THE SURVIVAL OF *STAPHYLOCOCCUS AUREUS* ON MILITARY SOCK FABRIC LAUNDERED AT VARIOUS WATER TEMPERATURES AND DETERGENT CONCENTRATIONS

by 500

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INTRODUCTION

The prevention of disease and the transmission of pathogenic microorganisms is of great concern today because of the rapid population growth and compactness of modern living. Information dealing with the survival and transmission of harmful microorganisms would be of benifit in decreasing the opportunity for infection to occur. Textiles are transmitters of disease causing microorganisms from person to person (1). Knowledge that clothing and textiles are a means of bacterial transference affects the laundering of contaminated clothing and indicates that the laundry is one area of the home environment where sanitation is an important factor.

An increase in the use of cold water, which is recommended for the laundering of synthetic fabrics and colored garments, and an increase in the use of public facilities such as launderomats cause a concern about sanitary laundry procedures. Often it is difficult to obtain high enough temperatures to destroy harmful microorganisms in the laundry. Even though modern detergents have many additives, detergents or soaps alone do not remove a significant amount of bacteria to give a sanitary wash (7, 26). Complete removal is needed to guarantee a sanitary wash since the presence of only one virulent pathogen is necessary to start infection or cause disease. It is important to know how the relationship between the amount of detergent and the water temperature affect the removal of pathogenic microorganisms present in the fabric and affect the prevention of their transference to other garments in the laundry.

The objectives of the present study were to determine the effect of water temperature and detergent concentration upon the survival of a specific microorganism in a home laundry situation and to determine the transference of the specific organism during the washing process. The microorganism used was Staphylococcus aureus A. T. C. C. # 6538, which is commonly found in the environment, causes infection, is easy to detect, and has a very great possibility of contamination.

The survival of a specific microorganism is determined by Staphylococcus aureus survival counts on the fabric after washing and drying and the Staphylococcus aureus count in the wash water. Transference during the washing process is determined by the redeposition of Staphylococcus aureus on the fabric during washing and its survival in the laundry equipment at the end of the washing cycle and drying period.

Definitions of Terms Related to the Present Study

Bacterial count or survival refers only to the microorganism Staphylococcus aureus A. T. C. C. # 6538. No other organism was considered in the analysis of bacterial survival on the fabric, bacterial count of the water, and bacterial removal during the wash cycle and drying period in this research.

Fabric swatch indicates a 12" x 8" rectangle of fabric cut from the original form of tubes and used for laundering.

Fabric sample indicates a one-inch square which was removed from the fabric swatches for determination of bacterial counts.

Washing period refers to a ten minute agitation period in which the detergent is in solution.

Wash cycle refers to the entire washing operation including washing period, spray rinses, and deep rinse period.

Washing treatment defines the procedural sequence using one water temperature, one detergent concentration, and one fabric.

Experimental design defines three repetitions of the procedural sequence for all combinations of water temperatures, detergent concentrations, and fabric.

REVIEW OF LITERATURE

The importance of the laundry in disease transference is apparent from research of previous work in the area. The review of literature has been divided into sections examining the characteristics of the microorganism, Staphylococcus aureus; analyzing the effect of water temperature in cleaning; describing the effect of detergent concentration upon the soil removal; and finally, a comparison of previous studies in which the removal of bacteria in the laundry has been investigated.

Characteristics of Staphylococcus aureus

A knowledge of staphylococcus is helpful in understanding many of the problems of cross infection with bacteria that arise in the laundry process (6). The following life requirements (10) for bacteria were given as: [1]. proper nourishment, [2]. air or oxygen (or the lack of it, depending upon the bacteria), [3]. moisture, [4]. proper temperature, and [5]. the absence of direct sunlight, retarding chemicals, and antagonistic organisms. Bacteria are dependent on animal and/or vegetable matter for nourishment and are provided with enzymes to digest complex substances such as blood, urine, some soaps, and food. Staphylococcus aureus grows best in a humidity of 60% or higher and at body temperature of 98.6° F. The bacteria can survive dry and cold conditions for several months and are able to multiply quickly when heat and moisture are again present. They are capable of producing disease under proper conditions causing boils, infection around cuts, food poisoning, mastitis, child bed fever, and may lead to death where infection becomes serious.

The control of this organism is difficult because staphylococcial infections are highly contagious and some strains have become resistant to antibiotics. Means of control are high temperatures, chemical attack, and ultra violet light or nuclear irritations. "Depending upon the strain moist staphylococci can be killed by anywhere from temperatures of 140° F. for ten minutes to 175° F. for thirty minutes. Dry staphylococci require higher temperatures and greater periods of time to be destroyed. Since laundries deal with all strains at some time or another, a water temperature of 175° F. held for thirty minutes must be a minimum for laundry." (6-240) Chemicals which kill bacteria are germicides and include halogens such as chlorine bleach.

Some of the paths of infection are controlled by and in the laundry. Cross infection from an infected garment to one not infected arises in the laundry process. Burrows (4) reported that staphylococci are constantly present on the skin and in the upper respiratory tract. A transitory drop from a carrier's nasal passage, in the air, or on a textile item is sufficient to allow local invasion and establishment of infection. The American Public Health Association (1) reported that staphylococcial infection can be transmitted by contact with articles recently soiled with moist discharges of infected skin lesions.

Effect of Water Temperature

Many studies (2, 9, 13, 16) have found that soil removal increases as the water temperature increases. Little sanitizing was found after washing with a water temperature below 120° F. (7, 15, 18, 21, 22, 26).

The United States Department of Agriculture (28) recommended that water temperatures for home laundering of 140° F. gave the most soil removal and sanitizing and were ideal for white cottons and linens as well as heavily soiled articles of washfast colors. A water temperature of 120° F. was recommended for lightly soiled articles but the lower water temperature gave no sanitizing. A warm water temperature of 100° F. was recommended for washable woolens and hand washing. Cool water temperatures of 60° F. to 80° F. required the use of greater amounts of detergent and gave the least cleaning, no sanitizing, and minimum wrinkles.

The detergent's effectiveness in soil removal was influenced by water temperature. Kohler (16) found that increasing the maximum water temperature within the interval of 65° C. to 95° C. (117° F. to 171° F.) resulted in a continuous increase in the detergent's efficiency for soil removal. Anderson (2) used water temperatures of 70° F., 100° F., 120° F., 140° F., and 160° F. in studying the cleaning ability of a washing medium. It was found that temperature was a significant factor and the greatest cleaning was obtained at 160° F. Galbraith (9) also found that increasing the washing temperature from 70° F. to 140° F. increased the percentage of soil removed. Hodam (13) found that an increase in temperature yielded an increase in soil removal. At 70° F., 26.3% of the soil was removed and at 120° F., 53.4% of the soil was removed.

Effect of Detergent Concentration

The relationship of the amount of detergent to the effectiveness of the cleaning solution is a significant one. There is an optimum concentration which gives the most efficient cleaning. Too little detergent does not have enough power to hold the soil particles in suspension and too great a concentration tends to increase redeposition.

Kohler (16) found that an increase of the soap concentration over and above that required for the dispersion of the dirt did not appreciably improve the detergent effect. Suds in the soap solution was usually an indication that the soap concentration was enough.

Anderson (2) tested the effect of detergent concentration on the cleaning ability of the washing medium. Concentrations of .075%, .15%, .30%, .60%, and 1.20% at five water temperatures were used. Results showed that the greatest cleaning was at the 1.2% detergent concentration and 160° F. The lowest cleaning efficiency occured at 100° F. with a .07% (the lowest) detergent concentration. With a decrease in water hardness, there was a decrease in the concentration of detergent needed. No pattern existed in the amount of redeposition on unsoiled samples washed with soiled samples.

Galbraith (9) evaluated the effectiveness of twenty-four detergents on natural and synthetic fabrics. It was found that heavy duty or built detergents have superior soil removal as compared to unbuilt detergents. Results indicated that increasing the detergent concentration from .1% to .2% increased the percentage of soil removed. Increasing the concentration to .3% did not give greater soil removal except in the wool fabric.

