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UTILIZATION OF SANITIZING WIPES ON SELECTED COATED NONSTICK FOOD CONTACT SURFACES

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14. ABSTRACT
The objective of this study was to find alternatives to military field sanitation methods that will reduce the amount of greywater produced during sanitation operations and the logistical requirements of field sanitation. The cleaning and bactericidal efficacy of commercial-off-the-shelf (COTS) sanitizing wipes was tested on specially coated non-stick food contact surfaces, also under investigation by the Army. A biofilm was produced on the surfaces by soiling them with a food substrate contaminated by multiple strains of *S.Aureus* and *E.Coli*. One COTS wipe was effective on all four surfaces tested. The average counts of bacteria on Hycheck contact slides were reduced from too numerous to count per square inch before application of the sanitizing wipe (gross food residues removed), to 0 to 11 colony-forming units per square inch after application of the sanitizing wipes. While effective on the four surfaces tested, it cannot be concluded that these wipes will clean and disinfect all surfaces found in a food service facility, or be reliable for removal of all possible soils and biofilms, without pre-cleaning the surface.

15. SUBJECT TERMS

SANITARY WIPE	FIELD FEEDING	FOOD SERVICE EQUIPMENT	WATERLESS SANITATION
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CLEANING	ALUMINUM SURFACES	NONSTICK COATINGS	DISINFECTION
SANITATION			

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PREFACE

This report explores the potential for using cloths infused with quaternary ammonium compounds to both clean and sanitize aluminum surfaces. Various nonstick coatings were used on the aluminum surfaces to determine their role and efficacy in this system. Water is a primary logistical burden for the Army and field sanitation operations use approximately 250 gallons of water per day. A waterless or nearly waterless sanitation system would greatly decrease the logistical burden of field-feeding and sanitation operations. Work for this project was conducted in-house at the Natick Army Soldier Systems Center under a Tech Base project, "Water Conservation", from July 2001 to September 2001 by the authors. Edmund Powers was employed as a contract microbiologist under contract number DAAD16-01-P-0054 and Chris Hoogeboom was a summer intern through the George Washington University Science and Engineering Apprentice Program (SEAP) for High School Students. Chad Haering and Claire Lee were employees of the Natick Soldier Systems Center.

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UTILIZATION OF SANITIZING WIPES ON SELECTED COATED NONSTICK FOOD CONTACT SURFACES

INTRODUCTION

Utilization of commercial off-the-shelf (COTS) sanitizing wipes as an alternative to military field sanitation methods (1) was the goal of a 6.2 Tech Base project entitled "Water Conservation" and has the potential to greatly reduce water consumption and conserve energy. For example, if effectively implemented, sanitizing wipes will reduce the amount of greywater produced during sanitation operations, conserve water for drinking and other uses, reduce energy required to heat water and finally they will serve as a backup sanitation system when either hot water or a potable water supply is unavailable.

After conducting a market search, two COTS wipes were selected which were Environmental Protection Agency (EPA) registered and designed for safely sanitizing food processing facilities and equipment. One was the Simple Solutions™ all surface sanitizing wipes produced by Atlantic Mills Inc., Lakewood, N. J. They were dry, and pretreated to create a gallon of non-rinse solution when re-hydrated/activated in water. The other was Sani-Quat 200 wipes produced by The Way Inc., P. O. Box 968, Picayune, MS 39466. They were non-rinse pre-saturated wipes that were ready to use. Both wipes were rated to provide 200ppm quaternary ammonium chloride (QAC). The QAC's found in these wipes were in compliance with the Code of Federal Regulations 21 CFR 178.1010, which permits their use on food processing equipment, utensils and other food contact surfaces (2).

Each year food-borne diseases kill 5200 Americans, hospitalize hundreds of thousands of others (CDC, Atlanta, GA) and continue to gain increasing attention. Food-borne illness is also a serious threat to military services. Contributing factors include poor hygiene, and dirty, contaminated equipment and utensils. Effective foodservice sanitation can have a significant impact on reducing food-borne outbreaks by preventing or eliminating contamination of kitchenware and food contact surfaces. Conventional practice is to clean the surface first with a suitable detergent to remove the biofilm and then expose the microorganism to the sanitizer, (3, 4, 5, 6, 7).

