimicrobial technology was applied in the atmospheric paddle dye machine during the softener application cycle. The bath was buffered to a pH of 5.5 with acidic acid; the temperature was raised to 115°F and run for 25 minutes. The machine was then drained and the socks removed. The socks were dried in a gas-fired tumble dryer at a temperature of 300°F for 45 minutes, removed from the dryer and boarded.

As with the other applications, initial screening was done at 2% Microban MLF 9200-200 with 4% R10800-0 for triclosan, 2% Arch Reputex 20 for PHMB, and 3% Microban MLF SiS 7200 AM for quat silane, based on dry weight of the socks. For the field evaluation triclosan and PHMB were used together, using 1.5% Microban MLF 9200-200, 3.0% Microban R10800-0, and 2.0% Arch Reputex 20, based on the dry weight of the socks.

Test Protocols

Microbiological Efficacy Testing

The organisms used for laboratory testing of unworn items were:

Streptococcus pyogenes (ATCC 8669);
Staphylococcus aureus (ATCC 6538);
Pseudomonas aeruginosa (ATCC 15152);
Corynebacterium diphtheriae (ATCC 19409);
Tricophyton mentagrophytes (ATCC 9533).

Bacterial testing was done using a modified version of the AATCC (American Association of Textile Chemists & Colorists) Test Method 100 (TM 100) and the AATCC Test Method 147 (TM 147). The TM 100 is a quantitative, inoculate and recover method while the TM 147 is a qualitative, zone of inhibition method. Antibacterial testing was done via both methods in order to

compare results from the two test methods. The TM 100 was modified to accommodate the use of a spiral plater and colony counter.

Initially the American Association of Textile Chemist and Colorists (AATCC) TM 100 was further modified by using a lower nutrient level for the inoculum consistent with test methods developed for silver-based antimicrobials. The lower nutrient level was the same as described in the JIS (Japan Industrial Standard) 2801. For the AATCC TM 100 the nutrient broth is BHIB (brain heart infusion broth) with 462 g of nutrient per 1 liter of total broth. The JIS method uses1 part of the BHIB to 500 parts of water so that the nutrient level is $1/500^{th}$ of that for the TM 100. The reason for running at this lower level, consistent with the JIS 2801, was to have a test that could be applicable to silver as well as to organic antimicrobials. At this level of nutrient, however, we were unable to reproducibly run the tests, particularly with *Corynebacteriu*m and *Streptococcus*; the level apparently being insufficient to maintain the viability of the organisms even without the presence of an antimicrobial. This problem has been encountered in other testing on plastic, where MPC has found that the higher nutrient levels in the AATCC TM 100 lead to better reproducibility in the testing. Ultimately, the testing was run with the nutrient levels as specified in the AATCC TM 100.

Tryptic soy agar (TSA) plates were used rather than standard nutrient agar plates because it was determined that the *Corynebacterium* and *Streptococcus* would not reproducibly grow on the standard nutrient agar. The TSA was used for both the AATCC TM 100 for plating the recovered inoculum and the AATCC TM 147, for all organisms.

The AATCC TM 147 calls for 1 ml of an overnight culture to be diluted into 9 ml of saline, for a 1:10 dilution, before streaking onto the plate. This extra dilution, particularly with the *Corynebacterium* and *Streptococcu*s, which were slow growing, added further variability to the testing. As a result the overnight cultures were used for streaking.

Sterile glass beads were added to the *Corynebacterium* overnight culture and the tube vortexed thoroughly before standardization. Without this step it was almost impossible to ensure an accurate standardization because the organism tends to clump and form a slime coat, making it difficult to separate without the glass beads.

Fungal testing for *Tricophyton mentagrophytes* was done using the AATCC Test Method 30, part III (TM 30).

The test method protocols are included in Appendix IV for the above test methods.

Chemical Analysis

The antimicrobial actives were quantitatively analyzed on treated fabrics by cutting up swatches of the material into pieces roughly 5 mm square and extracting accurately weighed portions (0.2 to 1 gram) with 25 ml of methanol using a microwave accelerated reaction system (MARS-X CEM Corporation, Matthews, NC). Conditions for the extraction are as follows:

Power:	1200 watts
Temperature Program:	Ambient to 100º C @ 5º C per minute with a 15 minute holding time
Stirring Setting:	3

The extracts were collected and analyzed using HPLC and LC-MS conditions dependent upon the specific analyte present.

- For Triclosan:

Column:	C18, 15 cm x 4 mm, 5 µm (Supelco Discovery)
Column Temperature:	40º C
Solvent System:	20% acetonitrile, 80% water, isocratic
Flow Rate:	0.6 ml/minute
Injection Volume:	2 µl
Detection:	LC-Mass Spectrometer (Shimadzu LC-MS 2010 EV), Electrospray Ionization, negative ion mode, selective ion monitoring at 286.9 amu.
Detection Limit:	approximately 200 ppb in solution, or 25 ppm per solid sample.

Under these conditions, Triclosan was detected at approximately 4.7 minutes.

- For PHMB:

Column:	25 cm x 0.46 cm Discovery C18 (Supelco)
Column Temp:	Ambient room temperature
Solvent System:	5% ACN/95% 20 mM potassium phosphate dibasic buffer, pH 7.3, containing 40 mM tetrabutylammonium bromide
Flow Rate:	1.0 ml/min.
Injection Vol:	5 µl
Detection:	Perkin-Elmer Series 200 HPLC System, UV at 237 nm
Detection Limit:	approximately 10 ppm in solution, or 1250 ppm per solid sample, so the ND level was very high for PHMB.

Under these conditions, PHMB was detected at approximately 10 minutes.

Microban could not develop an analytical technique for the quaternary ammonium silane. This antimicrobial actually crosslinks onto the textile, reacting through the Si-OH groups on the hydrolyzed silane. Digesting the material typically results in fragmentation of the silane into a number of components, such that it is difficult to quantify the original amount. Compounding the problem is the fact that the quat moiety is not detectable with UV, the primary detection method for HPLC and GC.

Analytical test methods are described in Appendix V. Analysis for triclosan is described in Appendix V.a, while the general HPLC method is outlined in Appendix V.b.

Durability

Durability of treatment was tested using wash wheel Method 5556.1, Federal Test Method Std. No. 191A, Title: "Mobile Laundry Evaluation for Textile Materials."

This method is intended for use where it is desired to reproduce, by means of a laboratory procedure, changes in dimensions of woven or knitted cloth and measure the durability and efficacy of functional finishes which simulate field conditions. A 20 pound standard load of material and ballast is required. A cylindrical wash wheel of reversing type is used. The wheel (cage) is 24 inches in diameter and 24 inches inside length. The temperature of the water used is 140º F. The liquid laundry detergent was provided by American Association of Textile Chemists and Colorists (AATCC), AATCC 8804. Following a 10 minute wash cycle and a 24 minute rinse cycle the material is extracted in a laundry type centrifugal extractor approximately 11 inches deep by 17 inches in diameter with an operating speed of 1500 rpm. The material is dried in a rotary, tumble type drier at 160º F for 45 minutes.

Samples of ACU fabric, t-shirt fabric, and socks were tested for microbiological efficacy and treatment level after 5, 10, 25, and 50 washes.

Field Evaluations

Field Evaluation I

(Excerpted from "Antimicrobial Treated Clothing Items: Field user Evaluation Report," by Chuck Greene, U. S. Army Natick Soldier Center. See Appendix I.)

In August 2005 the Natick Soldier RD&E Center conducted a field user evaluation of antimicrobial treated uniforms, T-shirts, and socks with Soldiers from the 1st Battalion, 31st Air Defense Artillery (ADA) Brigade at Fort Bliss, Texas. The treatments evaluated were targeted against common, but harmful, bacteria routinely encountered by Soldiers in the field. The goal of the evaluation was to determine if the protection provided by these treatments would manifest itself in ways that would be perceivable to Soldiers through an impact on a wide range of problems and conditions that could be caused by the targeted microbes as well as though other more subjective measures.

All of the Soldiers participating were issued treated or untreated versions of either the uniform or the T-shirt. All of the participants were also issued one of three types of socks: untreated, treated type 1, or treated type 2. The items were used during a seven day field training exercise conducted at McGregor Range, Fort Bliss. While the primary mission of the unit involved is Air Defense, they left their Patriot launchers behind and trained as Infantry for future deployment in that role. Data was collected through a series of questionnaires that addressed criteria relevant to

the assessment of antimicrobial treatments. A total of seven different questionnaires were used, copies of which are included as Attachments A through G. Approximately 300 Soldiers from the 1st Battalion, 31st Air Defense Artillery (ADA) Brigade participated, with 207 completing all of the evaluation requirements.

Item Description

There were a total of seven items under evaluation: two versions of the standard uniform (treated and untreated), two versions of the standard polyester T-shirt (treated and untreated), and three versions of the standard cotton sock (untreated, treated 1, and treated 2). A description of each of the items is included below.

- Uniform: Two versions of the standard Advanced Combat Uniform (ACU), one featuring a Microban® antimicrobial treatment formulation (type A) and one untreated (type B). Both uniforms also featured a wrinkle resistance treatment, which is standard for this uniform. Soldiers evaluating this item were issued one complete uniform to evaluate.
- T-shirt: Two versions of the standard issue 100% Polyester T-shirt, one featuring a Microban® antimicrobial treatment formulation (Type A) and one untreated (Type B). Soldiers evaluating this item were issued four T-shirts of the same type.
- Socks: Three versions of the standard issue 100% cotton socks: an untreated standard sock (Type A); the current treated standard sock, CR/PD 03-18 (Type B); and an untreated standard sock that received a Microban® antimicrobial treatment (Type C).

Test Design & Procedures

The evaluation of the antimicrobial treated uniforms, T-shirts, and socks featured a between-groups design. The independent variable was item type (treated vs. untreated) and the dependent measures were Soldier responses to an array of survey questions designed to quantify experiences and opinions relevant to the performance of the treatment. Data was collected at the beginning (background), half-way point of the evaluation on day four (midpoint) and at the conclusion on day seven (final). This also allowed for a within-groups dimension to the evaluation to assess performance of the items over time.

Input on questionnaire items was provided by Dermatologists at William Beaumont Army Medical Center, Fort Bliss, TX to ensure that content was appropriate in terms of the skin problems and conditions that Soldiers would experience in the field. The key questions and the primary scale used on the questionnaire were based on existing symptom assessment scales with proven reliability and validity and designed for use to collect data in "self-reporting" scenarios. Other questions were developed as appropriate.

Because the number of items exceeded the number of Soldiers, multiple items would have to be issued to each participant. Evaluation groups were developed to minimize any potential cross-over effects from one item to another. Four groups were defined: A (antimicrobial treated uniform and untreated socks), B (untreated uniform and antimicrobial treated socks), C (antimicrobial treated T-shirts and antimicrobial treated socks), and D (untreated T-shirts and

antimicrobial treated socks). Assignment to any group was purely random and Soldiers did not know if any of the items issued to them had the antimicrobial treatment or not.

Participants were briefed several days before the evaluation began on the purpose and procedures. They were informed that they would be evaluating antimicrobial clothing treatments and that some would receive treated items and some would receive untreated items. At that time they completed a background questionnaire to obtain demographic information and data on past experiences that would be relevant to the evaluation. Participation was voluntary. Several days later the participants were issued the test items and were given the opportunity to try them on to make sure they fit. A few Soldiers had to change assigned groups at this point due to size availability, but the randomness of the assignment was maintained.

A final roster was maintained and each Soldier was double-checked to make sure that they had received the correct items and that they had completed the background questionnaire before they left the issue location. Once items were checked for type (treated or untreated), the Soldier initialed for the items received and the combination of Soldier and items was logged into the evaluation. The issue was conducted over a two-day period. Twelve hours after the last item was issued the unit deployed to the training area for a seven-day field training exercise. The weather observations for the evaluation period are presented below in Table 1.

Table 1

Weather Conditions at Fort Bliss, 17 to 23 August 2005

Reporting station: El Paso, TX International Airport

August:	**17**	**18**	**19**	**20**	**21**	**22**	**23**
Temp (max)	90	91	84	83	89	85	89
Temp (min)	72	70	66	67	68	72	70
Precipitation (inches)	.1”	.1”	.6”	T*	T*	T*	.1”
Relative Humidity (avg.)	59%	62%	73%	71%	61%	62%	57%

[1] From: NOAA, National Climatic Data Center
http://www.ncdc.noaa.gov/oa/ncdc.html
*T=Trace amount

The unit was visited briefly by the data collection team on the first day to determine their location in “the box” and to collect some initial informal feedback. The Soldiers then conducted training for the next 72 hours. During this time, the data collection team finalized the evaluation roster and prepared the individualized questionnaire sets to be completed by each participant. Midpoint data was collected on the evening of day four. The unit then conducted an additional 72 hours of training. The process was repeated on the evening of day seven, and the evaluation was completed at that time.

(End of excerpt.)

The details of the first field study, its design and results, are more fully described in Appendix I.

Field Evaluation II

(Excerpted from "Antimicrobial Treated Clothing Items – Part II: Field User Evaluation Report," by Chuck Greene, U. S. Army Natick Soldier Center. See Appendix ????.)

In April 2006 the Natick Soldier RD&E Center conducted a field user evaluation of antimicrobially treated uniforms, T-shirts, and socks with Soldiers from the 4th Brigade Combat Team, 1st Cavalry Division at Fort Bliss, Texas. The treatments evaluated were targeted against common, but harmful, bacteria routinely encountered by Soldiers in the field. The goal of the evaluation was to determine if the protection provided by these treatments would manifest itself in ways that would be noticeable to Soldiers. This field user evaluation was conducted as a follow-on to one conducted at Fort Bliss in August 2005 (see OFIG Report: **Antimicrobial Treated Clothing Items Field User Evaluation Report**, dated 21 November 2005). All of the members of the participating unit were issued either untreated (type A) or treated (type B) versions of the uniform and the T-shirt. All of the participants were also issued one of three types of socks: untreated (type A), treated (type B), or treated (type C). The items were used both in the field and in garrison at Fort Bliss during a two-week period. While the training schedule of the different companies within the unit varied, they all spent approximately one week in the field and one week in garrison. Data was collected through a series of questionnaires that addressed criteria relevant to the assessment of antimicrobial treatments. Two primary questionnaires were used to assess treatment performance, copies of which are included as Attachments A (background) and B (final). A total of 217 Soldiers from the unit were issued items, with 185 completing all of the data requirements of the field user evaluation.

Item Description

There were a total of seven items under evaluation: two versions of the standard Army Combat Uniform (untreated and treated), two versions of the standard polyester T-shirt (untreated and treated), and three versions of the standard cotton sock (untreated and two treated). A description of each of the items is included below. The basic garments and any formulations and treatments were the same as those used in the August 2005 evaluation.

- Uniform: Two versions of the standard Advanced Combat Uniform (ACU), one featuring a Microban® antimicrobial treatment formulation (type A) and one untreated (type B). Both uniforms also featured a wrinkle resistance treatment, which is standard for this uniform. Soldiers evaluating this item were issued one complete uniform to evaluate.

- T-shirt: Two versions of the standard issue 100% Polyester T-shirt, one featuring a Microban® antimicrobial treatment formulation (Type A) and one untreated (Type B). Soldiers evaluating this item were issued four T-shirts of the same type.

- Socks: Three versions of the standard issue 100% cotton socks: an untreated standard sock (Type A); an untreated standard sock which received a Microban® antimicrobial treatment (Type B), and the current treated standard sock, CR/PD 03-18 (Type C).

Results of the Previous Evaluation

The goal of the first evaluation was to determine if the protection provided by an antimicrobial clothing treatment would manifest itself in ways that would be noticeable and beneficial to Soldiers. Based on the results of the evaluation, it appeared that the T-Shirt was a promising candidate for application of an antimicrobial treatment. Soldiers who used the treated T-Shirt reported a significant reduction in odor as well as an overall reduction in other problems, which included general discomfort, heat rash, itching skin, and skin rash. The results for the antimicrobial treated uniform were less promising, with no apparent reduction in problems noted. However, significantly more Soldiers felt that the treated uniform was controlling their body odor at the end of the evaluation. Soldiers who evaluated the treated uniform were all issued standard T-shirts that were treated. It was felt that the T-shirt may have performed better than the uniform because it was a "next-to-the-skin" item. Those who used antimicrobial socks showed an overall reduction in the percentage reporting foot odor, general discomfort, and itching. Overall, it seemed that an antimicrobial treatment offered benefits in terms of sock performance.

The results of the August 2005 evaluation raised a number of questions which were addressed in this effort. It was felt that the antimicrobial treatments were providing some noticeable benefits for soldiers, but it seemed that the questionnaires were not providing a clear picture of item performance. In response to this, the questions were redesigned to assess the performance of the treatments in four areas: injury reduction, odor reduction, comfort, and impact on hygiene practices. Soldiers would be issued either all treated items or all untreated items to improve the possibility of measuring differences in performance between the groups. There was also discussion about the impact of a "placebo effect." The August 2005 evaluation was a blind study and it was unclear if the Soldiers' assumptions about whether they had a treated or untreated item had an impact on their perceptions of item performance. This second evaluation would also be a blind study; however, on the final page of the final questionnaire, it was revealed which type of items the Soldiers were evaluating. They were then asked to indicate how this changed their opinions about antimicrobial treatments in general, and the items they evaluated specifically.

Test Design & Procedures

The evaluation of the antimicrobial treated uniforms, T-shirts, and socks featured a between-groups design. The independent variable was item type (treated vs. untreated) and the dependent measures were Soldier responses to an array of survey questions designed to quantify experiences and opinions relevant to the performance of the treatment. Assignment to either evaluation group was done by company and Soldiers did not know if any of the items issued to them had the antimicrobial treatment or not. Data was collected in a background questionnaire before the field wear evaluation began, at the halfway point of the evaluation on day four (midpoint), and at the conclusion of the evaluation on day eight (final).

The questionnaires used were derived from those used previously, which had been developed with input provided by Dermatologists at William Beaumont Army Medical Center, Fort Bliss,

TX, to ensure that content was appropriate in terms of the skin problems and conditions that Soldiers would experience in the field. The 3 key questions and the primary scale used on the questionnaire were based on existing symptom assessment scales with proven reliability and validity and designed for use to collect data in "self-reporting" scenarios. Other questions were developed as appropriate. In addition, the questionnaires were revised and expanded prior to the evaluation based on lessons learned.

Participants were briefed the day before the evaluation began on the purpose and procedures. They were informed that they would be evaluating antimicrobial clothing treatments and that some would receive treated items and some would receive untreated items. At that time they completed a background questionnaire to obtain demographic information as well as data on past experiences and their opinions on antimicrobial products and treatments. They were then issued the test items and were given the opportunity to try them on to make sure they fit. A few Soldiers had to change assigned groups at this point due to size availability of the various clothing items.

A final roster was maintained and each Soldier was double-checked to make sure that they had received the correct items and that they had completed the background questionnaire before they left the issue location. The issue was conducted on a Friday and the initial plan, based on the unit training schedule, was for the entire unit to spend the following Monday through Thursday in the field. On Thursday evening they would complete the midpoint questionnaire. On Friday, they would come out of the field. Soldiers were instructed by an operation order issued by the unit not to launder the uniforms and to wear them again the following week where the same schedule would be repeated: in the field Monday through Thursday, with final data being collected on Thursday evening.

At some point, the unit training schedule changed and only one of the companies went to the field for the first week (Alpha – predominantly untreated items) with the remainder staying in garrison (Headquarters & Headquarters Company, Bravo, Delta, Echo, and Foxtrot – predominantly treated items). For week two the schedule would be reversed, with Alpha being in garrison and the remainder going to the field. If the evaluation team had been aware of this at the time of issue, changes could have been made to minimize the impact on the evaluation. Ultimately, we have sufficient valid data to draw conclusions about the performance of the antimicrobial treatment. However, certain adjustments had to be made to the test design and the subsequent reporting of results.

All of the companies spent a week in the field and a week in garrison. Most of the soldiers wearing the treated uniforms were in garrison for the first week and then spent the following week in the field. Most of the soldiers with the untreated uniforms did the opposite: in the field for the first week and then in garrison for the second week. This largely invalidates the data collected at the midpoint because of the radical difference between field and garrison training. The final questionnaire was revised extensively to reflect the changes to the training schedule and evaluation.

There were some minor variations in this revised training schedule. Some Soldiers had a training holiday on Friday, 7 April. Some of the Soldiers who spent the second week in the field deployed on Sunday, 9 April, which was an off day for Soldiers who spent the second week in

garrison. By the end of the evaluation all of the participants had worn the items for approximately eight days, four in garrison and four in the field. The data presented below in Table 2 shows that weather conditions were substantially similar for the entire two week period. In the end, we feel that as long as Soldiers did not wash the uniforms during the intervening weekend, the final questionnaire data is a valid measure of the performance of the antimicrobial treatments over an extended wear period.

Table 2
Weather Conditions at Fort Bliss, 3 to 13 April 2006

Reporting station: El Paso, TX International Airport[1]

April:	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**
Temp (max)	81	88	83	71	76	77	87	82	84	89	91
Temp (min)	50	53	65	51	51	46	47	63	57	51	60
Precipitation (inches)	0	0	0	0	0	0	0	T*	T*	0	T*
Relative Humidity (avg.)	16%	20%	19%	10%	23%	22%	9%	30%	18%	15%	17%

[1] From: NOAA, National Climatic Data Center
http://www.ncdc.noaa.gov/oa/ncdc.html
* T=Trace amount

Results

Microban Laboratory Testing

Results from the laboratory microbiological testing are shown in Tables 3-5. These are divided by clothing item, with the results for each active shown with each level of laundering. The values shown for the AATCC TM 100 are the average of three replicates, and are the percentages of colony forming units (CFU) remaining after 24 hours versus the original inoculum. For the AATCC TM 147 non-numerical values, I (Inhibition) or NZ/NI (No Zone/No Inhibition) is shown if at least two of the three values were at that level, or a numerical value is shown if at least two of the three replicates had a zone of inhibition.

For all three items of clothing there was initial efficacy for the unwashed materials. This frequently occurs in textiles due to residual by-products such as formaldehyde from wrinkle-free finishes and often to the finishes themselves, such as softeners, which may be cationic materials or quaternary amines. Here, the residual efficacy was gone by 5 launderings for the ACU and T-shirt, and by 10 launderings for the boot socks.

None of the antimicrobials that were tried was effective against *Pseudomonas aeruginosa*. *Pseudomonas* is an opportunistic pathogen common in wet environments; frequently it is a problem in cases of severe burns, biofilm formation on invasive devices such as catheters, and

immune suppressed patients. *Pseudomonas* is naturally resistant to a broad range of antimicrobials, typically having a very high MIC (minimum inhibitory concentration).

The results from the AATCC TM 147 were poorly correlated with the AATCC TM 100 results. Not all antimicrobials will give a zone of inhibition, which requires the antimicrobial to diffuse into the agar, with silver being a common example of one that does not. It was surprising that the quat silane and PHMB gave zones for some tests because they normally do not have a zone of inhibition. The AATCC TM 100 is considered the more universal test because an antimicrobial that will yield a zone typically will result in a reduction in the TM 100, but MPC's experience has been that even triclosan, well known for providing a zone of inhibition, does not always show a corresponding reduction in a TM 100.

Triclosan was primarily effective against *Staphylococcus aureus* and effective against *Streptococcus pyogenes* at very high levels. It was generally ineffective against *Corynebacterium diptheriae*. The triclosan-based finish was not durable for the ACU and boot socks, lasting less than 10 washes, which was surprising since it is completely contrary to experience in commercial applications. (The problem with triclosan durability will be discussed in more detail below.) The triclosan was highly durable for the polyester T-shirt. The triclosan level on the boot sock was unexpectedly multiples of the targeted amounts. We believe that this is due to exhaustion onto the sock, which we normally do not see with this finish but may occur due to the characteristics of the softener that is being used by this supplier, rather than application levels proportional to the wet pickup of the finish onto the sock.

The quat silane showed poor durability to laundering going to less than 25 launderings on the ACU, less than 10 launderings on the T-shirt, but greater than 25 launderings for the boot sock, for *Strep*. There was some activity against *Corynebacterium* on the boot sock and ACU, but none on the T-shirt. There was no efficacy against *Staph*. As mentioned above, MPC was unable to develop an effective analytical procedure to measure the amount of quat silane on the treated textile.

PHMB gave strong results for *Strep* and *Corynebacterium* with a surprising degree of durability. Normally PHMB is expected to be durable to no more than 25 launderings, but in this case had reasonable durability to as many as 50 launderings for all three applications with *Strep*. As washings increased, efficacy against *Corynebacterium* dropped the fastest, going out to 50 launderings for the ACU, possibly >25 for the boot sock, but less than 10 for the T-shirt. The analytical results for PHMB are not meaningful because the detection limits were higher than the application levels; PHMB is a straight chain molecule and does not absorb well in UV light, which is the mode of detection in HPLC.

Given the efficacy of the PHMB against two, *Strep* and *Corynebacterium*, of the four organisms and its durability, the PHMB was the obvious top performer of the three antimicrobials in this laboratory study. Triclosan was effective against the *Staph*, but had unexpectedly poor durability. Despite that, MPC recommended that the triclosan and PHMB be combined to treat the clothing items for the field study. The reasoning was that by combining the two, we could achieve broader efficacy and a possible synergistic effect, and MPC believed that it needed to

control *Staph*, which is the organism most commonly associated with skin irritation, itching, and normal skin infections.

As stated earlier the poor durability of the triclosan finish was a surprise, since normally MPC has obtained greater than 50 home launderings on cotton and polycotton blends, confirmed by outside and customer testing. The first samples were run on small pilot line, and the primary question is whether or not the treated products reached minimum temperatures for curing the binder.

Table 3

Laboratory Test Results for Antimicrobial Advanced Combat Uniform

		Active Conc.		AATCC Test Method 100					AATCC Test Method 147			
Active	**Durability**	**ppm**		**Staph**	**Strep**	**Coryn**	**Pseud**		**Staph**	**Strep**	**Coryn**	**Pseud**
Triclosan	Unwashed	1060		99.9%	99.8%	99.9%	66.6%		6.7	2.3	2.0	NZ/NI
	5	1067		95.2%	20.1%	NR	NR		8.7	I*	I	NZ/NI
	10	173		21.2%	53.3%	NR	NR		3.3	I	I	NZ/NI
	25	0		NR	NR	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	50	27		NR	NR	NR	NR		I	NZ/NI	NZ/NI	NZ/NI
Quat silane	Unwashed	N/A		99.9%	99.8%	99.9%	99.9%		NZ/NI	1.7	1.0	NZ/NI
	5	N/A		NR	99.8%	21.4%	NR		NZ/NI	3.7	2.0	NZ/NI
	10	N/A		NR	99.9%	99.9%	NR		I	2.3	2.7	NZ/NI
	25	N/A		NR	66.3%	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	50	N/A		NR	NR	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
PHMB	Unwashed	ND**		99.9%	99.8%	99.9%	NR		1.7	3.0	2.0	NZ/NI
	5	ND		99.8%	99.8%	99.9%	NR		1.0	2.7	7.0	NZ/NI
	10	ND		97.2%	99.9%	99.9%	NR		1.0	2.0	2.7	NZ/NI
	25	ND		66.6%	99.9%	99.9%	NR		NZ/NI	1.7	1.0	NZ/NI
	50	ND		NR	99.9%	99.1%	NR		NZ/NI	I	1.0	NZ/NI
Control	Unwashed			66.4%	99.8%	95.2%	NR		NZ/NI	2.0	NZ/NI	NZ/NI
	5			NR	NR	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	10			NR	NR	99.9%	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	25			NR	25.5%	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	50			NR	64.4%	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI

* I = Contact inhibition under sample

** <1250 ppm. Detection limits for PHMB were higher than target application levels, so results were not meaningful.

Table 4
Laboratory Test Results for Antimicrobial Polyester T-Shirt

		Active Conc.		AATCC Test Method 100					AATCC Test Method 147			
Active	**Durability**	**ppm**		**Staph**	**Strep**	**Coryn**	**Pseud**		**Staph**	**Strep**	**Coryn**	**Pseud**
Triclosan	Unwashed	3800		98.1%	99.8%	NR	NR		6.3	NZ/NI	NZ/NI	NZ/NI
	5	3700		33.2%	99.4%	NR	NR		7.0	NZ/NI	NZ/NI	NZ/NI
	10	3233		91.9%	99.0%	NR	NR		6.3	NZ/NI	NZ/NI	NZ/NI
	25	3233		99.4%	33.3%	NR	NR		5.0	NZ/NI	NZ/NI	NZ/NI
	50	950		99.3%	NR	NR	NR		5.0	NZ/NI	NZ/NI	NZ/NI
Quat silane	Unwashed	N/A		99.9%	99.8%	99.9%	NR		NZ/NI	1.0	1.0	NZ/NI
	5	N/A		NR	33.0%	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	10	N/A		NR	96.4%	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	25	N/A		NR	NR	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	50	N/A		NR	NR	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
PHMB	Unwashed	ND**		99.9%	99.9%	99.9%	99.9%		NZ/NI	1.7	1.7	NZ/NI
	5	ND		66.7%	99.8%	99.8%	NR		NZ/NI	NZ/NI	I*	NZ/NI
	10	ND		99.9%	99.9%	11.1%	11.1%		NZ/NI	NZ/NI	2.0	NZ/NI
	25	ND		NR	99.5%	33.0%	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	50	ND		NR	99.9%	33.3%	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
Control	Unwashed			66.6%	99.9%	66.6%	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	5			NR	NR	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	10			NR	NR	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	25			NR	NR	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	50			NR	NR	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI

* I = Contact inhibition under sample

** <1250 ppm. Detection limits for PHMB were higher than target application levels, so results were not meaningful.

Table 5
Laboratory Test Results for Antimicrobial Boot Socks

		Active Conc.		AATCC Test Method 100					AATCC Test Method 147			
Active	**Durability**	**ppm**		**Staph**	**Strep**	**Coryn**	**Pseud**		**Staph**	**Strep**	**Coryn**	**Pseud**
Triclosan	Unwashed	6333		99.9%	99.9%	99.9%	99.9%		12.7	2.3	3.0	NZ/NI
	5	6333		99.4%	99.8%	8.0%	NR		8.7	1.0	NZ/NI	NZ/NI
	10	80		NR	75.4%	NR	NR		2.7	NZ/NI	NZ/NI	NZ/NI
	25	80		NR	62.5%	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	50	25		NR	NR	NR	NR		2.5	NZ/NI	NZ/NI	NZ/NI
Quat silane	Unwashed	N/A		99.9%	99.9%	99.9%	99.9%		NZ/NI	1.0	NZ/NI	NZ/NI
	5	N/A		NR	99.8%	76.1%	23.0%		NZ/NI	6.0	2.0	NZ/NI
	10	N/A		NR	97.1%	NR	NR		NZ/NI	2.5	1.3	NZ/NI
	25	N/A		NR	96.4%	98.6%	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	50	N/A		NR	63.3%	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
PHMB	Unwashed	ND**		99.9%	99.9%	99.9%	NR		2.0	3.7	4.7	NZ/NI
	5	ND		99.9%	99.8%	99.9%	NR		NZ/NI	I*	I	NZ/NI
	10	ND		NR	99.9%	NR	NR		NZ/NI	I	I	NZ/NI
	25	ND		99.4%	99.2%	99.9%	NR		NZ/NI	NZ/NI	I	NZ/NI
	50	ND		NR	99.9%	85.4%	NR		NZ/NI	NZ/NI	I	NZ/NI
Control	Unwashed			99.9%	99.9%	99.9%	NR		NZ	3.0	3.7	NZ
	5			NR	97.9%	74.1%	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	10			NR	7.7%	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI
	25			NR	NR	NR	NR		NZ/NI	I	NZ/NI	NZ/NI
	50			NR	NR	NR	NR		NZ/NI	NZ/NI	NZ/NI	NZ/NI

* I = Contact inhibition under sample
** <1250 ppm. Detection limits for PHMB were higher than target application levels, so results were not meaningful.

Other observations:

- Wash fading of the pattern in the ACU was much reduced with the MLF 9200-200 (triclosan) and MLF R10800-0 in durability testing with the wash wheel. Color retention has been one of the benefits that MPC has observed in commercial uses of this technology.

- "Pilling" on the polyester T-shirts with MLF 9200-200 (triclosan) was much reduced compared with no antimicrobial treatments and the other antimicrobial treatments under

consideration. This was an unexpected benefit of the triclosan treatment. Pilling can, affect the durability and appearance of the clothing item and reduce wear comfort, too.

- After wash durability testing, socks treated with the quaternary silane showed accelerated fraying and fading along the boarding lines. (The boarding lines are essentially creases from the socks being "boarded" during manufacturing and finishing. The socks are pulled over boards or forms shaped like feet, which tends to set in creases.)

Second Pilot Trial

To assess the feasibility of running the combination of triclosan and PHMB, a second pilot trial was run on the ACU fabric. Samples with MLF 9200-200 and MLF R10800-0 and samples with MLF 9200-200, MLF R10800-0, and Reputex (PHMB) were prepared. In order to speed up the durability testing, the samples were laundered using an accelerated test, the AATCC TM 61-2A, in which one cycle typically represents five home launderings, instead of the wash wheel used in Natick testing and used for the first part of the laboratory testing. A sample of ACU fabric from the original trials was run in order to compare results of the 61-2A versus results for the wash wheel. We used only the AATCC TM 100 for microbiological testing. The results are shown in Table 5.

For the original triclosan samples, the microbiology results (Table 6) were comparable to the results from the wash wheel for the 25 and 50 launderings (equivalent), but analytical results show much improved retention of the triclosan. Based on analytical results, we would have expected better micro performance, but again, we often experience problems with the TM 100 on these types of samples.

The triclosan-only sample from the second trial had only slightly better microbiology performance with the TM 100 than the original samples, with comparable retentions, even though the starting addition level was substantially higher.

The combination of triclosan and PHMB, however, was outstanding with excellent performance against Staph, Strep, and Corynebacterium at 50 launderings (equivalent).

Clearly, based on the second pilot trial, the combination of triclosan and PHMB offered the best option with regard to efficacy spectrum and durability, and was the choice for treating the clothing items for the field evaluation.

Table 6

Laboratory Test Results for Second Trial with Antimicrobial Advanced Combat Uniform

				AATCC Test Method 100			
Active	**Durability**	**Triclosan conc. (ppm)**		**Staph.**	**Strep**	**Coryn**	**Pseud.**
Triclosan	Unwashed	1333		NR	NR	31.0%	NR
(Original)	25	690		NR	NR	NR	NR
	50	810		NR	NR	NR	NR
Triclosan	Unwashed	2133		99.8%	99.4%	99.8%	NR
(2^{nd} trial)	25	973		31.4%	84.7%	NR	NR
	50	1200		24.0%	NR	12.0%	NR
Triclosan	Unwashed	2267		99.9%	99.9%	99.7%	NR
+ PHMB	25	1200		98.3%	64.1%	33.2%	NR
(2^{nd} trial)	50	1567		99.9%	99.5%	99.2%	NR

Manufacturing

Fabrics and items manufactured for the field wear evaluation were manufactured in the respective manufacturing facilities using standard manufacturing methods and on normal manufacturing equipment and production lines at Delta Mills (Advanced Combat Uniform), Akwatek (T-shirts), and Pickett Hosiery (boot socks), with the normal finishes. No issues with treatment or with manufacturing were reported by any of the three facilities. Delta Mills and Akwatek ran their normal tests on the finished products and reported that all products met standard specifications for the fabrics.

Field Evaluations

Field Evaluation I

(Excerpted from "Antimicrobial Treated Clothing Items: Field user Evaluation Report," by Chuck Greene, U. S. Army Natick Soldier RD&E Center. See Appendix I.)

The goal of the evaluation was to determine if the protection provided by an antimicrobial clothing treatment would manifest itself in ways that would be noticeable and beneficial to Soldiers. It is critical to keep in mind that we were looking for perceptible benefits of the use of

this type of treatment. Soldiers might be completely unaware of the primary benefit: protection from harmful microbes which could cause illness and render a Soldier ineffective and unable to complete his mission. While we feel that the results of the field evaluation do make a case in favor of antimicrobial technologies, the results must be evaluated alongside laboratory and technical data to gain a complete picture of the performance and benefits of the treatments.

Based on the results of this evaluation, it would appear that the T-Shirt is a promising candidate for application of an antimicrobial treatment. Soldiers who used the treated T-Shirt reported a significant reduction in odor as well as an overall reduction in other problems, which included general discomfort, heat rash, itching skin, and skin rash. A decrease in intensity of these problems was also noted across the board as experienced on this exercise and when compared to past exercises. The percentage of Soldiers in the treated group who reported an increase in general discomfort was only one-fourth that of the untreated group (7% vs. 26%). Soldiers in the treated group also had only half the rate of body odor and heat rash increase (21% and 7%, respectively) than did Soldiers in the untreated group (39% and 16%, respectively). A few Soldiers in each group also noted a reduction in chronic skin problems and conditions that they had 16% for the treated (n=4 out of 25) and 7% for the untreated (n=2 out of 31). Soldiers also rated the comfort and performance of the treated T-shirt higher than the untreated T-Shirt and a higher percentage felt that it was comfortable to wear for an extended period when compared to the untreated item. This seems to indicate that the overall acceptability of the item could benefit from the use of an antimicrobial treatment beyond the reduction of common skin problems. While few individual problems met the requirements of statistical significance, collectively the trend is positive.

The two antimicrobial socks showed an overall reduction in the percentage of Soldiers who reported foot odor, general discomfort, and itching when compared to the untreated item. A minor reduction was noted in the percentage of Soldiers experiencing athlete's foot. In terms of comparison to problems encountered on previous exercises, the B sock was rated significantly better for itching feet, blisters, and skin inflammation. Collectively, it was also rated better than the standard for all other problems in this area. The type C sock also received better ratings than the standard in this context, but a significant improvement was not noted in any specific area. A significantly higher percentage of sock C users did feel that the sock was controlling their foot odor at the midpoint than did Soldiers in the control or sock B group. While both candidate treatments performed well, the field evaluation suggests that the type B sock offered more in the way of perceptible benefits. Overall it would seem that an antimicrobial treatment offers benefits in terms of sock performance.

The results for the antimicrobial treated uniform, as evaluated, were not that promising. We did not see a reduction in problems reported or their intensity on either an individual or collective basis. We also did not see a difference in ratings comparing experiences with the treated uniform to past experience with an untreated uniform. However, significantly more Soldiers felt that the treated uniform was controlling their body odor at the end of the evaluation. We also noted a collective increase in acceptability ratings for the treated uniform, so it is still possible to see a minimal benefit from the antimicrobial treatment as it was evaluated. The key difference between the findings related to the uniform and the T-shirt or sock might be that the former were

"next to the skin" items and may have provided a greater observable benefit than treating an outer layer of clothing. Soldiers in the uniform groups did not receive antimicrobial treated undergarments. This may have somewhat reduced the ability of the participants to detect a benefit from the treatment in a short-term trial.

(End of excerpt.)

Please see Appendix I for data and analysis for this field trial.

Field Evaluation II

(Excerpted from "Antimicrobial Treated Clothing Items – Part II: Field user Evaluation Report," by Chuck Greene, U. S. Army Natick Soldier RD&E Center. See Appendix II.)

The goal of this evaluation was to determine if the protection provided by an antimicrobial clothing treatment would manifest itself in ways that would be noticeable and beneficial to Soldiers. Numerous significant differences were detected along these lines, particularly in relation to the uniform and the T-shirt. While we feel that the results of this field evaluation do make a case in favor of the application of antimicrobial technologies to military clothing items, the results must be evaluated alongside laboratory and technical data to gain a complete picture of the performance and benefits of the treatments.

Numerous significant differences were detected between the treated and untreated groups for uniform and T-shirt performance. Significantly more soldiers felt that the treated uniform and T-shirt were controlling their body odor than those in the untreated group. Significantly more Soldiers felt that the treated uniform and T-shirt decreased their discomfort, how dirty they felt, and the amount of time spent on personal hygiene. A related significant decrease in the frequency of use of certain hygiene products was also noted in the treated group over the untreated group. Significantly more Soldiers felt that the treated uniform and the T-shirt could be worn longer before needing to be changed; additional days were estimated at three for the uniform and two for the T-shirt compared to one each for the untreated uniform and T-shirt. Significantly more Soldiers felt that the treated uniform was comfortable for extended wear than those with the untreated items.

No significant differences were detected for the rate or intensity of various physical problems for those wearing treated or untreated items; however we feel that this may have more to do with the format of the question than the properties of the uniform treatment. This is based on the lack of variability apparent in the data (see Table 9 in Appendix II). Also, there was some data that seemed to indicate that the opposite may be true and that the treatments did have an impact on these problems. Significantly more Soldiers in the treated group felt that they experienced a reduction in chronic problems over those in the untreated group. Also, a significantly higher percentage of soldiers in the treated group felt that the T-shirt was safe to wear compared to those in the untreated group. The same trend was noted for the uniform, but the difference was not statistically significant.

Findings for the sock are somewhat muted by the low number of respondents in each of the two treated groups (type B: n=37, type C: n=24). Also, we did not collect as much data on the sock as

we did on the other items – it was considered secondary on the questionnaire to the uniform and T-shirt. However, there seemed to be some impact of the sock treatments on foot odor, hygiene practices, comfort and suitability for extended wear, and safety. A significantly higher percentage of soldiers with the type B sock felt it could be worn longer before needing to be changed than those with the untreated sock. A significantly higher percentage of Soldiers also felt that the type B sock reduced foot problems than those in the untreated group. In general, we feel that the results of the previous evaluation are more important to assessing treatment performance when used on socks. This data should be viewed as complimentary to that. At some point it might be useful to do a separate dedicated evaluation of antimicrobial socks. This evaluation could be designed specifically to address sock criteria as a primary objective.

Some additional data was collected on Soldier opinions related to the general effectiveness of antimicrobial products as well as some additional measures of acceptability and performance. Overall, 75% of the survey group feels that antimicrobial products are effective. This was true both before and after the evaluation. Interestingly, a significantly higher percentage of soldiers in the treated group believed in the general effectiveness of these products at the end of the evaluation when compared to the treated group. Approximately the same percentage of respondents (73%) indicated they would be in favor of adopting an antimicrobial treatment if it was proven to kill "germs" but did not necessarily reduce odor. Also, a high percentage soldiers in both groups felt that it was a good idea to treat field uniforms (84%) and T-shirts (81%) with antimicrobial treatments. There was also some evidence that the antimicrobial treatments may have a beneficial impact on the mood state of the wearer. This may be an area worth some follow-up in the future.