Detergent levels between .125% and .2% were recommended by Davis (8). Soil removal increased with increased concentrations up to .5%. Beyond that there was a sharp decrease in efficiency due to excess sudsing. Excess sudsing reduced the mechanical action of agitation or hindered the floating away of soil resulting in redeposition (8, 27). Data has shown that soil removal proceeded very rapidly, occuring mostly during the first five minutes of washing. Included in the forces that promote redeposition are a high amount of soil, adverse temperature conditions, low solution volume, and a low detergent concentration (29).

Hunter, et al. (14) found that redeposition was greater in fabrics laundered with the detergent concentration half that of normal concentration. Market research surveys have shown that housewives tend to under use detergents in actual practice, which was particularity detrimental and probably accounted for the extremely large amounts of redeposition sometimes observed in home laundering.

Importance of Laundry in Disease Transference

Previous studies (3, 5, 7, 15, 18, 21, 22, 23, 24, 25, 26) have shown the importance of the laundry in disease transference. Interest in removal of pathogenic organisms from contaminated textiles began in the laundering of hospital linens and commercial laundering and has evolved to the home laundry.

Oliphant, et al. (23) analyzed six cases of infection found in laundry employees handling soiled linens from a laboratory doing work with a pathogenic organism. Only the workers who handled the soiled clothing before laundering were infected. Unlike Oliphant, et al., Perry, et al. (24) found no evidence that a pathogenic organism caused respiratory infections when transferred by unlaundered and laundered blankets.

Beck (3) stated that textile products were barriers for the passage of bacteria but only when completely dry. When they become moistened, microorganisms were immediately carried through them. Research concerning the effect of high humidities upon the bacterial permeability of textiles found that the absorbtion of moisture under states of high humidity did not of itself cause textiles to pass bacterial organisms except after reaching the dew point.

Ridenour (26) did a bacteriological study of automatic clothes washing to establish the extent of microorganism survival on clothes after various laundry operations. It was found that warm temperatures exerted no germicidal action on the organisms remaining on the clothes. To prevent redeposition of bacteria, organisms must be removed by dilution (adequate rinse) or by chemical treatment (detergent). An extra-ordinary desorbant was needed to render cloth bacteria-free. This was not possible with the detergents available at the time. The use of soap as a detergent without heat, killed or removed 95% of the inoculated organisms. Removal increased with the optimum soap concentration of .4% and was made easier when soil was present with the organism.

Approximately 95% of the bacteria were removed in a complete cycle (26). Fifty-eight percent of the inoculated bacteria were removed during the wash period, ten percent were removed during the rinse period, with the remaining bacterial removal due to other factors. With an increase in soap concentration there was an increase in bacterial removal. The maximum removal by soap was limited to 85% and greater soap concentrations than .1% showed no increase in bacterial removal.

Ridenour concluded that the amount of bacterial removal was dependent upon the presence or absence of soil, type of organism, and the amount of detergent. Practically all organisms were destroyed at 145° F. within three minutes with the water at a pH of 8.0 or above. Ninty percent of the viable organisms were removed by wash action with a detergent, 90% to 99% of the remainder were destroyed by hot water, giving from 99.00% to 99.99% total reduction.

When heavily contaminated material was washed in the same load with lightly soiled materials, an equilibrium was approached for all material in the load due to redeposition. The amount of redeposition depended on the length of the wash cycle. Cross-contamination also occurred between succeeding batches of clothes in the same washer due to the redeposition factor. The data indicated that sanitation by a dryer cannot be considered as a substitute for good detergency. Good washing action and an effective detergent were found to be the primary assets in laundry sanitation.

Crone (7) examined the survival of pathogenic organisms in laundering under certain conditions and found reliance on heat was preferred to reliance on chemicals for destroying the microorganisms. Staphylococcus aureus was found to survive low wash water temperatures of 46° C. to 53° C. (81° F. to 98° F.). He recommended that if laundering is to give some hygienic protection, it should be carried out at a temperature of at least 60° C. (140° F.).

A study (22) of a short-time (five minute wash), low-temperature (100° F.) washing procedure proved inadequate in removing pathogenic bacteria from linen. It was recommended that only high-temperature of 160° F. and long time processing of thirty minutes with the proper concentration of chemicals can result in 100% kill of pathogens. When using the short-time, low-temperature formula, the main means of removing microorganisms from the fabric was by physical action of the water under agitation.

Tumble drying after wash did not amply decrease the bacterial count in the inoculated fabric. Sanitation by a dryer cannot be relied upon as a substitute for good detergency or chemical sanitation in the wash (22, 26). Jerram (15) concluded that a hot air (tumble) dryer does not have as great a bactericidal effect as calenders found in commercial laundries.

Summary of Review of Literature

Research has indicated that hot water (140° F. or higher), .1% to .2% detergent concentration, and proper agitation are needed to remove soil and bacteria from fabric during laundering resulting in a sanitary wash. There was some disagreement on the amount of cross contamination or redeposition occuring in the washing process. The tendency was to believe that drying has little, if any, effect on the bacterial removal.

PROCEDURE

Laundry Equipment

The equipment used in this research consisted of a home laundry automatic top loading washer and automatic tumble dryer. Since small loads were washed, a small inner-tub called a "mini-basket" was used in the washer with a low water level of eighteen liters. The wash period lasted ten minutes with delicate agitation or 85 rpm. of the agitator. The water was spun out of the fabric with a medium spin speed while a spray rinse removed the suds. After a minute's pause a deep rinse period lasted for three minutes and then a final spin left the fabric damp dry. The entire washing cycle took thirty minutes.

The dryer time was regulated by an automatic electronic sensor. A delicate setting was used having an air temperature of 126° F. and the drying period lasted approximately thirty minutes.

Water Temperature

Three water temperature settings used for the washings were cold wash with a cold rinse at 60° F. ± 4°, a warm wash with a warm rinse at 100° F. ± 2°, and a hot wash of 140° F. ± 2° with a warm rinse. The water temperatures used for the wash and rinse were regulated as the water entered the washer and varied somewhat due to pressure changes of the water as it entered the machine. Similar variation would be found in a home situation. The pH of the tap water ranged from six to seven.

Detergent

The detergent used was a built, enzyme containing, all purpose synthetic detergent. Surveys of the supermarkets in the Manhattan area found the detergent used to be the most frequently purchased. Detergent concentrations of 0% (none), .1%, .2%, and .4% by weight were used. The washer manufacturer recommended a detergent amount of 1/3 cup for the "mini-load". This was found equilivant to 36 grams and produced a .2% detergent concentration in solution. One half and double the recommended amounts of detergent were also used.

Fabric Preparation and Sampling

A terry knit fabric of 50% wool, 30% nylon, and 20% cotton meeting military specification MIL S-486 (appendix C, p. 68) and a rib knit fabric of 60% nylon and 40% cotton meeting military specification MIL S-12549E (appendix C, p. 66) were used. Both United States Air Force sock fabrics were black and knitted in the form of seamless tubes 7 to 8 inches in circumference and approximately 24 to 36 inches long.

The tubes were split and cut into twelve inch long swatches. One inch squares were marked on 360 nylon and cotton swatches and 360 wool, nylon, and cotton swatches. The swatches were selected at random and washed for ten minutes with hot water and detergent, then dryed, to remove any finish remaining from the fabric construction.

Experimental Sequence

Half of the washed swatches of each fabric were soiled in a synthetic soil used for bacteria-soil mixtures (appendix D, p. 71). The soil allowed maximum bacterial growth on the fabric. Each swatch was soaked in 15 to 20 cc. of soil for approximately a minute, wrung damp dry by hand, and placed on a wire rack.

The soiled swatches were then suspended in a Chromato cab air-tight chamber and inoculated with a suspension of Staphylococcus aureus A. T. C. C. # 6538 by aerosal exposure. One and one half millititers of the test organism suspension was used per fabric swatch. (See Tables I and III, appendix B, pp. 55 and 57 for original inoculum counts.) The inoculated swatches were put into plastic bags and held from 24 to 36 hours before washing to allow stabilization of the inoculum on the fabric.