Three specially coated aluminum cooking surfaces and uncoated aluminum were selected for testing the sanitizing efficacy of the wipes. The coatings, produced by General Magnaplate Corporation, 1331 Route I, Linden, NJ 07036, were Lectofluor (main ingredient of Teflon), Tufram and Plasmadize. These are all non-stick coatings similar to Teflon. All surfaces were soiled with contaminated food to produce a biofilm before attempts were made to sanitize them with the wipes.

The objective of this study was to simplify conventional practice and reduce logistical support by attempting to use a single sanitizing wipe to both clean and sanitize select food contact surfaces, even though previous investigators were not successful with this approach (4, 5, 6, 7). However, the feasibility of applying the wipes selected, with their unique combination of chemical sanitizers, improved reliability, stability and safety, to determine their sanitizing efficacy on the unique coated surfaces, was yet to be demonstrated and was of sufficient interest to warrant an investigation.

MATERIALS AND METHODS

Sanitizing Wipes

Sani-Quat 200 wipes are pre-saturated 6" x 7" towelettes packaged in a cylindrical plastic dispenser and designed for sanitizing food processing facilities, meat and dairy plants and mass feeding kitchens. They contained N-alkyl-dimethy benzyl ammonium chloride (0.0175%) as the active ingredient, glycol ether (6.5%), ethanol, tetrasodium ethylenediaminetetraacetate (EDTA) and 2-(2-butoxythoxy)-tetrasodium EDTA .

Simple Solutions™ wipes are dry, pretreated foodservice wipes (16.75" x 13.25") that instantly creates a gallon of non-rinse sanitizing solution containing 200 ppm quaternary ammonium chlorides (QAC) by re-hydrating the wipe in a gallon of water. The wipes contained N-alkyl-dimethy benzyl ammonium chloride, N-alkyl-dimethyl ethyl benzyl ammonium chloride, propylene glycol, nonoxynol-10 and turquoise A1X-4.

Wipes used as a non-bactericidal control for comparison were paper towels saturated with deionized water. They were wrapped with double layers of Kraft® paper and sterilized by autoclaving at 121°C for 15 minutes. Before wiping the test surfaces, the paper towels were dipped in sterile deionized water and excess water was squeezed out manually.

Application of Sanitizing Wipes

Commercial wipes were applied to test surfaces according to manufacturers instructions as follows: Wearing sterile latex gloves, Sani-quat 200 wipes were applied to test surfaces using normal pressure and by wiping in concentric circles for ten seconds. The wiped surface areas were allowed to dry for ten minutes before sampling with Hycheck monitors.

Simple Solutions™ wipes were re-hydrated in a gallon of water for at least one minute and "squeezed" to activate the green sanitizing solution. Then they were applied with gloved hands to each test surface in concentric circles using normal pressure for ten seconds. The surface was kept wet for 60 seconds by re-wiping as necessary and then air dried for 60 seconds or longer before sampling with Hycheck monitors.

Control wipes were applied in the same manner as the Sani-Quat 200 wipes.

Coated Surfaces

Aluminum squares (4"x 4") were coated with different, unique non-stick coatings including Lectrofluor (poly-tetra-fluoro-ethylene or PTFE, the main ingredient in Teflon); Tufram (anodized coating infused with PTFE); and Plasmadize (matrix consisting of ceramic, metallic and PTFE). All coatings were produced by General Magnaplate Corporation.

Cleaning and Sterilizing Test Surfaces

Coated and uncoated aluminum test surfaces (squares) were washed and soaked in RBS 35 detergent (Pierce, P.O. Box 117, Rockford, IL 61105) and tap water at 50° C, for 15 min., rinsed three times in tap water and then distilled water. Each square was placed in a disposable aluminum foil tray (two/tray), covered with aluminum foil and sterilized by autoclaving at 121°C for 20 min.

Test Food

Canned chili with beans (Hormel Foods Corporation, Austin, MN 55912) was selected as the substrate for inoculation and soiling of the test surfaces. The chili was blended by stomaching for one minute (8), then 50 g was transferred to beakers and sterilized by autoclaving at 121° C for 15 minutes. Ability of test bacteria to grow in the chili was pre-determined by inoculating the sterilized chili separately with *Staphylococcus aureus* (ATCC 6538) and *Eschericia coli* (ATCC 11229) at 10,000 CFU/g.

Test Bacteria

Test bacteria were *S. aureus strains* ATCC 6538, ATCC 13567, ATCC 8095 and *E. coli* strains ATCC 11229, ATCC 45827,and nontoxic *E.coli* 0157:H7 (culture #B6-914, Communicable Disease Center, Atlanta, GA). Each culture was activated by making three transfers in trypticase soy yeast extract broth (TSYB, Difco, Detroit, MI) incubated at 35C for 24 hours.