(End of excerpt.)

Please see Appendix II for a detailed discussion of the results and analysis for the second field study.

Discussion

MPC demonstrated that the ACU, polyester T-shirt, and boot sock could be antimicrobially treated during normal manufacturing operations without affecting performance standards for the articles. The combination of triclosan and PHMB was particularly efficacious, giving a broader spectrum of efficacy against three of the four organisms in the test panel of organisms requested by Natick Soldier RD&E Center and having somewhat enhanced durability.

Additional benefits of antimicrobial treatment with triclosan were reduced wash fading of the ACU and reduced pilling for the polyester T-shirt. The reduced fading may be due to a combination of the MLF 9200-20 and R10800-0 rather than just the use of triclosan, and it is unclear whether the reduced pilling is due to the triclosan or the use of triclosan in the MLF 9200-200 carrier system.

Part of the study was to compare the AATCC TM 100 and TM 147 and evaluate them as a basis for establishing specifications for antimicrobial treatment in military textiles. The results of this study would suggest that neither quantitative nor qualitative test is a complete answer for establishing specifications, though the TM 100 is probably the most useful. One caveat is that only organic antimicrobials were used in this study, and the TM 100 uses higher levels of nutrient in the initial inoculum than is commonly used in testing silver-based antimicrobial additives, because silver tends to be neutralized by proteins in the nutrient. It is possible that silver-based products, including some currently in use for antimicrobial boot socks could not pass a TM 100. Probably some follow-up work is needed to determine if a modified AATCC TM 100 would be appropriate for both organic and inorganic antimicrobial treatments for textiles. A successful field evaluation could provide the basis for deciding which one is the most appropriate laboratory test by establishing which antimicrobials are effective in the field.

Unfortunately, the results from the first field wear evaluation were statistically inconclusive. There may be a number of factors that contributed to this outcome.

- Assessing odor and comfort in the field is notoriously difficult to do, particularly self-assessing odor.
- Sample size in a study of this type is critical, particularly with the type of data being generated. Given the learning from this trial, a larger sample might be more likely to show results.
- One week may be insufficient to measure the benefit of antimicrobial treatments, particularly benefits beyond odor control.
- This study was designed to independently evaluate each item of antimicrobially treated clothing versus evaluating the concept of antimicrobially treated clothing versus standard clothing. The problem was outlined particularly with the uniform since in this case the clothing item closest to the skin, the T-shirt, was not antimicrobially treated. A study comparing Soldiers wearing clothing where most, if not all, items are antimicrobially treated versus Soldiers wearing clothing where none are antimicrobially treated might be more effective in evaluating the antimicrobial treatment.
- One of the major confounding factors in the trial was the use of antibacterial hygiene products by the Soldiers in the field. In the surveys three-fourths (75%) reported that they used some form of antibacterial medication or hygiene item, ranging from to deodorant to foot powder to lotion and hand sanitizers. Slightly less than a fourth of the Soldiers (23%) used an antibacterial first-aid or hygiene product on their feet. In an ideal study, such use would not be allowed so as not to interfere with the results of the study; of course in this case the Soldiers were not in the field for the purposes of the study, but to train for their primary mission, and the study was incorporated onto that training. It could be argued that since the Soldiers will normally be using such hygiene products, then the results are valid for the purposes of assessing the benefits of the technology for military applications, but the question is whether or not such items are always available under field operating conditions, which is when antimicrobial treatments would actually be most likely to come into their own as a benefit. *(See "Antimicrobial Treated Clothing Items: Field user Evaluation Report," by Chuck Greene, U. S. Army Natick Soldier RD&E Center, Appendix I.)*

Despite these problems and seeing few comparisons that meet the requirement of statistical significance, there were trends that suggested that the T-shirt and the boot socks could benefit from the use of antimicrobial technology. As noted above, Soldiers using the antimicrobial T-shirts reported reductions in odor as well as other problems such as general discomfort, heat rash, itching skin, and skin rash, with a reported decrease in intensity over the course of the study and compared with past exercises. Similarly the antimicrobial socks showed an overall reduction in the percentage of Soldiers who reported foot odor, general discomfort, and itching.

Because of questions concerning the design of the first field study and observations that seemed to show benefits of the technology but without having statistical significance, a second study was planned that focused specifically on the question of whether or not there were observable benefits of antimicrobially treated clothing versus standard issue clothing. With focus on the specific question of antimicrobial treatment versus no treatment, statistically significant results were obtained showing that Soldiers observed reductions in odor, felt less uncomfortable and dirty, and felt that their clothing was more comfortable during periods of extended wear than with untreated clothing. Soldiers reported that the uniform could be worn for an additional three days and the T-shirt for an additional two days before changing versus one day for the untreated items.

"The application of an antimicrobial treatment, particularly to the T-shirt and the uniform, seemed to offer a range of benefits to the user. These included improved odor control, comfort, hygiene, and wear time. Other benefits, to include those related to physical problems and mood state are possible but could not be validated based on the available questionnaire data. There is also a great deal of interest amongst the Soldiers in the use of antimicrobial products as a treatment for field uniforms (84%) and T-shirts (81%). Three-fourths of the Soldiers believe in the effectiveness of these products and nearly three-fourths would be in favor of using them if they were proven to be effective in the lab but offered no immediately perceptible benefit to them." *(Excerpted from "Antimicrobial Treated Clothing Items – Part II: Field user Evaluation Report," by Chuck Greene, U. S. Army Natick Soldier RD&E Center. See Appendix II.)*

Aside from morale improvements from improved comfort and hygiene, other potential benefits include lower water usage where water might be in short supply or require significant effort to maintain a supply and fewer personnel diverted to laundering. Potential reductions in health problems or skin problems in the field could not be validated in this study; the results did not show statistical significance. Analysis, however, suggests that this may be due to the format of the questions, and some data did suggest that there might be benefits. Longer periods in the field than used in these studies would also tend to show such benefits more clearly since the probability of occurrence and the degree of severity increase with time.

Conclusions & Recommendations

- The combination of triclosan and PHMB had broad spectrum efficacy, durability to 50 launderings, and could be applied easily during normal manufacturing without additional processing steps or equipment. We would recommend this combination for future developments and field evaluations.

- The AATCC Test Method 100 appears to be the most universal of the test methods, but is not necessarily definitive, i.e. it can result in false negatives. In our evaluation of the tests we observed zones of inhibition for triclosan using the AATCC Test Method 147, clearly indicating inhibition and efficacy, but for reasons that are not understood at this time, the same samples often gave poor results in the Test Method 100.

  The Test Method 100 is also the more difficult and expensive test to run. Microban Products Company's commercial experience in using quantitative testing is that most laboratories are not able to run the test dependably. We would recommend that laboratories be certified on a regular basis via blind testing prior to accepting results as evidence of performance or non-performance.

- Results from the field evaluation show benefits to the Soldier in improved odor control, comfort, hygiene, and wear time. Statistically significant results were obtained from surveys conducted after field evaluations of antimicrobially treated uniforms and T-shirts, worn together, versus untreated, standard issue uniforms and T-shirts. Not only did Soldiers show statistically significant results in the survey that was conducted, but there was a clear interest in the use of antimicrobially treated uniforms.

  - Different antimicrobial treatments were not studied in detail, and the one area, socks, where there was a straight comparison, the number of samples were relatively small. The results tended to show that Soldiers observed benefits with both a silver treatment and with the triclosan/PHMB treatment with a slight edge to the triclosan/PHMB treatment. A larger scale comparison would be needed to draw clear conclusions as to the relative merits of the different treatments.

  - Recommendations from the field evaluations:

    a) Implementation of antimicrobial treatments in field uniforms, T-shirts, and socks, or at least a larger scale evaluation if necessary to support implementation. The results from the second field evaluation support benefits to the Soldier in improved comfort and hygiene as well as show significant interest in the technologies.

    b) Longer term, possibly larger scale evaluation to determine if there is an impact on microbial related health problems, particularly skin problems that result from prolonged activity in the field.

c) As related in the Field Evaluation report, a larger evaluation of antimicrobially treated socks would probably be useful. The sample size for socks in the second field evaluation was very small.

In addition to the results summarized in this work and report, there is a follow-up study in progress, Project W911QY-05-C-0087, "Advanced Antimicrobial & Comfort Technologies for Military Applications." The second study involves the combination of antimicrobial treatment and moisture management finishes on cotton T-shirts as an alternative to polyester T-shirts. An additional part of the study involves the antimicrobial treatment of sleeping bag systems to protect against fungal organisms that damage the fabric as well as to reduce bacterial loadings on the fabric.

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APPENDIX I: Field User Evaluation Report

Chuck Greene
U.S. Army Natick Soldier RD&E Center

U.S. ARMY NATICK SOLDIER CENTER
Antimicrobial Treated Clothing Items
Field User Evaluation Report

Chuck Greene

21 November 2005

U.S. Army Natick Soldier Center
Natick, MA 01760

Executive Summary

Background

In August 2005 the Natick Soldier Center conducted a field user evaluation of antimicrobial treated uniforms, T-shirts, and socks with Soldiers from the 1st Battalion, 31st Air Defense Artillery Brigade at Fort Bliss, Texas. The treatments evaluated were targeted against common, but harmful, bacteria routinely encountered by Soldiers in the field. The goal of the evaluation was to determine if the protection provided by these treatments would manifest itself in ways that would be noticeable to Soldiers. Effectiveness of the treatments was measured through questionnaires which assessed relevant problems and conditions encountered by the participants during the exercise, their intensity, and how they compared to past experiences, along with perceptions of comfort, odor reduction, and performance.

Evaluation Design

The evaluation of the antimicrobial treated uniforms, T-shirts, and socks featured a between-groups design. The independent variable was item type (treated vs. untreated) and the dependent measures were Soldier responses to an array of survey questions designed to quantify experiences and opinions relevant to the performance of the treatment. All of the members of the participating unit were issued treated or untreated versions of either the uniform or the T-shirt. All of the participants were also issued one of three types of socks: untreated, treated type B, or treated type C. The items were used during a seven day field training exercise conducted in the training area at Fort Bliss. While the primary mission of the unit involved is Air Defense, they trained as Infantry for potential future deployment in that role. Data was collected through a series of questionnaires on day four (midpoint) and on day seven (final). This also allowed for a within-groups dimension to the evaluation to assess performance of the items over time.

Survey Sample

The survey group consisted of 207 male (88%, n=183) and female (12%, n=24) Soldiers from the 1st Battalion, 31st Air Defense Artillery Brigade. The average age of the participants was 25 and the average length of military service was five years. The breakdown by rank was E-1 to E-3 (23%, n=48 out of 207), E-4 to E-6 (59%, n=122 out of 207), E-7 to E-9 (5%, n=10 out of 207), O-1 to O-3 (11%, n=23 out of 207), with the remainder being senior officers and Warrant Officers. If Soldiers had not participated in this evaluation, most reported that they would have worn the Desert Camouflage Uniform ("DCU") (67%, n=139 out of 207), the standard cotton T-shirt (87%, n=180 out of 207), and the standard black wool socks (55%, n=113 out of 207).

Key Findings

The goal of the evaluation was to determine if the protection provided by an antimicrobial clothing treatment would manifest itself in ways that would be noticeable and beneficial to Soldiers. It is critical to keep in mind that we were looking for perceptible benefits of the use of this type of treatment. Soldiers might be completely unaware of the primary benefit: protection from harmful microbes which could cause illness and render a Soldier ineffective and unable to complete his mission. While we feel that the results of the field evaluation do make a case in favor of antimicrobial technologies, the results must be evaluated alongside laboratory and technical data to gain a complete picture of the performance and benefits of the treatments.

Based on the results of this evaluation, it would appear that the T-Shirt is a promising candidate for application of an antimicrobial treatment. Soldiers who used the treated T-Shirt reported a significant reduction in odor as well as an overall reduction in other problems, which included general discomfort, heat rash, itching skin, and skin rash. A decrease in intensity of these problems was also noted across the board as experienced on this exercise and when compared to past exercises. Soldiers also rated the comfort and performance of the treated T-shirt higher than the untreated T-Shirt and a higher percentage

felt that the it was comfortable to wear for an extended period when compared to the untreated item. This seems to indicate that the overall acceptability of the item could benefit from the use of an antimicrobial treatment beyond the reduction of common skin problems. While few individual problems met the requirements of statistical significance, collectively the trend is impressive.

The two antimicrobial socks showed an overall reduction in the percentage of Soldiers who reported foot odor, general discomfort, and itching when compared to the untreated item. A minor reduction was noted in the percentage of Soldiers experiencing athlete's foot. In terms of comparison to problems encountered on previous exercises, the B sock was rated significantly higher for itching feet, blisters, and skin inflammation. Collectively, it was also rated higher than the standard for all other problems in this area. The type C sock also received higher ratings than the standard in this context, but a significant improvement was not noted in any specific area. A significantly higher percentage of sock C users did feel that the sock was controlling their foot odor at the midpoint than did Soldiers in the control or sock B group. While both candidate treatments performed well, the field evaluation suggests that the type B sock offered more in the way of perceptible benefits. Overall it would seem that an antimicrobial treatment offers benefits in terms of sock performance.

The results for the antimicrobial treated uniform, as evaluated, were not that promising. We did not see a reduction in problems reported or their intensity on either an individual or collective basis. We also did not see a difference in ratings comparing experiences with the treated uniform to past experience with an untreated uniform. However, significantly more Soldiers felt that the treated uniform was controlling their body odor at the end of the evaluation. We also noted a collective increase in acceptability ratings for the treated uniform, so it is still possible to see a minimal benefit from the antimicrobial treatment as it was evaluated. The key difference between the findings related to the uniform and the T-shirt or sock might be that the former were "next to the skin" items and may have provided a greater observable benefit than treating an outer layer of clothing. Soldiers in the uniform groups did not receive antimicrobial treated undergarments. This may have somewhat reduced the ability of the participants to detect a benefit from the treatment in a short-term trial.

There is also some anecdotal evidence that emerges from the data to lend weight to the argument in favor of the antimicrobial treatments. For example, three Soldiers in the treated uniform group reported that they did not experience diarrhea in the field when they normally do. None of the respondents in the treated T-Shirt group reported that they developed a problem that they would not normally have compared to 13% in the untreated group. Fifty percent more Soldiers in the treated sock groups reported a reduction in chronic foot problems when compared to Soldiers in the untreated groups. There are other examples, these three are cited here in order to be brief.

In addition, three-fourths of our evaluation participants reported that they buy and use products branded as "antimicrobial" for use in the field. More than two-thirds of the survey group urged the adoption of antimicrobial treated clothing items either as standard or optional purchase items and one-fourth felt that the treatments should continue to be researched. Only 6% felt that there was no merit to the technology. It is clear that Soldiers in general are interested in antimicrobial products. They endorse the use of these treatments on military clothing items. The fact that Soldiers in the untreated group felt the exact same way as those in the treated group also seems to indicate that they recognize that the benefits of these types of treatments may not always be apparent. All of this, when taken together, offers a clear basis for further consideration of the antimicrobial technologies evaluated.

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INTRODUCTION

Background

In August 2005 the Natick Soldier Center conducted a field user evaluation of antimicrobial treated uniforms, T-shirts, and socks with Soldiers from the 1st Battalion, 31st Air Defense Artillery Brigade at Fort Bliss, Texas. The treatments evaluated were targeted against common, but harmful, bacteria routinely encountered by Soldiers in the field. The goal of the evaluation was to determine if the protection provided by these treatments would manifest itself in ways that would be noticeable to Soldiers through an impact on a wide range of problems and conditions that could be caused by the targeted microbes as well as though other more subjective measures.

All of the members of the participating unit were issued treated or untreated versions of either the uniform or the T-shirt. All of the participants were also issued one of three types of socks: untreated, treated type 1, or treated type 2. The items were used during a seven day field training exercise conducted in the training area at Fort Bliss. While the primary mission of the unit involved is Air Defense, they left their Patriot launchers behind and trained as Infantry for potential future deployment in that role. Data was collected through a series of questionnaires that addressed criteria relevant to the assessment of antimicrobial treatments. A total of seven different questionnaires were used, copies of which are included as Attachments A through G. Approximately 300 Soldiers from the 1st Battalion, 31st Air Defense Artillery Brigade participated, with 207 completing all of the evaluation requirements.

Item Description

There were a total of seven items under evaluation: two versions of the standard uniform (treated and untreated), two versions of the standard polyester T-shirt (treated and untreated), and three versions of the standard cotton sock (untreated, treated 1, and treated 2). A description of each of the items is included below.

- Uniform: Two versions of the standard Advanced Combat Uniform (ACU), one featuring a Microban® antimicrobial treatment formulation (type A) and one untreated (type B). Both uniforms also featured a wrinkle resistance treatment, which is standard for this uniform. Soldiers evaluating this item were issued one complete uniform to evaluate.

- T-shirt: Two versions of the standard issue 100% Polyester T-shirt, one featuring a Microban® antimicrobial treatment formulation (Type A) and one untreated (Type B). Soldiers evaluating this item were issued four T-shirts of the same type.

- Socks: Three versions of the standard issue 100% cotton socks: an untreated standard sock (Type A); the current treated standard sock, CR/PD 03-18 (Type B); and an untreated standard sock which received a Microban® antimicrobial treatment (Type C).

Test Design & Procedures

The evaluation of the antimicrobial treated uniforms, T-shirts, and socks featured a between-groups design. The independent variable was item type (treated vs. untreated) and the dependent measures were Soldier responses to an array of survey questions designed to quantify experiences and opinions relevant to the performance of the treatment. Data was collected at the half-way point of the evaluation on day four (midpoint) and at the conclusion on day seven (final). This also allowed for a within-groups dimension to the evaluation to assess performance of the items over time.

Input on questionnaire items was provided by Dermatologists at William Beaumont Army Medical Center, Fort Bliss, TX to ensure that content was appropriate in terms of the skin problems and conditions that Soldiers would experience in the field. The key questions and the primary scale used on the questionnaire were based on existing symptom assessment scales with proven reliability and validity and designed for use to collect data in "self-reporting" scenarios. Other questions were developed as appropriate.

Because the number of items exceeded the number of Soldiers, multiple items would have to be issued to each participant. Evaluation groups were developed to minimize any potential cross-over effects from one item to another. Four groups were defined: A (antimicrobial treated uniform and untreated socks), B (untreated uniform and antimicrobial treated socks), C (antimicrobial treated T-shirts and antimicrobial treated socks), and D (untreated T-shirts and antimicrobial treated socks). Assignment to any group was purely random and Soldiers did not know if any of the items issued to them had the antimicrobial treatment or not.

Participants were briefed several days before the evaluation began on the purpose and procedures. They were informed that they would be evaluating antimicrobial clothing treatments and that some would receive treated items and some would receive untreated items. At that time they completed a background questionnaire to obtain demographic information and data on past experiences that would be relevant to the evaluation. Participation was voluntary. Several days later the participants were issued the test items and were given the opportunity to try them on to make sure they fit. A few Soldiers had to change assigned groups at this point due to size availability, but the randomness of the assignment was maintained.

A final roster was maintained and each Soldier was double-checked to make sure that they had received the correct items and that they had completed the background questionnaire before they left the issue location. Once it was sure that everything was in order, the Soldier initialed for the items received and was logged into the evaluation. The issue was conducted over a two-day period. Twelve hours after the last item was issued the unit deployed to the training area for a seven-day field training exercise. The weather observations for the evaluation period are presented below in Table 1.

Table 1
Weather Conditions at Fort Bliss, 17 to 23 August 2005

Reporting station: El Paso, TX International Airport

August:	**17**	**18**	**19**	**20**	**21**	**22**	**23**
Temp (max)	90	91	84	83	89	85	89
Temp (min)	72	70	66	67	68	72	70
Precipitation (inches)	.1”	.1”	.6”	T*	T*	T*	.1”
Relative Humidity (avg.)	59%	62%	73%	71%	61%	62%	57%

[1] From: NOAA, National Climatic Data Center
http://www.ncdc.noaa.gov/oa/ncdc.html
* T=Trace amount

The unit was visited briefly by the data collection team on the first day to determine their location in “the box” and to collect some initial informal feedback. The Soldiers then conducted training for the next 72 hours. During this time, the data collection team finalized the evaluation roster and prepared the individualized questionnaire sets to be completed by each participant. Midpoint data was collected on the evening of day four. The unit then conducted an additional 72 hours of training. The process was repeated on the evening of day seven, and the evaluation was completed at that time.

Data Handling

The raw data was returned to Natick where it was scanned, cleaned, and assembled into a series of three interim data sets: one each for the uniform, T-shirt, and sock. Extensive preliminary analyses were conducted to determine the rules for inclusion in the final data set. Various adjustments to the data did not have an impact on the ultimate outcome of any of the various item evaluation, so it was decided to accept the most stringent handling of the data to provide the maximum flexibility for data analysis and the highest level of data integrity possible.

In order for a respondent to be included in the final version of the three data sets, they had to meet a series of requirements. They had to complete the background, midpoint, and final questionnaires with a minimal amount of missing data so that we knew they were actually in the field for the entire seven day period. They had to stay in their assigned groups throughout the evaluation so that we knew they used the items they were assigned for the duration of the evaluation. Finally, we had to be able to trace the respondent from the original issue roster to the final questionnaire so we could be sure that the integrity of the data sets was as secure as possible.

We started with a total of 305 respondents. After the retention rules were applied, we were left with a total of 207. Each Soldier could be represented in two separate data sets, either the uniform or the T-shirt and the sock. The final uniform data set included 136 Soldiers: 75 with the treated item and 61 with the untreated item. The final T-shirt data set consisted of 59 Soldiers: 28 in the treated group and 31 in the untreated group. The final sock data set consisted of 172 Soldiers: 59 in the untreated group, 52 in the first treated group, and 61 in the second treated group.

Data Analysis

Descriptive statistics used to describe the data are the number of Soldiers responding (n) and the percentage of the total responding to a certain option in a "yes - no" or multiple-choice question. Please note that the n reported for specific questions is based on the number of valid responses to that question which results in some variation from question to question in the total number of respondents. The mean (X) is reported for scale-ended questions or estimates of time or frequency. The data was analyzed using a variety of statistical procedures. In all instances the .05 criterion level was used as the minimum probability level to determine significance for all statistical procedures. This indicates that, on a statistical level, there is a less than 5% chance that the differences observed are attributable to error or normal variation. If a certain statistical procedure could not detect a significant difference it is notated as "ns" ("not significant") in the relevant table.

Student's t-test for independent samples was used for scale-ended data when only two groups were involved and data was analyzed on a between-groups basis. The paired samples version was used when data was analyzed on a within-groups basis. This test compares the actual difference between two means in relation to the variation in the data to determine if they are equal or not. The results are expressed by the "t' statistic and an associated significance level. Data analysis for the sock, which featured three groups, required the use of the Analysis of Variance (ANOVA) and post-hoc test, which is

essentially an extension of the t-test, to test the hypothesis that several means are equal or not. The results are expressed by the "F" statistic and its associated significance level.

The Sign test was used to test for differences in the distribution of scale-ended items on a between-groups basis. This procedure is used with two related samples to test the hypothesis that two variables have the same distribution. Finally, the Chi-square test was used to analyze all dichotomous data. This procedure tabulates a variable into categories and computes a Chi-square statistic. It compares the observed and expected frequencies in each category to test either that all categories contain the same proportion of values or that each category contains a user-specified proportion of values. The Chi-square test is expressed by the "X^2" statistic along with the corresponding significance level.

Survey Sample

The survey group consisted of 207 male (88%, n=183) and female (12%, n=24) Soldiers from the 1st Battalion, 31st Air Defense Artillery Brigade. The average age of the participants was 25 and the average length of military service was five years. The most common Military Occupation Specialties (MOSs) were 14T (26%, n=53 out of 207), 14E (20%, n=42 out of 207), 92A (8%, n=17 out of 207), 14J (7%, n=15 out of 207), 63B (6%, n=12 out of 207), and 91W (5%, n=11 out of 207). The remainder held a wide variety of other MOSs. The breakdown by rank was E-1 to E-3 (23%, n=48 out of 207), E-4 to E-6 (59%, n=122 out of 207), E-7 to E-9 (5%, n=10 out of 207), O-1 to O-3 (11%, n=23 out of 207), with the remainder being senior officers and Warrant Officers.

If Soldiers had not participated in this evaluation, most reported that they would have worn the Desert Camouflage Uniform ("DCU") (67%, n=139 out of 207), the standard cotton T-shirt (87%, n=180 out of 207), and the standard black wool socks (55%, n=113 out of 207) for a typical August field training exercise at Fort Bliss. Other Soldiers reported that they would wear the Hot Weather BDU (25%, n=52 out of 207) or the standard green cotton socks (32%, n=67 out of 207). A few noted that they would wear a commercial sock (13%, n=26 out of 207) or a polyester "wicking" T-shirt (12%, n=25 out of 207) in the field.

The survey group was divided into two major sub groups, with two-thirds (66%, n=136 out of 207) participating in the uniform evaluation and one-third (29%, n=59 out of 207) participating in the T-shirt evaluation. Most Soldiers also completed the sock evaluation (83%, n=172 out of 207). Because of various data handling procedures, 12 subjects who had been dropped from either the uniform or T-shirt evaluations were retained in the sock test group and 35 Soldiers who had participated in either the

uniform or T-shirt evaluation did not complete the sock evaluation. This is why the number of Soldiers in the three groups do not add up to the total number of participants.

Table 2
Demographic Characteristics: Uniform and T-shirt Sub-Groups
(n=195)

	Uniform (n=136)		T-shirt (n=59)	
	Treated Uniform (n=75)	**Untreated Uniform (n=61)**	**Treated T-shirt (n=28)**	**Untreated T-shirt (n=31)**
Age	27	26	24	22
Gender (M/F)	89%/11%	90%/10%	86%/14%	87%/13%
Time in (months)	76	69	46	39
MOS	14E (28%)	14T (25%)	14T (32%)	14T (36%)
Median Rank	E-4	E-4	E-4	E-4
Uniform*	DCU (76%)	DCU (62%)	68%	61%
T-shirt*	Cotton (88%)	Cotton (90%)	79%	84%

* Normally worn in the field

The two uniform groups did not differ significantly from each other on age, time in, rank, uniform, or t-shirt. This was also true for the two T-shirt groups. In terms of these basic demographic factors, there are no differences between the Soldiers who received the treated or untreated uniform or the treated or untreated T-shirts that could skew the results.

Table 3
Demographic Characteristics: Sock Sub-Groups
(n=172)

	Untreated Sock "A" (n=59)	**Treated Sock "B" (n=52)**	**Treated Sock "C" (n=61)**
Age	27	26	23
Gender (M/F)	91%/9%	86%/14%	85%/15%
Time in (months)	74	69	40
MOS	14E (31%)	14T (25%)	14T (30%)
Median Rank	E-4	E-4	E-4
Sock*	Std. Wool (58%)	Std. Wool (54%)	Std. Wool (52%)

* Normally worn in the field

There were significant differences in the make up in the three sock groups. An ANOVA found that Soldiers in Group C were significantly younger ($F=7.84$, $p=.001$) and had spent significantly less

time in the military ($F=6.65$, $p<.01$) than Soldiers in Groups A or B. A Chi-square analysis found that Group C also featured a rank structure skewed significantly to the lower enlisted grades when compared to Groups A and B ($X^2=20.94$, $p<.01$). No significant differences were detected for the sock type typically worn in the field.

Soldier Experience

The background questionnaire was completed by Soldiers at the time the evaluation items were issued to them or at a pre-briefing a few days before. The content of this survey dealt with issues that would be relevant to the evaluation of an antimicrobial clothing treatment: the history and experience level of Soldiers with a variety of minor issues and conditions that could result or be impacted by microbes encountered in the field. These questions were also used on the midpoint and final questionnaires to assess the impact of the treatments used. Results from the background survey provide a frame of reference for the total evaluation. At this point, however, we can use this data to determine if the Soldiers in the various experimental groups were essentially similar at the start of the evaluation in terms of their response to these key questions.

An analysis of the data from this survey found only two significant differences amongst the respondents in any of the evaluation groups: one for the uniform and one for the sock. These will be discussed at the appropriate time in this section. The major trend in this data indicates that the groups were well randomized and, as far as we can tell, there does not seem to be anything in their makeup that would have undue influence on item performance. Therefore, where appropriate, the results for the total survey group are presented in this section. However, since we want to present as complete a picture as possible, we will also show some of the data broken out for the various evaluation groups.

Overall, 8% (n=16 out of 207) of the survey group reported that they have been diagnosed with some form of chronic skin problem. These were identified as hyper-hydrosis (n=4), eczema (n=2), and an allergy to starch (n=1). The remainder did not describe what their problem was. Just under half (47%, n=97 out of 202) reported that during their normal field duties they usually come in contact with something that may cause a skin rash - with biting insects (n=85) and caustic substances (n=80) being mentioned most frequently. Three-fourths (75%, n=156 out of 206) reported that they usually use some form of anti-bacterial medication or hygiene item in the field. These items were deodorant (n=117), foot powder or spray (n=52), medicated powder (n=43), soap (n=28), “baby wipes” (n=20), lotion (n=14), and hand sanitizer (n=5).

Some Soldiers noted that they have experienced rashes and irritation that they believed were caused by standard issue clothing items. Fourteen percent (n=19 out of 136) of the Soldiers in the uniform group reported that they have had problems with standard BDUs or DCUs, specifically with rash (n=10), heat rash (n=8), chaffing (n=8), excessive sweating (n=1), and an allergic reaction to starch (n=1). Problems typically developed at the groin (n=7), arms (n=6), neck (n=5), lower legs (n=4), thighs (n=3), or "all over" (n=3). The actions taken to address these problems were to use medicated powder or cream (n=8), to do nothing (n=3), or to clean the area with soap (n=2). None of the Soldiers in the T-shirt group reported they have had problems with the standard item.

Fourteen percent of the Soldiers in the sock evaluation group (15%, n=24 out of 159) reported having problems that they believe were caused by a standard issue item. For the most part, these seem to be related to the standard issue wool sock. Problems were identified as athlete's foot (n=8), extreme sweating (n=8), rash or irritation (n=6), burning and itchy feet (n=3), and an allergy to wool (n=3). There was as significant difference for this data identified amongst the three sock sub-groups. Soldiers who received sock types A or B reported a significantly higher rate of problems with standard socks than those Soldiers in group C (A = 23%, n=13 out of 56; B = 18%, n = 8 out of 45; C = 5%, n = 3 out of 58; $X^2=7.59$, $p<.05$). It seems doubtful that this would have a major impact on the outcome of the evaluation since the sock type issued was based on the standard issue cotton sock and not the wool item.

Soldiers were presented with a list of common maladies and symptoms that could be caused or influenced by bacteria and microbes encountered in the field. They used a four-point scale (0 = "N/A," 1="Mildly," 2="Moderately," and 3="Severely") to indicate to what extent they experience these problems under the standard BDU or DCU during a typical seven-day field exercise. The results obtained are presented below in Table 4 and are combined for all of those Soldiers who participated in the uniform or T-shirt evaluation (n=195). The data is summarized two ways: by experience and intensity. "Experience" was derived from the scale and simply indicates the percentage of the population that typically experiences a certain problem. "Intensity" is the mean rating on the scale adjusted only for those Soldiers who actually do experience that problem. The last column summarizes the results of various paired comparisons for those Soldiers receiving the treated or untreated uniforms or T-shirts.

Table 4
"Typical" Problems and Conditions Experienced in the Field
(n=195)

Condition	Experience (n=195)	Intensity (Adjusted)	Significant Differences by Group?[1]
Body odor	93%	1.7	NO FOR ALL
General discomfort	72%	1.5	NO FOR ALL
Heat Rash	37%	1.6	UNIFORM=YES[2], T-SHIRT=NO
Itching skin	38%	1.6	NO FOR ALL
Chafing	53%	1.5	NO FOR ALL
Skin rash/irritation	31%	1.6	NO FOR ALL
Skin inflammation/redness	24%	1.5	NO FOR ALL
Skin lesions or sores	14%	1.2	NO FOR ALL
Acne/pimples	51%	1.5	NO FOR ALL
Infected or inflamed hair follicles	29%	1.3	NO FOR ALL
Infected cuts or scrapes	20%	1.3	NO FOR ALL

[1] Summary of four different comparisons: Chi-square for "experience" (T-shirt group, Uniform group) AND
the t-test for the mean "intensity" (T-shirt Group, Uniform group).

[2] Uniform: Significant difference for mean intensity (Treated=1.8, Untreated=1.4, $t=2.49$, $p<.05$).

As can be seen above, out of the 44 possible paired comparisons only one significant difference was found for the uniform groups and none for the T-shirt. The specific difference (a slightly higher mean intensity for "heat rash" amongst Soldiers in the treated uniform group) seems of minor importance since the Soldiers are so similar on all other criteria evaluated. It would seem safe to assume that Soldiers in the various evaluation groups were essentially identical in perceptions as to how likely they were to experience certain problems in the field.

In addition to the conditions listed above, a small number of Soldiers (5%, n=11) felt that there were other types of problems that they experienced during a typical seven-day field exercise that could be bacterial in nature. These were identified as diarrhea (n=5), eye irritation (n=4), allergies (n=1), itchy scalp (n=1), and urinary tract infections (n=1). There were no significant differences in the responses received from any of the evaluation groups (either uniform or T-shirt).

A similar approach was taken for the sock evaluation: Soldiers were presented with a list of common foot problems and they used a four-point scale (0 = "N/A," 1="Mildly," 2="Moderately," and 3="Severely") to indicate to what extent they experience these problems during a typical seven-day field exercise. The results obtained are presented below in Table 5. The data includes only those Soldiers who successfully completed the sock evaluation (n=172) and is summarized in the same way as in Table 4 (by "experience" and "intensity"). "Experience" was derived from the scale and simply indicates the percentage of the population that typically experiences a certain problem. "Intensity" is the mean rating on the scale adjusted only for those Soldiers who actually do experience that problem. The last column summarizes the results of various statistical comparisons for the three sock groups.

Table 5
"Typical" Foot Problems and Conditions Experienced in the Field
(n=172)

Condition	Experience (n=172)	Intensity (Adjusted)	Significant Differences by Group?[1]
Foot odor	93%	1.9	YES[2]
General discomfort	76%	1.6	NO FOR ALL
Itching feet	59%	1.6	NO FOR ALL
Athletes Foot	40%	1.7	NO FOR ALL
Toe nail fungus	19%	1.8	NO FOR ALL
Skin rash/irritation	26%	1.6	NO FOR ALL
Skin inflammation/redness	30%	1.6	NO FOR ALL
Blisters or calluses	65%	1.5	NO FOR ALL

[1] Summary of two different comparisons: Chi-square for "experience", ANOVA for mean "intensity"
[2] Sock A=2.0, Sock B=1.9, Sock C=1.7; A is significantly higher than C (F=3.09, p=.05).

Again, it would seem that we can assume that Soldiers in the three sock groups all experience the same rate of foot problems at the same relative intensity. The one significant difference detected, a slightly higher "foot odor" intensity for sock group A compared to sock group C seems inconsequential. Overall, it would seem that we can be reasonably certain that none of our evaluation groups (uniform, T-shirt, or socks) feature any kind of significant imbalances that could impact the outcome of the evaluation.

UNIFORM FINDINGS

Background

A total of 136 Soldiers completed the evaluation of the uniform: 75 had the treated uniform and 61 had the untreated uniform. There were no significant differences between the two groups for any of the background data discussed in this section, therefore, the results will be presented for the total survey group. Soldiers reported that they wore the uniform for an average of seven days for 22 hours per day. Eight percent (n=11 out of 136) reported that they had to stop wearing the uniform by the end of the evaluation. Specific reasons were for sleeping (n=4), the uniform was extremely dirty (n=3), or the Soldier was assigned to the role of the "opposition force" and wore civilian clothes for a time (n=3). These Soldiers reported an average of five to six days of wear compared to seven days for the remainder of the group. It was decided to keep them in the database since they only lost an average of one day of wear during the evaluation and because their data did not differ significantly on any of the key measures from those who did not stop wearing the uniform. Twenty percent (n=27 out of 134) of the respondents had a durability problem with the uniform during the evaluation. Specific problems identified were various rips, tears, and holes (n=10), crotch seam failure (n=8), and seams fraying (n=5).

The most common type of T-shirt worn with the uniform was the standard brown cotton T-shirt (94%, n=126 out of 134). Soldiers estimated that they had six T-shirts with them and that they changed approximately every two days. The most common type of underwear worn was standard cotton (69%, n=92 out of 134). Soldiers estimated that they had six pairs with them and that they changed their underwear every one to two days. A few Soldiers (2%, n=3 out of 134) noted that they did not usually wear underwear during the exercise. Only 9% (n=12 out of 135) reported wearing any other type of undergarment other than T-shirts and underwear. These were primarily the female Soldiers noting that they also wore a bra under the uniform.

Almost two-thirds (63%, n=85 out of 135) reported that they used some kind of hygiene or first aid product under the uniform that was branded as "anti-bacterial." The specific products identified were: deodorants or anti-perspirant (n=61), baby wipes (n=12), soap (n=10), medicated powder (n=10), foot powder or spray (n=9), along with various lotions (n=8), creams (n=5), and sprays (n=3). These items were typically used under the arms (n=61) or on the face (n=15), arms (n=9), feet (n=9), crotch (n=5), legs (n=3), hands (n=3), or chest (n=1). Some also noted that they used the items "everywhere" (n=14). If someone used something it was generally used daily.

Less than one-fourth (22%, n=30 out of 136) reported that they came in contact with anything that might, by itself, cause a problem for their skin. The most common sources identified were biting insects (n=24), caustic substances like gasoline or battery acid (n=15), plants (n=10), and "irritants" like insect repellent or camouflage face paint (n=5).

Performance

Soldiers rated the types of problems and conditions that they were experiencing at the midpoint and end of the evaluation. As a reminder, these ratings were based on the self-reports provided by the Soldiers themselves. The list of problems and conditions were the same as those used on the background questionnaire (see Table 4) and the same four-point scale was also used (0 = "N/A," 1="Mildly," 2="Moderately," and 3="Severely"). The data generated was analyzed two ways. The first was on a between-groups basis - comparing the treated and untreated uniforms against each other at the midpoint and final. The second was on a within-groups basis - comparing the results for each uniform from the midpoint to the final.

Before getting into the in-depth analysis of the results obtained, it may be useful to look at some general statistics related to the use of this question on the background, midpoint, and final questionnaires. On the background questionnaire, Soldiers had estimated the types of problems that they would expect to have during a week in the field. These expectations were accurate 75% of the time. In other words, 75% of the time a problem anticipated on the background also showed up during the evaluation on the midpoint or final. Fifteen percent of the time an anticipated problem did not appear and only 10% of the time did an unanticipated problem appear. The average number of problems reported during the evaluation by any particular Soldier was four and the problems lasted an average of four of the seven days.

On both the midpoint and final questionnaires, Soldiers were presented with a list of common maladies and symptoms that could be caused or influenced by bacteria and microbes encountered in the field. They used a four-point scale (0 = "N/A," 1="Mildly," 2="Moderately," and 3="Severely") to indicate to what extent they were experiencing each problem. Table 6, below shows the percentage of Soldiers who experienced each problem, regardless of its intensity. Also included are the results of a Chi-square analysis comparing the results for the treated and untreated uniforms at each point in time.

Table 6
Problems and Conditions Experienced in the Field: Treated vs. Untreated Uniform
(n=136)

Problem:	**Midpoint A[1] vs. B[2]**		**X^2 Result**	**Final A[1] vs. B[2]**		**X^2 Result**
Body odor	93%	87%	X^2=1.62, ns	95%	93%	X^2=0.09, ns
General discomfort	65%	72%	X^2=0.72, ns	69%	69%	X^2=0.04, ns
Heat Rash	12%	18%	X^2=0.98, ns	19%	18%	X^2=0.01, ns
Itching skin	35%	34%	X^2=0.01, ns	32%	26%	X^2=0.54, ns
Chafing	23%	25%	X^2=0.07, ns	24%	8%	X^2=5.98, p<.01
Skin rash/irritation	20%	31%	X^2=2.23, ns	24%	16%	X^2=1.19, ns
Skin inflammation/redness	11%	18%	X^2=1.52, ns	17%	13%	X^2=0.46, ns
Skin lesions or sores	3%	12%	X^2=4.22, p<.05	9%	7%	X^2=0.35, ns
Acne/pimples	25%	34%	X^2=1.34, ns	23%	28%	X^2=0.49, ns
Infected or inflamed hair follicles	15%	20%	X^2=0.60, ns	16%	16%	X^2=0.04, ns
Infected cuts or scrapes	5%	5%	X^2=0.61, ns	8%	10%	X^2=0.14, ns

[1] A=Treated Uniform
[2] B= Untreated Uniform

As can be seen above, there were essentially no significant differences between uniform types for the percentage of Soldiers reporting certain problems at either the midpoint or end of the evaluation. Only two significant differences were detected at all: a significant decrease in "skin lesions and sores" for the treated uniform group at the midpoint and a significant decrease in chafing for the untreated uniform at the final. Overall, there did seem to be a trend toward a lower percentage of reported problems for the treated uniform at the midpoint. However, this trend is not apparent in the results from the final questionnaire.

The ratings data from Soldiers who reported experiencing problems is presented below in Table 7. The means reported are derived from the four-point intensity scale. The data is presented in the same manner as it is above: by uniform type on a between-groups basis for both the midpoint and end of the evaluation. Also included is the result of a t-test to test for differences between uniform types. In this instance, the number of respondents is also included to reinforce the point that the ratings do not apply to the total survey group, but only to those who had a specific problem.