Before washing, an inch square was removed from two of the inoculated swatches for an initial Staphylococcus aureus count (Tables I and III, appendix B, pp. 55 and 57). Five soiled, inoculated and five unsoiled, non-inoculated swatches of the same fabric were put into the "mini-basket", detergent was added, and the washer was started. A sample of the water was taken after a minute of agitation, at the end of the wash period, and at the end of the rinse period for ascertaining pH and bacterial count of the water. At the end of the final spin, the damp swatches were removed from the washer and a single one-inch square was taken from each of the ten swatches in the wash. The bacterial count obtained from the five soiled, inoculated one-inch square samples was used to determine the survival of the test organism on the fabric after wash. The bacterial count obtained from the five unsoiled, non-inoculated fabric samples was used to ascertain the redeposited bacteria.

The laundered 12" x 8" swatches were then dryed. Another one-inch square was removed from six of the swatches drawn at random from the dryer. The entire sequence was repeated three times for each of the two fabrics using a different set of swatches.

The inch square samples removed from the fabric swatches before washing, at the end of the wash cycle, and at the end of the drying period were put into a test tube containing 9.0 cc. of Trypticase Soy Broth. The tubes were agitated on a Vortex stirrer for two minutes to remove the bacteria from the fabric and suspend it in broth. Broth dilutions of 1:10, 1:1000, and 1:100,000 were made. A milliliter of each dilution of the agitated fabric samples and of the three wash water samples was pipetted onto two petri dishes and Mannitol Salt Agar, a selective medium for the isolation of staphylococci, was added with a swirling motion to distribute the organisms evenly. The petri dishes were incubated for 48 hours at 37° C. and 60% relative humidity. Two plates of the same dilution having readily countable colonies of Staphylococcus aureus were selected and the number of colonies were counted and multiplied by the dilution factor. A mean of the ten plate counts of survival after washing, redeposition, and the mean of six plate counts of survival after drying were calculated (Fig. 1, p. 19).

The washer tub and dryer drum were swabed with sterile cotton tip sticks after each washing cycle. The swabs were diluted and plated in a similar manner to the fabric samples to determine the extent of bacterial survival in the washer and dryer. The washer was disinfected by using one-half cup of chlorine bleach with hot water in a regular wash cycle. The dryer was disinfected by allowing it to run at a regular setting (196° F.) for thirty minutes.

Statistical Analysis

Statistical test of the mean of three repetitions of the washings, an F test for variance, and a test of least standard difference between the means were calculated for the three variables of water temperature, detergent concentration, and fabric as well as interactions of the variables. Nine factors were analyzed for results: the bacterial survival on the fabric at the end of the wash cycle and drying period, the bacterial redeposition during the wash, the

EXPLANATION OF FIGURE 1

The experimental sequence was carried out for each washing treatment. The following combinations of water temperature and detergent concentration were used for each of the two fabrics:

1. hot water, no detergent
2. hot water, .1% detergent concentration
3. hot water, .2% detergent concentration
4. hot water, .4% detergent concentration
5. warm water, no detergent
6. warm water, .1% detergent concentration
7. warm water, .2% detergent concentration
8. warm water, .4% detergent concentration
9. cold water, no detergent
10. cold water, .1% detergent concentration
11. cold water, .2% detergent concentration
12. cold water, .4% detergent concentration

Each washing treatment was repeated three times. A mean of the three treatments was used in analysis.

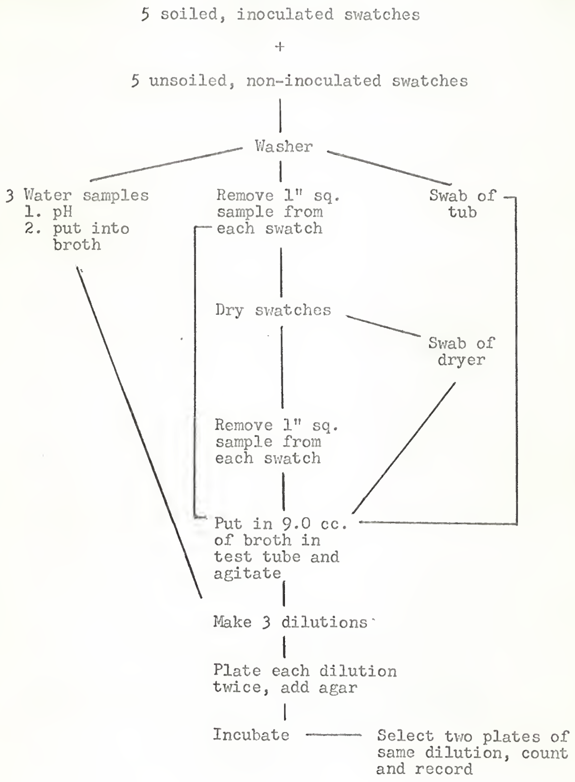

FIGURE 1

FLOW CHART OF EXPERIMENTAL SEQUENCE

bacterial count found in the wash water after one minute of agitation, the bacterial count found in the wash water at the end of the washing period, the bacterial count of the water at the end of the rinse period, the pH of the wash water, and representative bacterial survival in the washer and in the dryer.

RESULTS AND DISCUSSION

There was some bacterial survival on the fabric after washing and drying even with the use of hot water and the washer manufacturer's recommended concentration of detergent. Some survival of the test organism, Staphylococcus aureus, occurred with all washing procedures carried out except five instances out of twenty-four hot water washings and seventy-two total washings.

The fluctuation of the water pressure and inadequacy of hot water at times resulted in difficulty in controlling the water temperatures especially during the rinse periods. Thus, the rinse water temperatures varied. This situation is similar to that found in the home, but complicated evaluation of bacterial removal as a result of the water temperature (Tables II and IV, appendix B, pp. 56 & 58).

Effect of Water Temperature

The water temperature was found to be the most significant variable by an F test of significant variance in the bacterial removal and redeposition (Tables VI, VII, and VIII, appendix B, pp. 60, 61). Using hot water of 140° F. at all detergent levels removed 99.99% of the Staphylococcus aureus and resulted in traces of bacterial redeposition. Evaluation of the effectiveness of water temperature was made irregardless of the detergent concentrations.

A statistically significant difference at the 95% level in the variance of the bacterial survival on the fabric at the end of the wash cycle and drying period was shown by an F test to be affected by the water temperature (Tables VI and VII, pp.60). A least significant difference test at the 95% level resulted in significant differences between the water temperatures for bacterial survival on the fabric. At the end of the wash cycle, the 140° F. wash water was significantly different from the two lower water temperatures of 100° F. and 60° F. No significant difference was shown between the lower water temperatures of 100° F. and 60° F. in the bacterial removal at the end of the wash cycle. Figure 2 illustrated that an increase in water temperature resulted in a decrease in bacterial survival.

The water temperature showed a significant difference in the variance of the bacterial count found in the wash water after a minute of agitation, at the end of the wash period, and at the end of the rinse period when an F test was used. As the washing temperature increased, there was a decrease in bacterial count found in the wash waters (Fig. 3). A significant difference was shown between using 140° F. wash water and the two lower wash water temperatures of 100° F. and 60° F. in the bacterial count of the wash water after a minute of agitation, at the end of the wash period, and at the end of the rinse period. As the temperature of the wash water increased, there was a decrease in bacterial survival in the washer and dryer.

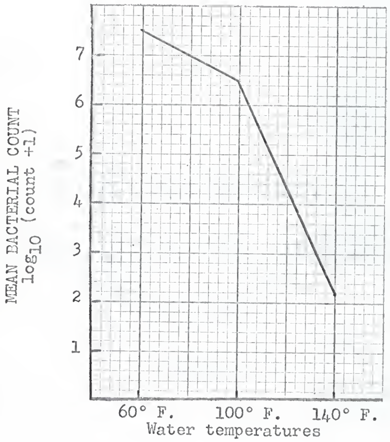

Figure 2. Mean bacterial survival count on fabric after washing at three water temperatures with no detergent. (Significant variance at the 95% level.)