Inoculum Preparation

Activated TSYB cultures were each diluted in Butterfields phosphate buffer (8) and adjusted turbidimetrically (Ratio/XR Turbidimeter, Model 43900), Hach Company, P.O. Box 389, Loveland, Colorado), to achieve 500,000 CFU's (colony forming units). Three strain cocktails were prepared by mixing equal volumes of each diluted culture. Then 1 ml of the cocktail was added to 50 grams of sterilized chili and mixed well to achieve 10,000 CFU/g.

Inoculation of Surfaces

Ten grams of the inoculated chili were spread on each of the 4"x 4" test surfaces with a glass spreading rod and incubated at 35° C for 24 hours, to develop a biofilm (attached bacteria) on the surfaces. Tests surfaces were protected by placing them into aluminum-foil food trays (10"W x 12"L), covered with aluminum foil. To prevent drying of the food during incubation, sterile paper towels were moistened to the saturation point with sterile distilled water and placed under each test surface in the tray. A sterile control for each test surface consisted of sterile chili inoculated with 1 ml of sterile distilled water (SDW).

Assessing Microbial Growth in the Biofilm and the Cleansing Efficacy of Wipes

Following incubation of inoculated chili on all test surfaces, gross residues of the spoiled chili were removed from each surface by wiping with a dry sterile paper towel. Bacteria remaining on the surfaces, before and after wiping with sanitizing wipes, were monitored by randomly taking duplicate surface samples with Hycheck contact slides containing D/E Neutralizing Agar (Difco). Surfaces were allowed to dry for up to 30 minutes before applying the Hycheck monitors.

Bacterial Monitors

Hycheck contact slides contained D/E Neutralizing Agar (Difco) on both sides of the slide (paddle). D/E Neutralizing Agar will neutralize a broad spectrum of disinfectant chemicals including quaternary ammonium compounds. Duplicate samples were taken from the test surfaces by using both sides of the slides. The surface area of each side was one square-inch.

RESULTS

Bacterial counts shown in Tables 1 to 3 are the averages of duplicate counts on each test surface. Reduction of the counts to 12.5/sq. in. was considered an effective reduction by the wipes and a suitably sanitized surface for food service (9).

Growth of Bacteria in Chili With Beans

Excellent growth of both test bacteria was achieved in the chili with beans. Preliminary studies indicated that test organisms grew rapidly in this substrate, reaching levels ranging from 4.3 to 7.0 x 10^8 CFU/g within 24 hours.

Bactericidal Efficacy of Sani–Quat 200 Wipes

Table 1 shows the efficacy of Sani-Quat-200® wipes on the four test surfaces after soiling with the chili and beans inoculated separately with three strain cocktails of *S. aureus*, and *E. coli*. The wipes did not effectively remove *S. aureus* or *E. coli* after wiping test surfaces as per the manufactures instructions, except on Lectrofluor where *E. coli* counts were reduced from too numerous to count (TNTC) before wiping, to 9/sq. in. after wiping. The wipes were more effective against *E. coli* than they were against *S. aureus*. These pre-moistened wipes contained only a single QAC and the concentration was only 100 ppm as measured by test strips for QAC's (Atlantic Mills, 129 Towbin Avenue, Lakewood, N.J), instead of 200 ppm as specified.

Table 1. Bactericidal Efficacy of Sani-Quat 200 Wipes

	Average[a] cfu/in^2			
	S. aureus		E. coli	
Surface	Before[b]	After[b]	Before[b]	After[b]
Lectrofluor	TNTC	TNTC	TNTC	9
Tufram	TNTC	TNTC	TNTC	23
Plasmadize	TNTC	TNTC	TNTC	TNTC
Aluminum	TNTC	TNTC	TNTC	67

[a] Average of two samples
[b] Before and after wiping with sanitizing wipe

Bactericidal Efficacy of Simple Solution™ Wipes

Table 2 shows that the Simple Solution™ sanitizing wipes were effective on all four test surfaces soiled with chili inoculated separately with three strain cocktails of both test bacteria. The counts of both test bacteria were reduced from TNTC before application of the sanitizing wipes (gross food residues removed with a dry paper towel), to 0 to 11 on average, after application of the sanitizing wipes. Unlike the previous wipes, these wipes were dry and pretreated, moistened just before use in a gallon of tap water that instantly created a non-rinse sanitizing solution containing two QAC's at a concentration of 200 ppm, as verified with test strips from Atlantic Mills, Inc.