Table 7
Problems and Conditions Experienced in the Field: Treated vs. Untreated Uniform
(n=136)

Midpoint to Final – Intensity Between-groups	**Midpoint A[1] vs. B[2]**		**n, t, p**	**Final A[1] vs. B[2]**		**n, t, p**
Body odor	1.6	1.7	123, t=0.58, ns	1.9	1.9	128, t=0.03, ns
General discomfort	1.5	1.6	93, t=0.57, ns	1.4	1.5	94, t=0.58, ns
Heat Rash	1.2	1.6	20, t=1.58, ns	2.0	1.6	25, t=1.41, ns
Itching skin	1.6	1.6	47, t=0.20, ns	1.5	1.3	40, t=0.98, ns
Chafing	1.5	1.3	32, t=0.87, ns	1.6	1.6	23, t=0.13, ns
Skin rash/irritation	1.3	1.4	34, t=0.17, ns	1.7	1.5	28, t=0.68, ns
Skin inflammation/redness	1.5	1.5	19, t=0.19, ns	1.9	1.8	21, t=0.24, ns
Skin lesions or sores	1.0	1.3	9, t=0.79, ns	1.4	1.8	11, t=0.98, ns
Acne/pimples	1.5	1.3	40, t=1.41, ns	1.4	1.4	34, t=0.00, ns
Infected or inflamed hair follicles	1.3	1.3	23, t=0.25, ns	1.3	1.2	22, t=0.22, ns
Infected cuts or scrapes	1.0	1.3	7, t=1.20, ns	1.3	1.3	12, t=0.00, ns

[1] A=Treated Uniform
[2] B= Untreated Uniform

Overall, the intensity ratings generally fell in the range of "slight" to "moderate," and never exceeded the upper end of this range. There were also no significant differences detected between the treated and untreated uniform groups for any of these criteria. There also seems to be no overall trend across all of the problems in favor of one uniform or the other at either the midpoint or final. Ultimately, we would have to conclude that these results are mixed and do not favor either uniform type.

The ratings data presented in Table 7 were also analyzed on a within groups basis using the Sign test to evaluate whether either of the two uniform groups showed changes in their ratings of problem intensities from the midpoint to final questionnaires. This procedure calculates the differences between two variables (e.g. chafing at midpoint versus final) on an individual basis and classifies the changes observed as either positive, negative, or tied. If the two variables are similarly distributed (i.e. little change occurred), the numbers of positive and negative differences will not be significantly different. We included those who answered "0" or "N/A" since this was essential to show the direction of change in intensity ratings for the total survey group. The three resulting categories were decreasing intensity, increasing intensity, and no change. Changes in intensity ratings were also analyzed on a between group

basis by using the Chi-square test to determine if the distribution of observed changes varied by treatment level. The results obtained are presented below in Table 8. For each treatment group the percentage showing decreases, increases, or no change for each problem is shown followed by the Sign test to determine if the overall change was significant for that treatment group. The last column shows the Chi-square test comparing the change for the two groups.

Table 8
Problems and Conditions Experienced in the Field: Direction of Intensity Change
(n=136)

	Treated - A				Untreated - B				
	Decrease	No Change	Increased	Sign Test Results (Within-groups)	Decrease	No Change	Increased	Sign Test Results (Within-groups)	X^2 Results (Between-groups)
Body odor	9%	61%	29%	p<.05	5%	67%	28%	p<.01	ns
General discomfort	20%	55%	24%	ns	28%	51%	21%	ns	ns
Heat Rash	5%	80%	15%	ns	7%	88%	5%	ns	ns
Itching skin	15%	77%	8%	ns	18%	75%	7%	ns	ns
Chafing	13%	73%	13%	ns	18%	80%	2%	p<.01	*
Skin rash/irritation	8%	76%	16%	ns	20%	75%	5%	p<.05	**
Skin inflammation/redness	3%	85%	12%	ns	12%	80%	8%	ns	ns
Skin lesions or sores	1%	91%	8%	ns	7%	90%	3%	ns	ns
Acne/pimples	12%	83%	5%	ns	10%	82%	8%	ns	ns
Infected or inflamed hair follicles	4%	92%	4%	ns	12%	82%	7%	ns	ns
Infected cuts or scrapes	5%	87%	8%	ns	2%	92%	6%	ns	ns

*X^2=6.38, p<.05
**X^2=7.21, p<.05

The Sign test revealed that the distribution for both the treated and untreated groups showed a significant difference for odor, with this difference being a larger than expected number of respondents noting an increase in body odor. A significant difference was also detected for chafing and irritation in the treated group. Significantly more Soldiers than expected in the untreated group reported a decrease in chafing and irritation. This was not observed for the treated group. The results for the remaining problems show no significant change between the distribution of the midpoint and final ratings for either

uniform. The Chi-square test did identify a significant difference between uniform groups for chafing and irritation, specifically that Soldiers in the untreated group were skewed more towards "no change" or a decrease in the intensity of the problems. Initially, the same percentage of Soldiers in both groups reported chafing. For some reason, the percentage for the untreated group dropped significantly at the end of the evaluation while it stayed the same in the treated group. No other significant differences were detected.

During the course of the evaluation, the same percentage of Soldiers in both groups reported that they developed a problem that they would not normally have (treated: 17%, n=13 out of 75; untreated: 15%, n=9 out of 61). The specific problems identified by Soldiers in the treated group were rashes (n=4), general irritation (n=4), chafing (n=2), itching (n=1), and a sore (n=1). These problems occurred at the crotch (n=3), feet (n=3), arms (n=2), legs (n=2), ankle (n=2), or "all over" (n=1). Most treated these problems themselves (n=7), although a few saw a medic (n=2) or did nothing (n=2). Soldiers in the untreated group reported similar problems: rash (n=3), general irritation (n=2), chafing (n=1), itching (n=1), and "acne" (n=1). Problems were noted at the arm (n=2), crotch (n=1), armpit (n=1), neck (n=1), shoulders (n=1), and "all over" (n=1). Soldiers either treated the problem themselves (n=2), went to a medic (n=2), or did nothing (n=2).

A few Soldiers in each group also noted a reduction in chronic skin problems and conditions that they had (treated: 7%, n=5 out of 73; untreated: 9% (n=5 out of 59), with no significant difference detected between uniform groups. Soldiers in the treated group reported that these problems were skin rash (n=2) and athlete's foot (n=1), while Soldiers in the untreated group reported less dry skin (n=1), sweating (n=1), and chafing (n=1). A few Soldiers in each group noted what they felt was a reduction in other bacteria-related problems (treated: 8%, n=6 out of 73; untreated: 7%, n=4 out of 58). For the treated group these were identified as diarrhea (n=3) and body odor (n=2), the untreated group noted diarrhea (n=1), body odor (n=1), and general discomfort (n=1). There were no significant differences detected for these variables by uniform group.

Only a handful of Soldiers in each group reported that they had to consult medical personnel for a skin-related problem during the course of the evaluation (treated: 7%, n=5 out of 75; untreated: 5%, n=3 out of 60). This was not a significant difference. Soldiers in the treated group indicated that this was for a rash (n=2), blister on the foot (n=1), and to have cactus needles removed (n=1). According to the respondents, they did not lose any duty time to have these problems addressed. Soldiers in the untreated

group reported an infected cut (n=1) and a "staph" infection (n=1), with no time lost for the former and three hours lost as an outpatient for the latter.

Comparison

On the final questionnaire, Soldiers compared the problems that they experienced during the evaluation to what they would expect when wearing the BDU or DCU under similar conditions. A "not applicable" option was provided for Soldiers who did not experience a certain problem, so the number of respondents for each criteria varies to some extent. The scale used and results obtained are presented below.

Table 9
Problems and Conditions Compared: This Exercise With Past Experience
(n=136)

Problems on this exercise have been…

MUCH WORSE	MODERATELY WORSE	SLIGHTLY WORSE	ABOUT THE SAME	SLIGHTLY BETTER	MODERATELY BETTER	MUCH BETTER
1	2	3	4	5	6	7

Comparison: Problems on this exercise w/this uniform and past exercises with the BDU / DCU	**Treated - A (n=75)**	**Untreated - B (n=61)**	**n, t, p**
Body odor	4.3	4.1	131, t=0.58, ns
General discomfort	4.3	4.0	121, t=1.18, ns
Heat Rash	4.2	4.3	76, t=0.08, ns
Itching skin	4.2	4.2	85, t=0.09, ns
Chafing	4.4	4.6	74, t=0.83, ns
Skin rash/irritation	4.2	4.3	79, t=0.51, ns
Skin inflammation/redness	4.2	4.4	70, t=0.42, ns
Skin lesions or sores	4.5	4.3	63, t=0.38, ns
Acne/pimples	4.5	4.3	80, t=1.07, ns
Infected or inflamed hair follicles	4.5	4.3	73, t=0.59, ns
Infected cuts or scrapes	4.6	4.4	65, t=0.73, ns
Overall	4.6	4.5	116, t=0.57, ns

There were no significant differences detected between either the treated or untreated groups for any of the problems rated. All of the ratings fell between "about the same" and "slightly better." The treated uniform did seem to do slightly better for some criteria (body odor, general discomfort, skin lesions, acne, and infected hair follicles, infected cuts, and overall). However, the trend is not that strong since some of the differences were small and the untreated uniform was rated higher for heat rash, chafing, skin rash, and skin inflammation, and the two were tied for itching.

Soldiers were asked if they felt that the uniforms they were issued for the evaluation were hotter than various versions of the standard BDU or DCU. Since there were no significant differences detected by uniform group, we will report these percentages for the total survey group combined. A little more than a third of the survey group felt that the uniforms that they evaluated were hotter than the Hot Weather BDU (36%, n=47 out of 130) or the Regular BDU (38%, n=49 out of 130). More than half felt that the uniform they were issued was hotter than the standard DCU (57%, n=73 out of 129), which was the uniform that most would normally wear in the field (see Table 2). Comments indicate that these Soldiers felt there was less air circulation in the new uniform (n=10), it seemed to "hold in" heat (n=9), the material felt heavier than the DCU (n=3), and that it seems to hold more sweat than the DCU (n=2).

It is difficult to know what impact, if any, this perception would have on the measures taken of the effectiveness of the antimicrobial treatment. It did not seem to show up in the data presented in Table 9 – Soldiers rated the problems encountered with the new uniform between "about the same" and "slightly better" when compared to what they would expect when wearing the BDU or DCU. A check of the data did find one significant difference for the problems and conditions evaluated in the previous section (see Table 6) based on heat perception. Those Soldiers who felt the new uniform was hotter reported a significantly higher rate of "general discomfort" than those Soldiers who did not (84%, n=61 out of 73 vs. 55%, n=31 out of 56, $X^2=12.32$, $p<.001$). This was true regardless of treatment level – nearly identical results were obtained when the data was analyzed separately for the treated group (83%, n=35 out of 42 vs. 53%, n=17 out of 32, $X^2=7.93$, $p<.01$) and the untreated group (84%, n=26 out of 31 vs. 58%, n=14 out of 24, $X^2=4.45$, $p<.05$). It is also important to remember that this was their first experience with this uniform and they had worn it for one week, so there is some question about how pervasive this perception would be over the long term.

Impact

We asked Soldiers if they felt that the uniform they were issued was noticeably decreasing their body odor as well as the body odor of others during the exercise. This question was included on both the midpoint and final questionnaires. The results obtained at each data collection point are presented below in Table 10.

Table 10
Is the Uniform Noticeably Decreasing Body Odor? Midpoint and Final
(n=136)

	Treated –A	**Untreated - B**	
Midpoint: Your body odor?	51% (38/74)	43% (26/60)	X^2=.85, ns
Midpoint: Other Soldier's body odor?	58% (43/74)	47% (28/60)	X^2=1.74, ns
	Treated- A	**Untreated -B**	
Final: Your body odor?	58% (43/74)	37% (22/60)	X^2=6.10, p<.01
Final: Other Soldier's body odor?	59% (43/73)	38% (23/61)	X^2=5.98, p<.01

No significant differences were detected in the results from the midpoint survey. However, data from the final questionnaires shows that significantly more respondents with the treated uniform felt that body odor was being controlled in themselves and others than did those with the untreated uniform. This data shows that more than half of the Soldiers in the treated group did notice a reduction in odor. This presents us with a contradiction in the data since multiple other ways of measuring odor reduction (see Tables 6, 8, and 9) failed to detect a significant impact of treatment level. It is also interesting that more than a third of the Soldiers in the untreated group felt that the uniform was doing something to control their body odor. Since Soldiers did not know which type of uniform they were receiving (treated or untreated) it is possible that there is a "placebo effect" at work. It could also be that some feature or property of the uniform aside from the treatment did have an impact on odor, possibly even the "newness" of the uniforms theselves.

The majority of the Soldiers felt that the uniform they were issued either had no impact (50%, n=67 out of 133) or a positive impact (41%, n=54 out of 133) on their performance in the field. Only 10% (n=12 out of 133) felt that it had a negative impact. These results are reported for the total survey group because they were identical for both the treated and untreated uniform sub-groups. Furthermore, 93% (n=124 out of 134) felt that the uniform was safe to wear, with no significant differences detected by uniform type. Those who did not feel it was safe generally did not offer much in the way of comments, other than to note that the uniform felt hot (n=3).

Finally, about two thirds of the Soldiers (64%, n=85 out of 132) felt the uniform was comfortable for wearing over an extended period of time. No significant differences were detected by uniform type. Soldiers in the treated uniform group who did not think it was comfortable commented that it was too hot (n=5), was not any different than the DCU (n=4), that seven days was too long without a clean uniform (n=4), and that the material was stiff with dried sweat (n=2). These comments were echoed by Soldiers in the untreated group: it was too hot (n=7), seven days was too long (n=3), and the material was stiff with dried sweat (n=2).

Comments from Soldiers who felt that the uniform was comfortable for extended wear do provide some interesting anecdotal evidence in support of the treated uniform. Soldiers in that group commented that "I do not feel dirty" (n=6), and noted that it was a very comfortable uniform overall (n=5) or that it was more comfortable than the DCU (n=4). Those in the untreated group noted that they liked the uniform (n=3), it was more comfortable than the DCU (n=3), and a few noted that it was comfortable if you ignored the smell (n=2). No one in the untreated group commented that they did not feel dirty and two who felt that it was comfortable added the caveat about how bad it smelled.

Acceptability

Soldiers rated their overall satisfaction level with the uniform they were issued on a variety of criteria. The data, which was obtained exclusively on the final questionnaire, was analyzed by uniform group. The scale used and results obtained are presented below in Table 11.

Table 11
Uniform Satisfaction Ratings
(n=136)

VERY DISSATISFIED 1	MODERATELY DISSATISFIED 2	SOMEWHAT DISSATISFIED 3	NEITHER DISSATISFIED NOR SATISFIED 4	SOMEWHAT SATISFIED 5	MODERATELY SATISFIED 6	VERY SATISFIED 7

	Treated - A (n=75)	**Untreated - B (n=61)**	**t, p**
Comfort of the uniform – start	5.4	5.1	t=0.73, ns
Comfort of the uniform – end	4.4	4.0	t=1.19, ns
Overall comfort	4.9	4.4	t=1.70, ns
Ability to reduce body odor	4.5	4.0	t=1.70, ns
Ability to reduce skin problems	4.5	4.2	t=0.99, ns
Overall performance	4.9	4.6	t=0.76, ns

In general, both uniforms received ratings in the "somewhat satisfied" range. The highest rated criteria, between "somewhat satisfied" and "moderately satisfied" was comfort at the start of the test for the treated uniform. The lowest rated criteria was comfort at the end of the test for the untreated uniform ("neither dissatisfied nor satisfied"). No significant differences were detected for any of the criteria. However, there is a clear overall trend apparent – the treated uniform was rated higher than the untreated uniform for all of the criteria. Soldiers with the antimicrobial treatment tended to be more satisfied with the performance of their uniform than Soldiers who did not have the treatment.

T-SHIRT FINDINGS

Background

A total of 59 Soldiers completed the T-shirt evaluation: 28 had the treated and 31 had the untreated items. There were no significant differences between the two groups for any of the data discussed in this section, so these results will be presented for the total survey group. Soldiers reported that they wore the T-shirts for an average of 23 hours per day for six to seven days and they changed T-shirts every two days. While no one had to stop wearing the T-shirts during the course of the evaluation, 15% (n=9 out of 59) reported that they did wear another type of T-shirt, most of which were identified as the standard brown cotton T-shirt (n=5). There were no durability problems reported any of the test items.

Most Soldiers (81%, n=47 out of 58) wore the DCU during the evaluation, with the remainder wearing either the Hot Weather BDU (14%, n=8 out of 58) or the regular BDU (5%, n=3 out of 58). Regardless of type, Soldiers had an average of two to three uniforms with them and changed twice during the exercise. Some reported wearing additional items under their uniform top and T-shirt (17%, n=10 out of 58). These were generally female Soldiers who wore a bra. More than half (60%, n=35 out of 58) used some kind of hygiene or first aid product under the T-shirt that was branded as "anti-bacterial." This was almost exclusively deodorant that was used under the arms on a daily basis (n=33). Less than one-fourth (21%, n=12 out of 58) reported that they came in contact with something that might cause a problem for their skin. The most common sources identified were biting insects (n=17), other natural irritants (n=9), caustic substances like gasoline or battery acid (n=7), and "irritants" like insect repellent or camouflage face paint (n=5).

Performance

Soldiers rated the types of problems and conditions that they were experiencing at the midpoint and end of the evaluation. As a reminder, these ratings were based on the self-reports provided by the Soldiers themselves. The list of problems and conditions were the same as those used on the background questionnaire (see Table 4) and the same four-point scale was also used (0 = "N/A," 1="Mildly," 2="Moderately," and 3="Severely"). Overall, 71% of the time that a Soldier indicated he would expect to experience a problem on the background questionnaire he also indicated that it was actually experienced on the midpoint or final. Nineteen percent of the time the problem identified on the background questionnaire did not appear during the evaluation and 10% of the time an unexpected problem developed. The average number of problems reported was four per Soldier and the problems lasted an average of five of the seven days.

The data from the midpoint and final questionnaires was analyzed two ways. The first was on a between-groups basis - comparing the treated and untreated T-shirts against each other at the midpoint and final. The second was on a within-groups basis - comparing the results for each T-shirt from the midpoint to the final. Table 12, below shows the percentage of Soldiers who experienced each problem, regardless of its intensity. Also included are the results of a Chi-square analysis comparing the results for the treated and untreated items at each point in time.

Table 12
Problems and Conditions Experienced in the Field: Treated vs. Untreated T-shirt
(n=59)

Problem:	**Midpoint A[1] vs. B[2]**		**X^2 Result**	**Final A[1] vs. B[2]**		**X^2 Result**
Body odor	79%	77%	X^2=0.11, ns	75%	94%	X^2=3.92, p=.05
General discomfort	54%	45%	X^2=0.41, ns	39%	52%	X^2=0.91, ns
Heat Rash	18%	16%	X^2=0.03, ns	18%	23%	X^2=0.20, ns
Itching skin	29%	32%	X^2=0.09, ns	36%	42%	X^2=0.24, ns
Chafing	14%	10%	X^2=0.30, ns	11%	7%	X^2=0.35, ns
Skin rash/irritation	11%	13%	X^2=0.07, ns	11%	19%	X^2=0.85, ns
Skin inflammation/redness	7%	10%	X^2=0.12, ns	11%	10%	X^2=0.02, ns
Skin lesions or sores	0%	7%	X^2=1.87, ns	0%	3%	X^2=0.92, ns
Acne/pimples	32%	36%	X^2=0.07, ns	36%	29%	X^2=0.30, ns
Infected or inflamed hair follicles	18%	7%	X^2=1.83, ns	18%	7%	X^2=1.83, ns
Infected cuts or scrapes	0%	7%	X^2=1.87, ns	7%	3%	X^2=0.47, ns

[1] A=Treated T-shirt
[2] B= Untreated T-shirt

As can be seen above, there was a significant difference between the two T-shirts: a significantly lower rate of body odor for the treated T-shirt at the end of the evaluation. However, the final questionnaire data suggests that Soldiers who used the treated T-shirt were better off overall than those who used the untreated T-shirt, particularly in the areas of body odor, general discomfort, heat rash, skin rash and irritation, and skin lesions. Soldiers who used the untreated T-shirt seemed to derive some benefit in two areas (acne and infected hair follicles) and a possible benefit in one other (chafing).

The intensity ratings from Soldiers who reported experiencing problems is presented below in Table 13. The data is presented in the same manner as it is above: by T-shirt type on a between-groups basis for both the midpoint and end of the evaluation. Also included is the result of a t-test to test for differences between T-shirts, as well as the number of respondents to reinforce the point that the ratings apply only to those who had a specific problem.

Table 13
Problems and Conditions Experienced in the Field: Treated vs. Untreated T-shirt
(n=59)

Midpoint to Final – Intensity Between-groups	**Midpoint A[1] vs. B[2]**		**n, t, p**	**Final A[1] vs. B[2]**		**n, t, p**
Body odor	1.4	1.3	46, t=0.45, ns	1.4	1.4	50, t=0.32, ns
General discomfort	1.3	1.5	29, t=1.01, ns	1.4	1.6	27, t=0.95, ns
Heat Rash	1.2	2.0	10, t=4.00, p<.01	1.4	1.4	12, t=0.09, ns
Itching skin	1.4	1.6	18, t=0.76, ns	1.2	1.2	23, t=0.28, ns
Chafing	1.3	1.0	7, t=0.85, ns	1.0	1.5	5, t=1.34, ns
Skin rash/irritation	1.3	2.0	7, t=1.20, ns	1.3	1.3	9, n/a
Skin inflammation/redness	1.0	1.3	5, t=0.78, ns	1.3	2.0	6, t=2.00, ns
Skin lesions or sores	-	1.0	2, n/a	-	1.0	1, n/a
Acne/pimples	1.3	1.2	20, t=0.75, ns	1.6	1.4	19, t=0.65, ns
Infected or inflamed hair follicles	1.4	1.0	7, t=0.98, ns	1.4	1.0	7, t=0.98, ns
Infected cuts or scrapes	-	1.0	4, n/a	-	2.0	3, n/a

[1] A=Treated T-shirt
[2] B= Untreated T-shirt

All of the intensity ratings fell in the "slight" to "moderate" range for all of the conditions evaluated. One significant difference was detected: a significantly lower rating for heat rash at the midpoint for the Soldiers wearing the treated T-shirt. However, again it seems that the overall trend slightly favors the treated T-shirt beyond that single significant difference. The intensity ratings for the treated T-shirt tended to be lower than the untreated T-shirt at both the midpoint and final data collection points.

As in the previous section, we calculated the direction of change in intensity ratings for the T-shirt on an individual basis between the midpoint and final data collection. A respondent could fall into one of three categories – decreasing intensity, increasing intensity, and no change. This analysis includes all respondents, even those who reported that they never experienced a certain problem – which in this case is a valid data point. The Sign test was used to determine if changes within the T-shirt groups between the midpoint and final were significant. The Chi-square test was used to determine if the distribution of observed changes varied by treatment level. The results obtained are presented below in Table 14.

Table 14
Problems and Conditions Experienced in the Field: Direction of Intensity Change
(n=59)

	Treated - A				Untreated - B				
	Decrease	No Change	Increased	Sign Test Results (Within-groups)	Decrease	No Change	Increased	Sign Test Results (Within-groups)	X^2 Results (Between-groups)
Body odor	25%	54%	21%	ns	23%	39%	39%	ns	ns
General discomfort	18%	75%	7%	ns	19%	55%	26%	ns	ns
Heat Rash	7%	86%	7%	ns	16%	68%	16%	ns	ns
Itching skin	14%	68%	18%	ns	19%	58%	23%	ns	ns
Chafing	7%	89%	4%	ns	7%	90%	3%	ns	ns
Skin rash/irritation	7%	82%	11%	ns	13%	71%	16%	ns	ns
Skin inflammation/redness	3%	86%	11%	ns	10%	80%	10%	ns	ns
Skin lesions or sores	0%	100%	0%	ns	7%	90%	3%	ns	ns
Acne/pimples	11%	68%	21%	ns	16%	71%	13%	ns	ns
Infected or inflamed hair follicles	7%	86%	7%	ns	6%	88%	6%	ns	ns
Infected cuts or scrapes	4%	92%	4%	ns	7%	90%	3%	ns	ns

While the sign test did not detect any significant differences on a within-groups basis, and the Chi-Square test did not detect significant differences on a between groups basis, there does seem to be something going on here. A higher percentage of Soldiers with the treated T-shirt reported that body

odor, general discomfort, and heat rash decreased or stayed the same than those in the untreated group. The percentage of Soldiers in the treated group who reported an increase in general discomfort was only one-fourth that of the untreated group (7% vs. 26%). Soldiers in the treated group also had only half the rate of body odor and heat rash increase (21% and 7%, respectively) than did Soldiers in the untreated group (39% and 16%, respectively).

During the course of the evaluation, none of the Soldiers in the treated group reported that they developed a problem that they would not normally have. This was not the case in the untreated group (13%, n=4 out of 30). While this might seem significant, a Chi-square test determined that it was not statistically so. Soldiers with the untreated T-shirt noted a rash (n=2) or itchy skin (n=2). The location was usually "wherever the T-shirt touched" (n=3) or just on the back (n=1). No one reported taking any action to correct the problem other than "cleaning with baby wipes" (n=2). None of the Soldiers in either group reported that they had to consult medical personnel for a skin-related problem during the course of the evaluation.

A few Soldiers in each group also noted a reduction in chronic skin problems and conditions that they had (treated: 16%, n=4 out of 25; untreated: 7%, n=2 out of 31). Soldiers in the treated group reported that these problems were body odor (n=2) and skin irritation (n=1), while Soldiers in the untreated group reported either a decrease in odor (n=1) or sweating (n=1). A few Soldiers in each group noted what they felt was a reduction in other bacteria-related problems (treated: 15%, n=4 out of 26; untreated: 7%, n=2 out of 31). For the treated group these were identified as diarrhea (n=1), body odor (n=1), and sweating (n=1). Only one comment was received for the untreated group: body odor (n=1). While there were no significant differences detected for these variables by treatment group, the results reinforce the trend in favor of the treated T-shirt that we have seen so far.

Comparison

Soldiers compared the problems that they experienced during the evaluation to what they would expect from wearing the standard T-shirt under similar conditions, which for most would be the brown cotton T-Shirt. A "not applicable" option was provided for Soldiers who did not experience a certain problem, so the number of respondents for each criteria varies to some extent. The scale used and results obtained are presented below.

Table 15
Problems and Conditions Compared: This Exercise With Past Experience
(n=59)

Problems on this exercise have been…

MUCH WORSE 1	MODERATELY WORSE 2	SLIGHTLY WORSE 3	ABOUT THE SAME 4	SLIGHTLY BETTER 5	MODERATELY BETTER 6	MUCH BETTER 7

Comparison: Problems on this exercise w/this T-shirt and past exercises with the std. T-shirt	**Treated -A (n=28)**	**Untreated - B (n=31)**	**n, t, p**
Body odor	5.2	4.7	54, t=1.21, ns
General discomfort	4.9	4.4	48, t=1.43, ns
Heat Rash	4.8	4.4	33, t=1.02, ns
Itching skin	4.5	4.2	36, t=0.75, ns
Chafing	4.7	4.2	31, t=1.30, ns
Skin rash/irritation	4.6	4.5	30, t=0.13, ns
Skin inflammation/redness	4.8	4.5	28, t=0.65, ns
Skin lesions or sores	4.8	4.5	26, t=0.74, ns
Acne/pimples	4.6	4.5	32, t=0.27, ns
Infected or inflamed hair follicles	4.7	4.3	29, t=0.93, ns
Infected cuts or scrapes	4.6	4.2	28, t=1.21, ns
Overall	5.2	5.0	49, t=0.55, ns

There were no significant differences detected between either the treated or untreated groups for any of the problems rated. Most of the ratings fell between "about the same" and "slightly better" – with the exception of body odor and "overall," which the treated group rated in the "slightly better" to "moderately better" range. It should be noted that Soldiers in the treated group provided higher ratings than those in the untreated group for all of the criteria evaluated, which would seem to strengthen the case for an emerging underlying trend in favor of this item.

Soldiers were asked if they felt that the T-shirts they were issued for the evaluation felt hotter than the standard cotton T-shirt. Overall, 23% (n=13 out of 57) of the Soldiers in the combined groups felt that they were. While the difference was not significant, only half the percentage of respondents felt that this was true about the treated compared to the untreated T-shirt (15%, n=4 out of 26 vs. 29%, n=9 out of 31). Comments for the untreated T-shirt were: it felt hotter (n=2), the fabric stuck to the skin (n=1), and it did not breathe as well as the standard (n=1). Only one comment was received for the treated T-shirt: it did not seem to absorb sweat as well as the standard (n=1). Since this was such a relatively small percentage of the respondents, no further data analysis was conducted along these lines.

Impact

We asked Soldiers if they felt that the T-shirts they were issued were noticeably decreasing their body odor as well as the body odor of others during the exercise. This question was included on both the midpoint and final questionnaires. The results obtained at each data collection point are presented below in Table 16.

Table 16
Is the T-shirt Noticeably Decreasing Body Odor? Midpoint and Final
(n=59)

	Treated – A	**Untreated - B**	
Midpoint: Your body odor?	61% (17/28)	68% (21/31)	X^2=.32, ns
Midpoint: Other Soldier's body odor?	50% (14/28)	57% (17/30)	X^2=1.74, ns

	Treated – A	**Untreated - B**	
Final: Your body odor?	73% (19/26)	58% (18/31)	X^2=1.40, ns
Final: Other Soldier's body odor?	62% (16/26)	53% (16/30)	X^2=0.38, ns

While no significant differences were detected it is clear that more Soldiers in the treated group felt that the T-shirt was reducing odor than did those in the untreated group. Nearly three-fourths of the Soldiers with the treated T-shirt felt that it was controlling their body odor. However, it is also interesting that more than half of the Soldiers in the untreated group felt the same way. As noted in the uniform section, this could be due either to a "placebo effect" or due to some feature or property of the T-shirt that

could, in itself, have an impact on odor. Open-ended comments show no clear trend in why Soldiers felt the way they felt. Some interesting individual comments from the treated group were: "[odor reduced] just for the first two days, after that body odor was same" and "[the] shirt dried up quick, odor did not become noticeable." Individual Soldiers in the untreated group noted: "decreased body odor because of [decreased] sweat" and "I wore each one two days in a row and the shirt did not smell noticeably terrible."

The majority of the Soldiers felt that the T-shirts they were issued either had a positive impact (51%, n=26 out of 57) or no impact (46%, n=26 out of 57) on their performance in the field. Only 3% (n=2 out of 57) felt that it had a negative impact. These results are reported for the total survey group because they were identical for both the treated and untreated T-shirt sub-groups.

Furthermore, 96% (n=25 out of 26) of the Soldiers in the treated group and 81% (n=25 out of 31) of Soldiers in the untreated group felt that the T-shirts were safe to wear. While this difference is interesting, it was not statistically significant. Only one negative comment was received for the untreated T-shirt, and that was "people were complaining of rashes." This problem was noted earlier amongst the untreated group and could have caused either those Soldiers who experienced or those who heard of it to conclude that the T-shirt was not safe. However, it seemed to be a problem exclusive to the untreated group since none of the Soldiers in the treated group experienced the rash and no negative safety comments were received.

More Soldiers in the treated group (86%, n=24 out of 28) compared to Soldiers in the untreated group (65%, n=20 out of 31) also felt the T-shirt was comfortable for wearing over an extended period of time, which was nearly a significant difference (X^2=3.49, p=.06). Comments from each group were nearly identical. Soldiers with the treated T-shirt felt that it dried quicker than the standard (n=2), retained less odor (n=2), and improved their comfort level in the field (n=2). Soldiers with the untreated T-shirt felt that it dried faster (n=3) and retained less odor (n=2) than the standard cotton T-shirt.

Acceptability

Soldiers rated their overall satisfaction level with the T-shirts they were issued on a variety of criteria. The data, which was obtained exclusively on the final questionnaire, was analyzed by treatment group. The scale used and results obtained are presented below in Table 17.

Table 17
T-shirt Satisfaction Ratings
(n=59)

VERY DISSATISFIED 1	MODERATELY DISSATISFIED 2	SOMEWHAT DISSATISFIED 3	NEITHER DISSATISFIED NOR SATISFIED 4	SOMEWHAT SATISFIED 5	MODERATELY SATISFIED 6	VERY SATISFIED 7

	Treated - A (n=28)	**Untreated - B (n=31)**	**t, p**
Comfort of the T-shirt – start	5.3	5.1	t=0.46, ns
Comfort of the T-shirt – end	5.3	4.9	t=1.07, ns
Overall comfort	5.4	5.0	t=1.03, ns
Ability to reduce body odor	5.2	4.7	t=1.24, ns
Ability to reduce skin problems	5.0	4.4	t=1.90, ns
Overall performance	5.3	5.0	t=0.71, ns

No significant differences were detected for any of the criteria. However, there is a clear overall trend for higher ratings for the treated T-shirts. In general, the treated T-shirt received ratings in the "somewhat satisfied" to "moderately satisfied" range. The untreated T-shirt received ratings in the neutral to "somewhat satisfied range." It is also interesting to note that the ratings for the comfort of the treated T-shirt stayed the same for the start and end of the test. Soldiers in the untreated group rated comfort at both the beginning and the end lower than those in the treated group and lower for there T-shirt from the start to the end. This further reinforces the trend that the treated T-shirt demonstrates a significant reduction in body odor over the untreated T-shirt, as well as an overall reduction in other types of problems encountered. Furthermore, the treated T-shirt also outperformed its untreated counterpart in terms of acceptability and comfort.

SOCK FINDINGS

Background

A total of 172 Soldiers completed the sock evaluation: 59 had the untreated control sock (type A), 52 had the standard sock with the standard treatment (type B), and 61 had the standard sock with a candidate treatment (type C). As noted with the uniform and T-shirt, there were no significant differences between the three groups for any of the background data discussed in this section – so the results will be presented for the total survey group. Soldiers estimated that they wore the socks for an average of six days and that they changed them once per day over the course of the evaluation. Twenty percent (n=34 out of 172) reported that they did run out of test socks during the evaluation . These Soldiers reported substituting the standard green sock (n=7), a commercial cotton sock (n=6), a commercial boot sock (n=5), or the standard black wool sock (n=4). Only a few durability problems were reported with the test sock (4%, n=6 out of 174). These were identified as holes (n=2), pilling (n=1), or they "fell apart" (n=1).

The socks were generally worn with standard issue boots (91%, n=153 out of 169), which was almost exclusively the Desert Boot (n=148). The remainder (9%, n=16) wore a commercial boot, identified as being Corcorans (n=7), Bellevilles (n=4), or jungle boots (n=3). Twelve percent (n=21 out of 172) also used an insole in their boots, these were identified as Dr. Scholl's (n=5), a generic gel insert (n=3), or a variety of other types (n=8). Less than a quarter of the group (23%, n=39 out of 171) used some kind of hygiene or first aid product on their feet that was branded as "anti-bacterial." Specifically, Gold Bond powder (n=11), a generic foot powder or spray (n=7), baby powder (n=6), an athlete's foot product (n=4), an Army issue foot powder (n=3), or Dr. Scholl's powder (n=2). Only a few (6%, n=10 out of 171) reported that they came in contact with something that might cause a problem for their feet – with insects (n=2), sweat (n=2), and cactus needles (n=2) being mentioned.

Performance

Soldiers rated the types of foot problems and conditions that they were experiencing at the midpoint and end of the evaluation. These ratings were based on the self-reports provided by the Soldiers themselves. This list was the same as those used on the background questionnaire (see Table 5) and the same four-point scale was also used (0 = "N/A," 1="Mildly," 2="Moderately," and 3="Severely"). The average number of problems reported was two to three per Soldier and the problems lasted an average of five of the seven days. The relationship to problems anticipated on the background questionnaire and those actually observed at the midpoint or final demonstrate that Soldiers were accurate in their expectations 65% of the time. Twenty-eight percent of the time an anticipated problem did not develop and 7% of the time an unanticipated problem appeared.

On both the midpoint and final questionnaires, Soldiers were presented with a list of common maladies and symptoms that could be caused or influenced by bacteria and microbes encountered in the field. They used a four-point scale (0 = "N/A," 1="Mildly," 2="Moderately," and 3="Severely") to indicate to what extent they were experiencing each problem. Table 18 shows the percentage of Soldiers who experienced each problem, regardless of its intensity. Also included are the results of a Chi-square analysis comparing the results for the treated and untreated socks at each point in time.

Table 18
Problems and Conditions Experienced in the Field: Untreated & Treated Socks
(n=172)

	Midpoint				Final			
Problem:	**A[1]**	**B[2]**	**C[3]**	**X^2**	**A[1]**	**B[2]**	**C[3]**	**X^2**
Foot odor	77%	67%	59%	X^2=4.07, ns	85%	71%	69%	X^2=4.63, ns
General discomfort	36%	29%	30%	X^2=0.74, ns	37%	27%	25%	X^2=2.58, ns
Itching Feet	31%	33%	20%	X^2=2.85, ns	29%	25%	21%	X^2=0.90, ns
Athlete's Foot	9%	14%	10%	X^2=0.77, ns	10%	8%	12%	X^2=0.46, ns
Toe Nail Fungus	5%	10%	7%	X^2=0.90, ns	9%	15%	7%	X^2=2.66, ns
Skin rash/irritation	12%	6%	5%	X^2=2.41, ns	12%	8%	10%	X^2=0.54, ns
Skin inflammation/redness	10%	12%	8%	X^2=0.36, ns	12%	6%	5%	X^2=2.41, ns
Blisters or Calluses	15%	17%	10%	X^2=1.43, ns	14%	17%	10%	X^2=1.35, ns

[1] A=Untreated Standard Sock
[2] B=Sock (standard treated sock)
[3] C=Sock (standard sock with candidate treatment)

As can be seen above, no significant differences amongst the sock groups were detected for any of the problems evaluated. There is a trend for a lower percentage of reported problems for Soldiers who used the treated socks at both the midpoint and end of the evaluation. This is apparent for both socks B and C for foot odor, general discomfort, and skin inflammation (final only). It is also apparent for sock C for itching and blisters. Individually, there is an uncomfortably high level of probability that any one of these differences may be caused by chance. Collectively, when taken as a trend both over time and across all of the problems evaluated, it is possible to see that the antimicrobial treatments did seem to have some beneficial impact on the health of the Soldiers in the field.

The intensity ratings from Soldiers who reported experiencing problems is presented below in Table 19. The data is presented in the same manner as it is above: by sock type on a between-groups basis for both the midpoint and end of the evaluation. Also included is the result of an ANOVA to test for differences between sock types. The number of respondents is also included to show that the ratings apply only to those who had a specific problem.

Table 19
Problems and Conditions Experienced in the Field: Untreated & Treated Socks
(n=172)

	Midpoint				Final			
Problem:	**A[1]**	**B[2]**	**C[3]**	**n, F, p**	**A[1]**	**B[2]**	**C[3]**	**n, F, p**
Foot odor	1.3	1.3	1.2	116, 0.14, ns	1.3	1.4	1.4	129, 0.27, ns
General discomfort	1.3	1.1	1.3	54, 0.65, ns	1.2	1.4	1.5	51, 1.13, ns
Itching Feet	1.4	1.2	1.3	47, 0.29, ns	1.4	1.4	1.6	43, 0.63, ns
Athlete's Foot	1.2	1.6	1.2	18, 0.92, ns	1.3	1.5	1.7	17, 0.34, ns
Toe Nail Fungus	1.0	1.2	1.5	12, 1.13, ns	1.4	1.3	2.0	17, 1.60, ns
Skin rash/irritation	1.6	1.3	1.0	13, 1.46, ns	1.6	1.5	1.3	17, 0.16, ns
Skin inflammation/redness	1.3	1.2	1.0	17, 0.98, ns	1.6	1.7	2.0	13, 0.30, ns
Blisters or Calluses	1.4	1.2	1.5	24, 0.39, ns	1.3	1.2	1.5	23, 0.48, ns

[1] A=Untreated Standard Sock
[2] B=Sock (standard treated sock)
[3] C=Sock (standard sock with candidate treatment)

All of the intensity ratings fell in the "slight" to "moderate" range for all of the conditions evaluated and no significant differences were detected. The numbers of respondents for some of these problems are small and are divided across three groups. This can lead to a high level of variability in the responses that can both confound the scale and the statistical tests used to evaluate the results.

The direction of change in intensity ratings between the midpoint and final data collection was calculated on an individual basis. A respondent could fall into one of three categories – decreasing intensity, increasing intensity, and no change. This analysis includes all respondents, even those who reported that they never experienced a certain problem – which in this case is a valid data point. The Sign test was used to determine if changes within the sock groups between the midpoint and final were significant. The Chi-Square test was used to test for differences in the distribution across all three sock groups. The results obtained are presented below in Table 20.

Table 20
Problems and Conditions Experienced in the Field: Direction of Intensity Change from the Midpoint to the Final Questionnaire
(n=172)

	Untreated - A				Treated - B				Treated - C				
	Decrease	No Change	Increased	Sign Test Results (Within-groups)	Decrease	No Change	Increased	Sign Test Results (Within-groups)	Decrease	No Change	Increased	Sign Test Results (Within-groups)	X^2 Results (Between-groups)
Foot odor	14%	64%	22%	ns	19%	60%	21%	ns	12%	61%	28%	ns	ns
General discomfort	15%	71%	14%	ns	15%	66%	19%	ns	18%	66%	16%	ns	ns
Itching Feet	20%	60%	20%	ns	17%	71%	12%	ns	12%	75%	13%	ns	ns
Athlete's Foot	7%	85%	8%	ns	11%	85%	4%	ns	5%	87%	8%	ns	ns
Toe Nail Fungus	2%	93%	5%	ns	4%	86%	10%	ns	5%	88%	7%	ns	ns
Skin rash/irritation	7%	86%	7%	ns	4%	90%	6%	ns	3%	89%	8%	ns	ns
Skin inflammation/redness	5%	85%	10%	ns	10%	86%	4%	ns	7%	90%	3%	ns	ns
Blisters or Calluses	8%	85%	7%	ns	10%	79%	11%	ns	7%	89%	5%	ns	ns

Neither the within-groups nor between groups statistical analysis detected a significant difference in the distribution of intensity change for any of the problems evaluated. This table is difficult to assess with three groups due to the sheer volume of numbers presented. It seems that whatever problem someone was experiencing at the midpoint, they tended to be at the same level at the end of the evaluation. The remainder tend to split evenly between those who note an increase in a problem's intensity and those who note a decrease. It is possible that foot problems did not have an adequate time to either develop or change given the duration of the test. The timeframe reflected here is extremely short for foot problems – it was only three days from the midpoint to the end of the evaluation. The presence of problems as displayed in Table 18, coupled with some of the other measures remaining to be discussed, may be more appropriate in terms of evaluating the performance of the antimicrobial socks.