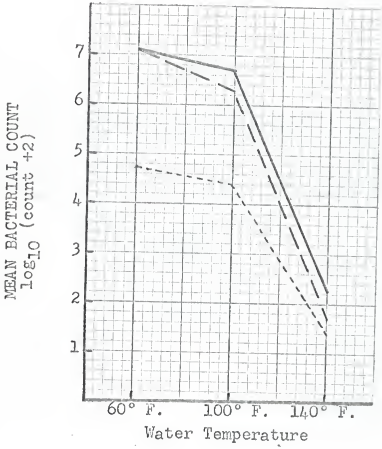

Figure 3. Mean bacterial count of the wash waters at three water temperatures with no detergent. (Significant variance at the 95% level.)

Key:
after one minute of agitation ________
end of agitation — — —
end of rinse - - - -

Effect of Detergent Concentration

The detergent concentration was found to be statistically significant in bacterial survival on fabric at the end of the wash cycle and drying period (Tables VI and VII, appendix B, pp. 60). In staphylococcial removal from the fabric after washing and in the redeposition during washing, a significant difference between no detergent and the three detergent concentrations was found. No significant differences was found between .1%, .2%, and .4% detergent concentrations in bacterial survival after washing and bacterial redeposition. However, there was a significant difference between use of .1% detergent concentration and a .4% detergent concentration in the bacterial survival on the fabric after it was dryed. As the detergent concentration increased, there was a decrease in bacterial survival on the fabric at the end of the wash cycle, drying period and a decrease in bacterial redeposition (Figs. 4, 5, and 6).

The detergent concentration showed a significant difference in the bacterial count found in the wash and rinse waters (Tables IX, X, and XI, appendix B, pp. 61, 62). An L. S. D. test found differences between the detergent concentrations in the bacterial counts of the wash waters (Fig. 7). Statistically significant differences using L. S. D. test occured between no detergent and detergent concentrations of .2% and .4% in bacterial count of the wash water after a minute of agitation. No difference was found between 0% and

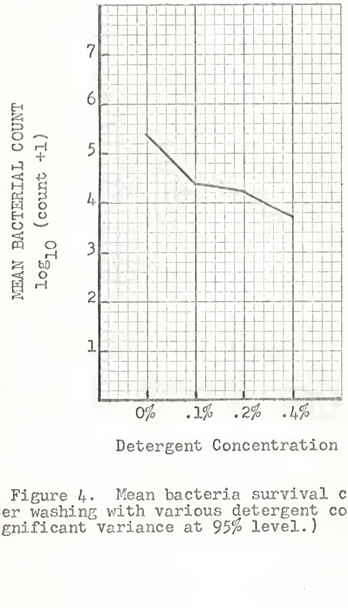

Figure 4. Mean bacteria survival count on fabric after washing with various detergent concentrations. (Significant variance at 95% level.)

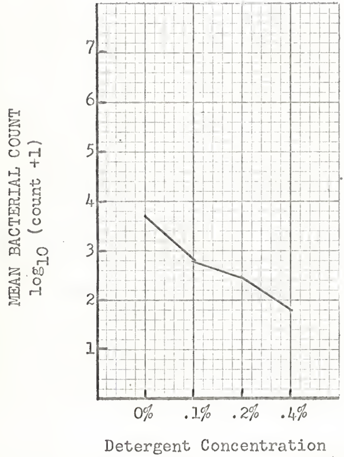

Figure 5. Mean bacterial survival count after drying inoculated fabric washed with various detergent concentrations. (Significant variance at 95% level.)

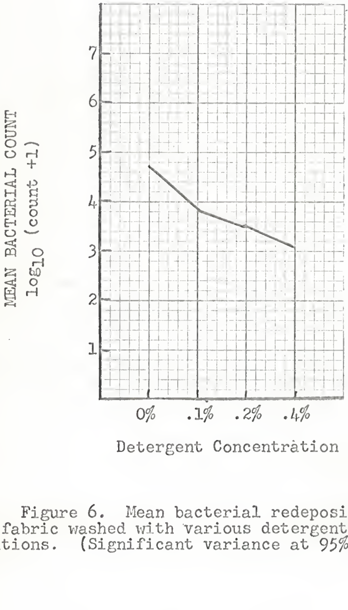

Figure 6. Mean bacterial redeposition count on fabric washed with various detergent concentrations. (Significant variance at 95% level.)

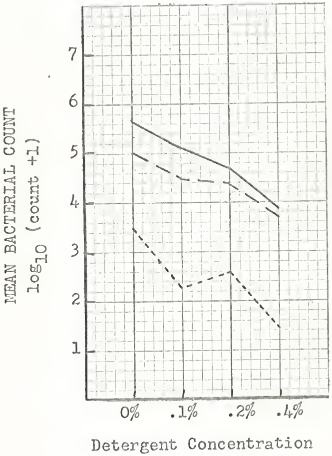

Figure 7. Mean bacterial count in wash waters with various detergent concentrations. (Significant variance at 95% level.)

Key:
after one minute of agitation ————
end of agitation — — — —
end of rinse - - - -

.1%, between .1% and .2%, and between .2% and .4% detergent concentrations. At the end of the wash period the only significant difference in bacterial count of the wash water occured between no detergent and a .4% detergent concentration. In the rinse water, a significant difference was found between no detergent and detergent concentrations of .2% and .4% in the bacterial count of the water. Swabs of the washer and dryer showed no statistically significant difference in bacterial survival in the washer and dryer between the detergent concentrations.

Effect of Fabric

The type of fabric was statistically significant at the 95% level in the variance of the bacterial survival on the fabric at the end of the wash cycle and drying period, of the bacterial redeposition, and the bacterial survival in the washer tub (Tables VI, VII, VIII, and XIII, appendix B, pp.60, 61, 63). In most cases the 50% wool, 30% nylon, 20% cotton terry knit fabric had more bacterial survival and bacterial redeposition than the rib knit 60% nylon and 40% cotton fabric. Figure 8 illustrated that with the bacterial survival on the fabric after washing, using hot water, similar patterns were shown between the two fabrics. The bacterial survival decreased with an addition of detergent when hot water was used on both fabrics.

When warm water was used, a .2% detergent concentration

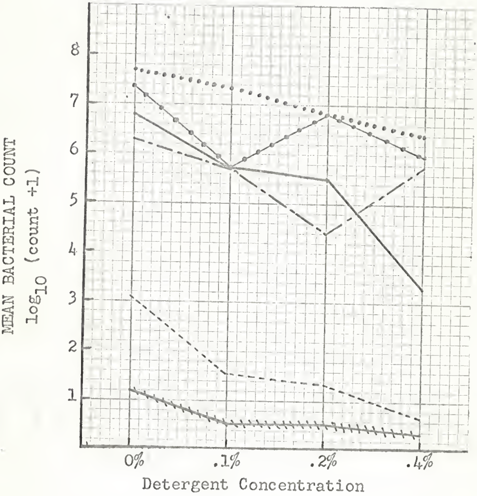

Figure 8. Mean bacterial survival count after washing two fabrics at hot, warm, and cold water temperatures with various detergent concentrations.

Key:

cold water, wool blend fabric · · · · ·
cold water, nylon and cotton fabric_._._._._
warm water, wool blend fabric— — — — — —
warm water, nylon and cotton fabric ————————
hot water, wool blend fabric - - - - - -
hot water, nylon and cotton fabric +++++++++++

resulted in the greatest bacterial removal in the wool blend fabric but a .4% detergent concentration resulted in the greatest bacterial removal of the nylon, cotton fabric.

When cold water was used on the wool blend fabric, there was a steady decrease in bacterial survival as the detergent concentration increased. In the cotton and nylon fabric, however, a different pattern occurred in the bacterial survival on the fabric at the end of the wash cycle. This pattern reoccurred in the survival after drying and in the redeposition of bacteria (Figs. 13 and 14, appendix A, pp. 49, 50). Figure 8 showed that bacterial survival first decreased with an increase in detergent concentration from 0% to .1%, then increased with a detergent concentration of .2%, and decreased again with a .4% detergent concentration. An L. S. D. test found no statistical difference between the fabrics in bacterial count of the wash and rinse waters and bacterial survival in the laundry equipment.