Table 2. Bactericidal Efficacy of Simple Solutions™ Wipes

	Average[a] cfu/in^2			
	S. aureus		*E. coli*	
Surface	Before[b]	After[b]	Before[b]	After[b]
Lectrofluor	TNTC	3	TNTC	5
Tufram	TNTC	0	TNTC	0
Plasmadize	TNTC	6	TNTC	3
Aluminum	TNTC	3	TNTC	11

[a] Average of two samples
[b] Before and after wiping with sanitizing wipe

Bactericidal Efficacy of Deionized Water Wipes

The deionized water–paper towel wipes which served as number controls were not effective in sanitizing the four test surfaces, soiled and contaminated as above, as shown in Table 3. Colonies on the Hycheck monitors remained TNTC after wiping, as was expected, since there was no sanitizing agent present in the wipes.

Table 3. Deionized Water / Paper Towel Wipes

	Average[a] cfu/in^2			
	S. aureus		***E. coli***	
Surface	Before[b]	After[b]	Before[b]	After[b]
Lectrofluor	TNTC	TNTC	TNTC	TNTC
Tufram	TNTC	TNTC	TNTC	TNTC
Plasmadize	TNTC	TNTC	TNTC	TNTC
Aluminum	TNTC	TNTC	TNTC	TNTC

[a] Average of two samples
[b] Before and after wiping with the surface

DISCUSSION

In this study, biofilms were produced in a 24-hour period to simulate what might occur in food service facilities. Bacteria grew and spoiled the chili substrate and were successfully attached to all four surfaces (Lectrofluor, Tufram, Plasmadize and aluminum) as evidenced by the fact that the counts were TNTC after gross food residues were removed by wiping with sterile paper towels.

The concept of a one-step operation to clean and sanitize selected surfaces was investigated in this study. Typically in a food service operation, cleaners (detergents) are used to remove soil from the surfaces of equipment and sanitizers are used to inactivate microorganisms that remain behind. Other investigators found that in general, chemical cleaners were much more effective than sanitizers on stainless steel, plastic (4) and surfaces in a milk processing system (7). This is because cleaners contain, among other agents, surfactants that suspend and remove greasy soil. They are able to penetrate biofilms, formed by the attachment of microorganisms and the accumulation of layers of fat, protein, polysaccharides and other products of metabolism, as well as food debri (6). Unlike cleaners, sanitizers cannot penetrate biofilms and thus cannot reach the attached cells. Adherent cells are also more resistant to chemicals (3, 4, 7, 11, 15, 16).

Simple Solutions™ wipes were more effective than Sani-Quat 200 wipes, probably because they had some detergent activity as noted by the manufacturer. They contained a nonionic surfactant (nonoxynol-10) and propylene glycol that is a solvent and emulsifier. Other factors that may account for the different bactericidal efficacies of the two wipes include different contact times, method of application, and different concentrations and number of QAC's in the wipes.

The data does not seem to indicate that resistance of *S. aureus or E coli* to the sanitizers is related to the surface to which the bacteria were attached. Neither organism showed resistance to Simple Solution wipes on any of the four test surfaces. *S. aureus* was more resistant than *E. coli* to Sani-Quat 200 on all four surfaces. The variable resistance exhibited by *E. coli* on the four surfaces could have been due to the sanitizer, as well as to the surface. This study could not confirm results reported by other investigators that microorganisms become more resistant to sanitizers once they become attached to a surface (4, 7, 10, 11, 12, 13 14), or that resistance was related to the surface to which the organism is attached (4). Some reasons for this may have been due to the fact that some of their surfaces were more difficult to clean and sanitize, biofilms were produced differently, substrates differed, application of sanitizers differed, and the condition and materials of surfaces differed.

A wipe that can be used in a one step operation to clean and sanitize is needed to save time as well as to conserve water. Simple Solutions™ wipes may be such a wipe since it has the attributes of both a detergent and a sanitizer and was effective on the test surfaces used. However more data is needed before its use can be recommended for removal of all possible soils, bacteria and biofilms found in a foodservice facility, without pre-cleaning the surface as recommended by the manufacturer. Although Simple Solutions™ wipes are not a true waterless system, because they must be activated in a gallon of water, they will still save a considerable amount of water.

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