During the course of the evaluation some Soldiers in each group reported that they developed a foot problem that they would not normally have. Soldiers in the untreated sock group reported a slightly higher percentage of these types of problems (10%, n=6 out of 58) than did Soldiers with either sock B (2%, n=1 out of 52) or sock C (3%, n=2 out of 61). Problems in the untreated sock group were identified

as a rash (n=2), itching (n=1), irritation (n=1), blisters (n=1), or athlete's foot (n=1) which occurred all over the foot (n=4), at the toes (n=2), or on the ankles (n=1). Three Soldiers in this group reported taking some action against these problems, noting that they used foot powder (n=1), cleaned their feet more frequently (n=1), or used alcohol wipes and antibacterial gel (n=1). Only one Soldier in each of the treated sock groups identified a problem: itching ankles with sock B and blisters with sock C. Neither of these respondents noted taking corrective action. While a Chi-square test did not detect a significant difference for these results it does represent an interesting anecdotal finding in favor of antimicrobial socks.

A number of Soldiers in each group reported that they saw a reduction in chronic foot problems when using the test socks. This ranged from 18% (n=10 out of 56) for the untreated sock to 29% (n=15 out of 52) and 30% (n=17 out of 57) for socks B and C, respectively. Soldiers in the untreated group reported less athlete's foot (n=4), discomfort (n=1), and excessive sweating (n=1). Sock B users noted either a general improvement in overall foot condition (n=3) or less athletes foot (n=2), odor (n=2), blisters (n=2), and excessive sweating (n=2). Soldiers who used sock C a reduction in athlete's foot (n=4), odor (n=3), irritation (n=1), and blisters (n=1). These differences were not statistically significant, but it is interesting that 50% more Soldiers in the treated sock groups reported a reduction in chronic foot problems over the untreated sock users.

Only one Soldier in each group reported that they had to consult medical personnel during the evaluation for a foot problem: a sock A user noted that this was because of "redness/sore", a sock B user "broke my toe - no time lost," and the sock C user did not specify what the problem was.

Comparison

Soldiers compared the problems that they experienced during the evaluation to what they would expect from wearing the standard socks under similar conditions. A "not applicable" option was provided for Soldiers who did not experience a certain problem, so the number of respondents for each criteria varies to some extent. The scale used and results obtained are presented below.

Table 21
Problems and Conditions Compared: This Exercise With Past Experience
(n=172)

Problems on this exercise have been…

MUCH WORSE	MODERATELY WORSE	SLIGHTLY WORSE	ABOUT THE SAME	SLIGHTLY BETTER	MODERATELY BETTER	MUCH BETTER
1	2	3	4	5	6	7

Comparison: Problems on this exercise w/these socks and past exercises with the std. sock	**Untreated - A (n=59)**	**Treated - B (n=52)**	**Treated - C (n=61)**	**n, F, p**
Foot odor	5.4	5.4	5.6	155, F=0.28, ns
General discomfort	5.3	5.4	5.4	135, F=0.05, ns
Itching Feet	4.7	5.5	5.0	115, F=2.47, ns*
Athlete's Foot	4.8	5.1	5.2	92, F=0.61, ns
Toe Nail Fungus	4.9	5.2	5.1	82, F=0.29, ns
Skin rash/irritation	4.8	5.4	4.8	84, F=1.79, ns
Skin inflammation/redness	4.6	5.6	4.9	83, F=3.10, p<.05
Blisters or Calluses	4.8	5.6	5.1	92, F=2.50, ns*

* No overall significant difference, but paired post-hoc comparisons did detect a significant difference between two groups. In both instances (itching feet and blisters or calluses), sock B was found to be rated significantly higher than sock A. This is also true for skin inflammation, where an overall significant difference was detected.

In general, Soldiers felt that the socks they evaluated had a beneficial impact on the problems listed above compared to their past experiences. For most, these past experiences would involve the black standard issue wool sock (see Table 3). As noted above in the table footnote, the post-hoc procedure identified three instances where sock B was rated significantly higher than the untreated sock: for athlete's foot, skin inflammation, and blisters.

It is interesting that these differences are only hinted at in Tables 18 and 19 (see above), yet they are so strong here. The source question for Table 21 is measuring something different than the previous tables – past compared to present experience. The magnitude of the differences, their statistical significance, and the fact that this was a blind study make it clear that there was some beneficial impact of one of the anti-microbial treatments in these areas. It is also important to note that sock C was rated higher than the untreated sock for all criteria, and higher than sock B for certain criteria (e.g. foot odor

and athlete's foot), but not by enough of a margin to satisfactorily account for chance for any individual problem. Overall, these socks reinforce the notion that the antimicrobial treated items did offer a benefit over the untreated items.

Soldiers were asked if they felt that the socks they were issued for the evaluation felt hotter than the standard black wool sock or the standard green cotton sock. Overall, 13% (n=21 out of 159) felt that the test socks were hotter than the black wool sock and 11% (n=16 out of 140) felt that the test socks were hotter than the standard green sock. Few comments were received on this question and there was only one comment made by multiple respondents: the test socks felt hotter than the standard wool sock because they are higher on the calf (n=3). There were no significant differences between any of the sock groups for this data.

Impact

We asked Soldiers if they felt that the socks they were issued were noticeably decreasing their foot odor as well as the foot odor of others during the exercise. This question was included on both the midpoint and final questionnaires. The results obtained at each data collection point are presented below in Table 22.

Table 22
Are the Socks Noticeably Decreasing Foot Odor? Midpoint and Final
(n=172)

	Untreated - A	Treated - B	Treated - C	
Midpoint: Your foot odor?	64% (38/59)	59% (30/51)	82% (50/61)	X^2=7.85, p<.05
Midpoint: Other Soldier's foot odor?	59% (33/56)	49% (25/51)	67% (38/57)	X^2=3.46, ns

	Untreated - A	Treated - B	Treated - C	
Final: Your foot odor?	73% (43/59)	66% (33/50)	76% (45/59)	X^2=1.45, ns
Final: Other Soldier's foot odor?	65% (37/57)	63% (30/48)	70% (40/57)	X^2=0.74, ns

At the midpoint, a significantly higher percentage of Soldiers in the sock C group felt that the socks were controlling their foot odor when compared to the other groups. This difference disappeared at the final, mainly because the other groups "caught up" with group C. We noted in the uniform and T-shirt

sections that a large percentage of Soldiers with untreated items were responding positively to these questions. This was also the case here. It is hard to know if this is because of some property of the sock in general or if it is due to a "placebo effect." It may be that these socks, regardless of treatment level, retained less odor than the type of socks that Soldiers are used to wearing. It may also be that Soldiers in the untreated group assumed that their socks were treated and that their feet did not smell as bad as usual. It could also be a combination of both of these factors. It is obvious that Soldiers believe the socks reduce odor, it is just hard to know if the treatment level had anything to do with it.

The majority of the Soldiers felt that the socks they were issued either had a positive impact (67%%, n=113 out of 169) or no impact (31%, n=53 out of 169) on their performance in the field. Only 2% (n=3 out of 169) felt that they had a negative impact. While their were no significant differences by sock type, it is interesting to note that nearly three-fourths of the Soldiers with sock B (73%, n=38 out of 52) felt that they had a positive impact on field performance compared to two-thirds for the untreated and sock C groups (64%, n=37 out of 58; 64%, n=38 out of 59, respectively).

Overall, 86% (n=146 out of 170) felt that the socks were comfortable to wear for an extended period of time, with no significant differences detected by sock type. There did not seem to be much of a trend in the comments from Soldiers who did not feel the socks were comfortable for extended wear. Untreated sock users noted that they were "itchy" (n=1), they "wear out" (n=1), and that "socks should NOT be worn for an extended period of time" (n=1). Similar comments were received from the sock B group: "you have to change your socks" (n=2), they "did not last" (n=1), and "they were too tight around my calves" (n=1). The C group noted that they were "just not comfortable" (n=1), they "start to smell" (n=1), and "nothing is comfortable to wear for an extended period of time" (n=1).

Nearly all of the Soldiers (94%, n=158 out of 169) felt that the socks were safe to wear regardless of type. The results for the three sock groups were similar and a significant difference was not detected. There were only two negative comments received in this area: one untreated sock user felt that they were too hot and one sock C user felt that the socks caused a rash. Positive comments were that no problems were experienced at all (n=11) and that the socks reduce foot problems (n=7).

Acceptability

On the final questionnaire, Soldiers rated their overall satisfaction level with the socks they were issued on a variety of criteria. The data was analyzed by treatment group. The scale used and results obtained are presented below in Table 23.

Table 23
Sock Satisfaction Ratings
(n=172)

VERY DISSATISFIED 1	MODERATELY DISSATISFIED 2	SOMEWHAT DISSATISFIED 3	NEITHER DISSATISFIED NOR SATISFIED 4	SOMEWHAT SATISFIED 5	MODERATELY SATISFIED 6	VERY SATISFIED 7

Acceptability Between –groups	**Untreated - A (n=59)**	**Treated - B (n=52)**	**Treated - C (n=61)**	**F, p**
Comfort of the socks – start	5.7	5.9	5.7	F=0.46, ns
Comfort of the socks – end	5.6	5.7	5.8	F=0.14, ns
Overall comfort	5.6	5.7	5.7	F=0.05, ns
Ability to reduce foot odor	5.5	5.6	5.5	F=0.10, ns
Ability to reduce foot problems	5.4	5.4	5.3	F=0.13, ns
Overall performance	5.6	5.8	5.6	F=0.19, ns

All of the socks received highly positive ratings for all of the criteria evaluated and no significant differences were detected by sock group. This table illustrates a point that we have not been able to make until now: that the socks were well received regardless of whether they were treated or not. The treated socks did slightly better than the untreated socks, but the point is clear – the socks by themselves are a highly acceptable item. In fact, practical experience shows that these ratings approach the higher end of the range in terms of acceptability for clothing and equipment items as measured by this type of scale. Ultimately, the solid performance of the sock in general may help to obscure an incremental improvement like antimicrobial protection.

PREFERENCE

Background

All of the final questionnaires featured a last question assessing overall Soldier opinion on the future of antimicrobial treatments for the items that they evaluated. At this point, it was still a blind study so the results obtained were not based on the specific knowledge that they had used either a treated or control item. While the question was phrased specifically towards the item they had just evaluated (e.g., the uniform or T-Shirt), the results were surprisingly similar regardless of the item evaluated or its treatment level. Therefore, it seemed like it would be a good gauge of Soldier opinion on antimicrobial clothing technologies in general. We calculated the results for this question based on membership in the uniform and T-Shirt sub-groups (n=195). Each Soldier was classified into two new groups based on the nature of the primary item that they evaluated – treated uniform and treated T-shirt (n=103) or untreated uniform and untreated T-shirt (n=92).

Result

Soldiers were given the opportunity on the final questionnaire to state their opinion as to the future of antimicrobial clothing treatments specific to the items that they had just evaluated. The data was limited to the Soldiers responding for the primary evaluation items (uniform and T-shirts) and divided into two overall groups based on the treatment level of the item received (treated or untreated). A Chi-square test conducted on this data did not detect a significant difference. Despite this, the results are presented below in Table 24 by group and overall since the outcome was interesting.

Table 24
What Should Be Done With Anti-microbial Uniform or T-Shirt Treatments?
(n=188)

Option	Treated (n=100)	Untreated (n=88)	Overall (n=188)
Reject them	5%	7%	6%
Adopt them as standard	34%	39%	36%
Make them optional purchase items	37%	33%	35%
Do more research	24%	21%	23%

As can be seen above, there seems to be broad support for antimicrobial treatments regardless of whether or not the Soldier evaluated a treated item or not. Nearly three-fourths of the group felt that they should either be adopted or made available as optional purchase item. About one-fourth felt that more research should be done along these lines and only 6% felt that the idea should be rejected. The

background questionnaire revealed that three-fourths of the total survey group (75%, n=156 out of 206) are routinely using some form of anti-bacterial medication or hygiene item in the field. We would argue that this represents a broad based interest and support amongst Soldiers for military applications of antimicrobial clothing treatments.

DISCUSSION

The goal of the evaluation was to determine if the protection provided by an antimicrobial clothing treatment would manifest itself in ways that would be noticeable and beneficial to Soldiers. All data was collected through subjective questionnaires completed by the participants. While Soldiers knew they were participating in an evaluation of antimicrobial treatments, no one knew for certain if the items they were issued to evaluate were treated or not. Effectiveness of the treatments was measured through a range of questions which assessed relevant problems and conditions encountered by the participants during the exercise, their intensity, and how they compared to past experiences, along with perceptions of comfort, odor reduction, and performance. It is critical to keep in mind that we were looking for perceptible benefits of the use of this type of treatment. Soldiers might be completely unaware of the primary benefit: protection from harmful microbes which could cause illness and render a Soldier ineffective and unable to complete his mission. While we feel that the results of the field evaluation do make a case in favor of antimicrobial technologies, it is critical to evaluate them alongside laboratory and technical data to gain a complete picture of the performance and benefits of the treatments.

Based on the results of this evaluation, it would appear that the T-Shirt is a promising candidate for application of an antimicrobial treatment. Soldiers who used the treated T-Shirt reported a significant reduction in odor as well as an overall reduction in other problems, which included general discomfort, heat rash, itching skin, and skin rash. A decrease in intensity of these problems was also noted across the board as experienced on this exercise and when compared to past exercises. Soldiers also rated the comfort and performance of the treated T-shirt higher than the untreated T-Shirt and a higher percentage felt that the it was comfortable to wear for an extended period when compared to the untreated item. This seems to indicate that the overall acceptability of the item could benefit from the use of an antimicrobial treatment beyond the reduction of common skin problems. While few individual problems met the requirements of statistical significance, collectively the trend is quite impressive.

Two types of antimicrobial socks were evaluated: one featured the standard or current antimicrobial treatment (type B) and one received a candidate antimicrobial treatment (type C). We feel that the overall strong performance of the sock hampered our ability to find a stronger trend than we did

in favor of the treated, it was clear that Soldiers felt the sock itself was a highly acceptable item. However, the antimicrobial socks showed an overall reduction in the percentage of Soldiers who reported foot odor, general discomfort, and itching. A minor reduction was noted in the percentage of Soldiers experiencing athlete's foot. In terms of comparison to problems encountered on previous exercises, the B sock was rated significantly better for itching feet, blisters, and skin inflammation. Collectively, it was also rated better than the standard for all other problems in this area. The type C sock also received better ratings than the standard in this context, but a significant improvement was not noted in any specific area. A significantly higher percentage of sock C users did feel that the sock was controlling their foot odor at the midpoint than did Soldiers in the control or sock B group. Our overall concern with this data is that the evaluation was not long enough to get an accurate measure of the treatment impact on foot problems. Past experience shows that the cycle of these problems is better measured in weeks compared to days. Overall, it would seem that an antimicrobial treatment offers benefits in terms of sock performance. While both candidate treatments performed well, the field evaluation suggests that the type B sock offered more in the way of perceptible benefits.

The results for the antimicrobial treated uniform were not as promising as those for the T-shirt and sock. We did not see a reduction in problems reported or their intensity on either an individual or collective basis. We also did not see a difference in ratings comparing experiences with the treated uniform to past experience with an untreated uniform. However, significantly more Soldiers felt that the treated uniform was controlling their body odor at the end of the evaluation. We also noted a collective increase in acceptability ratings for the treated uniform, so it is still possible to see a minimal benefit from the antimicrobial treatment as it was evaluated. We had considered some possible explanations for this in the main body of the report, from a "placebo" effect to the fact that the items were new for the Soldiers. However, it could be argued that these were factors for the T-Shirt and sock and we were still able to detect differences and trends in favor of the treated versions. The key difference might be that these were "next to the skin" items and may have provided a greater observable benefit than treating an outer layer of clothing. Soldiers in the uniform groups did not receive antimicrobial treated undergarments. They were wearing an untreated "next to the skin" layer that consisted of untreated cotton T-Shirts and underwear. We feel that this may have somewhat reduced the ability of the participants to detect a benefit from the treatment in a short-term trial.

While not immediately apparent, there is a series of anecdotal evidence that lends some weight to the argument in favor of the antimicrobial treatments. None of these could stand on their own, but in the light of the collective findings they are interesting. Three Soldiers in the treated uniform group reported

that they did not experience diarrhea in the field when they normally do compared to one in the untreated group. One Soldier in the untreated uniform group reported a "staph" infection compared to none for the treated group. Six Soldiers in the treated uniform group commented that they felt cleaner than usual in the field compared to none for the untreated group. None of the respondents in the treated T-Shirt group reported that they developed a problem that they would not normally have compared to 13% in the untreated group. A report of rashes as a safety concern in the untreated T-Shirt group which was not noted for the treated group. This corresponded with a lower percentage of Soldiers perceiving the untreated item as "safe" when compared to the treated item. Fifty percent more Soldiers in the treated sock groups reported a reduction in chronic foot problems when compared to Soldiers in the untreated groups.

Three-fourths of our evaluation participants reported that they buy and use products branded as "antimicrobial" for use in the field. Furthermore, more than two-thirds of the survey group urged the adoption of antimicrobial treated clothing items either as standard or optional purchase items and one-fourth felt that the treatments should continue to be researched. Only 6% felt that there was no merit to the technology. This opinion was held by both Soldiers who used the treated and untreated items. It is clear that Soldiers in general are interested in antimicrobial products. They endorse the use of these treatments on military clothing items. The fact that Soldiers in the untreated group felt the exact same way as those in the treated group also seems to indicate that they recognize that the benefits of these types of treatments may not always be apparent. All of this, when taken together, offers a clear basis for further consideration of the antimicrobial technologies evaluated.

Attachment A: Antimicrobial Field Uniform Background Questionnaire

User Name:_____ Evaluation ID Number:_____

Please answer the following questions based on your total experience in the military. Your answers will remain confidential. Thank you in advance for your participation!

Rank? E-____ O-____ WO-____ MOS, Branch, or Specialty?_____

Unit: _____ Battery: _____ Age? ____ years

Time in the military? _____ years _____ months

1. What type of uniform, t-shirt, and socks would you normally wear in the field this time of year? Circle one answer for each.

UNIFORM	**T-SHIRTS**	**SOCKS**
a. Hot Weather BDU	a. Standard Cotton	a. Standard Wool (black)
b. Regular BDU	b. Standard Polyester	b. Standard Cotton (green)
c. Other (specify below)	c. Other (specify below)	c. Other (specify below)
_____	_____	_____

2. Have you ever been diagnosed with any chronic skin problems like eczema, hyperhidrosis (excessive sweating), allergies to certain fibers (i.e. wool, etc.), and allergies to certain chemicals or substances that result in a rash or other skin reaction?

YES NO

If "YES," describe the problem.
Note: do not answer this part if you have privacy concerns.

3. In the field, do you routinely use any kind of antibacterial cream, lotion, liquid, or spray (i.e. first aid cream, deodorant, antibacterial soap, medicated powder, etc.) under areas covered by the uniform?

YES NO

If YES, list the items that you use.

4. In the normal course of your duties in the field do you come in contact with anything that might cause a skin reaction?

YES NO

If YES, what do you come in contact with? Circle one answer for each.

a. Irritants (insect repellent, camo face paint, etc.)? YES NO

b. Caustic substances (gasoline, battery acid, etc.)? YES NO

c. Biting insects (fleas, spiders, etc.)? YES NO

d. Other (poisonous plants, poisonous insects, etc.)? YES NO

If YES, specify what you come in contact with and any reaction you might have.

5. Do you feel that wearing the standard BDU causes you to develop physical problems (i.e. rashes, irritation, etc.) that you would not normally have?

YES NO

If NO, skip to question 6.
If YES, what type of problem have you had?

How much of your body is affected? Fill in one.

a. Specific areas (where:_____)

b. All over (wherever the uniform touched)

c. Other (specify:_____)

How many days does the problem last? _____ days

List any action that you take to address the problem:

6. Has wearing either of the following caused you to develop physical problems (i.e. rashes, irritation, etc.) that you would not normally have? Circle one answer for each.

a. Standard socks	YES	NO
b. Standard T-shirts	YES	NO

If YES, what type of problem have you had?

7. During a typical seven-day field exercise, to what extent do you experience the following under the standard BDU? Circle one answer for each. Circle "N/A" if you do not experience a certain problem.

To what extent did you experience any of the following during the exercise?	**N/A**	**Mildly**	**Moderately**	**Severely**
a. Body odor	0	1	2	3
b. General discomfort	0	1	2	3
c. Heat Rash	0	1	2	3
d. Itching skin	0	1	2	3
e. Chafing	0	1	2	3
f. Skin rash/irritation	0	1	2	3
g. Skin inflammation/redness	0	1	2	3
h. Skin lesions or sores	0	1	2	3
i. Acne/pimples	0	1	2	3
j. Infected or inflamed hair follicles	0	1	2	3
k. Infected cuts or scrapes	0	1	2	3
l. Other (list below)	0	1	2	3

Comments?

8. During a typical seven-day field exercise, to what extent do you experience the following **foot** problems? Circle one answer for each. Circle "N/A" if you do not experience a certain problem.

To what extent did you experience any of the following during the exercise?		**N/A**	**Mildly**	**Moderately**	**Severely**
a.	Foot odor	0	1	2	3
b.	General discomfort	0	1	2	3
c.	Itching feet	0	1	2	3
d.	Athletes Foot	0	1	2	3
e.	Toe nail fungus	0	1	2	3
f.	Skin rash/irritation	0	1	2	3
g.	Skin inflammation/redness	0	1	2	3
h.	Blisters or calluses	0	1	2	3
i.	Other (list below)	0	1	2	3

Comments?

9. Are there any other problems that you experience during a typical seven-day field exercise that could be related to bacteria or other microbes (eye infection, diarrhea, etc.)?

YES NO

If YES, describe the problem and how often you have it.

Attachment B: Antimicrobial Field Uniform Midpoint Questionnaire

Last Name:_____ Evaluation ID Number:_____

Please answer all of the questions based on your experience during this test period (17 August 05 – 20 August 05) while wearing the test uniform that you were issued. Your responses to the following questions will influence decisions on this item, so please consider each question carefully before answering. Your answers will remain confidential. Thank you in advance for your participation!

Rank? E-____ O-____ WO-____ Unit: _____ Battery: _____

1. Were you issued a uniform to evaluate? YES NO

   b. If YES, did you wear it? YES NO

   If NO, why not? Be specific.

IF YOU ANSWERED NO TO EITHER 1a or 1b, HAND IN THIS QUESTIONNAIRE.

2. How many total days did you wear the uniform since it was issued to you? _____ days

   How many hours per day did you usually wear the uniform? ____ hours per day

3. Have you had any durability problems with the uniform so far? YES NO

   If YES, explain.

4. During the time you were wearing the test uniform did you use any kind of antibacterial cream, lotion, liquid, or spray (i.e. first aid cream, deodorant, antibacterial soap, medicated powder, etc.)?

   YES NO

   If YES, what have you used?

5. While wearing the uniform did you come in contact with anything that might have caused a skin reaction?

YES NO

If YES, what did you come in contact with and how did it affect you?

6. Up to this point have you experienced any of the following under the uniform? In part a, circle one answer for each. Fill in the circle for "N/A" if you did not experience a certain problem. In part b, indicate if you would normally expect to have each problem after three days in the field. Circle an answer in part b even if you are not having that problem now.

a. So far, to what extent are you experiencing the following?	**N/A**	**Mildly**	**Moderately**	**Severely**	**b. Would you normally expect to experience these problems after three days in the field?**
a. Body odor	0	1	2	3	YES NO
b. General discomfort	0	1	2	3	YES NO
c. Heat Rash	0	1	2	3	YES NO
d. Itching skin	0	1	2	3	YES NO
e. Chafing	0	1	2	3	YES NO
f. Skin rash/irritation	0	1	2	3	YES NO
g. Skin inflammation/redness	0	1	2	3	YES NO
h. Skin lesions or sores	0	1	2	3	YES NO
i. Acne/pimples	0	1	2	3	YES NO
j. Infected or inflamed hair follicles	0	1	2	3	YES NO
k. Infected cuts or scrapes	0	1	2	3	YES NO
l. Other (list below)	0	1	2	3	YES NO

Comments?

7. Overall, do you feel that the uniform is noticeably decreasing (circle one answer for each.)….

a. your body odor?	YES	NO
b. other soldiers body odor?	YES	NO

Explain your answer.

8. Overall, how do the physical problems (with body odor, skin, etc.) that you are experiencing on THIS FIELD EXERCISE compare to what you would normally expect when wearing the standard BDU in the field? Fill in one circle.

Problems on this exercise have been…

MUCH WORSE	MODERATELY WORSE	SLIGHTLY WORSE	ABOUT THE SAME	SLIGHTLY BETTER	MODERATELY BETTER	MUCH BETTER
1	2	3	4	5	6	7

9. At any time during this evaluation did you have to consult medical personnel for a problem related to the skin or any type of bacterial or fungal infection?

YES NO

If YES, list the problem, the level of treatment you received (field, outpatient, etc.) and any duty time lost.
NOTE: YOU DO NOT HAVE TO ANSWER THIS PART IF YOU HAVE PRIVACY CONCERNS!

10. Does this uniform feel hotter than standard BDUs do under similar circumstances? Circle one answer for each. Circle "N/A" if you have never worn a certain uniform.

Does the test uniform feel hotter than…			
a. Hot Weather BDU (woodland, rip-stop)?	YES	NO	N/A
b. Regular BDU (woodland)	YES	NO	N/A
c. Desert BDU?	YES	NO	N/A

If YES, explain.

11. Please rate how satisfied or dissatisfied you are with the following aspects of the uniform?. Circle one answer for each.

	VERY DISSATISFIED 1	MODERATELY DISSATISFIED 2	SOMEWHAT DISSATISFIED 3	NEITHER DISSATISFIED NOR SATISFIED 4	SOMEWHAT SATISFIED 5	MODERATELY SATISFIED 6	VERY SATISFIED 7
a. Comfort of the uniform (at the beginning of the exercise)	1	2	3	4	5	6	7
b. Comfort of the uniform (after three days in the field)	1	2	3	4	5	6	7
c. Overall comfort	1	2	3	4	5	6	7
d. Ability to reduce body odor	1	2	3	4	5	6	7
e. Ability to reduce skin problems	1	2	3	4	5	6	7
f. Overall performance	1	2	3	4	5	6	7

12. Do you have any other comments on the uniform?

Attachment C: Antimicrobial Field Uniform Final Questionnaire

Last Name:_____ Evaluation ID Number:_____

Please answer all of the questions based on your experience during this test period (17 August 05 – 24 August 05) while wearing the test uniform that you were issued. Your responses to the following questions will influence decisions on this item, so please consider each question carefully before answering. Your answers will remain confidential. Thank you in advance for your participation!

Rank? E-____ O-____ WO-____ Unit: _____ Battery: _____

1a. Were you issued a uniform to evaluate? YES NO

b. If YES, did you wear it? YES NO

If NO, why not? Be specific.

IF YOU ANSWERED NO TO EITHER 1a or 1b, HAND IN THIS QUESTIONNAIRE.

2. How many total days did you wear the uniform since it was issued to you? _____ days

How many hours per day did you usually wear the uniform? ____ hours per day

3. Have you had any durability problems with the uniform so far?

YES NO

If YES, explain.

4. Have you had to stop wearing any of the uniforms or components (i.e. shirt or pants) for any reason?

YES NO

If YES, explain.

5. Did you launder, wash or clean the test uniform in any way during the evaluation? YES NO

If YES, how many times? _____

6. What type of T-shirt did you usually wear under the uniform? Fill in one circle. In the space to the right, write in how many clean t-shirts of each type you brought with you to the field.

a. Standard Brown T-shirt	how many? _____
b. Commercial cotton T-shirt (list below)	how many? _____
c. Commercial Polyester T-shirt (list below)	how many? _____
d. Other (list below)	how many? _____

How often did you change T-shirts during the evaluation period?

Every _____ days or _____ times a day

7. What type of underwear did you usually wear under the uniform? Fill in one circle. In the space to the right, write in how many clean pairs of each type you brought with you to the field.

a. Standard cotton underwear	how many? _____
b. Other (list below)	how many? _____
c. None	

How often did you change underwear during the evaluation period?

Every _____ days or _____ times a day

8. Did you wear any additional undergarments under the uniform? YES NO

If YES, list the type, the number of days that you wore them, and how frequently you changed them.

9. During the time you were wearing the test uniform did you use any kind of antibacterial cream, lotion, liquid, or spray (i.e. first aid cream, deodorant, antibacterial soap, medicated powder, etc.) under areas covered by the uniform?

YES NO

If YES, list the type (include brand name if known), the number of times you used it, and where.

TYPE/BRAND NAME	TIMES USED	WHERE (ON THE BODY)

10. While wearing the uniform did you come in contact with anything that might have caused a skin reaction?

YES NO

If YES, what did you come in contact with? Circle one answer for each.

a. Irritants (insect repellent, camo face paint, etc.)?	YES	NO
b. Caustic substances (gasoline, battery acid, etc.)?	YES	NO
c. Biting insects (fleas, spiders, etc.)?	YES	NO
d. Other (poisonous plants, poisonous insects, etc.)?	YES	NO

If YES, specify what you came in contact with and any reaction you might have had.

11. Did you experience any of the following under the uniform? Circle one answer for each. Circle "N/A" if you did not experience a certain problem. If you did experience a certain problem, use the space at the right to indicate how many days you had it during the exercise (maximum = 7 days).

So far, to what extent are you experiencing the following?	**N/A**	**Mildly**	**Moderately**	**Severely**	**If you answered 1,2, or 3… How many days did you experience it during the exercise? (max=7 days)**
a. Body odor	0	1	2	3	_____ Days
b. General discomfort	0	1	2	3	_____ Days
c. Heat Rash	0	1	2	3	_____ Days
d. Itching skin	0	1	2	3	_____ Days
e. Chafing	0	1	2	3	_____ Days
f. Skin rash/irritation	0	1	2	3	_____ Days
g. Skin inflammation/redness	0	1	2	3	_____ Days
h. Skin lesions or sores	0	1	2	3	_____ Days
i. Acne/pimples	0	1	2	3	_____ Days
j. Infected or inflamed hair follicles	0	1	2	3	_____ Days
k. Infected cuts or scrapes	0	1	2	3	_____ Days
l. Other (list on next page)	0	1	2	3	_____ Days

Comments?

12. During the course of the evaluation did you develop any physical problems (i.e. rashes, irritation, etc.) that you do not normally have?

YES NO

If NO, skip to question 13.
If YES, what type of problem did you have?

How much of your body was affected? Fill in one.

a. Specific areas (where:_____)
b. All over (wherever the uniform touched)
c. Other (specify:_____)

List any action that you took to address the problem:

13. Overall, did you see any reduction in any chronic skin conditions that you have been diagnosed with? This would include skin problems like eczema, hyperhidrosis (excessive sweating), allergies to certain fibers (i.e. wool, etc.), and allergies to certain chemicals or substances that result in a rash or other skin reaction. Circle "N/A" if you do not have a history of these types of problems.

YES NO N/A

If "YES," explain.
Note: DO NOT ANSWER THIS PART IF YOU HAVE PRIVACY CONCERNS.

14. Did you see a reduction in any other problems that you normally experience which could be related to bacteria or other microbes (eye infection, diarrhea, etc.)?

YES NO

If YES, explain.

15. Overall, do you feel that the uniform is noticeably decreasing (circle one answer for each)….

a. your body odor?	YES	NO
b. other soldiers' body odor?	YES	NO

Explain your answer.

16. At any time during this evaluation did you have to consult medical personnel for a problem related to the skin or any type of bacterial or fungal infection?

YES NO

If YES, list the problem, the level of treatment you received (field, outpatient, etc.) and any duty time lost.
NOTE: YOU DO NOT HAVE TO ANSWER THIS PART IF YOU HAVE PRIVACY CONCERNS!

17. What impact did the test uniform have on your performance in the field? Fill in one circle.

a. positive impact (improved performance)
b. no impact
c. negative impact (decreased performance)

Explain your answer.

18. Does this uniform feel hotter than standard BDUs do under similar circumstances? Circle one answer for each. Circle "N/A" if you have never worn a certain uniform.

Does the test uniform feel hotter than…			
a. Hot Weather BDU (woodland, rip-stop)?	YES	NO	N/A
b. Regular BDU (woodland)	YES	NO	N/A
c. Desert BDU?	YES	NO	N/A

If YES, explain.

19. Overall, how did the problems that you experienced on THIS FIELD EXERCISE compare to what you would normally expect when wearing the standard BDU? Circle one answer for each. Circle "N/A" if you have never experienced a certain problem.

Problems on this exercise have been…

MUCH WORSE 1	MODERATELY WORSE 2	SLIGHTLY WORSE 3	ABOUT THE SAME 4	SLIGHTLY BETTER 5	MODERATELY BETTER 6	MUCH BETTER 7

a.	Body odor	**N/A**	1	2	3	4	5	6	7
b.	General discomfort	**N/A**	1	2	3	4	5	6	7
c.	Heat Rash	**N/A**	1	2	3	4	5	6	7
d.	Itching skin	**N/A**	1	2	3	4	5	6	7
e.	Chafing	**N/A**	1	2	3	4	5	6	7
f.	Skin rash/irritation	**N/A**	1	2	3	4	5	6	7
g.	Skin inflammation/redness	**N/A**	1	2	3	4	5	6	7
h.	Skin lesions or sores	**N/A**	1	2	3	4	5	6	7
i.	Acne/pimples	**N/A**	1	2	3	4	5	6	7
j.	Infected or inflamed hair follicles	**N/A**	1	2	3	4	5	6	7
k.	Infected cuts or scrapes	**N/A**	1	2	3	4	5	6	7
l.	Other (list below)	**N/A**	1	2	3	4	5	6	7
m.	Overall	**N/A**	1	2	3	4	5	6	7

Comments?

20. Overall, do you feel that the uniform is comfortable for wearing over an extended period of time?

YES NO

Explain your answer.

21. Overall, do you feel that the uniform is safe to wear? YES NO

Explain your answer.

22. Please rate how satisfied or dissatisfied you are with the following aspects of the uniform. Circle one answer for each.

	VERY DISSATISFIED 1	MODERATELY DISSATISFIED 2	SOMEWHAT DISSATISFIED 3	NEITHER DISSATISFIED NOR SATISFIED 4	SOMEWHAT SATISFIED 5	MODERATELY SATISFIED 6	VERY SATISFIED 7
a. Comfort of the uniform (at the beginning of the exercise)	1	2	3	4	5	6	7
b. Comfort of the uniform (at the end of the exercise)	1	2	3	4	5	6	7
c. Overall comfort	1	2	3	4	5	6	7
d. Ability to reduce body odor	1	2	3	4	5	6	7
e. Ability to reduce skin problems	1	2	3	4	5	6	7
f. Overall performance	1	2	3	4	5	6	7

23. What would your decision be on the future of anti-microbial treated uniforms? Fill in one circle.

a. Reject them.
b. Adopt them as standard.
c. Make them available as an optional purchase item.
d. Do more research on them.

Explain your answer.

Attachment D: Antimicrobial T-shirt Midpoint Questionnaire

Last Name:_____ Evaluation ID Number:_____

Please answer all of the questions based on your experience during this test period (17 August 05 – 20 August 05) while wearing the test t-shirt that you were issued. Your responses to the following questions will influence decisions on this item, so please consider each question carefully before answering. Your answers will remain confidential. Thank you in advance for your participation!

Rank? E-____ O-____ WO-____ Unit: _____ Battery: _____

1a. Were you issued T-shirts to evaluate? YES NO

b. If YES, have you worn them? YES NO

If NO, why not? Be specific.

IF YOU ANSWERED NO TO EITHER 1a or 1b, HAND IN THIS QUESTIONNAIRE.

2. How many total days have you worn the T-shirts since they were issued to you? _____ days

How many hours per day did you usually wear the T-shirts? ____ hours per day

How often have you changed T-shirts so far? Every _____ days or _____ times a day

3. Have you worn a T-shirt other than those that were issued for the evaluation? YES NO

If YES, list the type of T-shirt and the number of days you wore it.

4. Have you had any durability problems with the T-shirts so far? YES NO

If YES, explain.

5. During the time you were wearing the T-shirts, have you used any kind of antibacterial cream, lotion, liquid, or spray (i.e. first aid cream, deodorant, antibacterial soap, medicated powder, etc.)?

YES NO

If YES, what have you used?

6. While wearing the T-shirts have you come in contact with anything that might have caused a skin reaction?

YES NO

If YES, what did you come in contact with and how did it affect you?

7. Up to this point have you experienced any of the following under the T-shirts? In part a, circle one answer for each.. Fill in the circle for "N/A" if you did not experience a certain problem. In part b, indicate if you would normally expect to have each problem after three days in the field. Circle an answer in part b even if you are not having that problem now.

a. So far, to what extent are you experiencing the following?	**N/A**	**Mildly**	**Moderately**	**Severely**	**b. Would you normally expect to experience these problems after three days in the field?**
a. Body odor	0	1	2	3	YES NO
b. General discomfort	0	1	2	3	YES NO
c. Heat Rash	0	1	2	3	YES NO
d. Itching skin	0	1	2	3	YES NO
e. Chafing	0	1	2	3	YES NO
f. Skin rash/irritation	0	1	2	3	YES NO
g. Skin inflammation/redness	0	1	2	3	YES NO
h. Skin lesions or sores	0	1	2	3	YES NO
i. Acne/pimples	0	1	2	3	YES NO
j. Infected or inflamed hair follicles	0	1	2	3	YES NO
k. Infected cuts or scrapes	0	1	2	3	YES NO
l. Other (list below)	0	1	2	3	YES NO

8. Overall, do you feel that the T-shirts are noticeably decreasing (circle one answer for each)….

   a. your body odor? YES NO

   b. other soldiers' body odor? YES NO

   Explain your answer.

9. Overall, how do the physical problems (with body odor, skin, etc.) that you are experiencing on THIS FIELD EXERCISE compare to what you would normally expect when wearing the standard T-shirts in the field? Fill in one answer.

   Problems on this exercise have been…

MUCH WORSE	MODERATELY WORSE	SLIGHTLY WORSE	ABOUT THE SAME	SLIGHTLY BETTER	MODERATELY BETTER	MUCH BETTER
1	2	3	4	5	6	7

10. At any time during this evaluation did you have to consult medical personnel for a problem related to the skin or any type of bacterial or fungal infection?

    YES NO

    If YES, list the problem, the level of treatment you received (field, outpatient, etc.) and any duty time lost. **NOTE: YOU DO NOT HAVE TO ANSWER THIS PART IF YOU HAVE PRIVACY CONCERNS!**

11. Do the test T-shirts feel hotter than the standard cotton T-shirt?

    YES NO

    If YES, explain.

11. Please rate how satisfied or dissatisfied you are with the following aspects of the T-shirts. Circle one answer for each.

	VERY DISSATISFIED 1	MODERATELY DISSATISFIED 2	SOMEWHAT DISSATISFIED 3	NEITHER DISSATISFIED NOR SATISFIED 4	SOMEWHAT SATISFIED 5	MODERATELY SATISFIED 6	VERY SATISFIED 7
a. Comfort of the T-shirt (at the beginning of the exercise)	1	2	3	4	5	6	7
b. Comfort of the T-shirt (after three days in the field)	1	2	3	4	5	6	7
c. Overall comfort	1	2	3	4	5	6	7
d. Ability to reduce body odor	1	2	3	4	5	6	7
e. Ability to reduce skin problems	1	2	3	4	5	6	7
f. Overall performance	1	2	3	4	5	6	7

12. Do you have any other comments on the T-shirts?

Attachment E: Antimicrobial T-shirt Final Questionnaire

Last Name:_____ Evaluation ID Number:_____

Please answer all of the questions based on your experience during this test period (17 August 05 – 24 August 05) while wearing the test T-shirt that you were issued. Your responses to the following questions will influence decisions on this item, so please consider each question carefully before answering. Your answers will remain confidential. Thank you in advance for your participation!

Rank? E-____ O-____ WO-____ Unit: _____ Battery: _____

1a. Were you issued T-shirts to evaluate? YES NO

b. If YES, have you worn them? YES NO

If NO, why not? Be specific.

IF YOU ANSWERED NO TO EITHER 1a or 1b, HAND IN THIS QUESTIONNAIRE.

2. How many total days have you worn the test T-shirts since they were issued to you? _____ days

How many hours per day did you usually wear the test T-shirts? ____ hours per day

How often have you changed test T-shirts so far? Every _____ days or _____ times a day

3. Have you worn a T-shirt other than those that were issued for the evaluation? YES NO

If YES, list the type of T-shirt and the number of days you wore it.

4a. What type of uniform did you wear during this field training exercise? Fill in one circle. In the space to the right, write in how many clean uniforms you brought with you to the field.

a. Hot Weather BDU how many sets? _____
b. Regular BDU how many sets? _____
c. Other (specify below) how many sets? _____

4b. How many times did you change this uniform and put on a clean one during the exercise? _____ times

5. Have you had any durability problems with the test T-shirts so far? YES NO

   If YES, explain.

6. Did you launder, wash or clean the test T-shirts in any way during the evaluation? YES NO

   If YES, how many times? _____

7. Did you wear any additional undergarments either under or over the test T-shirts? YES NO

   If YES, list the type, the number of days that you wore them, and how frequently you changed them.

8. During the time you were wearing the test T-shirt did you use any kind of antibacterial cream, lotion, liquid, or spray (i.e. first aid cream, deodorant, antibacterial soap, medicated powder, etc.) under the T-shirt?

   YES NO

   If YES, list the type (include brand name if known), the number of times you used it, and where.

TYPE/BRAND NAME	TIMES USED	WHERE (ON THE BODY)

9. While wearing the test T-shirt did you come in contact with anything that might have caused a skin reaction?

   YES NO

   If YES, what did you come in contact with? Circle one answer for each.

   a. Irritants (insect repellent, camo face paint, etc.)? YES NO
   b. Caustic substances (gasoline, battery acid, etc.)? YES NO
   c. Biting insects (fleas, spiders, etc.)? YES NO
   d. Other (poisonous plants, poisonous insects, etc.)? YES NO

   If YES, specify what you came in contact with and any reaction you might have had.