Some of the variances noted between the bacterial removal and bacterial redeposition on the two fabrics may have been a result of the degree of saturation of the initial inoculum (Tables I and III, appendix B, pp.55, 57). Since the wool, nylon, and cotton fabric was of a thick, terry construction; more inoculum was needed to infect the fabric. This may have resulted in less removal of the test organism during washing because of the fabric's construction.

Effect of Drying

Staphylococcus aureus counts on the fabric after drying followed a similar pattern to the bacterial counts on the fabric after washing, except that drying decreased the counts (Fig. 9). The bacterial survival on the fabric occurring after drying indicated that tumble drying at a delicate setting cannot be relied upon for sanitation. These findings were in agreement with the previous findings of Meyers (22) and Ridenour (26). The delicate setting and low temperature of drying was not hot enough to destroy the bacteria.

The survival of Staphylococcus aureus in the dryer at the end of the drying period was also an indication of the inadequacy of the tumble dryer to provide sanitation. Since viable test organisms remained in the dryer, a chance existed for the transfer of the organisms to items dryed in subsequent loads.

Bacterial Redeposition

Redeposition of the Staphylococcus aureus from the inoculated fabric to the non-inoculated fabric during the wash followed the same pattern as bacterial survival on the inoculated fabric after washing (Fig. 10). The bacterial transference was nearly the same as the bacterial survival count on the inoculated fabric after washing (Tables I and II, appendix B, pp. 55-56). In a previous study (26), it was found that an equilibrium in bacterial count on the

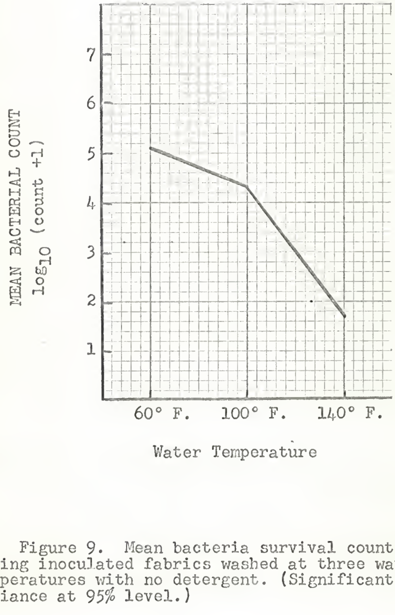

Figure 9. Mean bacteria survival count after drying inoculated fabrics washed at three water temperatures with no detergent. (Significant variance at 95% level.)

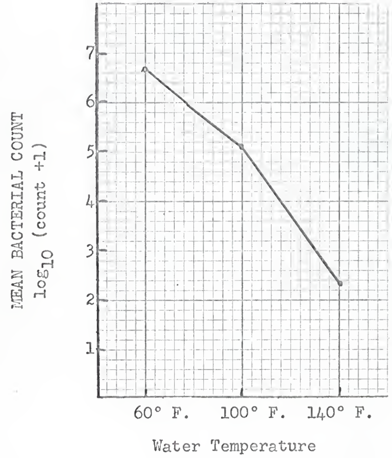

Figure 10. Mean bacterial redeposition count on fabric washed at three water temperatures with no detergent. (Significant variance at 95% level.)

fabric was approached for all materials in the load due to redeposition. McNeil (20, 21) also found evidence of bacterial redeposition during agitation and recommended further study be done on redeposition.

The water temperature, detergent concentration, and type of fabric were found to cause statistically significant differences in variance of the bacterial redeposition by an F test (Tables VIII-X, appendix B, pp. 61-62). There was a significant difference between each of the water temperatures in bacterial redeposition. Figures 5 and 10 showed that as the water temperature and detergent concentration increased, there was a decrease in bacterial redeposition during washing.

The Staphylococcus aureus survival in the washer at the end of the wash cycle was also a source of bacterial redeposition (Tables II and IV, appendix B, pp. 56, 58). Bacterial survival in the washer decreased with an increase in the water temperature. Survival of bacteria in the washer tub at the end of the washing cycle would be a means of bacterial transference to a suceeding load.

Affect on pH

A reaction between the soil and the bacteria was observed on the soiled, inoculated fabric which was held for one or two days. A "sour" odor indicated that the soil was metabolized by the bacteria, causing the fabric to become acidic before washing. The detergent had to have sufficient alkalinity to neutralize the soil-bacteria mixture and then remove the soil from the fabric. The acidity of the mixture on the fabric explained the neutral range of pH occurring at detergent concentrations of .1% and .2% (Tables II and IV, appendix B, 56, 58).

An F test showed statistically significant variance in pH with detergent concentrations, water temperatures, interactions of detergent concentration and water temperature, interactions of detergent concentration and fabric, interactions of water temperature and fabric, as well as interactions of all three variables (Table XII, appendix B, p. 63). As the water temperature increased, there was an increase in pH of the washing solution (Fig. 11). The pH of the solution did not increase until a .2% detergent concentration was used (Fig. 12).

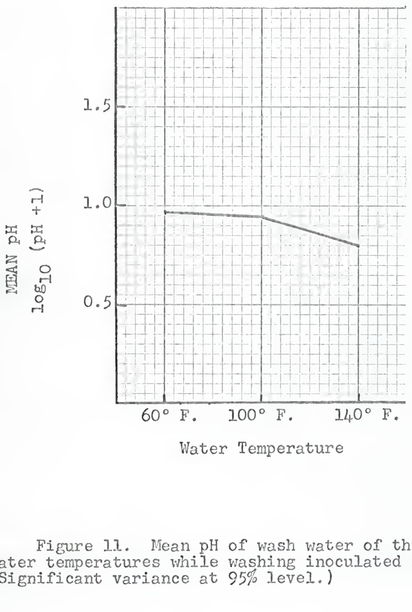

Figure 11. Mean pH of wash water of three water temperatures while washing inoculated fabric. (Significant variance at 95% level.)

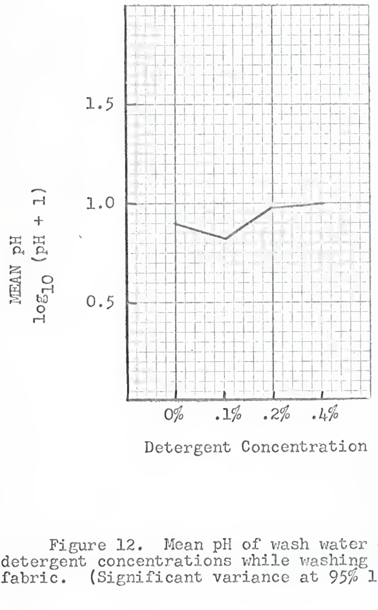

Figure 12. Mean pH of wash water of various detergent concentrations while washing inoculated fabric. (Significant variance at 95% level.)

SUMMARY AND CONCLUSIONS

The mean of three washes at each of the water temperatures of 140° F., 100° F., and 60° F. with detergent concentrations of 0%, .1%, .2%, and .4% did not result in 100% removal of the test organism, Staphylococcus aureus. Even with the use of 140° F. hot wash water, traces of bacterial survival on the fabric after washing and drying and redeposition of bacteria during washing occurred on both fabrics. A statistically significant difference in variance of bacterial survival was observed between the two fabrics. The bacterial removal from the nylon and cotton rib knit fabric was higher than from the wool blend terry knit fabric, probably due to the construction and thickness of the terry fabric.

As the water temperature and detergent concentration increased, there was a decrease in bacterial survival and redeposition. Bacterial redeposition during the wash showed similar trends as bacterial survival on the fabric after washing. A .2% detergent concentration used in cold water with the nylon and cotton fabric tended to increase bacterial survival and redeposition when compared to a .1% concentration. Survival of Staphylococcus aureus decreased slightly with tumble drying for each washing treatment.

The Staphylococcus aureus found in the wash and rinse waters decreased as the water temperature and detergent concentration increased. There was no statistically significant variance between the two fabrics in the staphylococci count of the wash and rinse waters. Bacterial survival in the water at the end of the agitation period was decreased from the bacterial survival after a minute of agitation. Bacterial survival in the rinse water was less than the bacterial counts of the wash water samples.