10. Did you experience any of the following under the test T-shirt? Circle one answer for each. Circle "N/A" if you did not experience a certain problem. If you did experience a certain problem, use the space at the right to indicate how many days you had it during the exercise (maximum = 7 days).

So far, to what extent are you experiencing the following?	**N/A**	**Mildly**	**Moderately**	**Severely**	**If you answered 1,2, or 3… How many days did you experience it during the exercise? (max=7 days)**
a. Body odor	0	1	2	3	_____ Days
b. General discomfort	0	1	2	3	_____ Days
c. Heat Rash	0	1	2	3	_____ Days
d. Itching skin	0	1	2	3	_____ Days
e. Chafing	0	1	2	3	_____ Days
f. Skin rash/irritation	0	1	2	3	_____ Days
g. Skin inflammation/redness	0	1	2	3	_____ Days
h. Skin lesions or sores	0	1	2	3	_____ Days
i. Acne/pimples	0	1	2	3	_____ Days
j. Infected or inflamed hair follicles	0	1	2	3	_____ Days
k. Infected cuts or scrapes	0	1	2	3	_____ Days
l. Other (list on next page)	0	1	2	3	_____ Days

Comments?

11. During the course of the evaluation did you develop any physical problems (i.e. rashes, irritation, etc.) that you do not normally have?

YES NO

If NO, skip to question 12.
If YES, what type of problem did you have?

How much of your body was affected? Fill in one.

a. Specific areas (where:_____)
b. All over (wherever the test T-shirt touched)
c. Other (specify:_____)

List any action that you took to address the problem:

12. Overall, did you see any reduction in any chronic skin conditions that you have been diagnosed with? This would include skin problems like eczema, hyperhidrosis (excessive sweating), allergies to certain fibers (i.e. wool, etc.), and allergies to certain chemicals or substances that result in a rash or other skin reaction. Circle "N/A" if you do not have a history of these types of problems.

YES NO N/A

If "YES," explain.
Note: DO NOT ANSWER THIS PART IF YOU HAVE PRIVACY CONCERNS.

13. Did you see a reduction in any other problems that you normally experience which could be related to bacteria or other microbes (eye infection, diarrhea, etc.)?

YES NO

If YES, explain.

14. Overall, do you feel that the test T-shirt is noticeably decreasing (circle one answer for each)….

a. your body odor? YES NO

b. other soldiers' body odor? YES NO

Explain your answer.

15. At any time during this evaluation did you have to consult medical personnel for a problem related to the skin or any type of bacterial or fungal infection?

YES NO

If YES, list the problem, the level of treatment you received (field, outpatient, etc.) and any duty time lost.
NOTE: YOU DO NOT HAVE TO ANSWER THIS PART IF YOU HAVE PRIVACY CONCERNS!

16. Do the test T-shirts feel hotter than the standard cotton T-shirt? YES NO

If YES, explain.

17. Overall, how did the problems that you experienced on THIS FIELD EXERCISE compare to what you would normally expect when wearing the standard T-shirt? Circle one answer for each. Circle "N/A" if you have never experienced a certain problem.

Problems on this exercise have been…

MUCH WORSE 1	MODERATELY WORSE 2	SLIGHTLY WORSE 3	ABOUT THE SAME 4	SLIGHTLY BETTER 5	MODERATELY BETTER 6	MUCH BETTER 7

a.	Body odor	**N/A**	1	2	3	4	5	6	7
b.	General discomfort	**N/A**	1	2	3	4	5	6	7
c.	Heat Rash	**N/A**	1	2	3	4	5	6	7
d.	Itching skin	**N/A**	1	2	3	4	5	6	7
e.	Chafing	**N/A**	1	2	3	4	5	6	7
f.	Skin rash/irritation	**N/A**	1	2	3	4	5	6	7
g.	Skin inflammation/redness	**N/A**	1	2	3	4	5	6	7
h.	Skin lesions or sores	**N/A**	1	2	3	4	5	6	7
i.	Acne/pimples	**N/A**	1	2	3	4	5	6	7
j.	Infected or inflamed hair follicles	**N/A**	1	2	3	4	5	6	7
k.	Infected cuts or scrapes	**N/A**	1	2	3	4	5	6	7
l.	Other (list below)	**N/A**	1	2	3	4	5	6	7
m.	Overall	**N/A**	1	2	3	4	5	6	7

Comments?

18. What impact did the test T-shirt have on your performance in the field? Fill in one circle.

a. positive impact (improved performance)

b. no impact

c. negative impact (decreased performance)

Explain your answer.

19. Overall, do you feel that the test T-shirt is comfortable for wearing over an extended period of time?

YES NO

Explain your answer.

20. Overall, do you feel that the test T-shirt is safe to wear? YES NO

Explain your answer.

21. Please rate how satisfied or dissatisfied you are with the following aspects of the test T-shirts. Circle one answer for each.

	VERY DISSATISFIED 1	MODERATELY DISSATISFIED 2	SOMEWHAT DISSATISFIED 3	NEITHER DISSATISFIED NOR SATISFIED 4	SOMEWHAT SATISFIED 5	MODERATELY SATISFIED 6	VERY SATISFIED 7
a. Comfort of the test T-shirt (at the beginning of the exercise)	1	2	3	4	5	6	7
b. Comfort of the test T-shirt (at the end of the exercise)	1	2	3	4	5	6	7
c. Overall comfort	1	2	3	4	5	6	7
d. Ability to reduce body odor	1	2	3	4	5	6	7
e. Ability to reduce skin problems	1	2	3	4	5	6	7
f. Overall performance	1	2	3	4	5	6	7

22. What would your decision be on the future of anti-microbial treated T-shirts? Fill in one circle.

a. Reject them.
b. Adopt them as standard.
c. Make them available as an optional purchase item.
d. Do more research on them.

Explain your answer.

Attachment F: Antimicrobial Sock Midpoint Questionnaire

Last Name:_____ Evaluation ID Number:_____

Please answer all of the questions based on your experience during this test period (17 August 05 – 20 August 05) while wearing the test socks that you were issued. Your responses to the following questions will influence decisions on this item, so please consider each question carefully before answering. Your answers will remain confidential. Thank you in advance for your participation!

Rank? E-____ O-____ WO-____ Unit: _____ Battery: _____

1a. Were you issued socks to evaluate? YES NO

b. If YES, did you wear them? YES NO

If NO, why not?

IF YOU ANSWERED NO TO EITHER 1a or 1b, HAND IN THIS QUESTIONNAIRE NOW.

2. How many total days did you wear the socks since they were issued to you? _____ days

   How many times per day did you change the socks? ____ times per day

3. Did you ever run out of the test socks during the evaluation period? YES NO

   If YES, what other socks did you wear and how often?

4. What type of boot did you usually wear with the Anti-Microbial Socks? Fill in one circle.

   - ✔ A standard issue boot (specify type:_____)
   - A commercial boot (specify type:_____)

5. Did you wear any kind of insole in the boots? YES NO

   If YES, list the brand and specific type.

6. Have you used any kind of powder or ointment on your feet or in your boots? YES NO

   If YES, what have you used?

7. While wearing the socks did your feet come in contact with anything that might have caused a skin reaction or other problem?

   YES NO

   If YES, what did you come in contact with and what was the reaction?

8. Up to this point have you experienced any of the following with your feet? In part a, circle one answer for each. Fill in the circle for "N/A" if you did not experience a certain problem. In part b, indicate if you would normally expect to have each problem after three days in the field. Circle an answer in part b even if you are not having that problem now.

a. So far, to what extent are you experiencing the following?	**N/A**	**Mildly**	**Moderately**	**Severely**	**b. Would you normally expect to experience these problems after three days in the field?**
a. Foot odor	0	1	2	3	YES NO
b. General discomfort	0	1	2	3	YES NO
c. Itching feet	0	1	2	3	YES NO
d. Athletes Foot	0	1	2	3	YES NO
e. Toe nail fungus	0	1	2	3	YES NO
f. Skin rash/irritation	0	1	2	3	YES NO
g. Skin inflammation/redness	0	1	2	3	YES NO
h. Blisters or calluses	0	1	2	3	YES NO
i. Other (list below)	0	1	2	3	YES NO

9. Overall, do you feel that the socks are noticeably decreasing (circle one answer for each.)….

   a. your foot odor? YES NO

   b. other soldiers' foot odor? YES NO

   Explain your answer.

10. Overall, how do the foot problems that you are experiencing on THIS FIELD EXERCISE compare to what you would normally expect when wearing standard socks in the field? Fill in one circle.

    Problems on this exercise have been…

MUCH WORSE	MODERATELY WORSE	SLIGHTLY WORSE	ABOUT THE SAME	SLIGHTLY BETTER	MODERATELY BETTER	MUCH BETTER
1	2	3	4	5	6	7

11. At any time during this evaluation did you have to consult medical personnel for a foot problem?

    YES NO

    If YES, list the problem, the level of treatment you received (field, outpatient, etc.) and any duty time lost.
    NOTE: YOU DO NOT HAVE TO ANSWER THIS PART IF YOU HAVE PRIVACY CONCERNS!

12. Do the test socks feel hotter than the standard cotton and wool socks under similar circumstances? Circle one answer for each.

    Does the test socks feel hotter than…

    a. Standard Wool (black)? YES NO N/A

    b. Standard Cotton (green) YES NO N/A

    If YES, explain.

13. Please rate how satisfied or dissatisfied you are with the following aspects of the socks. Circle one answer for each..

	VERY DISSATISFIED 1	MODERATELY DISSATISFIED 2	SOMEWHAT DISSATISFIED 3	NEITHER DISSATISFIED NOR SATISFIED 4	SOMEWHAT SATISFIED 5	MODERATELY SATISFIED 6	VERY SATISFIED 7
a. Comfort of the socks (at the beginning of the exercise)	1	2	3	4	5	6	7
b. Comfort of the socks (after three days in the field)	1	2	3	4	5	6	7
c. Overall comfort	1	2	3	4	5	6	7
d. Ability to reduce foot odor	1	2	3	4	5	6	7
e. Ability to reduce foot problems	1	2	3	4	5	6	7
f. Overall performance	1	2	3	4	5	6	7

14. Do you have any other comments on the socks?

Attachment G: Antimicrobial Sock Final Questionnaire

Last Name:_____ Evaluation ID Number:_____

Please answer all of the questions based on your experience during this test period (17 August 05 – 24 August 05) while wearing the test socks that you were issued. Your responses to the following questions will influence decisions on this item, so please consider each question carefully before answering. Your answers will remain confidential. Thank you in advance for your participation!

Rank? E-____ O-____ WO-____ Unit: _____ Battery: _____

1a. Were you issued socks to evaluate? YES NO

b. If YES, did you wear them? YES NO

If NO, why not?

IF YOU ANSWERED NO TO EITHER 1a or 1b, HAND IN THIS QUESTIONNAIRE NOW.

2. How many total days did you wear the socks since they were issued to you? _____ days

How many times per day did you change the socks? ____ times per day

3. Did you ever run out of the test socks during the evaluation period? YES NO

If YES, what other socks did you wear and how often?

4. What type of boot did you usually wear with the Anti-Microbial Socks? Fill in one circle.

a. A standard issue boot (specify type:_____)

b. A commercial boot (specify type:_____)

5. Did you wear any kind of insole in the boots? YES NO

If YES, list the brand and specific type.

6. While wearing the test socks did you use any kind of powder or ointment on your feet or in your boots?

YES NO

If YES, list below the type (include brand name if known) and the number of times you used it.

TYPE/BRAND NAME TIMES USED

7. Did you launder, wash or clean the socks in any way during the evaluation? YES NO

If YES, how many times? _____

8. Have you had any durability problems with the test socks so far? YES NO

If YES, explain.

9. While wearing the socks did your feet come in contact with anything that might have caused a skin reaction or other problem?

YES NO

If YES, what did you come in contact with and what was the reaction?

10. During the course of the evaluation did you develop any physical problems with your feet (i.e. rashes, irritation, etc.) that you do not normally have?

YES NO

If NO, skip to question 11.
If YES, what type of problem did you have?

How much of your body was affected? Fill in one.

a. Specific areas (where:_____)
b. All over (wherever the test socks touched)
c. Other (specify:_____)

List any action that you took to address the problem:

11. Overall, did you see any reduction in any chronic foot conditions that you have been diagnosed with? This would include problems like athletes foot, fungus, etc.? Circle "N/A" if you do not have a history of these types of problems.

YES NO N/A

If "YES," explain.
Note: DO NOT ANSWER THIS PART IF YOU HAVE PRIVACY CONCERNS.

12. Did you experience any of the following with your feet? Circle one answer for each. Circle "N/A" if you did not experience a certain problem. If you did experience a certain problem, use the space at the right to indicate how many days you had it during the exercise (maximum = 7 days).

So far, to what extent are you experiencing the following?	**N/A**	**Mildly**	**Moderately**	**Severely**	**If you answered 1,2, or 3… How many days did you experience it during the exercise? (max=7 days)**
a. Foot odor	0	1	2	3	_____ Days
b. General discomfort	0	1	2	3	_____ Days
c. Itching feet	0	1	2	3	_____ Days
d. Athletes Foot	0	1	2	3	_____ Days
e. Toe nail fungus	0	1	2	3	_____ Days
f. Skin rash/irritation	0	1	2	3	_____ Days
g. Skin inflammation/redness	0	1	2	3	_____ Days
h. Blisters or calluses	0	1	2	3	_____ Days
i. Other (list below)	0	1	2	3	_____ Days

13. At any time during this evaluation did you have to consult medical personnel for a foot problem?

YES NO

If YES, list the problem, the level of treatment you received (field, outpatient, etc.) and any duty time lost.
NOTE: YOU DO NOT HAVE TO ANSWER THIS PART IF YOU HAVE PRIVACY CONCERNS!

14. Overall, do you feel that the socks are noticeably decreasing (circle one answer for each)….

a. your foot odor? YES NO

b. other soldiers' foot odor? YES NO

Explain your answer.

15. Overall, how do the foot problems that you are experiencing on THIS FIELD EXERCISE compare to what you would normally expect when wearing standard socks in the field? Fill in one circle.

Problems on this exercise have been…

	MUCH WORSE 1	MODERATELY WORSE 2	SLIGHTLY WORSE 3	ABOUT THE SAME 4	SLIGHTLY BETTER 5	MODERATELY BETTER 6	MUCH BETTER 7
a.	Foot odor			**N/A**	1 2 3	4 5 6	7
b.	General discomfort			**N/A**	1 2 3	4 5 6	7
c.	Itching feet			**N/A**	1 2 3	4 5 6	7
d.	Athletes Foot			**N/A**	1 2 3	4 5 6	7
e.	Toe nail fungus			**N/A**	1 2 3	4 5 6	7
f.	Skin rash/irritation			**N/A**	1 2 3	4 5 6	7
g.	Skin inflammation/redness			**N/A**	1 2 3	4 5 6	7
h.	Blisters or calluses			**N/A**	1 2 3	4 5 6	7

16. Do the test socks feel hotter than the standard cotton and wool socks under similar circumstances? Circle one answer for each.

Do the test socks feel hotter than the…			
a. Standard Wool (black)?	YES	NO	N/A
b. Standard Cotton (green)?	YES	NO	N/A

If YES, explain.

17. Please rate how satisfied or dissatisfied you are with the following aspects of the socks. Circle one answer for each.

	VERY DISSATISFIED 1	MODERATELY DISSATISFIED 2	SOMEWHAT DISSATISFIED 3	NEITHER DISSATISFIED NOR SATISFIED 4	SOMEWHAT SATISFIED 5	MODERATELY SATISFIED 6	VERY SATISFIED 7
a. Comfort of the socks (at the beginning of the exercise)	1	2	3	4	5	6	7
b. Comfort of the socks (at the end of the exercise)	1	2	3	4	5	6	7
c. Overall comfort	1	2	3	4	5	6	7
d. Ability to reduce foot odor	1	2	3	4	5	6	7
e. Ability to reduce foot problems	1	2	3	4	5	6	7
f. Overall performance	1	2	3	4	5	6	7

18. What impact did the test socks have on your performance in the field? Fill in one circle.

a. positive impact (improved performance)

b. no impact

c. negative impact (decreased performance)

Explain your answer.

19. Overall, do you feel that the test socks are comfortable for wearing over an extended period of time?

YES NO

Explain your answer.

20. Overall, do you feel that the test socks are safe to use? YES NO

Explain your answer.

21. What would your decision be on the future of anti-microbial treated socks? Fill in one circle.

a. Reject them.
b. Adopt them as standard.
c. Make them available as an optional purchase item.
d. Do more research on them.

Explain your answer.

APPENDIX II: Field User Evaluation Report – Part II

Chuck Greene
U.S. Army Natick Soldier RD&E Center

U.S. ARMY NATICK SOLDIER CENTER
Antimicrobial Treated Clothing Items – Part II
Field User Evaluation Report

Chuck Greene

16 June 2006

U.S. Army Natick Soldier Center
Natick, MA 01760

Executive Summary

Background

In April 2006 the Natick Soldier Center conducted a field user evaluation of antimicrobial treated uniforms, T-shirts, and socks with Soldiers from the 4th Brigade Combat Team, 1st Cavalry Division at Fort Bliss, Texas. The treatments evaluated were targeted against common, but harmful, bacteria routinely encountered by Soldiers in the field. The goal of the evaluation was to determine if the protection provided by these treatments would manifest itself in ways that would be noticeable to Soldiers. This field user evaluation was conducted as a follow-on to one conducted at Fort Bliss in August 2005 (see OFIG Report: **Antimicrobial Treated Clothing Items Field User Evaluation Report**, dated 21 November 2005).

Evaluation Design

All of the members of the participating unit were issued either untreated (type A) or treated (type B) versions of the uniform and the T-shirt. All of the participants were also issued one of three types of socks: untreated (type A), treated (type B), or treated (type C). The items were used both in the field and in garrison at Fort Bliss during a two-week period. While the training schedule of the various companies within the unit varied, they all spent approximately one week in the field and one week in garrison. Data was collected through a series of questionnaires that addressed criteria relevant to the assessment of antimicrobial treatments.

Survey Sample

The survey group consisted of 136 Soldiers from 2nd Battalion, 12th Cavalry Regiment. Most were males (98%, n=133 out of 136) and had been in the military for an average of four years. The breakdown by age was: 20 or less (41%, n=56 out of 136), 21 to 25 (28%, n=38 out of 136), 26 to 30 (18%, n=24 out of 136), and 31 or over (13%, n=18). The breakdown by rank was: E-1 to E-3 (60%, n=81 out of 136), E-4 to E-6 (33%, n=45 out of 136), E-7 to E-9 (4%, n=6 out of 136), and Officers (3%, n=4 out of 136). The most common career fields were Infantry (n=81), Supply (n=10), Combat Engineer (n=8), Mechanic (n=7), and Armor (n=6).

Key Findings

The goal of this evaluation was to determine if the protection provided by an antimicrobial clothing treatment would manifest itself in ways that would be noticeable and beneficial to Soldiers. Numerous significant differences were detected along these lines, particularly in relation to the uniform and the T-shirt. While we feel that the results of the field evaluation do make a case in favor of the application of antimicrobial technologies to military clothing items, the results must be evaluated alongside laboratory and technical data to gain a complete picture of the performance and benefits of the treatments.

Numerous significant differences were detected between the treated and untreated groups for uniform and T-shirt performance. Significantly more soldiers felt that the treated uniform and T-shirt were controlling their body odor than those in the untreated group. Significantly more Soldiers felt that the treated uniform and T-shirt decreased their discomfort, how dirty they felt, and the amount of time spent on personal hygiene. A related significant decrease in the frequency of use of certain hygiene products was also noted in the treated group over the untreated group. Significantly more Soldiers felt that the treated uniform and the T-shirt could be worn longer before needing to be changed; additional days were estimated at three for the uniform and two for the T-shirt compared to one each for the untreated uniform and T-shirt. Significantly more Soldiers felt that the treated uniform was comfortable for extended wear than those with the untreated items.

No significant differences were detected for the rate or intensity of various physical problems for those wearing treated or untreated items, however we feel that this may have more to do with the format of the question than the properties of the uniform treatment. This is based on the lack of variability apparent in the data (see Table 9). Also, there was some data that seemed to indicate that the opposite may be true and that the treatments did have an impact on these problems. Significantly more Soldiers in the treated group felt that they experienced a reduction in chronic problems over those in the untreated group. Also, a significantly higher percentage of soldiers in the treated group felt that the T-shirt was safe to wear compared to those in the untreated group. The same trend was noted for the uniform, but the difference was not statistically significant.

Findings for the sock are somewhat muted by the low number of respondents in each of the two treated groups (type B: n=37, type C: n=24). Also, we did not collect as much data on the sock as we did on the other items – it was considered secondary on the questionnaire to the uniform and T-shirt. However, there seemed to be some impact of the sock treatments on foot odor, hygiene practices, comfort and suitability for extended wear, and safety. A significantly higher percentage of soldiers with the type B sock felt it could be worn longer before needing to be changed than those with the untreated sock. A significantly higher percentage of Soldiers also felt that the type B sock reduced foot problems than those in the untreated group. In general, we feel that the results of the previous evaluation are more important to assessing treatment performance when used on socks. This data should be viewed as complimentary to that. At some point it might be useful to do a separate dedicated evaluation of antimicrobial socks. This evaluation could be designed specifically to address sock criteria as a primary objective.

Some additional data was collected on Soldier opinions related to the general effectiveness of antimicrobial products as well as some additional measures of acceptability and performance. Overall, three-quarters of the survey group feel that antimicrobial products are effective. This was true both before and after the evaluation. Interestingly, a significantly higher percentage of soldiers in the treated group believed in the general effectiveness of these products at the end of the evaluation when compared to the treated group. Approximately the same percentage of respondents (73%) indicated they would be in favor of adopting an antimicrobial treatment if it was proven to kill "germs" but did not necessarily reduce odor. Also, a high percentage soldiers in both groups felt that it was a good idea to treat field uniforms (84%) and T-shirts (81%) with antimicrobial treatments. There was also some evidence that the antimicrobial treatments may have a beneficial impact on the mood state of the wearer. This may be an area worth some follow-up in the future.

The application of an antimicrobial treatment, particularly to the T-shirt and the uniform, seemed to offer a range of benefits to the user. These included improved odor control, comfort, hygiene, and wear time. Other benefits, to include those related to physical problems and mood state are possible but could not be validated based on the available questionnaire data. There is also a great deal of interest amongst the Soldiers in the use of antimicrobial products as a treatment for field uniforms (84%) and T-shirts (81%). Three-fourths of the Soldiers believe in the effectiveness of these products and nearly three-fourths would be in favor of using them if they were proven to be effective in the lab but offered no immediately perceptible benefit to them.

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INTRODUCTION

Background

In April 2006 the Natick Soldier Center conducted a field user evaluation of antimicrobial treated uniforms, T-shirts, and socks with Soldiers from the 4th Brigade Combat Team, 1st Cavalry Division at Fort Bliss, Texas. The treatments evaluated were targeted against common, but harmful, bacteria routinely encountered by Soldiers in the field. The goal of the evaluation was to determine if the protection provided by these treatments would manifest itself in ways that would be noticeable to Soldiers. This field user evaluation was conducted as a follow-on to one conducted at Fort Bliss in August 2005 (see OFIG Report: **Antimicrobial Treated Clothing Items Field User Evaluation Report**, dated 21 November 2005). All of the members of the participating unit were issued either untreated (type A) or treated (type B) versions of the uniform and the T-shirt. All of the participants were also issued one of three types of socks: untreated (type A), treated (type B), or treated (type C). The items were used both in the field and in garrison at Fort Bliss during a two-week period. While the training schedule of the various companies within the unit varied, they all spent approximately one week in the field and one week in garrison. Data was collected through a series of questionnaires that addressed criteria relevant to the assessment of antimicrobial treatments. Two primary questionnaires were used to assess treatment performance, copies of which are included as Attachments A (background) and B (final). A total of 217 Soldiers from the unit were issued items, with 185 completing all of the data requirements of the field user evaluation.

Item Description

There were a total of seven items under evaluation: two versions of the standard Army Combat Uniform (untreated and treated), two versions of the standard polyester T-shirt (untreated and treated), and three versions of the standard cotton sock (untreated and two treated). A description of each of the items is included below. The basic garments and any formulations and treatments were the same as those used in the August 2005 evaluation.

- Uniform: Two versions of the standard Advanced Combat Uniform (ACU), one featuring a Microban® antimicrobial treatment formulation (type A) and one untreated (type B). Both uniforms also featured a wrinkle resistance treatment, which is standard for this uniform. Soldiers evaluating this item were issued one complete uniform to evaluate.

- T-shirt: Two versions of the standard issue 100% Polyester T-shirt, one featuring a Microban® antimicrobial treatment formulation (Type A) and one untreated (Type B). Soldiers evaluating this item were issued four T-shirts of the same type.

- Socks: Three versions of the standard issue 100% cotton socks: an untreated standard sock (Type A); an untreated standard sock which received a Microban® antimicrobial treatment (Type B), and the current treated standard sock, CR/PD 03-18 (Type C).

Results of the Previous Evaluation

The goal of the first evaluation was to determine if the protection provided by an antimicrobial clothing treatment would manifest itself in ways that would be noticeable and beneficial to Soldiers. Based on the results of the evaluation, it appeared that the T-Shirt was a promising candidate for application of an antimicrobial treatment. Soldiers who used the treated T-Shirt reported a significant reduction in odor as well as an overall reduction in other problems, which included general discomfort, heat rash, itching skin, and skin rash. The results for the antimicrobial treated uniform were less promising, with no apparent reduction in problems noted. However, significantly more Soldiers felt that the treated uniform was controlling their body odor at the end of the evaluation. Soldiers who evaluated the treated uniform were all issued standard T-shirts. It was felt that the T-shirt may have performed better than the uniform because it was a "next-to-the-skin" item. Those who used antimicrobial socks showed an overall reduction in the percentage reporting foot odor, general discomfort, and itching. Overall, it seemed that an antimicrobial treatment offered benefits in terms of sock performance.

The results of the August 2005 evaluation raised a number of questions which were addressed in this effort. It was felt that the antimicrobial treatments were providing some noticeable benefits for soldiers, but it seemed that the questionnaires were not providing a clear picture of item performance. In response to this, the questions were redesigned to assess the performance of the treatments in four areas: injury reduction, odor reduction, comfort, and impact on hygiene practices. Soldiers would be issued either all treated items or all untreated items to improve the possibility of measuring differences in performance between the groups. There was also a lot of discussion about the impact of a "placebo effect." It was a blind study and it was unclear if soldiers assumptions about whether they had a treated or untreated item had an impact on their perceptions of item performance. This would also be a blind study, however, on the final page of the final questionnaire, it was revealed which type of items the Soldiers were evaluating. They were then asked to indicate how this changed their opinions about antimicrobial treatments in general, and the items they evaluated specifically.

Test Design & Procedures

The evaluation of the antimicrobial treated uniforms, T-shirts, and socks featured a between-groups design. The independent variable was item type (treated vs. untreated) and the dependent measures were Soldier responses to an array of survey questions designed to quantify experiences and opinions relevant to the performance of the treatment. Assignment to either evaluation group was done by company and Soldiers did not know if any of the items issued to them had the antimicrobial treatment or not. Data was collected at the half-way point of the evaluation on day four (midpoint) and at the conclusion on day eight (final).

The questionnaires used were derived from those used previously, which had been developed with input provided by Dermatologists at William Beaumont Army Medical Center, Fort Bliss, TX, to ensure that content was appropriate in terms of the skin problems and conditions that Soldiers would experience in the field. The

key questions and the primary scale used on the questionnaire were based on existing symptom assessment scales with proven reliability and validity and designed for use to collect data in "self-reporting" scenarios. Other questions were developed as appropriate. In addition, the questionnaires were revised and expanded prior to the evaluation based on lessons learned.

Participants were briefed several days before the evaluation began on the purpose and procedures. They were informed that they would be evaluating antimicrobial clothing treatments and that some would receive treated items and some would receive untreated items. At that time they completed a background questionnaire to obtain demographic information as well as data on past experiences and their opinions on antimicrobial products and treatments. They were then issued the test items and were given the opportunity to try them on to make sure they fit. A few Soldiers had to change assigned groups at this point due to size availability of the various clothing items.

A final roster was maintained and each Soldier was double-checked to make sure that they had received the correct items and that they had completed the background questionnaire before they left the issue location. The issue was conducted on a Friday and the initial plan, based on the unit training schedule, was for the entire unit to spend the following Monday through Thursday in the field. On Thursday evening they would complete the midpoint questionnaire. On Friday, they would come out of the field. Soldiers were instructed by an operation order issued by the unit not to launder the uniforms and to wear them again the following week where the same schedule would be repeated: in the field Monday through Thursday, with final data being collected on Thursday evening.

At some point, the unit training schedule changed and only one of the companies went to the field for the first week (Alpha – predominantly untreated items) with the remainder staying in garrison (Headquarters, Bravo, Delta, Echo, and Fox – predominantly treated items). For week two the schedule would be reversed, with Alpha being in garrison and the remainder going to the field. If the evaluation team had been aware of this at the time of issue, changes could have been made to minimize the impact on the evaluation. Ultimately, we have sufficient valid data to draw conclusions about the performance of the antimicrobial treatment. However, certain adjustments had to be made to the test design and the subsequent reporting of results.

All of the companies spent a week in the field and a week in garrison. Most of the soldiers wearing the treated uniforms were in garrison for the first week and then spent the following week in the field. Most of the soldiers with the untreated uniforms did the opposite: in the field for the first week and then in garrison for the second week. This largely invalidates the data collected at the midpoint because of the radical difference between field and garrison training. The final questionnaire was revised extensively to reflect the changes to the training schedule and evaluation.

There were some minor variations in this revised training schedule. Some Soldiers had a training holiday on Friday, 7 April. Some of the Soldiers who spent the second week in the field deployed on Sunday, 9 April, which was an off day for Soldiers who spent the second week in garrison. By the end of the evaluation all of the participants had worn the items for approximately eight days, four in garrison and four in the field. The data presented below in Table 1 shows that weather conditions were substantially similar for the entire two-week period. In the end, we feel that as long as Soldiers did not wash the uniforms during the intervening weekend, the final questionnaire data is a valid measure of the performance of the antimicrobial treatments over an extended wear period.

Table 1
Weather Conditions at Fort Bliss, 3 to 13 April 2006

Reporting station: El Paso, TX International Airport[1]

April:	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**
Temp (max)	81	88	83	71	76	77	87	82	84	89	91
Temp (min)	50	53	65	51	51	46	47	63	57	51	60
Precipitation (inches)	0	0	0	0	0	0	0	T*	T*	0	T*
Relative Humidity (avg.)	16%	20%	19%	10%	23%	22%	9%	30%	18%	15%	17%

[1] From: NOAA, National Climatic Data Center
http://www.ncdc.noaa.gov/oa/ncdc.html
* T=Trace amount

Data Handling and Analysis

The raw data was returned to Natick where it was scanned, cleaned, and assembled into a final data set that contained the information from the background and final questionnaires. This data set contained a total of 185 respondents. Any respondents who reported that they laundered the uniforms were excluded (n=49). This left a remainder of 136: 74 in the untreated group and 62 in the treated group. Soldiers in the treated group had either the Type B (n=37) or the Type C (n=25) treated socks. There were no differences across the groups in terms of demographic factors (age, rank, gender, etc.).

Descriptive statistics used to describe the data are the number of Soldiers responding (n) and the percentage of the total responding to a certain option in a "yes - no" or multiple-choice question. Please note that the number of respondents reported for specific questions is based on the number of valid responses to that question, which results in some variation from question to question in the total number of respondents. The mean (X) is reported for scale-ended questions or estimates of time or frequency. The data was analyzed using a variety of statistical procedures. In all instances the .05 criterion level was used as the minimum probability level to determine significance for all statistical procedures. This indicates that, on a statistical level, there is a less than 5% chance that the differences observed are attributable to error or normal variation. If a certain

statistical procedure could not detect a significant difference the abbreviation "ns" ("not significant") is used in the relevant table.

Student's t-test for independent samples was used for scale-ended data when only two groups were involved and data was analyzed on a between-groups basis. This test compares the actual difference between two means in relation to the variation in the data to determine if they are equal or not. The results are expressed by the "t' statistic and an associated significance level. Data analysis for the sock, which featured three groups, required the use of the Analysis of Variance (ANOVA) and post-hoc test, which is essentially an extension of the t-test, to test the hypothesis that several means are equal or not. The results are expressed by the "F" statistic and its associated significance level. The Chi-square test was used to analyze all dichotomous data. This procedure tabulates a variable into categories and computes a Chi-square statistic. It compares the observed and expected frequencies in each category to test either that all categories contain the same proportion of values or that each category contains a user-specified proportion of values. The Chi-square test is expressed by the "X^2" statistic along with the corresponding significance level.

Survey Sample

The survey group consisted of 136 Soldiers from 2nd Battalion, 12th Cavalry Regiment stationed at Fort Bliss, TX. Most were males (98%, n=133 out of 136) and had been in the military for an average of four years. The breakdown by age was: 20 or less (41%, n=56 out of 136), 21 to 25 (28%, n=38 out of 136), 26 to 30 (18%, n=24 out of 136), and 31 or over (13%, n=18). The breakdown by rank was: E-1 to E-3 (60%, n=81 out of 136), E-4 to E-6 (33%, n=45 out of 136), E-7 to E-9 (4%, n=6 out of 136), and Officers (3%, n=4 out of 136). The most common career fields were Infantry (n=81), Supply (n=10), Combat Engineer (n=8), Mechanic (n=7), Armor (n=6), and Field Artillery (n=5).

Half of the respondents have experience with the ACU (50%, n=68 out of 136) and approximately two-thirds have experience with the polyester T-shirt (62%, n=84 out of 136) and the standard green cotton sock (71%, n=96 out of 136). Only 4% (n=6 out of 136) reported that any of these items have caused some kind of physical problem for them in the past, specifically athlete's foot (n=3) or excessive sweating (n=2). In addition, 4% (n=6 out of 136) also reported that they have been diagnosed with some kind of chronic skin or foot problem. These included individual two instances of hyperhidrosis and individual instances of athlete's foot, eczema, and allergies to certain types of detergents. Soldiers estimated that in the field they change their uniform every five days, their T-shirt every three days, and their socks every two days. In garrison, they change their uniform, T-shirt, and socks every three days, two days, and one day (respectively). The longest these Soldiers have ever worn the same item while in training or deployed was estimated at twelve days for the uniform, seven days for the T-shirt, and five days for the socks.

FINDINGS: UNIFORM AND T-SHIRT COMBINATION

Usage Profile

Soldiers estimated that they wore the uniform for an average of fifteen hours per day over nine days and that they changed T-shirts an average of every two days. The most common type of underwear worn were standard cotton briefs (43%, n=58 out of 136) or boxers (16%, n=22 out of 136), which were also changed an average of every two days. About one-third (34%, n=46 out of 136) reported that they did not wear underwear at all. As noted previously, anyone who had laundered or cleaned the uniform or T-shirt in any way had already been dropped from the data set. Four percent (n=5 out of 136) reported that they wore additional undergarments with the uniform, which were not identified. No one reported that they came in contact with anything that might have caused a skin reaction during the evaluation (insect repellent, gasoline, poisonous plants, etc.).

The use of hygiene products over the course of the entire evaluation was assessed on a five-point frequency of use scale. The results are presented below by uniform type (treated vs. untreated). Note that a series of t-tests were conducted and that significant differences were detected between groups.

Table 2
Frequency of use of Hygiene Products During the Evaluation
(n=134)

Several Times a Day 1	Once a Day 2	Once Every Other Day 3	Once or Twice This Week 4	Never 5
Product	**Untreated- A (n=72)**	**Treated - B (n=62)**	**t**	**p**
Regular wet wipes	3.2	3.2	0.07	ns
Antibacterial wet wipes	3.8	3.8	0.16	ns
Hand sanitizer	3.2	3.0	0.75	ns
Soap	2.3	3.1	3.70	p<.001
Deodorant / Anti-perspirant	2.3	2.8	2.70	p<.01

It is interesting that Soldiers in both groups used wet wipes and hand sanitizer at about the same rate, but that those in the treated group used soap and deodorant significantly less frequently than those in the untreated group. This represents a difference in using soap and deodorant every day (untreated) versus every other day (treated). This may be due to either the effectiveness of the clothing treatments. However, it could also be due to the different training scenario (untreated in garrison vs. treated in the field) experienced by the two groups during the second week. Even though the question was phrased to capture hygiene product use during the total two-week period, Soldier responses may have been more heavily influenced by what they were doing in the immediate past.

Odor Control

Odor control would obviously be one of the primary features of an antimicrobial treatment that would be observable to a test participant. Soldiers were asked directly if they felt that the clothing items they were issued were controlling their body and clothing odor. The results obtained, along with those of a Chi-square analysis, are presented below.

Table 3
Do the Clothing Items Control Your Body Odor?
(n=132)

	Untreated –A	Treated – B	X^2	p
Your body odor?	45% (32/71)	64% (39/61)	4.70,	<.05
Your Clothing odor?	56% (40/71)	66% (23/61)	1.17	ns

As can be seen above, significantly more Soldiers in the treated group felt that the clothing items were controlling their body odor. While the results for clothing odor were not significant, they still show a trend in favor of the treated uniform. It is interesting that about half of the Soldiers in the untreated group felt that the clothing items were doing something to control odor. This may have something to do with the baseline properties of the items themselves. It may also be related to the expectations of the evaluation participants as to the level of odor they would expect to experience after extended uniform wear compared to the reality of that which was actually experienced. However, using the untreated data as a baseline, it is clear that the treated clothing items did have an impact on body odor for significantly more Soldiers.

Hygiene

One of the areas not thoroughly addressed in the first antimicrobial evaluation was the impact of clothing treatments on various hygiene-related criteria. This shortcoming was addressed in this iteration. The broader concept of "hygiene" was defined in terms of perceptions of cleanliness, discomfort, the need for hygiene products, and the time spent on personal hygiene. A question was framed along these lines and the results obtained are presented below. Note that the data is presented by group and was analyzed using a Chi-square analysis.

Table 4
Clothing Item Impact on Hygiene Perceptions?

(n=132)

Did the clothing items decrease…

	Untreated –A	Treated – B	X^2	p
How unclean your skin feels?	19% (13/70)	32% (20/62)	3.29	=.05
How unclean your clothes feel?	21% (15/71)	34% (21/62)	2.72	ns
How uncomfortable you feel?	23% (16/71)	38% (24/62)	4.18	<.05
Your need to use hygiene products?	20% (14/71)	27% (17/62)	1.10	ns
Time spent on personal hygiene?	17% (12/71)	32% (20/62)	1.17	<.05

Significantly more Soldiers using the treated uniform felt the clothing items decreased how "unclean" their skin felt, how uncomfortable they felt, and the time spend on personal hygiene than those in the treated group. Additionally, there is a clear trend in favor of the treated items across all five criteria assessed. It is interesting to view these findings in light of those presented in Table 2 where it was found that Soldiers in the treated group used soap and deodorant significantly less frequently than those in the treated group. The potential problem with that data was that it could have been impacted by the most recent training scenario (garrison vs. field). However, in this instance, the questions were more general in nature and would seem less likely to be influenced by specific field or garrison hygiene practices.

When the questionnaire was revised on-site, an additional hygiene question was added. This question had to do with the perception of how long the clothing items could be worn before they needed to be changed. The data was analyzed by group using the Chi-square and the results obtained are presented below.

Table 5
Can the Clothing Items Be Worn Longer Before Needing to be Changed?

(n=134)

	Untreated –A	Treated - B	
Uniform	34% (24/71)	70% (42/60)	X^2=17.04, p<.001
T-shirt	28% (20/71)	58% (36/62)	X^2=12.14, p<.001

As can be seen above, significantly more Soldiers in the treated group felt that the uniform and the T-shirt could be worn longer before needing to be changed than those in the untreated group. In both instances, the margin was two-to-one in favor of the treated item. A second part of this question asked Soldiers to estimate how much longer they thought the item could be worn before needing to be changed. Those who felt that the uniform could not be worn longer were entered as "0." This was calculated into the Soldier estimates to provide a more comprehensive picture for the total group. The data was analyzed using the t-test and the results are presented below.

Table 6
Estimated Additional Days Before Laundering*

(n=134)

	Untreated –A	Treated – B	t
Uniform	1 day	3 days	2.41, p<.05
T-shirt	1 day	2 days	1.61, ns

***** If the respondent answered "no" in Table 5, additional days were calculated as 0 in this table.

Soldiers estimated that they could wear the treated uniform three additional days compared to one for the untreated uniform, which was a significant difference. While the estimates for the T-shirt were not significant, we would argue that two additional days (treated) compared to one (untreated) represented a practical difference.

Comfort

Comfort of the clothing items, specifically in terms of extended wear without laundering was addressed for both the uniform and the T-shirt. The results, analyzed using the Chi-square, are presented below.

Table 7
Are the Clothing Items Comfortable When Worn for Extended Periods Without Laundering?

(n=133)

	Untreated –A	Treated - B	
Uniform	47% (33/71)	68% (42/62)	X^2=6.01, p=.01
T-shirt	56% (40/71)	79% (49/62)	X^2=7.70, p<.01

Significantly more Soldiers using the treated uniform and T-shirt responded affirmatively than those using the untreated items. The results for the T-Shirt are particularly striking, with nearly 80% of the respondents wearing the treated item feeling that they were comfortable when worn for an extended period without laundering. The data presented in Table 7 compliments the data presented in Tables 5 and 6, so it would seem that there is a comfort factor to extended wear for the treated items and not just factors related to perceptions of "cleanliness."

Physical Problems

Soldiers were presented with a list of common maladies and symptoms that could be caused or influenced by bacteria and microbes. They used a ten-point scale to measure the intensity to which they were experiencing each of each of the problems listed (1 = "Not at All" to 10 = "Extreme"). The results were analyzed on a between-group basis using the t-test.

Table 8
Problems and Conditions Experienced During the Evaluation
(n=132)

<-Not at all								Extreme->
1	2	3	4	5	6	7	8	9	10

Problem	Untreated	Treated	n, t, p
Heat rash	1.44	1.23	133, 1.12, ns
Itching skin	1.58	1.68	134, 0.42, ns
Chafing	1.60	1.61	134, 0.07, ns
Skin rash or irritation	1.35	1.35	134, 0.05, ns
Skin lesions, sores, pimples	1.32	1.40	134, 0.52, ns
Infected cuts or scrapes	1.28	1.21	133, 0.48, ns

As can be seen above, no significant differences were detected between the groups for any of the problems listed. This could indicate either that the uniform does not have a noticeable impact on these problems or that we are not properly measuring the extent to which Soldiers are experiencing these problems. We feel the most likely explanation is the latter. Changes to the training schedule did impact this question more than any other. We had hoped to be able to do a direct comparison between the background questionnaire and the final. Originally, each question was phrased to provide a "snapshot" of these problems at the beginning and end of the evaluation. However, since the environment was different for half the group at the time of the final questionnaire (half in garrison and half in the field) this would have been invalid. So the background data had to be scrapped and the question on the final had to be rephrased to something more general (see question 12 in Attachment B).

We did have a backup to this question which asked Soldiers to assess the impact that the clothing items had on the types of physical problems they experienced in the field and the intensity of the problems that they did experience. The results are presented below in Table 9.

Table 9
Do the Clothing Items Reduce Physical Problems?
(n=132)

	Untreated –A	Treated - B	
Types of physical problems?	14% (10/72)	20% (12/60)	X^2=0.88, ns
Intensity of physical problems?	10% (7/71)	12% (7/60)	X^2=0.11, ns

There is a slight trend in favor of the treated items, but no significant difference detected between the evaluation groups. It is hard to avoid the conclusion that Soldiers could not perceive an impact of the uniform treatment on these larger physical issues. But the data also suggests that these problems are not that common, which we seriously doubt. So this reinforces the notion that we need to rethink how we measure this aspect of performance.

It is interesting to note that, in a separate question, four times as many soldiers in the treated group reported a reduction in chronic skin (or foot) problems than those in the untreated group (33%, n=6 out of 18 vs. 8%, n=2 out of 24; X^2=2.38, ns). This data is somewhat flawed because it is not exclusive to skin problems but also includes foot problems. Also, the number of Soldiers responding to this question is much greater than those reporting on the background questionnaire that they have been diagnosed with specific chronic problems in these areas (n=6). Soldiers were prompted to comment on their answer, but only one was received: someone with the treated uniform noted that he had "no skin problems at all with this uniform." This does suggest that the clothing treatments are doing something in this area, it is just not clear what.

Safety

None of the Soldiers in either the untreated or treated group reported that they had to consult medical personnel at any level for a skin problem during the evaluation. The evaluation participants were asked if they felt that the clothing items they were issued were safe to use. The results obtained, along with that of a Chi-square, are presented below.

Table 10
Are the Clothing Items Safe to Use?
(n=131)

	Untreated –A	Treated - B	
Uniform	90% (66/73)	95% (55/58)	X^2=0.89, ns
T-Shirt	89% (64/72)	100% (59/59)	X^2=6.98, p=.01

It is interesting that significantly more Soldier who used the treated T-shirt felt that it was safe to use when compared with the untreated T-shirt results. In fact, everybody who used the treated item felt that it was safe. This was true to a lesser extent for the uniform where no significant difference was detected. While Soldiers were given the opportunity to comment on this few did, and the comments do not shed any light as to why these differences exist. Perhaps it is a reflection of some or all of the differences detected so far (comfort, odor control, perceptions of cleanliness, etc.). Or perhaps it is a reflection of some other aspect of clothing performance that is not addressed adequately.

Comparison

Soldiers compared the problems that they experienced during the evaluation to what they would expect from wearing standard clothing items under similar conditions. The "standard" items for this group would be roughly split between evenly between the ACU/polyester T-shirt combination and the BDU/cotton T-shirt combination. A "not applicable" option was provided for Soldiers who did not experience a certain problem, so the number of respondents for each criteria varies to some extent. The scale used and results obtained are presented below.

Table 11
Problems and Conditions Compared: This Exercise With Past Experience
(n=132)

Problems on this exercise have been…

MUCH WORSE	MODERATELY WORSE	SLIGHTLY WORSE	ABOUT THE SAME	SLIGHTLY BETTER	MODERATELY BETTER	MUCH BETTER
1	2	3	4	5	6	7

Comparison: Problems on this exercise w/this uniform and past exercises	**Untreated A (n=72)**	**Treated - B (n=62)**	**n, t, p**
Body odor	4.3	4.5	130, t=0.81, ns
Uniform odor	4.4	4.8	132, t=1.43, ns
T-shirt odor	4.4	4.6	131, t=0.98, ns
Physical problems (body)	4.2	4.1	132, t=0.20, ns
How dirty you feel	4.2	4.3	132, t=0.38, ns
How dirty your clothes feel	4.2	4.5	132, t=1.31, ns
Overall discomfort in the field	4.3	4.3	131, t=0.16, ns
Overall discomfort in garrison	4.3	4.3	131, t=0.07, ns

There were no significant differences detected between either the treated or untreated groups for any of the problems rated. All of the ratings fell between "about the same" and "slightly better." It should be noted that Soldiers in the treated group generally provided higher ratings than those in the untreated group for all of the criteria evaluated, which would seem to be at least somewhat favorable for the treated items.

FINDINGS: SOCK

Usage Profile

The survey group was divided into three sock groups: untreated type A (n=74), treated type B (n=37), and treated type C (n=25). There were no significant differences detected for any of the usage variables by sock type. Soldiers estimated that they changed socks an average of every two days. The most common type of boot worn with the socks was the standard Desert Boot (78%, n=106 out of 136). Some wore a commercial item (18%, n=24 out of 136), which was identified as Belleville (n=18) or Altima (n=9). No one reported that they came in contact with anything that might have caused a skin reaction during the evaluation (insect repellent, gasoline, poisonous plants, etc.). As noted previously, anyone who had laundered or cleaned the socks in any way had already been dropped from the data set.

The use of foot care products was assessed on a five-point frequency of use scale. The results are presented below by sock type. Note that an ANOVA conducted on this data found no significant differences across the three groups.

Table 12
Frequency of use of Foot Care Products During the Evaluation
(n=134)

Several Times a Day		Once Every	Once or Twice	
1	Once a Day 2	Other Day 3	This Week 4	Never 5

Product	Mean
Foot powder	3.0
Foot spray	3.7
Anti-fungal cream	4.0

Soldiers in all three sock groups used foot care products at the same rate. The most common item used was foot powder ("every other day"). Foot spray and anti-fungal cream were not used to a great extent by the respondents.

Odor

Soldiers were asked to indicated if the socks seemed to control the level of foot odor that they experienced. The results obtained are presented below. The data from the three groups were analyzed using a Chi-square test, the results of which are also included.

Table 13
Do the Socks Control Your Foot Odor?
(n=132)

	Untreated –A	Treated - B	Treated C	X^2
Positive responses:	42% (30/71)	54% (19/35)	56% (14/25)	2.13, ns

While there were no significant differences detected, a higher percentage of Soldiers in each of the two treated sock groups felt that the socks controlled their foot odor. This represents a majority of respondents in the treated sock groups compared to a minority in the untreated group.

Hygiene

Questions related to hygiene were originally designed for the uniform and T-shirt. However analyzing them by sock group should give us some indication if the sock type had any impact on these issues.

Table 14

Sock Impact on Hygiene Perceptions

(n=132)

Did the clothing items decrease…

	Untreated –A	Treated - B	Treated C	X^2
How uncomfortable you feel?	23% (16/71)	49% (18/37)	24% (6/25)	8.43, $p<.05$
Your need to use hygiene products?	20% (14/71)	32% (12/37)	20% (5/25)	2.39, ns
Time spent on personal hygiene?	17% (12/71)	35% (13/37)	28% (7/25)	4.69, ns

Soldiers who used sock B felt that the items which they tested, including the sock, decreased their discomfort. This was a significantly higher percentage than that noted by the socks A and C users. No significant differences were detected for use of hygiene products or time spent on personal hygiene, although sock B did receive more positive responses than either of the other two socks.

Significantly more Soldiers in the treated sock groups felt that the socks could be worn longer before needing to be changed than those in the untreated group (see Table 15, below).

Table 15
Can the Socks Be Worn Longer Before Needing to be Changed?
(n=134)

	Untreated –A	Treated - B	Treated C	X^2
Socks	24% (24/71)	54% (42/60)	44% (6/25)	10.40, p<.01

The estimated additional days of wear before laundering was less than one for the untreated sock, two days for type B sock, and 1 day for the type C sock. An ANOVA detected a significant difference for these results (F=2.96, p<.05). A post-hoc test revealed that the type B sock could be worn significantly longer than the untreated sock. No difference was detected for the type C sock.

Comfort

Comfort of the socks in terms of extended wear without laundering was addressed. The results, analyzed using the Chi-square, are presented below.

Table 16
Are the Socks Comfortable When Worn for Extended Periods Without Laundering?
(n=133)

	Untreated –A	Treated - B	Treated C	X^2
Socks	39% (28/71)	62% (23/37)	52% (13/25)	5.22, ns

Clearly, both of the treated socks were considered comfortable for extended wear compared to the untreated sock. In this instance, the ability of the Chi-square to detect a difference may have been impacted by the smaller number of Soldiers in the two treated sock groups.

Physical Problems

Soldiers were presented with a list of common foot problems that could be caused or influenced by bacteria and microbes. They used a ten-point scale to measure the intensity to which they were experiencing each of each of the problems listed (1 = “Not at All” to 10 = “Extreme”). The results were analyzed on a between-group basis using the ANOVA.

Table 17
Problems and Conditions Experienced During the Evaluation
(n=132)

<-Not at all									Extreme->
1	2	3	4	5	6	7	8	9	10

Problem	Untreated	Treated B	Treated C	n, F, p
Itching feet	1.64	1.84	1.84	134, 0.36, ns
Athlete's Foot	1.67	1.84	1.92	134, 0.31, ns
Toe Nail Fungus	1.43	1.24	1.52	134, 0.47, ns
Skin rash or irritation	1.47	1.11	1.48	134, 1.24, ns
Blisters or calluses	1.42	1.35	2.00	134, 2.72, ns

There were no significant differences detected across the three sock groups for any of the problems listed. Data obtained through this question has all of the limitations identified previously in the section on the uniform and T-shirt. We feel that this question is flawed to some extent both because of its design and due to how it had to be changed after the unit training schedule changed. The results of the backup question, which asked Soldiers to assess the impact that the socks had on the types and intensity of foot problems experienced, is presented below.

Table 18
Do the Socks Reduce Foot Problems?
(n=132)

	Untreated –A	Treated - B	Treated C	X^2
Types of foot problems?	11% (8/70)	30% (11/37)	24% (6/25)	5.80, p=.05
Intensity of foot problems?	10% (7/71)	17% (6/36)	24% (6/25)	3.21, ns

There is a slight trend in favor of the treated items, with a significant difference detected in favor of the Type B sock over the untreated sock. As noted in the previous section, four times as many soldiers in the treated group reported a reduction in chronic skin or foot problems than those in the untreated group (33%, n=6 out of 18 vs. 8%, n=2 out of 24; X^2=2.38, ns). When analyzed by sock type, we find a similar trend (Treated B: 25%, n=2 out of 8; Treated C: 40%, n=4 out of 24; Untreated A: 8%, n=2 out of 24; X^2=4.82, ns). This data is somewhat flawed because it is not exclusive to foot problems and the number of Soldiers responding is much greater than those reporting chronic problems on the background questionnaire. However, we feel it is worth noting again here since it seems to indicate some type of positive impact of the treatments on physical problems.

Safety

None of the Soldiers in any of the sock groups reported that they had to consult medical personnel for a foot problem. The evaluation participants were asked if they felt that the socks they were issued were safe to use. The results obtained, along with that of a Chi-square, are presented below.

Table 19
Are the Clothing Items Safe to Use?
(n=131)

	Untreated –A	Treated - B	Treated C	X^2
Sock	89% (65/73)	94% (33/35)	100%, (24/24)	3.34, ns

While the Chi-square test did not detect a significant difference between any of the groups, it is interesting to note that all of the Soldiers who used the type C sock felt that it was safe to use. The untreated sock had the lowest percentage of those who felt it was safe to use, but it was still satisfactory at 89%.

Comparison

Soldiers compared the foot problems that they experienced during the evaluation to what they would expect from wearing standard socks under similar conditions. The scale used and results obtained are presented below.

Table 20
Problems and Conditions Compared: This Exercise With Past Experience
(n=132)

Problems on this exercise have been…

MUCH WORSE	MODERATELY WORSE	SLIGHTLY WORSE	ABOUT THE SAME	SLIGHTLY BETTER	MODERATELY BETTER	MUCH BETTER
1	2	3	4	5	6	7

Comparison: Problems on this exercise w/this uniform and past exercises	**Untreated A (n=71)**	**Treated - B (n=37)**	**Treated C (n=25)**	**n, F, p**
Foot odor	4.3	4.5	4.5	133, F=0.50, ns
Sock odor	4.2	4.4	4.4	131, F=0.28, ns
Foot problems	4.2	4.3	4.1	132, F=0.22, ns
Overall discomfort in the field	4.3	4.4	4.2	133, F=0.31, ns
Overall discomfort in garrison	4.3	4.3	4.2	133, F=0.02, ns

There were no significant differences detected between the untreated item and either of the treated items. All of the ratings fell between "about the same" and "slightly better." It should be noted that Soldiers in the sock B group generally provided higher ratings than those in the untreated group for all of the criteria evaluated, which was not the case with the sock C group. However, it should also be noted that the "standard" item in this case would most likely be the green cotton which does feature the same antimicrobial treatment as used in sock C. The data for the untreated item (sock A) in this table is interesting because it is identical to the treated standard sock.

ADDITIONAL FINDINGS

"Revelation Question"

Soldiers were informed of the type of item that they evaluated, either treated or untreated, at the top of the last page on the final questionnaire. The information was provided to the participant in the form of a question: "you received a (treated or untreated) uniform, T-shirts, and socks to evaluate. To what extent did you know or suspect this?" A five-point scale was used (1= "Convinced Item was Untreated," 2 = "Suspected Item was Untreated," 3=Had no Idea," 4= "Suspected Item was Treated," 5= "Convinced Item Was Treated") and an answer was solicited for each item (uniform, T-shirt, and sock). The raw data was transformed so that anyone indicating that they suspected or were convinced their uniform had been treated or untreated were reclassified into three groups: those who guessed what they had correctly, those who did not know, and those who guessed wrongly. The results were analyzed using a Chi-square analysis and are presented below.

Table 21
Did The Participants Guess What Type of Item They Had?
(n=134)

		Untreated (n=73)	Treated (n=61)	X^2	p
a. Uniform	Guessed right:	16%	36%		
	Did not know:	70%	44%		
	Guessed wrong:	14%	20%	9.51	p<.01
b. T-shirt	Guessed right:	12%	30%		
	Did not know:	74%	54%		
	Guessed wrong:	14%	16%	6.79	p<.05

		Untreated (n=73)	Treated B (n=37)	Treated C (n=24)	X^2	p
a. Sock	Guessed right:	15%	30%	34%		
	Did not know:	73%	57%	50%		
	Guessed wrong:	12%	13%	16%	6.05	ns

A significantly higher percentage of Soldiers in the treated group had guessed what they were issued than those in the untreated group. We are reasonably certain that the secret regarding which uniforms were treated and which were not held until the end of the evaluation. Given this, it could be argued that there was some noticeable benefit being provided by the treated items that led Soldiers to guess correctly what they had. The findings for the sock groups would lead us to the same conclusion for those items. However, the Chi-square test could not detect a significant difference across the three groups.

A follow-up question asked Soldiers if their opinion about the items that they had evaluated had changed now that they knew specifically what they were evaluating. Significantly more Soldiers in the treated group indicated that it had when compared to the untreated group (18%, n=11 out of 61 vs. 3%, n=2 out of 67; X^2=7.92, p<.01). The data was also broken out by sock type (untreated: 3%, n=2 out of 67; treated B: 22%, n=8 out of 37; treated C: 13%, 3 out of 24). In this instance, a significant difference was found (X2=9.25, p=.01). Specifically, more soldiers using the type B sock had their opinion changed that those using the untreated items. No differences were noted for the type C sock. Soldiers were invited to comment on how their opinion changed. Unfortunately, only a few did – with most indicating that their opinion of the treated items was positive or was enhanced somehow (n=7).

Acceptability

The participants opinion regarding the general effectiveness of antimicrobial was assessed on the background and the final questionnaire. The data was divided by group (untreated vs. treated) and analyzed using the Chi-square. The results obtained are presented below in Table 22. Note that only those who expressed an opinion are included.

Table 22
Belief in the General Effectiveness of Antimicrobial Products
(n=115)

	Background	
Untreated	71%, n=44 out of 62	
Treated	79%, n=42 out of 53	All = 75%, n=86 out of 115
X^2=1.04, ns		
	Final	
Untreated	66%, n=36 out of 55	
Treated	84%, n=46 out of 55	All = 75%, n=82 out of 110
X^2=4.79, p<.05		

In both instances, of those who have an opinion, three-fourths of the survey group believe in the general effectiveness of antimicrobial products. It is interesting that no significant difference was detected between the groups on the background questionnaire. However, a significance difference was detected between groups on

the final questionnaire. This data was collected after the identity of the test items had been revealed. At the end of the evaluation, a significantly higher percentage of Soldiers in the treated group believed in the effectiveness of antimicrobial products than those in the untreated group. It is also interesting to see that the treated group's confidence in these products increased after the evaluation experience while those with the untreated items decreased.

Soldiers were asked if they would be in favor of adopting an anti-microbial uniform treatment if it was proven to kill germs but did not necessarily reduce odor. Essentially, the same percentages were obtained as presented in Table 22: overall, 73% (n=96 out of 131) indicated that they would be in favor. The percentages within the two groups was also similar (untreated: 66%, n=46 out of 70; treated, 82%, n=50 out of 61; X^2=4.40, p<.05). Overall, those who in favor commented that the treatment would help prevent illness (n=4) and keep soldiers clean (n=3). Those who were against it felt that they would not really know if it worked if there was no way of detecting it (n=3) and that odor reduction was important (n=2).

Soldiers gave their opinions on whether or not antimicrobial treatments should be used on Army items. This included items that they evaluated as well as those they had not. The results obtained are presented below in Table 23. The data is presented by group (untreated vs. treated) and for the total population.

Table 23
Should Antimicrobial Treatments Be Used On Other Army Items?
(n=134)

	Positive Responses:			
	Untreated –A	**Treated – B**	**X^2**	**p**
Field uniforms?	77% (56/73)	92% (56/61)	5.52,	<.05
T-shirts?	75% (55/73)	89% (54/61)	3.81,	<.05
Other clothing items? (glove liners, polypros, etc.)	61% (43/71)	81% (46/57)	6.05,	<.05
Equipment items? (sleeping bags, poncho liner, etc.)	68% (47/69)	86% (48/56)	5.52,	<.05

	ALL
Field uniforms?	84% (112/134)
T-shirts?	81% (109/134)
Other clothing items? (glove liners, polypros, etc.)	70% (89/128)
Equipment items? (sleeping bags, poncho liner, etc.)	76% (95/125)

As can be seen above, a significantly higher percentage of the treated group were in favor of using antimicrobial treatments across the board than those who evaluated untreated items. This included approximately 90% of the respondents in the treated group recommending the use of these types of treatments on the uniform and T-shirt. Overall, three-fourths or more of the total group felt that antimicrobial treatments

should be applied to equipment items, field uniforms, and T-shirts. More than two-thirds felt that they should be applied to other clothing items like gloves and polypropylene underwear.

Mood Profile

After the first antimicrobial evaluation we felt that it would be worthwhile to investigate the impact, if any, that use of the treatment had on a Soldier's mood. There are a number of "mood inventories" available through which this could be accomplished. Perhaps the most venerable has been the Profile of Mood States (POMS). The POMS is a standardized psychological test. Respondents are presented with a list of 65 adjectives and asked to rate on five-point scale the extent to which they have been experiencing each over a recent period of time. The 65 items fall into six subscales which are scored separately, and then collectively. Since this was just a trial run of this concept, we did not want to use the full POMS. Two items were selected from each of the six scales, for twelve in all. The scale used is the same as called for in the standard test. The mean was calculated for each of the twelve items and analyzed by group using the t-test. The results obtained are presented below.

Table 24
Abbreviated Mood Profile by Treatment Level
(n=134)

Not At All	A Little	Moderately	Quite a Bit	Extremely
0	1	2	3	4

	Untreated (n=72)	**Treated (n=62)**	**n, t, p**
Energetic	1.7	1.9	134, 1.13, ns
Miserable	1.2	0.8	133, 2.05, <.05
Alert	1.9	2.1	133, 1.03, ns
Tense	1.3	0.9	133, 1.74, ns
Efficient	2.0	2.1	133, 0.59, ns
Bad Tempered	1.1	0.9	133, 1.09, ns
Fatigued	1.0	1.1	133, 0.22, ns
Forgetful	0.9	0.6	133, 1.00, ns
Restless	1.0	0.9	133, 0.56, ns
Sluggish	0.9	0.8	133, 0.91, ns
Unhappy	1.0	0.7	133, 1.84, ns
Annoyed	1.2	0.9	133, 1.29, ns

It is interesting that one significant difference was detected: Soldiers wearing the treated items reported that they were significantly less "miserable" than Soldiers wearing the treated uniform. However, this could just be a reflection of factors already discussed. The definition of "miserable" for these respondents probably had a lot more to do with the physical reality of being hot, dirty, and uncomfortable than it did with describing a state of mind. It is interesting that, for each of the negative words, the treated group always reported experiencing them to a lesser extent than the untreated group. The opposite is noted for the positive adjectives: the treated group always being experiencing them to a greater extent than the untreated group. We do not really know how much to make of this and are doubtful if a full POMS should be added to future antimicrobial evaluations. However, it does seem to provide support for the concept that treated clothing items could possibly have an impact on the "mood" of the wearer and to validate the use of similar questions in the future.

DISCUSSION

The goal of this evaluation was to determine if the protection provided by an antimicrobial clothing treatment would manifest itself in ways that would be noticeable and beneficial to Soldiers. Numerous significant differences were detected along these lines, particularly in relation to the uniform and the T-shirt. While we feel that the results of the field evaluation do make a case in favor of the application of antimicrobial technologies to military clothing items, the results must be evaluated alongside laboratory and technical data to gain a complete picture of the performance and benefits of the treatments.

Numerous significant differences were detected between the treated and untreated groups for uniform and T-shirt performance. Significantly more soldiers felt that the treated uniform and T-shirt were controlling their body odor than those in the untreated group. Significantly more Soldiers felt that the treated uniform and T-shirt decreased their discomfort, how dirty they felt, and the amount of time spent on personal hygiene. A related significant decrease in the frequency of use of certain hygiene products was also noted in the treated group over the untreated group. Significantly more Soldiers felt that the treated uniform and the T-shirt could be worn longer before needing to be changed; additional days were estimated at three for the uniform and two for the T-shirt compared to one each for the untreated uniform and T-shirt. Significantly more Soldiers felt that the treated uniform was comfortable for extended wear than those with the untreated items.

No significant differences were detected for the rate or intensity of various physical problems for those wearing treated or untreated items, however we feel that this may have more to do with the format of the question than the properties of the uniform treatment. This is based on the lack of variability apparent in the data (see Table 9). Also, there was some data that seemed to indicate that the opposite may be true and that the treatments did have an impact on these problems. Significantly more Soldiers in the treated group felt that they experienced a reduction in chronic problems over those in the untreated group. Also, a significantly higher

percentage of soldiers in the treated group felt that the T-shirt was safe to wear compared to those in the untreated group. The same trend was noted for the uniform, but the difference was not statistically significant.

Findings for the sock are somewhat muted by the low number of respondents in each of the two treated groups (type B: n=37, type C: n=24). Also, we did not collect as much data on the sock as we did on the other items – it was considered secondary on the questionnaire to the uniform and T-shirt. However, there seemed to be some impact of the sock treatments on foot odor, hygiene practices, comfort and suitability for extended wear, and safety. A significantly higher percentage of soldiers with the type B sock felt it could be worn longer before needing to be changed than those with the untreated sock. A significantly higher percentage of Soldiers also felt that the type B sock reduced foot problems than those in the untreated group. In general, we feel that the results of the previous evaluation are more important to assessing treatment performance when used on socks. This data should be viewed as complimentary to that. At some point it might be useful to do a separate dedicated evaluation of antimicrobial socks. This evaluation could be designed specifically to address sock criteria as a primary objective.

Some additional data was collected on Soldier opinions related to the general effectiveness of antimicrobial products as well as some additional measures of acceptability and performance. Overall, three-quarters of the survey group feel that antimicrobial products are effective. This was true both before and after the evaluation. Interestingly, a significantly higher percentage of soldiers in the treated group believed in the general effectiveness of these products at the end of the evaluation when compared to the treated group. Approximately the same percentage of respondents (73%) indicated they would be in favor of adopting an antimicrobial treatment if it was proven to kill "germs" but did not necessarily reduce odor. Also, a high percentage soldiers in both groups felt that it was a good idea to treat field uniforms (84%) and T-shirts (81%) with antimicrobial treatments. There was also some evidence that the antimicrobial treatments may have a beneficial impact on the mood state of the wearer. This may be an area worth some follow-up in the future.

The application of an antimicrobial treatment, particularly to the T-shirt and the uniform, seemed to offer a range of benefits to the user. These included improved odor control, comfort, hygiene, and wear time. Other benefits, to include those related to physical problems and mood state are possible but could not be validated based on the available questionnaire data. There is also a great deal of interest amongst the Soldiers in the use of antimicrobial products as a treatment for field uniforms (84%) and T-shirts (81%). Three-fourths of the Soldiers believe in the effectiveness of these products and nearly three-fourths would be in favor of using them if they were proven to be effective in the lab but offered no immediately perceptible benefit to them.

Attachment A: Field Uniform, T-shirt, and Sock Background Questionnaire

Please answer the following questions based on your total experience in the military. When you answer, fill in the circle COMPLETELY. Your answers will remain confidential. Thank you in advance for your participation!

Rank? E-____ O-____ WO-____ MOS, Branch, or Specialty?_____

Unit: _____ Company: _____ Platoon:_____ Gender? M F

Time in the military? _____ years _____ months Age? A 20 or less
B 21 to 25 years
C 26 to 30 years
D 31 to 35 years
E 36 or older

1. Are you currently using any of the following items? Fill in one circle for each. If you answer "YES," use the space to the right to indicate how many months you have been wearing these items.

a. New ACU	Y	N	_____ months
b. Standard Polyester T-shirt?	Y	N	_____ months
c. Standard green Cotton sock?	Y	N	_____ months

If NO, have you ever worn any of these items? Y N

If YES, list the item, where you wore it, and for how long.

Has wearing any of these items ever caused you to develop physical problems (i.e. rashes, irritation, etc.) that you would not normally have?

Y N

If YES, explain.

2. Have you ever been diagnosed with any chronic skin problems like eczema, hyperhidrosis (excessive sweating), allergies to certain fibers (i.e. wool, etc.), and allergies to certain chemicals or substances that result in a rash or other skin reaction?

Y N

If "YES," describe the problem.
Note: do not answer this part if you have privacy concerns.

3. How many days do you usually wear a uniform, T-shirt, or socks in the field or in garrison **before laundering them?** Fill in one answer for each.

	Field:	**Garrison:**
a. Uniform (ACU or BDU)	____ days	____ days
b. T-shirt (standard issue)	____ days	____ days
c. Socks (standard issue)	____ days	____ days

4. When in the field for either training or on deployment, what is the **LONGEST** you have ever worn one of these items **without laundering them?**

a. Uniform (ACU or BDU)	____ days
b. T-shirt (standard issue)	____ days
c. Socks (standard issue)	____ days

5. While in the field, how often do you usually use the following? Fill in one circle for each. Use the following scale:

Several Times a Day 1	Once a Day 2	Once Every Other Day 3	Once or Twice A Week 4	Never 5

a. Regular wet wipes	1	2	3	4	5
b. Antibacterial wet wipes	1	2	3	4	5
c. Hand sanitizer	1	2	3	4	5
d. Soap	1	2	3	4	5
e. Deodorant / Anti-perspirant	1	2	3	4	5
f. Foot powder	1	2	3	4	5
g. Foot spray	1	2	3	4	5
h. Anti-fungal cream	1	2	3	4	5
i. Other (list below)	1	2	3	4	5

6. AS OF TODAY, to what extent are you experiencing the following types of skin and foot problems? Fill in one circle for each. Use the scale of 1 to 10 to rate how intense the various types of problems are. As the numbers increase, so would the severity of the problem from 1 (not at all) to 10 (extreme).

		| <- Not at all ---------- Extreme->|									
		1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**
	Body:										
a.	Heat rash	1	2	3	4	5	6	7	8	9	10
b.	Itching skin	1	2	3	4	5	6	7	8	9	10
c.	Chafing	1	2	3	4	5	6	7	8	9	10
d.	Skin rash or irritation	1	2	3	4	5	6	7	8	9	10
e.	Skin lesions, sores, pimples	1	2	3	4	5	6	7	8	9	10
f.	Infected cuts or scrapes	1	2	3	4	5	6	7	8	9	10
	Feet:										
a.	Itching feet	1	2	3	4	5	6	7	8	9	10
b.	Athletes Foot	1	2	3	4	5	6	7	8	9	10
c.	Toe nail fungus	1	2	3	4	5	6	7	8	9	10
d.	Skin rash/irritation	1	2	3	4	5	6	7	8	9	10
e.	Blisters or calluses	1	2	3	4	5	6	7	8	9	10

7. During a **typical week in the field**, how often do you change your uniform, T-shirts and socks?

Uniform (ACU or BDU)	Every _____ days	or	_____ times a day
T-shirt:	Every _____ days	or	_____ times a day
Socks:	Every _____ days	or	_____ times a day

8. What type of underwear do you USUALLY wear in the field? Fill in one circle.

A. Standard cotton underwear

B. Other (Type:_____)

C. None

If you filled in "c," go to the next question.
How often do you change underwear in the field? Every _____ days or _____ times a day

9. Have you ever purchased a clothing item ("odor eater" socks, T-shirts, underwear, etc.) labeled "antibacterial" or "antimicrobial" for use in the field?

Y N

If YES, list the items that you have purchased.

10. In general, do you feel that antimicrobial or antibacterial products are effective? Y N

Why or why not?

11. Antibacterial and antimicrobial products are used for different reasons. How important are each of the following benefits of these types of products? Use the scale below and fill in one circle for each.

	NOT IMPORTANT AT ALL 1	SOMEWHAT IMPORTANT 2	MODERATELY IMPORTANT 3	VERY IMPORTANT 4
a. Reducing or preventing body odor	1	2	3	4
b. Reducing or preventing clothing odor	1	2	3	4
c. Reduce the risk of illness (diarrhea, urinary tract infection, etc.)	1	2	3	4
d. Reduce the risk of infected cuts, scrapes, wounds	1	2	3	4
e. Reduce the risk of skin infection, rashes, or disease	1	2	3	4

12. How much does each of the following impact the comfort of the clothing you wear in the field? Use the scale provided below and fill in one circle for each.

	NO IMPACT 1	SLIGHT IMPACT 2	MODERATE IMPACT 3	HIGH IMPACT 4
a. Odor	1	2	3	4
b. A dirt/dust coating	1	2	3	4
c. Mud/clay caked on	1	2	3	4
d. Dried sweat/salt stains	1	2	3	4
e. Other (specify:_____)	1	2	3	4

Comments?

Attachment B: Field Uniform, T-shirt, and Sock Final Questionnaire

For the week of **April 9 to April 13**, please answer the following questions:

a. How many times did you shower? _____ times

b. How many nights did you sleep in the field? _____ nights

c. How many nights did you sleep at home? _____ nights

d. How many days did you wear the test items in the field and in garrison? Fill in one answer for each.

	Field	Garrison
a. Test uniform (ACU)?	_____ days	_____ days
b. Test T-shirts?	_____ days	_____ days
c. Test socks?	_____ days	_____ days

e. While in the field what type of jobs or training did you perform?

f. What type of jobs or training did you perform in garrison?

IMPORTANT: ANSWER THE REST OF THE QUESTIONS BASED ON YOUR EXPERIENCE WITH THE UNIFORM, T-SHIRTS, AND SOCKS FOR THE ENTIRE EVALUATION PERIOD: APRIL 1 TO APRIL 13!

1. Have you had to stop wearing, or not wear, any of the issued items for any reason? Y N

   If YES, explain.

2a. How many total days did you wear the uniform since it was issued to you? _____ days for _____ hours per day

2b. How often did you change your T-shirts and socks during the evaluation period? T-shirt: Every _____ days

Socks: Every _____ days

3. Did you launder, wash or clean the test uniform, T-shirts, or socks during the evaluation? Y N

   Which ones did you launder and how did you launder them (i.e. machine washed, "wind washed," etc.)?

4. What underwear did you USUALLY wear? Fill in one. A. Standard cotton

   B. Other (Type:_____)

   C. None

   How often did you change underwear during the evaluation period? Every _____ days

5. Did you wear any additional undergarments under the uniform? Y N

   If YES, list the type, the number of days that you wore them, and how frequently you changed them.

6. What type of boot did you usually wear? Fill in one. A. Issue Desert Boot (type?:_____)
   B. A commercial boot (type?:_____ _____)

   Identify any insole worn in the boot:

7. Since you have been wearing the test items how often have you used the following? Fill in one circle for each.

	Several Times a Day 1	Once a Day 2	Once Every Other Day 3	Once or Twice This Week 4	Never 5
a. Regular wet wipes	1	2	3	4	5
b. Antibacterial wet wipes	1	2	3	4	5
c. Hand sanitizer	1	2	3	4	5
d. Soap	1	2	3	4	5
e. Deodorant / Anti-perspirant	1	2	3	4	5
f. Foot powder	1	2	3	4	5
g. Foot spray	1	2	3	4	5
h. Anti-fungal cream	1	2	3	4	5

8. While wearing the uniform, T-shirt, and socks, did you come in contact with anything that caused a skin reaction (i.e. insect repellent, gasoline, poisonous plants, etc.)?

   Y N

   If YES, what was it and what part of your body was affected?

9. During the course of the evaluation did you develop any physical problems (i.e. rashes, irritation, etc.) that you do not normally have?

   Y N

   If YES, identify the problem, where it occurred on your body and any action that you took to address it.

10. At any time during this evaluation did you have to consult medical personnel for a (fill in one circle for each)…

    a. skin problem? Y N
    b. foot problem? Y N

    If YES, list the problem, the level of treatment you received (field, outpatient, etc.) and any duty time lost.
    NOTE: YOU DO NOT HAVE TO ANSWER THIS PART IF YOU HAVE PRIVACY CONCERNS!

11. Overall, do you feel that the clothing items controlled (fill in one circle for each)….

a. YOUR Body odor?	Y	N
b. YOUR Clothing odor?	Y	N
c. YOUR Foot odor?	Y	N

Explain your answer.

12. Since you have been wearing the test items to what extent are you experiencing the following skin and foot problems? Fill in one circle for each. Use the scale of 1 to 10 to rate how intense the various types of problems are. As the numbers increase, so would the severity of the problem from 1 (not at all) to 10 (extreme).

		| <- Not at all ---------- Extreme->|									
		1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**
	Body:										
a.	Heat rash	1	2	3	4	5	6	7	8	9	10
b.	Itching skin	1	2	3	4	5	6	7	8	9	10
c.	Chafing	1	2	3	4	5	6	7	8	9	10
d.	Skin rash or irritation	1	2	3	4	5	6	7	8	9	10
e.	Skin lesions, sores, pimples	1	2	3	4	5	6	7	8	9	10
f.	Infected cuts or scrapes	1	2	3	4	5	6	7	8	9	10
	Feet:										
a.	Itching feet	1	2	3	4	5	6	7	8	9	10
b.	Athletes Foot	1	2	3	4	5	6	7	8	9	10
c.	Toe nail fungus	1	2	3	4	5	6	7	8	9	10
d.	Skin rash/irritation	1	2	3	4	5	6	7	8	9	10
e.	Blisters or calluses	1	2	3	4	5	6	7	8	9	10

13. Overall, do you feel that the clothing items reduced (fill in one circle for each)….

BODY:

a. the types of physical problems you are experiencing?	Y	N
b. the intensity of physical problems you are experiencing?	Y	N

If YES, which types of problems?

FEET:

c. the types of foot problems you are experiencing?	Y	N
d. the intensity of foot problems you are experiencing?	Y	N

If YES, which types of problems?

14. Overall, did you see any reduction in any chronic skin or foot conditions that you have been diagnosed with? This would include skin problems like eczema or allergies to certain fibers (i.e. wool, etc.) that result in a rash or other skin reaction. Fill in (/) for "N/A" if you do not have a history of these types of problems.

    Y N (/)

    If "YES," explain.
    Note: DO NOT EXPLAIN IF YOU HAVE PRIVACY CONCERNS.

15. Did you see a reduction in any other problems that you normally experience which could be related to bacteria or other microbes (eye infection, diarrhea, etc.)?

    Y N

    If YES, explain.

16. Overall, do you feel that the clothing items you were issued decreased (fill in one circle for each)….

a. how unclean your skin feels?	Y	N
b. how unclean your clothes feel?	Y	N
c how uncomfortable you feel?	Y	N
d. your need to use hygiene products?	Y	N
e. amount of time spent on personal hygiene?	Y	N

    If YES, explain.

17. Do you feel that the test items could be worn for longer periods of time than standard items before needing to be changed? Circle one answer for each.

a. Uniform	Y	N	If YES, how many days longer?	_____ days
b. T-shirt	Y	N	If YES, how many days longer?	_____ days
c. Socks	Y	N	If YES, how many days longer?	_____ days

    Explain your answers.

18. Do you feel that the test items are comfortable when worn for an extended period without laundering? Fill in one circle for each.

a. T-shirt	Y	N
b. Uniform	Y	N
c. Socks	Y	N

    Comments?

19. In general, describe how you have felt this past week, including today, by circling one answer after each of the words listed below. Use the following scale:

Not At All	A Little	Moderately	Quite a Bit	Extremely
1	2	3	4	5

a.	Energetic	1	2	3	4	5
b.	Miserable	1	2	3	4	5
c.	Alert	1	2	3	4	5
d.	Tense	1	2	3	4	5
e.	Efficient	1	2	3	4	5
f.	Bad Tempered	1	2	3	4	5
g.	Fatigued	1	2	3	4	5
h.	Forgetful	1	2	3	4	5
i.	Restless	1	2	3	4	5
j.	Sluggish	1	2	3	4	5
k.	Unhappy	1	2	3	4	5
l.	Annoyed	1	2	3	4	5

20. How do your experiences wearing the test items compare to what you would expect when wearing the standard uniform, T-shirts, and socks under similar circumstances (for the same length of time, etc.)? Fill in one circle for each.

MUCH WORSE	MODERATELY WORSE	SLIGHTLY WORSE	ABOUT THE SAME	SLIGHTLY BETTER	MODERATELY BETTER	MUCH BETTER
1	2	3	4	5	6	7

a.	Body odor	1	2	3	4	5	6	7
b.	Foot odor	1	2	3	4	5	6	7
c.	Uniform odor	1	2	3	4	5	6	7
d.	T-shirt odor	1	2	3	4	5	6	7
e.	Sock odor	1	2	3	4	5	6	7
f.	Physical problems (body)	1	2	3	4	5	6	7
g.	Foot problems	1	2	3	4	5	6	7
h.	How dirty you feel	1	2	3	4	5	6	7
i.	How dirty your clothes feel	1	2	3	4	5	6	7
j.	Overall discomfort in the field	1	2	3	4	5	6	7
k.	Overall discomfort in garrison	1	2	3	4	5	6	7

21. What impact did wearing the uniform, T-shirt, and socks have on your performance in the field? Fill in one circle.

1. POSITIVE IMPACT (improved performance)
2. No impact
3. NEGATIVE IMPACT (decreased performance)

Explain your answer.

22. Overall, do you feel that the test items are safe to use? Fill in one circle for each.

a. T-shirt?	Y	N
b. Uniform?	Y	N
c. Socks?	Y	N

If NO, explain your answer.

23. Please rate how satisfied or dissatisfied you are with the following aspects of the clothing items that you are evaluating. Fill in one circle for each.

	VERY DISSATISFIED 1	MODERATELY DISSATISFIED 2	SOMEWHAT DISSATISFIED 3	NEITHER DISSATISFIED NOR SATISFIED 4	SOMEWHAT SATISFIED 5	MODERATELY SATISFIED 6	VERY SATISFIED 7
Uniform:							
a. Comfort at the beginning of the exercise	1	2	3	4	5	6	7
b. Comfort as of today	1	2	3	4	5	6	7
c. How clean the uniform feels over time	1	2	3	4	5	6	7
d. Odor resistance	1	2	3	4	5	6	7
e. Reduction in expected skin problems	1	2	3	4	5	6	7
f. Overall performance	1	2	3	4	5	6	7
T-Shirts:							
a. Comfort at the beginning of the exercise	1	2	3	4	5	6	7
b. Comfort as of today	1	2	3	4	5	6	7
c. How clean the T-shirts feel over time	1	2	3	4	5	6	7
d. Odor resistance	1	2	3	4	5	6	7
e. Reduction in expected skin problems	1	2	3	4	5	6	7
f. Overall performance	1	2	3	4	5	6	7
Socks:							
a. Comfort at the beginning of the exercise	1	2	3	4	5	6	7
b. Comfort as of today	1	2	3	4	5	6	7
c. How clean the socks feel over time	1	2	3	4	5	6	7
d. Odor resistance	1	2	3	4	5	6	7
e. Reduction in expected foot problems	1	2	3	4	5	6	7
f. Overall performance	1	2	3	4	5	6	7

1. You received an ANTI-MICROBIAL TREATED uniform, T-shirts, and socks to evaluate. To what extent did you know or suspect this? Fill in one answer for each.

	CONVINCED ITEM WAS UNTREATED 1	SUSPECTED ITEM WAS UNTREATED 2	HAD NO IDEA 3	SUSPECTED ITEM WAS TREATED 4	CONVINCED ITEM WAS TREATED 5
a. Uniform	1	2	3	4	5
b. T-shirts	1	2	3	4	5
c. Socks	1	2	3	4	5

2. Now that you know this, does this change your opinions about the items or antimicrobial treatments?

Y N

If YES, which ones and how?