As the detergent concentration and the water temperature increased, the pH of the wash water increased. Swabs of the washer and dryer indicated that bacterial survival decreased as the detergent concentration and the water temperature increased. Bacterial survival in the washer was greater than in the dryer. No statistically significant variance was shown between the interaction of the three variables in bacterial survival after washing and drying, bacterial redeposition during washing, bacterial counts of the wash and rinse waters, and bacterial survival in the washer and dryer at the 95% level.

Since only 100% removal of the test organism, Staphylococcus aureus, was considered satisfactory; none of the combinations of washing temperatures and detergent concentrations were found adequate in providing a sanitary wash. The initial inoculum was considerably higher than bacteria counts found in naturally soiled clothing and may have accounted for the ineffective sanitation of the hot water and recommended amount of detergent. It is suggested that a similar study be carried out using less initial inoculum or actual wear garments for laundering.

Several other suggestions for future research are made. A suggestion would be the use of water temperatures of 160° F. or higher as used in commercial laundries. The hardness of the water was not considered in this study and future research may want to include it as well as determination of the effect of pH on bacterial removal. Further study is needed to determine the influence of the differences of fabric construction and fiber content upon bacterial removal and transference in the laundry. It would also be interesting to compare the bacterial removal and soil removal at various water temperatures and detergent concentrations, as well as a study of the effect (if any) of detergents containing enzymes on bacterial survival on fabric during washing.

The amount of redeposition and bacterial survival in the washer and dryer indicated the danger of cross-contamination within the wash load and with succeeding loads. This would be an important consideration in the home and in the use of public laundry facilities. It is suggested that the homemaker treat infected garments separately from the rest of the laundry and be cautious in the use of public facilities. For a sanitary wash, sufficiently hot water is recommended with the amount of detergent dependent upon the type of fabric. Proper care for the fabric may have to be sacrificed if a garment is greatly infected. This research has been a small beginning in the area of bacterial removal by laundering and many questions are left unanswered.

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APPENDIX A

EXPLANATION OF FIGURES

The figures in appendix A represent the interaction of the three variables of water temperature, detergent concentration, and fabric upon the mean bacterial survival on the fabric and mean bacterial count of the wash water of three washings. The mean survival has been converted into the $\log_{10}$ (count + 1). The interaction of the variables has been analysed for several factors, survival after drying, redeposition, bacterial count of the wash and rinse waters, and bacterial survival in the washer and dryer.

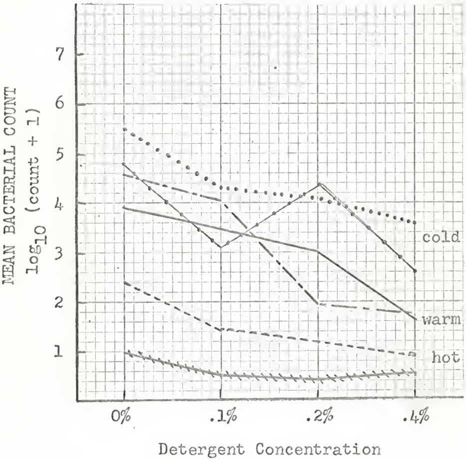

Figure 13. Mean bacterial survival count after drying two fabrics washed at three water temperatures with various detergent concentrations.

Key:

Wool blend, cold water · · · · ·
Nylon and cotton, cold water —.—.—.—.
Wool blend, warm water — – — – — –
Nylon and cotton, warm water ————
Wool blend, hot water – – – – – –
Nylon and cotton, hot water ⧵⧵⧵⧵⧵⧵⧵⧵⧵⧵

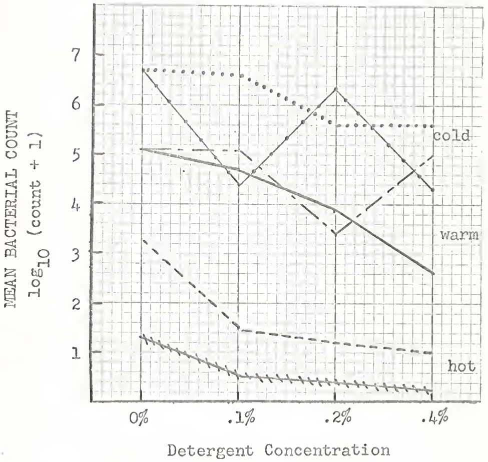

Figure 14. Mean bacterial redeposition count on two fabrics washed at three water temperatures with various detergent concentrations.

Key:

cold water, wool blend · · · · ·
cold water, nylon and cotton —·—·—·—·—·
warm water, wool blend — — — — — —
warm water, nylon and cotton ————————
hot water, wool blend - - - - -
hot water, nylon and cotton ⧵⧵⧵⧵⧵⧵⧵⧵⧵

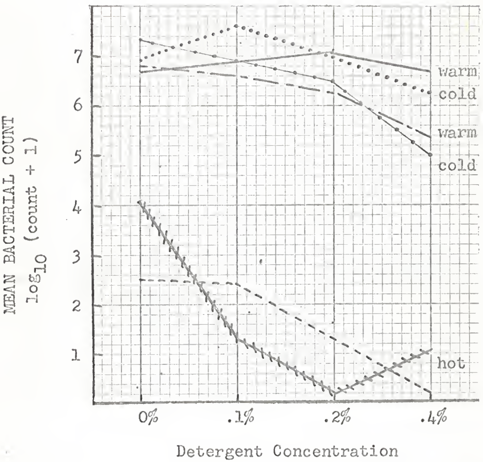

Figure 15. Mean bacterial count in wash water after one minute of agitation of two fabrics at three water temperatures with various detergent concentrations.

Key:

cold water, wool blend · · · · ·
cold water, nylon and cotton –·–·–·–·–·
warm water, wool blend — – — – — – —
warm water, nylon and cotton ————————
hot water, wool blend – – – – –
hot water, nylon and cotton +++++++++

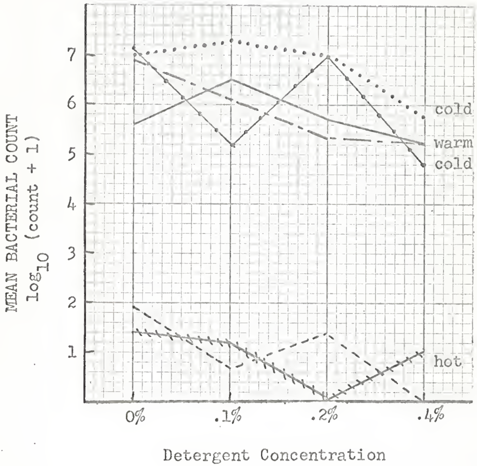

Figure 16. Mean bacterial count in wash water at the end of agitation of two fabrics at three water temperatures with various detergent concentrations.

Key:

cold water, wool blend · · · · ·
cold water, nylon and cotton —·—·—·—·—·
warm water, wool blend — – — – — –
warm water, nylon and cotton ————————
hot water, wool blend – – – – –
hot water, nylon and cotton ⧵⧵⧵⧵⧵⧵⧵⧵⧵

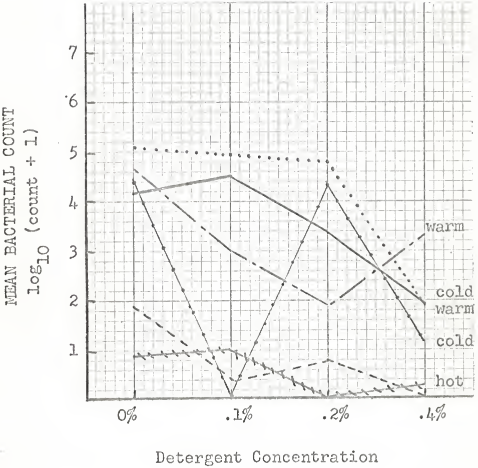

Figure 17. Mean bacterial count found in the rinse water while washing two fabrics at three water temperatures with various detergent concentrations.