3. Did wearing the uniform, T-shirt, or socks seem to prevent any physical problems that you would normally experience in the field (rash, sores, etc.)? Fill in one circle for each.

a. Uniform?	Y	N
b. T-shirt?	Y	N
c. Sock?	Y	N
d. Wearing all three together?	Y	N

If YES, explain.

4. Now that you know for certain, rate your level of satisfaction with the antimicrobial treatment when used on each item. Fill in one circle for each.

	VERY DISSATISFIED 1	MODERATELY DISSATISFIED 2	SOMEWHAT DISSATISFIED 3	NEITHER DISSATISFIED NOR SATISFIED 4	SOMEWHAT SATISFIED 5	MODERATELY SATISFIED 6	VERY SATISFIED 7
a. Uniform treatment	1	2	3	4	5	6	7
b. T-shirt treatment	1	2	3	4	5	6	7
c. Sock treatment	1	2	3	4	5	6	7

Comments?

5. In general, do you feel that antimicrobial or antibacterial products are effective? Y N

Why or why not?

6 Now that you know for certain, rate your OVERALL level of satisfaction with each of the items you evaluated. Fill in one circle for each.

	VERY DISSATISFIED 1	MODERATELY DISSATISFIED 2	SOMEWHAT DISSATISFIED 3	NEITHER DISSATISFIED NOR SATISFIED 4	SOMEWHAT SATISFIED 5	MODERATELY SATISFIED 6	VERY SATISFIED 7
a. Uniform	1	2	3	4	5	6	7
b. T-shirt	1	2	3	4	5	6	7
c. Sock	1	2	3	4	5	6	7

7. Antibacterial and antimicrobial products are used for different reasons. How important are each of the following benefits of these types of products? Use the scale below and fill in one circle for each.

	NOT IMPORTANT AT ALL 1	SOMEWHAT IMPORTANT 2	MODERATELY IMPORTANT 3	VERY IMPORTANT 4
a. Reducing or preventing body odor	1	2	3	4
b. Reducing or preventing clothing odor	1	2	3	4
c. Reduce the risk of illness (diarrhea, urinary tract infection, etc.)	1	2	3	4
d. Reduce the risk of infected cuts, scrapes, wounds	1	2	3	4
e. Reduce the risk of skin infection, rashes, or disease	1	2	3	4
f. Other (specify:_____)	1	2	3	4

8. Would you be in favor of adopting an anti-microbial uniform treatment if it was proven to kill germs but you could not necessarily tell if it was working (i.e. no odor reduction or only minimal odor reduction)?

Y N

Comments?

9. The Army is considering treating field uniforms and other items with antibacterial or antimicrobial treatments. Do you feel that this is a good idea? Fill in one circle for each.

a. Field uniforms? Y N

b. T-shirts? Y N

c. Other clothing items? (glove liners, polypros, etc. Y N

(specify type:_____)

d. Equipment items? (sleeping bags, poncho liner, etc.) Y N

(specify type:_____)

Explain your answer.

10. Do you have any final comments on either the items that you evaluated or antimicrobial treatments and products in general?

APPENDIX III: Experimental Application Conditions

(Conditions for application of antimicrobials
for preliminary microbiology study.)

Sample Description: *ACU Control*. 50/50 nylon/cotton ACU fabric 60" inside width

Trial No. 1

Sample ID: 10-D

Yardage: 15

Application Method: Pad 50% wet pickup 40 liter pad charge

Application Procedure:

1.) Prepare pad bath as follows:

Permafresh DM4	211 grams per liter
Metasoft ED23	48 grams per liter
Tyner 787-8	65 grams per liter
Soda ash	4 grams per liter

2.) Dry / Cure 170 C (340 F)

Sample Description: *ACU Triclosan*. 50/50 nylon/cotton ACU fabric 60" inside width

Trial No. 2

Sample ID: 10-A 2.0% Microban 9200-200, 4.0% Microban R10800-0

Yardage: 15

Application Method: Pad 50% wet pickup 40 liter pad charge

Application Procedure:

1.) Prepare pad bath as follows:

Microban 9200-200	40 grams per liter
Microban 10800-0	40 grams per liter
Permafresh DM4	211 grams per liter
Metasoft ED23	48 grams per liter
Tyner 787-8	65 grams per liter

2.) Adjust ph to 5-6

3.) Dry / Cure 170 C (340 F)

Amount of Microban 9200-200 needed: 3.6 lbs
Amount of Microban 10800-0 needed: 3.6 lbs.

Sample Description: *ACU Quat Silane*. 50/50 nylon/cotton BDU fabric 60" width

Trial No. 4

Sample ID: 10-B 3.0% Microban SiS 7200 AM OWG

Yardage: 15

Application Method: Pad 50% wet pickup 40 liter pad charge

Application Procedure:

1.) Prepare pad bath as follows:

Microban SiS 7200 AM	60 grams per liter
Permafresh DM4	211 grams per liter
Metasoft ED23	48 grams per liter
Tyner 787-8	65 grams per liter

2.) Adjust ph to 6-7

3.) Dry / Cure 170 C (340 F)

Amount of Microban SiS 7200 AM needed: 5.3 lbs

Sample Description: *ACU PHMB*. 50/50 nylon/cotton BDU fabric 60" inside width

Trial No.: 3

Sample ID: 10-C 2.0% Reputex OWG

Yardage: 15

Application Method: Pad 50% wet pickup 40 liter pad charge

Application Procedure:

1.) Prepare pad bath as follows:

Reputex 20	40 grams per liter
Permafresh DM4	211 grams per liter
Metasoft ED23	48 grams per liter
Tyner 787-8	65 grams per liter

2.) Adjust ph to 6-7

3.) Dry / Cure 170 C (340 F)

Amount of Reputex 20 needed: 3.6 lbs

Sample Description: *T shirt Control*. 20" tubular knit 22/1 MJS 100% Akwatek polyester

Trial No.

Sample ID: 20-D

Sample Size: 15 yards + 80 yards 22s Airjet polyester ballast

Application Method:

Application Procedure:

Controls were standard Akwatek finish on white, undyed, unfinished fabric.

Sample Description: *T shirt Triclosan*. 20" tubular knit 22/1 MJS 100% Akwatek polyester

Trial No. 8

Sample ID: 20-A 2.0% Microban 9200-200 OWG

Sample Size: 15 yards + 80 yards 22s Airjet polyester ballast

Application Method: Presure jet dying
School sample jet 40 lbs load
10:1 liquor ratio

Application Procedure:

1.) Load fabric into machine
2.) Add water to 10:1 liquor ratio
3.) Add 1% owg Foamaster 340 defoamer
4.) Adjust ph to 5.5 with citric acid
5.) Add 2% owg Microban 9200-200
6.) Raise temerature 3 deg/min to 130 deg C
7.) Hold at 130 deg C for 40 min
8.) Cool bath 2.5 deg/min to 65 deg C
9.) Rinse
10.) Unload
11.) Extract excess water and straighten fabric on pad
12.) Dry

Amount of Microban 9200-200 needed: 0.8 lbs

Sample Description: *T shirt Quat Silane*. 20" tubular knit 22/1 MJS 100% Akwatek polyester

Trial No. 9

Sample ID: 20-B 3.0% Microban SiS 7200 AM OWG

Yardage: 15

Application Method: Pad 75% wet pickup
60% pick up extraction pad
15% pickup applicator pad 40 lbs charge

Application Procedure:

1.) Prepare applicator pad bath as follows:

31.6 lbs water
8.4 lbs Microban SiS 7200 AM

2.) Adjust ph to 7

3.) Dry

Amount of Microban SiS 7200 AM needed: 8.4

Sample Description: *T shirt PHMB*. 20" tubular knit 22/1 MJS 100% Akwatek polyester

Trial No. 10

Sample ID: 20-C 2.0 % Reputex 20 OWG

Yardage: 15

Application Method: Pad 75% wet pickup
60% pick up extraction pad
15% pickup applicator pad 40 lbs charge

Application Procedure:

1.) Prepare applicator pad bath as follows:

34. 4 lbs water
5.6 lbs Arch Chemical Reputex 20

2.) Adjust ph to 7

3.) Dry

Amount of Arch Chemical Reputex 20 needed: 5.6 lbs.

Sample Description: *Cotton / Nylon Boot Sock Control*

Trial No.

Sample ID: 30-D

Sample Size: 4 dozen pairs (9.5 lbs.)

Application Method: Atmospheric Exhaust

Application Procedure:

Control Boot Sock was left unfinished.

Sample Description: *Cotton / Nylon Boot Sock Triclosan*

Trial No. 6

Sample ID: 30-A 2.0% Microban 9200-200, 4.0% Microban R10800-8

Sample Size: 4 dozen pairs (9.5 lbs.)

Application Method: Atmospheric Exhaust

Application Procedure:

1.) Set bath at 44 C (110 F)
2.) Adjust ph to 5-6
3.) Add 2% of Microban 9200-200 based on weight of bath
4.) Add 2% of Microban 10800-0 based on weight of bath
5.) Run 20 minutes
6.) Drop - Extract
7.) Dry 170 C (340 F)

Amount of Microban 9200-200 needed: TBD based on size of bath
Amount of Microban 10800-0 needed: TBD based on size of bath

Sample Description: *Cotton / Nylon Boot Sock Quat Silane*

Trial No. 7

Sample ID: 30-B 3.0% Microban SiS 7200 AM OWG

Sample Size: 4 dozen pairs (9.5 lbs.)

Application Method: Atmospheric Exhaust

Application Procedure:

1.) Set bath at 44 C (110 F)
2.) Adjust ph to 6-8
3.) Add 3% of Microban SiS 7200 AM based on weight of goods (130 grams)
4.) Run 20 minutes
5.) Drop - Extract
6.) Dry 170 C (340 F)

Amount of Microban SiS 7200 AM needed: 0.29 lbs

Sample Description: *Cotton / Nylon Boot Sock PHMB*

Trial No. 5

Sample ID: 30-C 2.0% Reputex 20 OWG

Sample Size: 4 dozen pairs (9.5 lbs.)

Application Method: Atmospheric Exhaust

Application Procedure:

1.) Set bath at 44 C (110 F)
2.) Adjust ph to 6-8
3.) Add 2% of Reputex 20 based on weight of goods (86.5 grams)
4.) Run 20 minutes
5.) Drop - Extract
6.) Dry 170 C (340 F)

Amount of Reputex 20 needed: 0.19 lbs

APPENDIX IV: Antimicrobial Application Conditions for Field Study

Sample Description: 50/50 nylon/cotton ACU fabric 60" inside width

Trial No. Field Trial Production - ACU - Delta Mills

Sample ID: 10-AC 1.5% Microban 9200-200 3.0% Microban R10800-0 2.0% Reputex 20

Yardage: 1000

Application Method: Pad 50% wet pickup 475 liter pad charge

Application procedure:

1.) Prepare pad bath as follows:

Microban 9200-200	30 grams per liter
Microban 10800-0	60 grams per liter
Reputex 20	40 grams per liter
Permafresh DM4	211 grams per liter
Metasoft ED23	48 grams per liter
Tyner 787-8	65 grams per liter

2.) Adjust ph to 5-6

3.) Dry / Cure 170 C (340 F)

Sample Description: T shirt tubular knit 22/1 MJS 100% Akwatek polyester

Trial No. Field Trial Production - T Shirts - Carolina Cotton Works

Sample ID: 20-AC 1.5% Microban 9200-200 OWG 2.0% Reputex 20 OWG

Sample Size: 1000 pounds

Application Method: Microban 9200-200 -Pressure jet dyeing
School Jet 1000 lbs load
10:1 liquor ratio
Reputex 20 - Pad with softener
Pad 75% wet pickup
60% pick up extraction pad
15% pickup applicator pad

Application Procedure:

1.) Load fabric into machine
2.) Add water to 10:1 liquor ratio
3.) Add 1% owg Foamaster 340 defoamer
4.) Adjust ph to 5.5 with citric acid
5.) Add 1.5% owg Microban 9200-200
6.) Raise temperature 3 deg/min to 130 deg C
7.) Hold at 130 deg C for 40 min
8.) Cool bath 2.5 deg/min to 65 deg C
9.) Rinse
10.) Unload
11.) Extract excess water
12.) Pad 2.0% Reputex 20 owg with softener
13.) Dry 210° F

Sample Description: Cotton / Nylon Boot Sock

Trial No. Field Trial Production - Boot Sock - Pickett Hosiery Mills

Sample ID: 30-AC 1.5% Microban 9200-200 3.0% Microban R10800-8
2.0% Reputex 20

Sample Size: 400 pairs

Application Method: Atmospheric Exhaust
Paddle Tub

Application Procedure:

1.) Set bath at 44 C (110 F)
2.) Adjust ph to 5-6
3.) Add 1.5% of Microban 9200-200 OWG
4.) Add 3% of Microban 10800-0 OWG
5.) Add 2.0% of Reputex 20
6.) Add softener
7.) Run 20 minutes
8.) Drop - Extract
9.) Dry 170 C (340 F)

APPENDIX V: Test Methods

Microbiological Test Methods for Study

APPENDIX V.a: AATCC Test Method 100

Quantitative test method for bacterial efficacy on treated textile products.

Antibacterial Finishes on Textile Materials: Assessment of

Developed in 1961 by AATCC Committee RA31; revised 1965, 1981, 1988 (with title change), 1993, 1999; editorially revised 1969, 1971, 1974, 1985; reaffirmed 1977, 1981, 1989, 1998; editorially revised and reaffirmed 1986.

1. Purpose and Scope

1.1 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 is necessary. Quantitative evaluation also provides a clearer picture for possible uses of such treated textile materials.

2. Principle

2.1 Swatches of test and control textile materials are tested qualitatively for antibacterial activity by AATCC Method 147. Those showing activity are evaluated quantitatively. Test and control swatches are inoculated with the test organisms. After incubation, the bacteria are eluted from the swatches by shaking in known amounts of neutralizing solution. The number of bacteria present in this liquid is determined, and the percentage reduction by the treated specimen is calculated.

3. Terminology

3.1 **activity**, n.—*of an antibacterial agent*, a measure of effectiveness of the agent.

3.2 **antibacterial agent**, n.—*in textiles*, any chemical which kills bacteria (bactericide) or interferes with the multiplication, growth or activity of bacteria (bacteriostat).

4. Safety Precautions

NOTE: These safety precautions are for information purposes only. The precautions are ancillary to the testing procedures and are not intended to be all inclusive. It is the user's responsibility to use safe and proper techniques in handling materials in this test method. Manufacturers MUST be consulted for specific details such as material safety data sheets and other manufacturer's recommendations. All OSHA standards and rules must also be consulted and followed.

4.1 Both the qualitative and quantitative tests should be carried out by persons with training and experience in the use of bacteriological techniques. The U.S. Department of Health and Human Services publication, *Biosafety in Microbiological and Biomedical Laboratories*, should be consulted (see 13.1).

4.2 CAUTION: Some of the bacteria used in this test are capable of infecting humans and producing disease. Therefore, every necessary and reasonable precaution must be taken to eliminate this risk to the laboratory personnel and to personnel in the associated environment. Wear protective clothing and respiratory protection that prevents penetration by the bacteria.

4.3 Good laboratory practices should be followed. Wear safety glasses in all laboratory areas.

4.4 All chemicals should be handled with care.

4.5 An eyewash/safety shower should be located nearby for emergency use.

4.6 Sterilize all contaminated samples and test materials prior to disposal.

4.7 Exposure to chemicals used in this procedure must be controlled at or below levels set by government authorities (e.g., Occupational Safety and Health Administration's [OSHA] permissible exposure limits [PEL] as found in 29 CFR 1910.1000 of January 1, 1989). In addition, the American Conference of Governmental Industrial Hygienists (ACGIH) Threshold Limit Values (TLVs) comprised of time weighted averages (TLV-TWA), short term exposure limits (TLV-STEL) and ceiling limits (TLV-C) are recommended as a general guide for air contaminant exposure which should be met (see 13.2).

5. Limitations

5.1 For a qualitative, relatively quick and easily executed method to determine residual antibacterial activity of textile materials, refer to AATCC Method 147, Antibacterial Activity Assessment of Textile Materials: Parallel Streak Method.

6. Test Organisms

6.1 Test bacteria.

6.1.1 *Staphylococcus aureus*, American Type Culture Collection No. 6538. Gram positive organism.

6.1.2 *Klebsiella pneumoniae*, American Type Culture Collection No. 4352. Gram negative organism.

6.1.3 Other suitable species can also be used.

7. Culture Medium

7.1 Suitable broth/agar media are Nutrient, Trypticase Soy and Brain-Heart Infusion.

Nutrient Broth:	
Peptone (Bacto-peptone) (see 13.3)	5 g
Beef extract (see 13.4)	3 g
Distilled water	to 1000 mL

7.2 Heat to a boil to disperse ingredients. Adjust to pH 6.8 ± 0.1 with 1*N* sodium hydroxide (NaOH) solution. (This is not necessary if prepared, dehydrated medium is used.)

7.3 Dispense in 10 mL amounts in conventional bacteriological culture tubes (i.e., 125 × 17 mm). Plug and sterilize at 103 kPa (15 psi) for 15 min.

7.4 Nutrient agar. Add 1.5% bacteriological agar to nutrient (or appropriate) broth (see 7.1). Heat to boiling. Check pH and adjust to 7.1 ± 0.1 using NaOH solution if necessary. Dispense in 15 ± 1 mL amounts in conventional bacteriological culture tubes. Plug and sterilize at 103 kPa (15 psi) for 15 min. (May be sterilized in 1000 mL borosilicate glass flasks and petri dishes poured from this.)

7.5 Slurry Inoculum Carrier (for hydrophobic fabrics) (see 7.2 and 7.3):

Sodium Chloride	8.5 g
Agar	3.0 g
Distilled Water	1000 mL

8. Maintenance of Culture of Test Organisms

8.1 Using a 4 mm inoculating loop, transfer the culture daily in nutrient (or appropriate medium) broth for not more than two weeks. At the conclusion of two weeks, make a fresh transplant from stock culture. Incubate cultures at 37 ± 2°C (99 ± 3°F) or other optimal temperature.

8.2 Maintain stock cultures on nutrient or appropriate agar slants. Store at 5 ± 1°C (41 ± 2°F) and transfer once a month to fresh agar (see 13.5).

9. Qualitative Test (Screening or Presumptive Test)

9.1 For detection of bacteriostatic activity use AATCC Method 147 on a test specimen and control specimen using the organisms referred to above. For demonstration of bactericidal activity, proceed to the quantitative test described below.

10. Quantitative Test (Reference or Confirmatory Test)

10.1 Preparation. The following description will be in terms of fabric swatches. Textile materials not in fabric form can likewise be tested with the appropriate modification.

10.1.1 Size and shape of treated swatches: Cut circular swatches 4.8 ± 0.1 cm (1.9 ± 0.03 in.) in diameter, from the test fabric (preferably with a steel die). Stack the swatches in a 250 mL wide-mouth glass jar with screw cap. The number of swatches to be used is dependent on the fiber type and fabric construction. Use that amount of fabric which will absorb the 1.0 ± 0.1 mL of inoculum, and leave no free liquid in the jar. For example, 4 swatches of cotton print cloth will absorb 1 mL. The number of swatches used per jar should be reported.

10.1.2 Controls. Swatches of the same fiber type and fabric construction as test sample but containing no antibacterial finish (negative control).

10.1.3 Sterilization of samples. This is optional. The method to be used depends on the type of fiber and finish. Cotton, acetate and many manmade fibers can be sterilized in the autoclave. Wool can be sterilized by ethylene oxide or by intermittent (fractional) sterilization in flowing steam. The latter is also least damaging to certain finishes. Report method of sterilization, if used.

10.1.4 Size of inoculum per sample. Apply 1.0 ± 0.1 mL of an appropriate dilution of a 24 h broth culture of the test organism so that recovery from (1) untreated control fabric swatches or (2) treated test fabric swatches at "0" contact time (plated as soon as possible after inoculation) will show counts of 1-2 × 10^5 organisms. The dilution of the test organism should be made in nutrient (or appropriate) broth (see 7.1, 7.5 and 13.6).

10.2 Procedure.

10.2.1 Inoculation of fabrics. When using *Staphylococcus aureus*, shake a 24 h culture and let stand for 15-20 min before preparing the inoculum.* Place the swatches separately in sterile petri dishes and use a microliter pipette to inoculate them making sure that there is even distribution of the inoculum (see 13.7). Transfer these swatches aseptically to the jar. Screw the jar tops on tightly to prevent evaporation.

10.2.2 As soon as possible after inoculation ("0" contact time), add 100 ± 1 mL of neutralizing solution to each of the jars containing the inoculated untreated control swatches, the inoculated treated test swatches and the uninoculated treated test swatches.

10.2.3 The neutralizing solution should include ingredients to neutralize the specific antibacterial fabric treatment and to take care of any pH requirements of the fabrics (from finishes, antibacterial agents, etc.). The neutralizing solution employed should be reported (see 13.8).

10.2.4 Shake the jars vigorously for one minute. Make serial dilutions with water and plate (in duplicate) on nutrient (or appropriate) agar. Dilutions of 10^0, 10^1, 10^2 are usually suitable.

10.2.5 Incubation over contact periods. Incubate additional jars containing inoculated untreated control swatches and jars containing inoculated treated test swatches at 37 ± 2°C (99 ± 3°F) for 18-24 h. Similar jars may be incubated over other periods (e.g., 1 or 6 h) to provide information about the bactericidal activity of the treatment over such periods.

10.2.6 Sampling of inoculated and incubated swatches. After incubation, add 100 ± 1 mL of neutralizing solution to jars containing untreated control swatches and to jars containing treated test swatches. Shake the jars vigorously for one minute. Make serial dilutions and plate (in duplicate) on nutrient (or appropriate) agar. Dilutions of 10^0, 10^1, 10^2 are usually suitable for treated test fabrics. Several different dilutions may be required for untreated control fabrics depending on the incubation period.

10.2.7 Incubate all plates for 48 h at 37 ± 2°C (99 ± 3°F) or other optimal temperature.

11. Evaluation

11.1 Report bacterial counts as the number of bacteria per sample (swatches in jar) not as the number of bacteria per mL of neutralizing solution. Report "0" counts at 10^0 dilution as "less than 100."

11.2 Calculate percent reduction of bacteria by the specimen treatments by one of the following formulas:

1) $100\,(B - A)/B = R$

where:

R = % reduction

A = the number of bacteria recovered from the inoculated treated test specimen swatches in the jar incubated over the desired contact period

B = the number of bacteria recovered from the inoculated treated test specimen swatches in the jar immediately after inoculation (at "0" contact time)

2) $100\,(C - A)/C = R$

where:

C = the number of bacteria recovered from the inoculated untreated control specimen swatches in the jar immediately after inoculation (at "0" contact time)

If "B and C" are not similar, the larger number should be used. If "B" and "C" are not significantly different, $(B + C)/2$ should be used as follows:

3) $100(D - A/D) = R$

where:

$D = (B + C)/2$

11.3 If an untreated control is not available, use the following calculation which allows for any background organisms that might interfere with the test.

$$Bg = 100\,[(B - E) - (A - F)/B - E]$$

where:

A, B = (see 11.2)

E = the number of bacteria initially recovered from the uninoculated treated test sample (existing background organisms)

F = The number of bacteria recovered from the uninoculated, pre-wet treated test sample after incubation in the jar over the desired contact period (existing background organisms after contact period)

Bg = background organisms

11.4 For a valid test there should be: (1) "0" colonies of test organism recovered from the uninoculated treated test specimen swatches and (2) a significant increase in the numbers of bacteria recovered from the inoculated untreated control specimen swatches incubated for the specified contact time over the numbers of bacteria recovered from the inoculated untreated specimen swatches at "0" contact time (immediately after inoculation). This applies only if dilution was made in broth (see 10.1.4 and 13.6).

11.5 Report percent reduction of bacteria by the specimen treatment against each test organism.

11.6 The criterion for passing the test must be determined by the interested parties.

11.7 Report the dilution medium used.

12. Precision and Bias

12.1 Studies (see 13.9) indicate the following *within-laboratory* precision of the Standard Plate Count (SPC) Test: (a) *among-analyst* variation of 18% and (b) *within-analyst* variation of 8%.

13. Notes and References

13.1 Publication available from U.S. De-

*Using a 1 mL pipette, pod the inoculum carefully onto the fabric. If a strain of *Pseudomonas* that forms a pellicle is used, avoid including fragments of the pellicle in the inoculum.

partment of Health and Human Services CDC/NIH-HHS Publication No. (CDC) 84-8395.

13.2 Booklet available from Publications Office, ACGIH, Kemper Woods Center, 1330 Kemper Meadow Dr., Cincinnati, OH 45240; tel: 513/742-2020.

13.3 Bacto-Peptone may be obtained from Difco Laboratories, 920 Henry St., Detroit MI 48201.

13.4 Beef extract may be obtained from Baltimore Biological Laboratories, 250 Schilling Cir., Cockeysville MD 21030; Difco Laboratories (address above); or Oxoid (USA) Ltd., 9017 Red Branch Rd., Columbia MD 21045.

13.5 Consistent and accurate testing requires maintenance of a pure, uncontaminated, nonmutant test culture. Avoid contamination by use of good sterile technique in plating and transferring. Avoid mutation by strict adherence to monthly stock transfers. Check culture purity by making streak plates periodically and observing for single species-characteristic type of colonies.

13.6 The dilution of the test organism may be prepared in sterile 0.85% saline solution or suitable buffer if a steady-state culture is desired during the contact period with a fabric or in the slurry inoculum carrier when hydrophobic fabrics are being tested.

13.7 A surfactant may be added to the dilution medium to enhance wetting of hydrophobic fabrics. The surfactant must be shown not to cause a reduction in bacterial numbers, by prior testing at the intended use concentration. Report the use and concentration of surfactant used.

13.8 If sterile distilled water is used in the place of a neutralizing solution, there will always be the possibility that some of the biocide will be carried over.

13.9 Peeler, J. T.; Leslie, J. W.; Messer, J. W. Replicate counting errors by analysts and bacterial colony counters. *J. Food Protection*, Vol. 45, 1982, pp 238-240.

APPENDIX V.b: AATCC Test Method 147

Qualtitative test method for bacterial efficacy on treated textile products.

Antibacterial Activity Assessment of Textile Materials: Parallel Streak Method

Developed in 1976 by AATCC Committee RA31; reaffirmed 1977, 1982, 1998; editorially revised 1980, 1982, 1983, 1986; revised 1987, 1988 (with title change), 1993.

Foreword

The Parallel Streak Method has filled a need for a relatively quick and easily executed qualitative method to determine antibacterial activity of diffusable antimicrobial agents on treated textile materials.

AATCC Method 100, Antibacterial Finishes on Textile Materials, Assessment of, is a quantitative procedure which is adequately sensitive but is cumbersome and time consuming for routine quality control and screening tests. Therefore, when the intent is to demonstrate bacteriostatic activity by the diffusion of the antibacterial agent through agar, Method 147 fulfills this need. In the Parallel Streak Method, the agar surface is inoculated making it easier to distinguish between the test organism and contaminant organisms which may be present on the unsterilized specimen. The Parallel Streak Method has proven effective over a number of years of use in providing evidence of antibacterial activity against both Gram positive and Gram negative bacteria.

1. Purpose and Scope

1.1 The objective is to detect bacteriostatic activity on textile materials. The results of using this procedure have been demonstrated by Committee RA31 to be reproducible by various laboratories working with materials containing residual amounts of antibacterial agents (as determined by chemical assay) after multiple standard washings. The method is useful for obtaining a rough estimate of activity in that the growth of the inoculum organism decreases from one end of each streak to the other and from one streak to the next resulting in increasing degrees of sensitivity. The size of the zone of inhibition and the narrowing of the streaks caused by the presence of the antibacterial agent permit an estimate of the residual antibacterial activity after multiple washings.

2. Principle

2.1 Specimens of the test material, including corresponding untreated controls of the same material, are placed in intimate contact with nutrient agar (see 7.1 and 7.4) which has been previously streaked with an inoculum of a test bacterium. After incubation, a clear area of interrupted growth underneath and along the sides of the test material indicates antibacterial activity of the specimen. A standard strain of bacteria is used which is specific to the requirements of the material under test. If no other bacterial species is specified, *Staphylococcus aureus* may be used as a representative Gram positive organism. Other recommended strains are listed below in Section 6.

3. Terminology

3.1 **activity**, n.—*of an antibacterial agent*, a measure of effectiveness of the agent.

3.2 **antibacterial agent**, n.—*in textiles*, any chemical which kills bacteria (bactericide) or interferes with the multiplication, growth or activity of bacteria (bacteriostat).

3.3 **zone of inhibition**, n.—clear area of no growth of a microorganism, cultured onto the surface of an agar growth medium, in proximity to the borders of a specimen placed in direct contact with this agar surface.

NOTE: A zone of inhibition occurs as a result of the diffusion of an antimicrobial agent from the specimen.

4. Safety Precautions

NOTE: These safety precautions are for information purposes only. The precautions are ancillary to the testing procedures and are not intended to be all inclusive. It is the user's responsibility to use safe and proper techniques in handling materials in this test method. Manufacturers MUST be consulted for specific details such as material safety data sheets and other manufacturer's recommendations. All OSHA standards and rules must also be consulted and followed.

4.1 This test should be performed only by trained personnel. The U.S. Department of Health and Human services publication *Biosafety in Microbiological and Biomedical Laboratories* should be consulted (see 13.1).

4.2 CAUTION: Some of the bacteria used in this test are pathogenic; i.e., capable of infecting humans and producing disease. Therefore, every necessary and reasonable precaution must be taken to eliminate this risk to the laboratory personnel and to personnel in the associated environment. Wear protective clothing and respiratory protection that prevents penetration by the bacteria.

4.3 Good laboratory practices should be followed. Wear safety glasses in all laboratory areas.

4.4 All chemicals should be handled with care.

4.5 An eyewash/safety shower should be located nearby for emergency use.

4.6 Sterilize all contaminated samples and test materials prior to disposal.

4.7 Exposure to chemicals used in this procedure must be controlled at or below levels set by government authorities (e.g., Occupational Safety and Health Administrations [OSHA] permissible exposure limits [PEL] as found in 29 CFR 1910.1000 of January 1, 1989). In addition, the American Conference of Governmental Industrial Hygienists (ACGIH) Threshold Limit Values (TLVs) comprised of time weighted averages (TLV-TWA), short term exposure limits (TLV-STEL) and ceiling limits (TLV-C) are recommended as a general guide for air contaminant exposure which should be met (see 13.2).

5. Uses and Limitations

5.1 The method is not suitable for materials which tend to encapsulate and prevent the diffusion of the antibacterial agent or contain antibacterial-neutralizing substances.

6. Test Organisms

6.1 Test bacteria:

6.1.1 *Staphylococcus aureus*, American Type Culture Collection No. 6538. Gram positive organism. (see 13.3)

6.1.2 *Klebsiella pneumoniae*, American Type Culture Collection No. 4352. Gram negative organism. (see 13.3)

6.1.3 Other suitable species can also be used depending on the intended end-use of the test sample.

6.2 Whenever possible, test the activity of the culture to be used against a standard control specimen (a positive control) with known antibacterial activity.

6.3 To determine whether the antibacterial activity is due to the antibacterial agent, test a specimen of the same material treated in exactly the same way with

whatever other finishing agents were used, but without the antibacterial agent. Many standard textile finishing chemicals, especially crease resistant and permanent press reagents, will often give strong antibacterial activity even after many washes.

7. Culture Medium

7.1 Suitable broth/agar media are Nutrient, Trypticase Soy and Brain-Heart Infusion.

Nutrient Broth:	
Peptone (Bacto-peptone) (see 13.5)	5 g
Beef extract (see 13.6)	3 g
Distilled water	to 1000 mL

7.2 Heat to a boil to disperse ingredients. Adjust to pH 6.8 ± 0.1 with 1*N* NaOH solution. (This is not necessary if prepared, dehydrated medium is used.)

7.3 Dispense in 10.0 ± 0.5 mL amounts in conventional bacteriological culture tubes (i.e., 125 × 17 mm). Plug and sterilize at 103 kPa (15 psi) for 15 minutes.

7.4 Nutrient agar (see 13.4). Add 1.5% bacteriological agar to nutrient (or appropriate) broth. Heat to boiling. Check pH and adjust to 7.1 ± 0.1 using NaOH solution if necessary. Dispense in 15.0 ± 0.5 mL amounts in conventional bacteriological culture tubes, plug, and sterilize at 103 kPa (15 psi) for 15 min. (May be sterilized in 1,000 mL borosilicate glass flasks and petri dishes poured from this.)

8. Maintenance of Culture of Test Organisms

8.1 Using a 4 mm inoculating loop, transfer the culture daily in nutrient (or appropriate medium) broth for not more than two weeks. At the conclusion of two weeks, make a fresh transplant from stock culture. Incubate cultures at 37 ± 2°C (99 ± 3°F).

8.2 Maintain stock cultures on nutrient or appropriate agar slants. Store at 5 ± 1°C (41 ± 2°F) and transfer once a month to fresh agar (see 13.7).

9. Test Specimens

9.1 Test specimens (non-sterile) are cut by hand or with a die. They may be any convenient size. Rectangular specimens cut 25 × 50 mm are recommended. A 50 mm length permits the specimens to lie across 5 parallel inoculum streaks each of diminishing width from about 8 mm to 4 mm wide.

10. Procedure

10.1 Dispense sterilized nutrient (or appropriate medium) agar [cooled to 47 ± 2°C (117 ± 4°F)] by pouring 15 ± 2 mL into each standard (15 × 100 mm) flat bottomed petri dish. Allow agar to gel firmly before inoculating.

10.2 Prepare inoculum by transferring 1.0 ± 0.1 mL of a 24 h broth culture into 9.0 ± 0.1 mL of sterile distilled water contained in a test tube or small flask. Mix well using appropriate agitation.

10.3 Using a 4 mm inoculating loop, load one loopful of the diluted inoculum and transfer to the surface of the sterile agar plate by making five streaks approximately 60 mm in length, spaced 10 mm apart covering the central area of a standard petri dish (see 10.1) without refilling the loop. Take care not to break the surface of the agar while making the streaks.

10.4 Gently press the test specimen transversely across the five inoculum streaks to ensure intimate contact with the agar surface. This may be accomplished more easily by pressing the specimen to the agar surface with a biological section lifter or with a spatula which has been sterilized by flaming and then air cooled immediately before use.

10.5 If the specimen curls, preventing intimate contact with the inoculated surface, place sterile glass slides on the ends of the specimen to hold it in place.

10.6 Incubate at 37 ± 2°C (99 ± 4°F) for 18-24 h.

11. Evaluation

11.1 Examine the incubated plates for interruption of growth along the streaks of inoculum beneath the specimen and for a clear zone of inhibition beyond its edge. The average width of a zone of inhibition along a streak on either side of the test specimen may be calculated using the following equation:

$$W = (T - D)/2$$

where:

W = width of clear zone of inhibition in mm
T = total diameter of test specimen and clear zone in mm
D = diameter of the test specimen in mm

11.2 The size of the zone cannot be construed as a quantitative evaluation of antibacterial activity. Treated materials should be compared to an untreated corresponding material and a material specimen with known bacteriostatic activity. Report of results will include an observation of zones of inhibition and growth under the specimen if present. The criterion for passing the test must be agreed upon by the interested parties. To constitute acceptable antibacterial activity, there must be no bacterial colonies directly under the sample in the contact area.

12. Precision and Bias

12.1 The precision and bias of this test method are being established.

13. Notes and References

13.1 Publication available from U.S. Department of Health and Human Services—CDC/NIH-HHS Publication No. (CDC) 84-8395.

13.2 Booklet available from Publications office, ACGIH, Kemper Woods Center, 1330 Kemper Meadow Dr., Cincinnati OH 45240; tel: 513/742-2020.

13.3 American Type Culture Collection, 12301 Parklawn Drive, Rockville MD 20852.

13.4 Nutrient Agar can be obtained from Difco Laboratories, 920 Henry St., Detroit MI 48201 and from Baltimore Biological Laboratories, 250 Schilling Circle, Cockeyville MD 21030.

13.5 Peptone from Difco Laboratories (address above), or Thiotone from Baltimore Biological Laboratories (address above).

13.6 Beef extract may be obtained from Baltimore Biological Laboratories (address above); Difco Laboratories (address above); or Oxoid USA Inc., 9017 Red Branch Road, Columbia MD 21045.

13.7 Consistent and accurate testing requires maintenance of a pure, uncontaminated, non-mutant test culture. Avoid contamination by using good sterile technique in plating and transferring. Avoid mutation by strict adherence to monthly stock transfers. Check culture purity by making streak plates periodically and observing for a single species-characteristic type of colonies.

APPENDIX V.c: AATCC Test Method 30, pt III

Qualtitative test method for anti-fungal efficacy on treated textile products.

Antifungal Activity, Assessment on Textile Materials: Mildew and Rot Resistance of Textile Materials

Developed in 1946 by AATCC Committee RA31; revised 1952, 1957, 1971, 1981, 1987, 1988 (with title change), 1993, 1999; reaffirmed 1970, 1974, 1979, 1989, 1998; editorially revised and reaffirmed 1986.

1. General Purpose and Scope

1.1 The two purposes of this test method are to determine the susceptibility of textile materials to mildew and rot and to evaluate the efficacy of fungicides on textile materials.

2. Principle

2.1 Tests I, II, III and IV can be used, singly or in combination, depending on the type of exposure to which the textile material will be subjected. For example, if the final product will come in contact with soil, Test I, which simulates this exposure, should be used; if the finished product will never come in contact with soil or tropical conditions, a much less severe test (e.g., II or III) should be used. Test II is specifically designed for cellulose-containing materials while Test III is for all others. For all materials intended for outdoor and above ground use, Test IV should be used. The two important considerations when evaluating textile materials in relation to fungal growth are (1) the actual deterioration of the textile product (rot), and (2) growth not necessarily deteriorating the product but making it unsightly (mildewy) often with an unpleasant and musty odor.

2.2 Certain pre-exposures of textile products may be indicated when specific end-uses are critical (see Appendix A). When the end-use will be near high temperature and the fungicide may be volatile, a preliminary oven exposure may be desired. When the end-use will be in tropical exposures or outside with rainfall present, a leaching exposure should be performed before mildew evaluation is made. When at all possible, the textile material should be exposed to the expected conditions of use prior to performing this test.

3. Terminology

3.1 **mildew resistance,** n.—*in textiles*, resistance to development of unsightly fungal growths and accompanying unpleasant, musty odors on textile materials exposed to conditions favoring such growths.

3.2 **rot resistance,** n.—*in textiles*, resistance to deterioration of a textile material as a result of fungal growth in or on it.

NOTE: Such deterioration is normally assessed by measuring loss in tensile strength.

4. Safety Precautions

NOTE: These safety precautions are for information purposes only. The precautions are ancillary to the testing procedures and are not intended to be all inclusive. It is the user's responsibility to use safe and proper techniques in handling materials in this test method. Manufacturers MUST be consulted for specific details such as material safety data sheets and other manufacturer's recommendations. All OSHA standards and rules must also be consulted and followed.

4.1 This test should be performed only by trained personnel. The U.S. Department of Health and Human Services publication *Biosafety in Microbiological and Biomedical Laboratories* should be consulted (see 24.1).

4.2 CAUTION: Some of the fungi used in these tests are allergenic and pathogenic; i.e., capable of infecting humans and producing disease. Therefore, every necessary and reasonable precaution must be taken to eliminate this risk to the laboratory personnel and to personnel in the associated environment. Wear protective clothing, respiratory protection, and impervious gloves when working with the organisms. NOTE: Choose respiratory protection that prevents penetration by the spores.

4.3 Good laboratory practices should be followed. Wear safety glasses in all laboratory areas.

4.4 All chemicals should be handled with care.

4.5 An eyewash/safety shower should be located nearby for emergency use.

4.6 Sterilize all contaminated samples and test materials prior to disposal.

4.7 Exposure to chemicals used in this procedure must be controlled at or below levels set by government authorities (e.g., Occupational Safety and Health Administration's [OSHA] permissible exposure limits [PEL] as found in 29 CFR 1910-1000 of January 1, 1989). In addition, the American Conference of Governmental Industrial Hygienists (ACGIH) Threshold Limit Values (TLVs) comprised of time weighted averages (TLV-TWA), short term exposure limits (TLV-STEL) and ceiling limits (TLV-C) are recommended as a general guide for air contaminant exposure which should be met (see 24.2).

Test I
Soil Burial

5. Scope

5.1 This procedure is generally considered to be the most severe test for textile products. Only those specimens that will come in direct contact with soil—such as sandbags, tarpaulins, tents—need to be tested by this procedure. It can also be used for testing experimental textile fungicides.

6. Test Specimens

6.1 Prepare the fabric specimens with dimensions 15.0 ± 1.0 × 4.0 ± 0.5 cm (6.0 ± 0.4 × 1.5 ± 0.2 in.) with the long dimension parallel to the warp and unraveling to 2.5 ± 0.1 cm width (1.0 ± 0.04 in.), or, in the case of fabric with less than 20 threads per 2.5 cm (1.0 in.) to a predetermined number of threads to give a specimen 2.5 ± 1.0 cm in width (1.0 ± 0.4 in.). A sample cutter can also be used (see 24.3). The number of specimens will vary according to the number of variables. The suggested number of specimens is five for each treatment, control and reference fabric.

7. Test Procedure

7.1 Viability control: Expose untreated cotton cloth 271 g/m^2 (8 oz/yd^2) in the soil bed for seven days during the test period to verify fungal activity. The soil bed shall be considered as satisfactory if the viability control fabric loses 90% breaking strength after seven days exposure.

7.2 Soil Bed: Place the air-dry test soil (see 24.4) in trays, boxes or suitable containers to a depth of 13.0 ± 1.0 cm (5.1 ± 0.4 in.) and bring to optimum moisture content by gradual addition of water accompanied by mixing to avoid puddling. After allowing it to stand for 24 h, sieve it through a 6.4 mm (0.25 in.) mesh screen. Maintain uniform moisture content by covering the soil container with a suitable lid. The moisture content of the soil during the test period shall be maintained between 25 ± 5% (based on dry weight). If the surrounding air is maintained at higher than 83 ± 3% relative humidity,

the loss of moisture is negligible.