Key:

cold water, wool blend · · · · ·
cold water, nylon and cotton —·—·—·—·—·
warm water, wool blend — — — — — —
warm water, nylon and cotton ————————
hot water, wool blend - - - - - - -
hot water, nylon and cotton ++++++++

APPENDIX B

TABLE I

ORIGINAL INOCULUM COUNT, INITIAL COUNT BEFORE WASH, SURVIVAL AFTER WASH, SURVIVAL AFTER DRYING, AND REDEPOSITION COUNT OF STAPHYLOCOCCUS AUREUS AT VARIOUS WATER TEMPERATURES AND DETERGENT CONCENTRATIONS ON NYLON AND COTTON FABRIC

(Numbers are counts per sq. inch of fabric)

Water Temp.	Det. Conc.	Wash #	O-inoc. $x 10^6$	Initial count $x 10^6$	Survival after wash $x 10^6$	Survival after dry $x 10^6$	Redeposition $x 10^6$
Hot	none	1	850	28	0	0	.000001
Hot	none	2	2500	191	.005000	.001000	.003000
Hot	none	3	235	375	0	0	.000001
Hot	.1%	1	4900	101	.000048	.000025	.000054
Hot	.1%	2	252	1231	.000001	.000001	.000002
Hot	.1%	3	236	2558	0	0	0
Hot	.2%	1	4900	142	.000014	.000008	.000014
Hot	.2%	2	1300	705	0	.000001	.000001
Hot	.2%	3	168	745	.000003	0	0
Hot	.4%	1	4900	143	.000005	.000007	.000005
Hot	.4%	2	1400	705	.000001	.000001	0
Hot	.4%	3	41	749	.000001	.000001	0
Warm	none	1	1450	133	13.000000	.006560	.270000
Warm	none	2	2500	191	29.770000	.102000	.235600
Warm	none	3	236	375	1.100000	.001320	.033000
Warm	.1%	1	6800	97	.193000	.007775	.055000
Warm	.1%	2	1500	521	2.030000	.001597	.100500
Warm	.1%	3	168	745	.540000	.004527	.041500
Warm	.2%	1	6800	97	.240000	.002915	.001200
Warm	.2%	2	1500	521	1.610000	.001195	.030500
Warm	.2%	3	168	745	.070800	.000524	.013900
Warm	.4%	1	6800	150	.000225	.000067	.000146
Warm	.4%	2	252	1232	.131900	.000842	.016800
Warm	.4%	3	41	749	.000301	.000001	.000042
Cold	none	1	2500	22	10.400000	.023266	1.400000
Cold	none	2	2.36	2557	73.000000	.478000	5.600000
Cold	none	3	41	372	34.320000	.254000	29.790000
Cold	.1%	1	252	931	1.900000	.000503	.061000
Cold	.1%	2	168	488	1.840000	.000502	.081400
Cold	.1%	3	90	160	.045600	.011000	.006300
Cold	.2%	1	252	123	31.500000	.013337	.113700
Cold	.2%	2	41	934	21.180000	.011494	30.230000
Cold	.2%	3	90	160	3.870000	.115000	5.460000
Cold	.4%	1	1400	705	.446900	.000091	.008400
Cold	.4%	2	4100	934	12.200000	.001526	.046500
Cold	.4%	3	895	160	28.340000	.000705	.039400

TABLE II

WASH AND RINSE WATER STAPHYLOCOCCUS AUREUS COUNTS AND pH OF WASH WATER AT VARIOUS WATER TEMPERATURES AND DETERGENT CONCENTRATIONS WITH NYLON AND COTTON FABRIC

(Numbers are counts per milliliter of water)

Water Temp.	Det. Conc.	Wash #	Actual wash	Temp. rinse	After min. agitation x 10^6	End of wash x 10^6	End of rinse x 10^6	pH
Hot	none	1	140°	96°	.012200	.000015	0	7.12
Hot	none	2	140°	100°	.614000	.000300	.000738	7.14
Hot	none	3	141°	100°	.000453	.000005	0	7.50
Hot	.1%	1	142°	94°	.010000	.007500	.001000	7.41
Hot	.1%	2	142°	60°	.001300	.000200	.000003	8.38
Hot	.1%	3	140°	140°	0	0	0	8.00
Hot	.2%	1	140°	130°	0	0	0	9.29
Hot	.2%	2	140°	140°	0	0	0	8.90
Hot	.2%	3	140°	100°	3	0	0	9.10
Hot	.4%	1	140°	136°	.001600	.002000	.000012	9.34
Hot	.4%	2	140°	140°	0	0	0	9.80
Hot	.4%	3	142°	100°	0	0	0	9.50
Warm	none	1	100°	60°	2.510000	4.670000	3.150000	7.05
Warm	none	2	100°	100°	7.520000	5.580000	.012800	7.12
Warm	none	3	100°	100°	7.900000	.800000	.000148	6.40
Warm	.1%	1	102°	60°	8.120000	3.670000	.925000	8.05
Warm	.1%	2	100°	100°	6.800000	3.000000	.000743	7.62
Warm	.1%	3	100°	100°	14.000000	5.100000	.050000	8.90
Warm	.2%	1	101°	59°	3.850000	3.620000	.000350	9.20
Warm	.2%	2	100°	100°	5.500000	4.400000	.049500	9.09
Warm	.2%	3	100°	100°	2.730000	1.400000	.001230	9.30
Warm	.4%	1	100°	62°	.009500	.043500	0	9.28
Warm	.4%	2	100°	80°	.800000	5.500000	.008300	9.10
Warm	.4%	3	100°	100°	.133000	.023400	.000088	9.20
Cold	none	1	62°	63°	6.500000	6.400000	.020300	7.18
Cold	none	2	62°	62°	89.370000	42.630000	.004125	6.90
Cold	none	3	60°	61°	26.300000	22.800000	.300000	7.65
Cold	.1%	1	60°	60°	6.150000	.060000	0	8.48
Cold	.1%	2	60°	60°	4.200000	.600000	0	8.10
Cold	.1%	3	64°	64°	.029300	.011500	0	9.37
Cold	.2%	1	60°	60°	71.150000	60.000000	.225000	8.59
Cold	.2%	2	60°	60°	18.050000	18.550000	.200000	8.60
Cold	.2%	3	62°	62°	2.100000	11.570000	.000483	9.32
Cold	.4%	1	62°	62°	.050300	.043200	.005500	9.30
Cold	.4%	2	60°	60°	4.970000	5.550000	0	9.50
Cold	.4%	3	62°	62°	.650000	.198000	0	9.67

TABLE III

ORIGINAL INOCULUM COUNT, INITIAL COUNT BEFORE WASH, SURVIVAL AFTER WASH, SURVIVAL AFTER DRYING, AND REDEPOSITION COUNT OF STAPHYLOCOCCUS AUREUS AT VARIOUS WATER TEMPERATURES AND DETERGENT CONCENTRATIONS ON WOOL, NYLON, AND COTTON FABRIC

(Numbers are counts per sq. inch of fabric)

Water Temp.	Det. Conc.	Wash #	O-inoc. x 10^6	Initial count x 10^6	Survival after wash x 10^6	Survival after dry x 10^6	Redeposition x 10^6
Hot	none	1	9900	16	0	0	0
Hot	none	2	202	658	.050000	.003040	.209500
Hot	none	3	41	341	.069700	.007485	.075000
Hot	.1%	1	9900	16	.000001	.000001	0
Hot	.1%	2	202	1293	.028957	.001037	.023540
Hot	.1%	3	33	1560	0	.000008	0
Hot	.2%	1	9900	1600	0	0	0
Hot	.2%	2	161	1073	0	0	0
Hot	.2%	3	34	16	.015000	.004539	.003240
Hot	.4%	1	9900	16	0	0	0
Hot	.4%	2	161	2337	0	.000003	.000370
Hot	.4%	3	41	104	.000148	.000001	.000402
Warm	none	1	1450	159	.252200	.016200	.003000
Warm	none	2	202	6586	.669200	.093000	.050800
Warm	none	3	41	341	53.150000	.050000	1.500000
Warm	.1%	1	1450	159	.333700	.009213	.053700
Warm	.1%	2	202	1293	.033700	.017000	.023600
Warm	.1%	3	33	1560	13.440000	.025240	2.574000
Warm	.2%	1	850	32	.014228	.000016	.001128
Warm	.2%	2	161	1073	.039000	.002919	.071000
Warm	.2%	3	41	310	.003771	.000016	.000337
Warm	.4%	1	850	164	.627770	.000188	.130810
Warm	.4%	2	161	2337	1.200000	.000002	.212000
Warm	.4%	3	41	104	.357600	.000163	.049300
Cold	none	1	202	66	48.450000	4.700000	6.960000
Cold	none	2	41	1700	48.400000	.090000	9.260000
Cold	none	3	41	10	57.860000	.803000	2.430000
Cold	.1%	1	202	1293	37.840000	.100000	5.490000
Cold	.1%	2	33	1560	21.880000	.002870	3.140000
Cold	.1%	3	41	104	31.260000	.015187	3.880000
Cold	.2%	1	161	1073	7.770000	.001336	.930000
Cold	.2%	2	41	310	3.435000	.001219	.060600
Cold	.2%	3	41	104	11.830000	.205192	2.040000
Cold	.4%	1	161	2337	26.040000	.007615	2.700000
Cold	.4%	2	41	310	3.510000	.004171	.710000
Cold	.4%	3	70	217	.183800	.035000	.038000