7.3 Incubation: Bury the specimens horizontally on 10.0 ± 1.0 cm (3.9 ± 0.4 in.) of soil, spaced at least 2.5 cm (1.0 in.) apart and then cover with 2.5 ± 0.5 cm (1.0 ± 0.2 in.) of test soil. Incubation periods can vary from 2-16 weeks, depending on severity of service requirements, and other factors of importance to the interested parties. Maintain the temperature at 28 ± 1°C (82 ± 2°F) during the test period.

8. Evaluation and Report

8.1 Strength loss determination: Remove specimens, gently wash with water, dry at room temperature for 22 ± 4 h and then condition in an atmosphere of 64 ± 2% humidity and a temperature of 24 ± 3°C (76 ± 6°F) for 24 h. Determine breaking strength by the method outlined in ASTM D 5035, Standard Test Method for Breaking Force and Elongation of Textile Fabrics (Strip Force), using 25 × 75 mm (1 × 3 in.) jaw faces. The gage length is determined as 25% of the specimen length. Testing can be performed every two weeks or as specified by the end-user.

8.2 Report: Report the length of exposure to the soil bed, percent retained breaking strength when compared to the unexposed textile, any pre-exposure of specimens before burying and percent retained breaking strength of untreated specimen and/or viability control.

Test II
Agar Plate, *Chaetomium Globosum*

9. Scope

9.1 This procedure is used for evaluating rot resistance of cellulose-containing textile materials that will not come in contact with soil. It may also be used for determining uniformity of fungicide treatment.

10. Test Specimens

10.1 Proceed as in Section 6, if strength loss is to be determined. If only a visual examination is performed, a minimum of five samples is required. However, any number can be tested depending on end-users request. Cut 3.8 ± 0.5 cm (1.5 ± 0.2 in.) diameter discs from both treated and untreated samples.

11. Test Procedure

11.1 Organism: *Chaetomium globosum*. American Type Culture Collection No. 6205 (see 24.5).

11.2 Culture medium (see 24.6): The mineral salts agar should have the following composition:

Ammonium nitrate, NH_4NO_3	3.0 g
Potassium dihydrogen phosphate, KH_2PO_4	2.5 g
Dipotassium hydrogen phosphate, K_2HPO_4	2.0 g
Magnesium sulfate, $MgSO_4 \cdot 7H_2O$	0.2 g
Ferrous sulfate, $FeSO_4 \cdot 7H_2O$	0.1 g
Agar	20.0 g
Distilled water	to 1000 mL

11.3 Inoculum: Place agar solution in any desired container such as test tube, French square bottle, Erlenmeyer flask, or petri dish. Sterilize in an autoclave at 103 kPa (15 psi) and 121°C (250°F) for 15 min and cool in a position which affords maximum inoculation surface. After the agar has hardened, under aseptic conditions, place on its surface a disc of filter paper previously sterilized by dry heat in an oven at 71 ± 3°C (160 ± 5°F) for 1 h. Streak the filter paper with spores of *Chaetomium globosum* by use of a sterile needle. Incubate at 28 ± 1°C (82 ± 2°F) for approximately 10-14 days to produce abundant growth. Remove the filter paper from the container and add it to 50 ± 2 mL of sterile distilled water containing a few glass beads and shake vigorously to bring the spores into suspension. Use this suspension for inoculum in 11.5.

11.4 Culture chamber: Melt mineral salts agar of the composition specified in 11.2 in an autoclave and distribute into any convenient container. Sterilize under conditions given in 11.3 and leave undisturbed until the agar hardens.

11.5 Inoculation: Pre-wet the specimens (but do not rub or squeeze) in water containing 0.05% of a nonionic wetting agent (see 24.7) and place in contact with the hardened agar medium in each container under aseptic conditions. Distribute 1.0 ± 0.1 mL of the inoculum evenly over the 15.0 ± 1.0 × 4.0 ± 0.5 cm (6.0 ± 0.4 × 1.5 ± 0.2 in.) specimens by means of a sterile pipette. Use 0.2 ± 0.01 mL of inoculum for the 3.8 ± 0.5 cm (1.5 ± 0.2 in.) discs. Set up a control specimen, cellulose filter paper or untreated control, in a similar way by using 1.0 ± 0.1 or 0.2 ± 0.01 mL of sterile water.

12. Evaluation and Report

12.1 Strength loss evaluation: Proceed as per 8.1 and report the change in breaking strength as compared to the sample before exposure or the control if available.

12.2 Visual assessment: Report the extent of fungal growth on the discs, using a microscope (50X) where necessary, in accordance with the following scheme:

Observed Growth

No growth

Microscopic growth (visible only under the microscope)

Macroscopic growth (visible to the eye)

Test III
Agar Plate, *Aspergillus Niger*

13. Score

13.1 Certain fungi, of which *Aspergillus niger* is one, can grow on textile products without causing measurable breaking strength loss within a laboratory experimental time frame. Nonetheless, their growth may produce undesirable and unsightly effects. This procedure is used to evaluate textile specimens where growth of these fungi is important.

14. Test Specimens

14.1 Cut duplicate 3.8 ± 0.5 cm (1.5 ± 0.2 in.) diameter discs from both treated and untreated samples. Other shapes and sizes can be used provided any anticipated size of the growth-free zone is taken into consideration.

15. Test Procedure

15.1 Organism: *Aspergillus niger*, American Type Culture Collection No. 6275 (see 24.5).

15.2 Culture medium: Proceed as per 11.2. Other suitable media are Czapek (Dox) Agar and Sabouraud Dextrose Agar (see 24.8).

15.3 Inoculum: Add scrapings from a ripe (7-14 days) fruiting culture of *Aspergillus niger* grown on the medium described in 11.2 containing 3.0 ± 0.1% glucose, to a sterile Erlenmeyer flask containing 50 ± 1 mL of sterile water and a few glass beads. Shake the flask thoroughly to bring the spores into suspension. Use this suspension as the inoculum.

15.4 Inoculation: If the test medium contains glucose, distribute evenly 1.0 ± 0.1 mL of the inoculum over the surface of the agar. Pre-wet the discs (but do not rub or squeeze) in water containing 0.05% of a nonionic wetting agent (see 24.7) and place on the agar surface. Distribute evenly over each disc 0.2 ± 0.01 mL of the inoculum by means of a sterile pipette. If the test medium does not contain glucose, a negative control fabric is required to ensure inoculum viability. Incubate all specimens at a temperature of 28 ± 1°C (82 ± 2°F) for 14 days when mineral salts agar is used and for seven days when 3% glucose is added to the mineral salts agar.

16. Evaluation and Report

16.1 At the end of the incubation period, report the percentage of surface area of the discs covered with the growth of *Aspergillus niger*, using a microscope (50X) where necessary, in accordance with the following scheme:

Observed Growth

No growth (if present, report the size of the growth-free zone in mm)

Microscopic growth (visible only under the microscope)

Macroscopic growth (visible to the eye)

Test IV
Humidity Jar, Mixed Spore Suspension

17. Scope

17.1 This test method is designed to determine the fungistatic effectiveness of treatments intended to control mildew and non-pathogenic fungal growth on articles or surfaces composed of textile materials intended for outdoor and above ground use and which are usually waterproofed.

17.2 For this test method visual assessment is used. Additionally, breaking strength may be determined by method as per 8.1.

18. Principle

18.1 Treated and untreated, nutrient-saturated strips of fabric are sprayed with a mixed spore suspension of mildew-causing organisms and incubated at 90 ± 2% relative humidity. Mildew growth on treated and untreated strips is rated at weekly intervals for up to four weeks.

19. Apparatus

19.1 Glassware: 500 mL French square jars or equivalent with fitting screw caps. Caps are modified by center drilling and inserting an appropriate size stainless steel or brass bolt to which a hook (formed from a 6.5 ± 0.5 cm length [2.6 ± 0.2 in.] of #22 nickel-chromium wire or other non-corrosive wire) is attached.

19.2 Plastic paper clips or nylon thread to suspend the specimens from the screw caps of the French jars.

19.3 Atomizer, DeVilbiss #152 (or equivalent) operated at 69 kPa (10 psi).

19.4 Counting chamber suitable for determining spore concentrations; e.g., hemocytometer.

20. Test Specimens

20.1 The specimens are prepared by cutting 2.5 ± 0.5 cm × 7.5 ± 0.5 cm (1.0 ± 0.2 × 3.0 ± 0.2 in.) strips from sample weighing 170.0 ± 34.0 g/m² (5.0 ± 1.0 oz/yd²). For heavier fabrics use strips 2.0 ± 0.5 cm × 2.0 ± 0.5 cm (0.8 ± 0.2 × 0.8 ± 0.2 in.).

20.2 Use at least four specimens of each treated and untreated fabric.

20.3 Untreated fabric strips, identical in all other respects to the treated specimens under test, are required to establish the test validity. If untreated fabrics are not available, use a control cloth with the following requirements:

Cotton:	American type, good middling
Warp:	18.5 tex z 886 × 2S748
Weft:	30 tex z 630 × 2S748
Weave:	Plain 34 ends/cm and 17 picks/cm
Mass/unit area:	230.0 g/m² (6.8 oz/yd²)
Finish:	Scoured only

21. Test Procedures

21.1 Organisms:

21.1.1 *Aspergillus niger*, American Type Culture Collection No. 6275.

21.1.2 *Penicillium funiculosum*, American Type Culture Collection No. 10509.

21.1.3 *Trichoderma viride*, American Type Culture Collection No. 28020 (see 24.5).

21.2 Culture medium:

21.2.1 Maintain stock culture of each of test tube slants of potato dextrose agar for *A. niger* and *P. funiculosum*, and malt extract agar for *T. viride* (see 24.6 and 24.8 for media).

21.2.2 Incubate new stock culture 7-10 days at 25 ± 1°C (77 ± 2°F), then store at 2-10°C (36-50°F).

21.3 Preparation of conidial suspensions:

21.3.1 Conidial suspensions of fungal organisms are prepared by adding 10 mL of a sterile 0.5% saline solution containing 0.05% of a non-fungicidal wetting agent (see 24.7) to a 7-10 day agar culture.

21.3.2 Scrape the surface of the culture gently with a platinum or nichrome wire to liberate the spores. Agitate the liquid slightly to disperse the spores without detaching mycelial fragments, and gently decant the mold suspension into a flask containing a few glass beads.

21.3.3 Shake the dispersion vigorously to break up any clumps of spores and then filter through a thin layer of sterile cotton or glass wool. Conidial suspensions may be stored at 6 ± 4°C (43 ± 7°F) for up to four weeks.

21.3.4 Inoculum for test should be adjusted using a hemocytometer or a Petroff-Hausser bacteria counter to contain five million conidia per mL on day of use by appropriate dilution of stock suspension with saline solution.

21.4 Preparation of test specimens:

21.4.1 To ensure luxuriant growth, both the test and control strips must be saturated with a sterilized glycerol nutrient solution of the following composition: 97.6% distilled water, 2.0% glycerol, 0.1% K_2HPO_4, 0.1% NH_4NO_3, 0.05% $MgSO_4 \cdot 7H_2O$, 0.1% yeast extract and 0.05% of a nonionic wetting agent (see 24.7). Adjust the pH to 6.3 ± 0.1. Sufficient nutrient solution should be prepared to saturate all the specimens used in a single test.

21.4.2 Soak each strip in nutrient for three minutes or until saturated. Squeeze excess liquid and allow fabric strips to air dry before proceeding with application of test fungi.

21.5 Pre-mix equal volumes of well agitated conidial suspensions of *A. niger*, *T. viride* and *P. funiculosum*. Evenly distribute 1.0 ± 0.1 mL of the above suspension onto both sides of each specimen either by spraying or by means of a pipette.

21.6 Suspend fabric strips using plastic paper clips or nylon thread from the caps of individual jars containing 90 ± 3 mL of water each. Hook position must be adjusted so that the bottom ends of attached strips are all at a uniform height above the water level. The caps are tightened, then backed off one-eighth turn to allow for some ventilation.

21.7 Incubate at 28 ± 1°C (82 ± 2°F) for 14 days (for non-coated cellulosic textiles) or 28 days (for non-cellulosic or coated cellulosic textiles).

22. Evaluation and Report

22.1 A record of the percent of surface area covered with fungal growth for each strip is made at weekly intervals, or until heavy growth occurs on each sample replicate. Using a microscope (50X) where necessary, assess each specimen in accordance with the scheme given in 12.2.

22.2 After seven days each control strip must show macroscopic growth. If this is not the case repeat the test since test conditions were not valid.

22.3 Any adverse effect of incubation on the fabric; e.g., color changes, flexibility, water repellency, should be qualitatively reported.

22.4 Strength loss determination can be carried out as per 8.1.

22.5 The results of this test method have to be correlated to claims and directions for use recommended for the mildew control product plus any other criteria agreed upon by the interested parties.

23. Precision and Bias

23.1 The precision and bias of this test method are being established. If the breaking strength loss is determined, then refer to the statement given in ASTM D 5035.

24. Notes

24.1 Publication available from U.S. Department of Health & Human Services—CDC/NIH-HHS Publication No. (CDC) 84-8395.

24.2 Booklet available from Publications Office, ACGIH, Kemper Woods Center, 1330 Kemper Meadow Dr., Cincinnati OH 45240; tel: 513/742-2020.

24.3 A JDC Precision sample cutter may be

purchased from Thwing-Albert Instrument Co., 10960 Dutton Road, Philadelphia PA 19154; tel: 215/637-0100; fax: 215/632-8370; Cat. #99 Model JOC25.

24.4 Types of soil which have been found satisfactory for this purpose include garden and naturally fertile topsoils, composts and non-sterile greenhouse potting soils. An equal blend of good topsoil, well rotted and shredded manure, and coarse sand should be used. These usually possess the proper physical characteristics, along with an organic content sufficient to ensure a high degree of microbial activity and the presence of cellulose destroying organisms. The optimum moisture content of these is about 30% moisture above oven dry weight.

24.5 *Chaetomium globosum* ATCC 6205, *Aspergillus niger*, ATCC 6275, *Penicillium funiculosum*, ATCC 10509 and *Trichoderma viride*, ATCC 28020, can be purchased from the American Type Culture Collection, 12301 Parklawn Dr., Rockville MD 20852.

24.6 Culture medium having composition prescribed in 11.2 (Mineral Salts) can be purchased from Baltimore Biological Laboratories, 250 Schilling Cir., Cockeysville MD 21030.

24.7 Triton™ X-100 (Rohm & Haas Co., Philadelphia PA 19104) has been found to be a good wetting agent. Suitable alternatives are dioctyl sodium sulfosuccinate or N-methyltauride derivatives.

24.8 These culture media can be bought either from Baltimore Biological Laboratories (see 24.6) or Difco Laboratories, 920 Henry St., Detroit MI 48201.

24.9 ASTM D 5035 can be used for yarn, thread, cordage or tape (see 12.1).

24.10 If testing is being performed for Federal Standards, use AATCC 30-2. Other organisms can be used: *Myrothecium verrucaria* ATCC 9095, QM 460; *Trichoderma* SP ATCC 9645, QM 365; *Memmoniella echinata* ATCC 11973, QM 1225; *Aspergillus niger* ATCC 6275, QM 458; *Aspergillus clavatus* ATCC 18214, QM 862.

Appendix A
Pre-Treatments

A1. Leaching

A1.1 The leaching should conform in principle to the following procedure: Water from a faucet is passed through a tubing into leaching vessels, care being taken that specimens having different amounts of the same treatment are in separate vessels. Flow is adjusted to ensure a complete change of water not less than three times in 24 h. The delivery tubes are inserted down through the center of wire mesh cylinders in the leaching vessels and held in the wire cylinders with rubber bands and leached for 24 h. The pH and temperature of the water is recorded and included in the report of the test results.

A2. Volatilization

A2.1 Standard specimens of the fabric to be tested are exposed continuously to dry heat at 100-105°C (212-221°F) for 24 h in a well ventilated oven.

A3. Weathering

A3.1 Portions of the material to be tested are exposed on a series of weathering racks at 45° to the horizontal facing South, between April 1 and October 1, in such a manner as to avoid sagging or flapping. It is recommended that such racks be set up in at least four locations within the United States; e.g., Washington DC; Miami FL; New Orleans LA; and suitable desert locations.

APPENDIX V.d: Microbiology Test Results

Complete test results for microbiological testing for preliminary studies.

Table I. Natick Testing Unwashed													
			Active Conc.		Test Method 100					Test Method 147			
Item	Sample ID	Active	ppm		Staph	Strep	Coryn	Pseud		Staph	Strep	Coryn	Pseud
ACU	10-A-0-R1	Triclosan	980		99.90%	99.80%	99.90%	NR		6	3	2	NZ
	10-A-0-R2		1100		99.90%	99.80%	99.90%	99.90%		7	2	2	NZ
	10-A-0-R3		1100		99.90%	99.80%	99.90%	99.90%		7	2	NZ	NZ
	10-B-0-R1	Quat silane			99.90%	99.80%	99.90%	99.90%		NA	1	NZ	NZ
	10-B-0-R2				99.90%	99.80%	99.90%	99.90%		NZ	2	1	NZ
	10-B-0-R3				99.90%	99.80%	99.90%	99.90%		NZ	2	1	NZ
	10-C-0-R1	PHMB			99.90%	99.80%	99.90%	NR		1	2	1	NZ
	10-C-0-R2				99.90%	99.80%	99.90%	NR		2	3	2	NZ
	10-C-0-R3				99.90%	99.80%	99.90%	NR		2	4	3	NZ
	10-D-0-R1	Control			99.60%	99.80%	99.90%	NR		NZ	2	NZ	NZ
	10-D-0-R2				99.60%	99.80%	98.90%	NR		NZ	NZ	NZ	NZ
	10-D-0-R3				NR	99.80%	86.70%	NR		NZ	2	NZ	NZ
T-shirt	20-A-0-R1	Triclosan	4500		99.90%	99.80%	NR	NR		7	NZ	NZ	NZ
	20-A-0-R2		3000		97.00%	99.80%	NR	NR		6	NZ	NZ	NZ
	20-A-0-R3		3900		97.50%	99.80%	NR	NR		6	NZ	NZ	NZ
	20-B-0-R1	Quat silane			99.90%	99.80%	99.90%	NR		NZ	1	NZ	NZ
	20-B-0-R2				99.90%	99.80%	99.90%	NR		NZ	1	NZ	NZ
	20-B-0-R3				99.90%	99.80%	99.90%	NR		NZ	NZ	1	NZ
	20-C-0-R1	PHMB			99.90%	99.90%	99.90%	99.90%		NZ	2	2	NZ
	20-C-0-R2				99.90%	99.90%	99.90%	99.90%		NZ	1	2	NZ
	20-C-0-R3				99.90%	99.90%	99.90%	99.90%		NZ	2	1	NZ
	20-D-0-R1	Control			99.90%	99.90%	99.90%	NR		NZ	NZ	NZ	NZ
	20-D-0-R2				99.90%	99.90%	NR	NR		NZ	NZ	NZ	NZ
	20-D-0-R3				NR	99.90%	99.90%	NR		NZ	NZ	NZ	NZ

Table I. Natick Testing Unwashed (cont'd)													
			Active Conc.		Test Method 100					Test Method 147			
Item	Sample ID	Active	ppm		Staph	Strep	Coryn	Pseud		Staph	Strep	Coryn	Pseud
Boot sock	30-A-0-R1	Triclosan	6300		99.90%	99.90%	99.90%	99.90%		13	2	4	NZ
	30-A-0-R2		5500		99.90%	99.90%	99.90%	99.90%		10	3	2	NZ
	30-A-0-R3		7200		99.90%	99.90%	99.90%	99.90%		15	2	3	NZ
	30-B-0-R1	Quat silane			99.90%	99.90%	99.90%	99.90%		NZ	1	NZ	NZ
	30-B-0-R2				99.90%	99.90%	99.90%	NR		NZ	1	1	NZ
	30-B-0-R3				99.90%	99.90%	99.90%	NR		NZ	1	NZ	NZ
	30-C-0-R1	PHMB			99.90%	99.90%	99.90%	NR		2	3	3	NZ
	30-C-0-R2				99.90%	99.90%	99.90%	NR		2	3	6	NZ
	30-C-0-R3				99.90%	99.90%	99.90%	NR		2	5	5	NZ
	30-D-0-R1	Control			99.90%	99.90%	99.90%	NR		NZ	2	3	NZ
	30-D-0-R2				99.90%	99.90%	99.90%	NR		1	3	1	NZ
	30-D-0-R3				99.90%	99.90%	99.90%	NR		NZ	4	7	NZ

Table II. Natick Testing 5 Washes													
			Active Conc.		Test Method 100					Test Method 147			
Item	Sample ID	Active	ppm		Staph	Strep	Coryn	Pseud		Staph	Strep	Coryn	Pseud
ACU	10-A-5-R1	Triclosan	1100		91.90%	60.20%	NR	NR		9	3	I	NZ
	10-A-5-R2		1000		96.60%	NR	NR	NR		12	I	I	NZ
	10-A-5-R3		1100		97.00%	NR	NR	NR		5	I	I	NZ
	10-B-5-R1	Quat silane			NR	99.80%	NR	NR		NZ	3	2	NZ
	10-B-5-R2				NR	99.80%	64.10%	NR		NZ	4	2	NZ
	10-B-5-R3				NR	99.80%	NR	NR		1	4	2	NZ
	10-C-5-R1	PHMB			99.90%	99.80%	99.90%	NR		1	3	6	NZ
	10-C-5-R2				99.90%	99.80%	99.90%	NR		1	3	4	NZ
	10-C-5-R3				99.50%	99.80%	99.90%	NR		1	2	11	NZ
	10-D-5-R1	Control			NR	NR	NR	NR		NZ	NZ	NZ	NZ
	10-D-5-R2				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	10-D-5-R3				NR	NR	NR	NR		NZ	12	NZ	NZ
T-shirt	20-A-5-R1	Triclosan	3700		99.50%	99.60%	NR	NR		7	NZ	NZ	NZ
	20-A-5-R2		3500		NR	99.30%	NR	24.40%		6	NZ	NZ	NZ
	20-A-5-R3		3900		NR	99.30%	NR	NR		8	NZ	NZ	NZ
	20-B-5-R1	Quat silane			NR	99.10%	NR	NR		NZ	NZ	NZ	NZ
	20-B-5-R2				NR	98.80%	NR	NR		NZ	NZ	NZ	NZ
	20-B-5-R3				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-C-5-R1	PHMB			99.90%	99.80%	99.60%	NR		NZ	NZ	NZ	NZ
	20-C-5-R2				NR	99.80%	99.90%	NR		NZ	NZ	I	NZ
	20-C-5-R3				99.90%	99.80%	99.90%	NR		NZ	I	I	NZ
	20-D-5-R1	Control			NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-D-5-R2				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-D-5-R3				NR	NR	NR	NR		NZ	NZ	NZ	NZ

Table II. Natick Testing 5 Washes (cont'd)													
			Active Conc.		Test Method 100					Test Method 147			
Item	Sample ID	Active	ppm		Staph	Strep	Coryn	Pseud		Staph	Strep	Coryn	Pseud
Boot sock	30-A-5-R1	Triclosan	6300		99.70%	99.80%	NR	NR		9	I	NZ	NZ
	30-A-5-R2		5500		99.50%	99.80%	15.00%	NR		9	1	NZ	NZ
	30-A-5-R3		7200		98.90%	99.80%	9.00%	NR		8	1	NZ	NZ
	30-B-5-R1	Quat silane			NR	99.80%	99.90%	NR		NZ	2	2	NZ
	30-B-5-R2				NR	99.80%	98.50%	68.90%		NZ	7	2	NZ
	30-B-5-R3				NR	99.80%	30.00%	NR		NZ	9	2	NZ
	30-C-5-R1	PHMB			99.90%	99.80%	99.90%	NR		NZ	NZ	NZ	NZ
	30-C-5-R2				99.90%	99.80%	99.90%	NR		NZ	I	I	NZ
	30-C-5-R3				99.90%	99.80%	99.90%	NR		NZ	4	I	NZ
	30-D-5-R1	Control			NR	99.80%	92.60%	NR		NZ	NZ	NZ	NZ
	30-D-5-R2				NR	99.80%	30.00%	NR		2	NZ	NZ	NZ
	30-D-5-R3				NR	94.20%	99.80%	NR		NZ	NZ	NZ	NZ

Table III. Natick Testing 10 Washes													
			Active Conc.		Test Method 100					Test Method 147			
Item	Sample ID	Active	ppm		Staph	Strep	Coryn	Pseud		Staph	Strep	Coryn	Pseud
ACU	10-A-10-R1	Triclosan	160		NR	85.90%	NR	NR		3	I	I	NZ
	10-A-10-R2		190		45.90%	NR	NR	NR		3	I	I	NZ
	10-A-10-R3		170		17.70%	74.10%	NR	NR		4	I	I	NZ
	10-B-10-R1	Quat silane			NR	99.90%	99.90%	NR		I	2	2	NZ
	10-B-10-R2				NR	99.90%	9.90%	NR		I	1	3	NZ
	10-B-10-R3				NR	99.90%	99.90%	NR		I	4	3	NZ
	10-C-10-R1	PHMB			93.20%	99.90%	99.90%	NR		1	2	2	NZ
	10-C-10-R2				98.60%	99.90%	99.90%	NR		1	2	2	NZ
	10-C-10-R3				99.90%	99.90%	99.90%	NR		1	2	4	NZ
	10-D-10-R1	Control			NR	NR	99.90%	NR		NZ	NZ	NZ	NZ
	10-D-10-R2				NR	NR	99.90%	NR		NZ	NZ	NZ	NZ
	10-D-10-R3				NR	NR	99.90%	NR		NZ	NZ	NZ	NZ
T-shirt	20-A-10-R1	Triclosan	3200		99.40%	97.10%	NR	NR		6	NZ	NZ	NZ
	20-A-10-R2		2900		99.90%	99.90%	NR	NR		7	NZ	NZ	NZ
	20-A-10-R3		3600		76.50%	99.90%	NR	NR		6	NZ	NZ	NZ
	20-B-10-R1	Quat silane			NR	99.00%	NR	NR		NZ	NZ	NZ	NZ
	20-B-10-R2				NR	90.40%	NR	NR		NZ	NZ	NZ	NZ
	20-B-10-R3				NR	99.90%	NR	NR		NZ	I	NZ	NZ
	20-C-10-R1	PHMB			99.90%	99.90%	33.40%	33.40%		NZ	NZ	2	NZ
	20-C-10-R2				99.90%	99.90%	NR	NR		NZ	NZ	I	NZ
	20-C-10-R3				99.90%	99.90%	NR	NR		NZ	NZ	I	NZ
	20-D-10-R1	Control			NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-D-10-R2				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-D-10-R3				NR	NR	NR	NR		NZ	NZ	NZ	NZ

Table III. Natick Testing 10 Washes (cont'd)													
			Active Conc.		Test Method 100					Test Method 147			
Item	Sample ID	Active	ppm		Staph	Strep	Coryn	Pseud		Staph	Strep	Coryn	Pseud
Boot sock	30-A-10-R1	Triclosan	74		NR	69.00%	NR	NR		4	NZ	NZ	NZ
	30-A-10-R2		84		NR	63.00%	NR	NR		2	NZ	NZ	NZ
	30-A-10-R3		81		NR	94.10%	NR	NR		2	NZ	NZ	NZ
	30-B-10-R1	Quat silane			NR	NR	NR	NR		NZ	I	1	NZ
	30-B-10-R2				NR	94.30%	NR	NR		NZ	2	2	NZ
	30-B-10-R3				NR	99.90%	NR	NR		NZ	3	1	NZ
	30-C-10-R1	PHMB			NR	99.90%	NR	NR		NZ	I	2	NZ
	30-C-10-R2				NR	99.90%	NR	NR		NZ	I	I	NZ
	30-C-10-R3				NR	99.90%	NR	NR		NZ	I	I	NZ
	30-D-10-R1	Control			NR	NR	NR	NR		NZ	NZ	NZ	NZ
	30-D-10-R2				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	30-D-10-R3				NR	23.00%	NR	NR		NZ	NZ	NZ	NZ

Table IV. Natick Testing 25 Washes													
			Active Conc.		Test Method 100					Test Method 147			
Item	Sample ID	Active	ppm		Staph	Strep	Coryn	Pseud		Staph	Strep	Coryn	Pseud
ACU	10-A-25-R1	Triclosan	0		NR	NR	NR	NR		NZ	NZ	NZ	NZ
	10-A-25-R2		0		NR	NR	NR	NR		NZ	NZ	NZ	NZ
	10-A-25-R3		0		NR	NR	NR	NR		NZ	NZ	NZ	NZ
	10-B-25-R1	Quat silane			NR	99.10%	NR	NR		NZ	NZ	NZ	NZ
	10-B-25-R2				NR	99.90%	NR	NR		NZ	NZ	2	NZ
	10-B-25-R3				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	10-C-25-R1	PHMB			99.90%	99.90%	99.90%	NR		NZ	2	1	NZ
	10-C-25-R2				NR	99.90%	99.90%	NR		NZ	2	1	NZ
	10-C-25-R3				99.90%	99.90%	99.90%	NR		NZ	1	NZ	NZ
	10-D-25-R1	Control			NR	76.40%	NR	NR		NZ	NZ	NZ	NZ
	10-D-25-R2				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	10-D-25-R3				NR	NR	NR	NR		NZ	NZ	NZ	NZ
T-shirt	20-A-25-R1	Triclosan	3200		99.40%	NR	NR	NR		5	NZ	NZ	NZ
	20-A-25-R2		2900		99.90%	NR	NR	NR		5	NZ	NZ	NZ
	20-A-25-R3		3600		99.80%	99.80%	NR	NR		5	NZ	NZ	NZ
	20-B-25-R1	Quat silane			NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-B-25-R2				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-B-25-R3				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-C-25-R1	PHMB			NR	99.30%	NR	NR		NZ	NZ	NZ	NZ
	20-C-25-R2				NR	99.30%	NR	NR		NZ	NZ	NZ	NZ
	20-C-25-R3				NR	99.80%	99.10%	NR		NZ	NZ	NZ	NZ
	20-D-25-R1	Control			NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-D-25-R2				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-D-25-R3				NR	NR	NR	NR		NZ	NZ	NZ	NZ

Table IV. Natick Testing 25 Washes (cont'd)													
			Active Conc.		Test Method 100					Test Method 147			
Item	Sample ID	Active	ppm		Staph	Strep	Coryn	Pseud		Staph	Strep	Coryn	Pseud
Boot sock	30-A-25-R1	Triclosan	74		NR	NR	NR	NR		NZ	NZ	NZ	NZ
	30-A-25-R2		84		NR	87.80%	NR	NR		NZ	NZ	NZ	NZ
	30-A-25-R3		81		NR	99.80%	NR	NR		NZ	NZ	NZ	NZ
	30-B-25-R1	Quat silane			NR	99.80%	99.00%	NR		NZ	I	NZ	NZ
	30-B-25-R2				NR	98.00%	97.00%	NR		NZ	NZ	NZ	NZ
	30-B-25-R3				NR	91.50%	99.90%	NR		NZ	NZ	NZ	NZ
	30-C-25-R1	PHMB			NR	99.80%	99.90%	NR		NZ	NZ	I	NZ
	30-C-25-R2				NR	99.80%	99.90%	NR		NZ	NZ	NZ	NZ
	30-C-25-R3				99.40%	98.00%	99.90%	NR		NZ	NZ	I	NZ
	30-D-25-R1	Control			NR	NR	NR	NR		NZ	NZ	NZ	NZ
	30-D-25-R2				NR	NR	NR	NR		NZ	I	NZ	NZ
	30-D-25-R3				NR	NR	NR	NR		NZ	I	NZ	NZ

Table V. Natick Testing 50 Washes													
			Active Conc.		Test Method 100					Test Method 147			
Item	Sample ID	Active	ppm		Staph	Strep	Coryn	Pseud		Staph	Strep	Coryn	Pseud
ACU	10-A-50-R1	Triclosan	27		NR	NR	NR	NR		I	I	NZ	NZ
	10-A-50-R2		24		NR	NR	NR	NR		I	NZ	NZ	NZ
	10-A-50-R3		31		NR	NR	NR	NR		I	NZ	I	NZ
	10-B-50-R1	Quat silane			NR	NR	NR	NR		NZ	NZ	NZ	NZ
	10-B-50-R2				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	10-B-50-R3				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	10-C-50-R1	PHMB			NR	99.90%	99.80%	NR		I	I	1	NZ
	10-C-50-R2				NR	99.90%	97.80%	NR		NZ	I	NZ	NZ
	10-C-50-R3				NR	99.90%	99.80%	NR		NZ	I	1	NZ
	10-D-50-R1	Control			NR	99.90%	NR	NR		NZ	NZ	NZ	NZ
	10-D-50-R2				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	10-D-50-R3				NR	93.20%	NR	NR		NZ	NZ	NZ	NZ
T-shirt	20-A-50-R1	Triclosan	880		98.10%	NR	NR	NR		5	NZ	NZ	NZ
	20-A-50-R2		87		99.90%	NR	NR	NR		5	NZ	NZ	NZ
	20-A-50-R3		1100		99.90%	NR	NR	NR		5	NZ	NZ	NZ
	20-B-50-R1	Quat silane			NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-B-50-R2				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-B-50-R3				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-C-50-R1	PHMB			5.30%	99.90%	NR	NR		NZ	NZ	NZ	NZ
	20-C-50-R2				NR	99.90%	NR	NR		NZ	NZ	NZ	NZ
	20-C-50-R3				NR	99.90%	99.90%	NR		NZ	NZ	NZ	NZ
	20-D-50-R1	Control			NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-D-50-R2				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	20-D-50-R3				NR	NR	NR	NR		NZ	NZ	NZ	NZ

Table V. Natick Testing 50 Washes (cont'd)													
			Active Conc.		Test Method 100					Test Method 147			
Item	Sample ID	Active	ppm		Staph	Strep	Coryn	Pseud		Staph	Strep	Coryn	Pseud
Boot sock	30-A-50-R1	Triclosan	24		NR	NR	NR	NR		I	NZ	NZ	NZ
	30-A-50-R2		20		NR	NR	NR	NR		3	NZ	NZ	NZ
	30-A-50-R3		30		NR	NR	NR	NR		2	NZ	NZ	NZ
	30-B-50-R1	Quat silane			NR	NR	NR	NR		NZ	NZ	NZ	NZ
	30-B-50-R2				NR	96.00%	NR	NR		NZ	NZ	NZ	NZ
	30-B-50-R3				NR	94.00%	NR	NR		NZ	NZ	NZ	NZ
	30-C-50-R1	PHMB			NR	99.90%	56.50%	NR		NZ	NZ	I	NZ
	30-C-50-R2				NR	99.90%	99.90%	NR		NZ	NZ	1	NZ
	30-C-50-R3				NR	99.90%	99.90%	NR		NZ	NZ	1	NZ
	30-D-50-R1	Control			NR	NR	NR	NR		NZ	NZ	NZ	NZ
	30-D-50-R2				NR	NR	NR	NR		NZ	NZ	NZ	NZ
	30-D-50-R3				NR	NR	NR	NR		NZ	NZ	NZ	NZ

Natick 2nd Trial - ACU -Triclosan + PHMB								
			Active Conc.		Test Method 100			
Item	Sample ID	Active	ppm		Staph	Strep	Coryn	Pseud
Triclosan	10-A-0 R1	Unwashed	1400		NR	NR	93.00%	NR
1st Trial	10-A-0 R2		1300		NR	NR	NR	NR
Wash wheel	10-A-0 R3		1300		NR	NR	NR	NR
	10-A-25 R1	25 Washes	690		NR	NR	NR	NR
	10-A-25 R2		650		NR	NR	NR	NR
	10-A-25 R3		720		NR	NR	NR	NR
	10-A-50 R1	50 Washes	800		NR	NR	NR	NR
	10-A-50 R2		780		NR	NR	NR	NR
	10-A-50 R3		850		NR	NR	NR	NR
Triclosan	10-A1-0 R1	Unwashed	2100		99.80%	99.90%	99.90%	NR
1st Trial	10-A1-0 R2		2100		99.90%	99.90%	99.90%	NR
61-2A Wash	10-A1-0 R3		2200		99.80%	98.50%	99.70%	NR
	10-A1-25 R1	25 Washes	1100		94.30%	64.90%	NR	NR
	10-A1-25 R2		940		NR	97.80%	NR	NR
	10-A1-25 R3		880		NR	91.40%	NR	NR
	10-A1-50 R1	50 Washes	1200		NR	NR	37.00%	NR
	10-A1-50 R2		1100		NR	NR	NR	NR
	10-A1-50 R3		1300		71.40%	NR	NR	NR
Triclosan	10-AC-0 R1	Unwashed	2300		99.90%	99.90%	99.90%	NR
+ PHMB	10-AC-0 R2		2300		99.90%	99.90%	99.40%	NR
2nd Trial	10-AC-0 R3		2200		99.90%	99.90%	99.90%	NR
61-2A Wash								
	10-AC-25 R1	25 Washes	1200		97.50%	92.30%	NR	NR
	10-AC-25 R2		1200		99.80%	99.90%	99.70%	NR
	10-AC-25 R3		1200		97.60%	NR	NR	NR
	10-AC-50 R1	50 Washes	1400		99.80%	99.90%	97.90%	NR
	10-AC-50 R2		1600		99.90%	99.90%	99.90%	NR
	10-AC-50 R3		1700		99.90%	98.80%	99.70%	NR

APPENDIX VI: Analytical Test Methods

APPENDIX V: Analytical Test Method - Triclosan

Methodology for the Extraction and Analysis of Microban Additive "B" From Textiles Using LC-MS

General Description of Methodology:

Textile samples containing Microban Additive "B" (Triclosan) are cut up and each is extracted in 20 ml of methanol using a microwave accelerated reaction system at 100º C with stirring for 15 minutes. The extract is then filtered into a 25 ml volumetric flask, brought to volume with methanol, and analyzed by LC-MS.

Required Materials:

1) Microban Additive "B" (Triclosan), >99.5% purity.
2) Acetonitrile, HPLC grade (Fisher OPTIMA or equivalent)
3) Water, HPLC Grade (Fisher OPTIMA or equivalent)
4) Microwave Accelerated Reaction System (MARS-X, CEM Corporation) equipped with teflon microwave vessels, sleeves, frames, stir bars and magnetic stirring option.
5) Analytical balance capable of weighing accurately to 0.0001 gram
6) Volumetric flasks for preparation of analytical standards. Typical capacities are 100, 50, 25 and 10 ml.
7) Funnels, glass, 5 cm. diameter
8) Filter paper, 7.5 cm, qualitative, (VWR 417 or equivalent)
9) Graduated cylinder, 50 ml or 100 ml capacity
10) HPLC autosampler vials, 1.5 ml fill volume, glass with Teflon septum caps.
11) LC-MS System (Shimadzu LC-MS 2010 or equivalent) configured as given in the procedure.

Sample Preparation:

1) Cut the material to be tested into pieces of approximately 5 mm by 5 mm.

2) Accurately weigh to the nearest 0.001 gram 0.1 to 0.2 gram of the cut material into the teflon microwave vessel.

3) Add a magnetic stir bar and pour 20 ml of methanol into the vessel.

4) Cap the vessel. Place it into the accompanying microwave absorbing sleeve and frame and tighten the frame to pressure seal the vessel.

5) Place the vessel in within the microwave oven along with the temperature control vessel and probe as directed by the system manual, close the door and program the system to ramp up over 5 minutes to a temperature of 100º C. The hold period at this temperature is set for 15 minutes. Stirring of the sample within the vessel is continuous throughout the heating and cooling.

6) Allow the system to cool down to approximately 25º C then remove the sample vessel, open, and quantitatively transfer the methanol extract into a 25 ml volumetric flask. Use additional methanol as necessary to facilitate the transfer and to bring the flask to volume.

7) Mix the flask contents well, then transfer approximately 1.5 ml of the extract solution into an autosampler vial and cap this vial securely. Set the vial aside for analysis by LC-MS.

Standard Preparation

1) Using the balance and volumetric flasks, prepare a stock solution by accurately weighing approximately 100 mg of Microban Additive "B" into a 100 ml volumetric flask. Dissolve the additive in 50 ml of methanol then bring the flask to volume with methanol and mix well.

2) Prepare a series of five working standards of Additive "B" in by volumetrically diluting measured aliquots of the stock solution with methanol. These standards are 0.2, 0.4, 0.8, 1.0 and 4.0 µg/ml.

Extract Analysis

1) Set up the LC-MS system. The following conditions are typically used.

Column:	15 cm x 4.6 mm i.d. C_{18} (e.g. Supelco Discovery) reverse phase column
Mobile Phase:	80% acetonitrile, 20 % water
Flow Rate:	0.5 ml/min
Temp:	40º C
Detector:	MS, Electrospray probe, single ion monitor, negative polarity, monitored at 286.9 amu.
Injection Volume:	2 µl

Under these conditions, Additive "B" has a retention time of approximately 4.5 to 5 minutes.

2) Make sufficient injections of pure methanol to ensure that the instrument baseline is flat and the system free of contaminants.

3) Make duplicate injections of each analytical standard beginning with the lowest concentration and ending with the highest.

4) Plot the average peak area for each standard versus the respective concentration in µg/ml and determine the calibration regression equation. This plot results in a straight line with a typical correlation coefficient (r^2) of approximately 0.995 or better.

5) Make sufficient injections of pure methanol to ensure that the instrument baseline is flat and the system free of analyte carryover and contaminants.

6) Make duplicate injections of each sample extract. Agreement for peak areas of each duplicate should be within 5%.

7) Complete the chromatographic run by repeating duplicate injections of one of the mid-range standards to ensure that neither column nor detector fluctuations have affected the integrity of the analysis.

8) Determine the average peak area for Microban Additive "B" in each sample extract, and then calculate the μg/ml value for those extracts using the calibration regression equation.

9) Calculate the parts per million (ppm) of Microban Additive "B" in the sample using the following equation:

$$ppm\ of\ Microban\ Additive\ "\mathbf{B}" = \frac{(25\,ml)(ug\ /\ ml\ Additive\ "\mathbf{B}"\ in\ extract)}{(grams\ \ sample\ weight)}$$