TABLE IV

WASH AND RINSE WATER STAPHYLOCOCCUS AUREUS COUNTS AND pH OF WASH WATER AT VARIOUS WATER TEMPERATURES AND DETERGENT CONCENTRATIONS WITH WOOL, NYLON, AND COTTON FABRIC

(Numbers are counts per milliliter of water)

Water Temp.	Det. Conc.	Wash #	Actual wash	Temp. rinse	After min. agitation x 10^6	End of wash x 10^6	End of rinse x 10^6	pH
Hot	none	1	140°	102°	.000065	0	0	7.00
Hot	none	2	140°	140°	.000500	.000572	.000775	6.90
Hot	none	3	140°	100°	.001500	.001500	.001000	7.00
Hot	.1%	1	140°	102°	.000005	0	0	8.00
Hot	.1%	2	140°	105°	.001100	.000120	.000020	7.30
Hot	.1%	3	140°	140°	.004760	0	0	8.10
Hot	.2%	1	141°	140°	0	0	0	9.00
Hot	.2%	2	144°	100°	0	0	0	8.40
Hot	.2%	3	140°	140°	.020000	.017400	.000500	8.10
Hot	.4%	1	141°	141°	0	0	0	9.50
Hot	.4%	2	140°	140°	0	0	0	9.00
Hot	.4%	3	140°	140°	.000003	0	.000003	9.55
Warm	none	1	100°	54°	4.700000	2.350000	.000662	6.28
Warm	none	2	101°	101°	3.050000	6.030000	.148800	7.30
Warm	none	3	100°	100°	25.300000	39.050000	1.550000	7.00
Warm	.1%	1	102°	60°	.650000	.970000	.006500	7.19
Warm	.1%	2	100°	99°	15.700000	.191000	.281000	7.20
Warm	.1%	3	100°	100°	7.320000	12.620000	0	7.30
Warm	.2%	1	101°	60°	.039000	.003000	.000075	9.01
Warm	.2%	2	102°	102°	17.450000	.236000	.000007	8.20
Warm	.2%	3	100°	100°	11.880000	11.000000	.001100	8.30
Warm	.4%	1	99°	60°	.031500	.000930	.035200	9.28
Warm	.4%	2	100°	100°	.550000	2.600000	.000030	9.10
Warm	.4%	3	100°	100°	1.000000	1.930000	.010000	9.47
Cold	none	1	62°	62°	11.570000	11.430000	.100000	6.60
Cold	none	2	60°	60°	11.620000	20.750000	.150000	7.10
Cold	none	3	61°	61°	5.480000	.134000	.134000	7.60
Cold	.1%	1	61°	61°	33.120000	38.700000	1.650000	7.40
Cold	.1%	2	62°	62°	12.750000	20.000000	.125000	7.70
Cold	.1%	3	61°	61°	212.500000	15.080000	.004500	8.85
Cold	.2%	1	62°	62°	27.400000	15.000000	.001920	8.70
Cold	.2%	2	62°	62°	7.030000	6.250000	.136800	8.94
Cold	.2%	3	60°	60°	7.980000	16.930000	1.630000	9.38
Cold	.4%	1	61°	60°	6.000000	.003360	0	9.30
Cold	.4%	2	62°	62°	2.550000	.650000	.550000	9.10
Cold	.4%	3	64°	64°	.980000	.165000	0	9.86

TABLE V

STAPHYLOCOCCUS AUREUS COUNTS REMAINING IN THE WASHER AND DRYER AFTER FABRIC WAS WASHED AT VARIOUS WATER TEMPERATURES AND DETERGENT CONCENTRATIONS*

Water Temp.	Det. Conc.	Wash #	Nylon, Cotton Fabric washer	dryer	Wool, Nylon, Cotton Fabric washer	dryer
Hot	none	1	1	1	1	1
Hot	none	2	1	1	101	0
Hot	none	3	235	2	1668	301
Hot	.1%	1	20	22	0	0
Hot	.1%	2	0	0	7	0
Hot	.1%	3	0	0	0	22
Hot	.2%	1	1	1	0	0
Hot	.2%	2	0	0	0	0
Hot	.2%	3	0	0	1034	6
Hot	.4%	1	1	0	0	0
Hot	.4%	2	0	0	2	1
Hot	.4%	3	0	0	1	1
Warm	none	1	101	0	506	39
Warm	none	2	1	1	1	0
Warm	none	3	225	1	1000	1
Warm	.1%	1	278	1	632	46
Warm	.1%	2	32	1	225	1
Warm	.1%	3	570	1	2120	5
Warm	.2%	1	42	0	2	0
Warm	.2%	2	186	1	443	0
Warm	.2%	3	289	1	0	1
Warm	.4%	1	7	19	228	2
Warm	.4%	2	97	0	300	0
Warm	.4%	3	2	1	330	1
Cold	none	1	602	1	680	8
Cold	none	2	1831	1	5150	1
Cold	none	3	34	924	640	4
Cold	.1%	1	32	1	1032	0
Cold	.1%	2	246	0	4640	8
Cold	.1%	3	80	8	2465	3
Cold	.2%	1	1845	10	1526	1
Cold	.2%	2	689	15	780	2
Cold	.2%	3	64	0	749	1
Cold	.4%	1	238	1	529	1
Cold	.4%	2	500	0	2268	1
Cold	.4%	3	197	0	222	1

*The counts were representative numbers taken from a swab of a particular area of the washer tub and dryer drum. The swabs did not include the total area and thus the counts were not of the total number of Staphylococcus aureus remaining in the washer and dryer.

TABLE VI

ANALYSIS OF VARIANCE FOR SURVIVAL AFTER WASH

Source of Variance	Degrees of Freedom	F Test for Significance
Detergent Concentration	3	5.54539 *
Water Temperature	2	127.39969 *
Fabric	1	4.07546 *
Det. Conc. x Water Temp.	6	0.24732
Det. Conc. x Fabric	3	0.68147
Water Temp. x Fabric	2	0.58908
Water Temp. x Det. Conc. x Fabric	6	1.32679
Error	48	
Total	71	

*Significant at 95% level.

TABLE VII

ANALYSIS OF VARIANCE FOR SURVIVAL AFTER DRYING

Source of Variance	Degrees of Freedom	F Test for Significance
Detergent Concentration	3	9.95473 *
Water Temperature	2	53.19865 *
Fabric	1	4.71354 *
Det. Conc. x Water Temp.	6	1.59295
Det. Conc. x Fabric	3	1.11587
Water Temp. x Fabric	2	1.11024
Det. Conc. x Water Temp. x Fabric	6	0.30779
Error	48	
Total	71	

*Significant at 95% level.

TABLE VIII

ANALYSIS OF VARIANCE FOR REDEPOSITION COUNT

Source of Variance	Degrees of Freedom	F Test for Significance
Detergent Concentration	3	5.18503 *
Water Temperature	2	85.37480 *
Fabric	1	6.90854 *
Det. Conc. x Water Temp.	6	0.66916
Det. Conc. x Fabric	3	1.43550
Water Temp. x Fabric	2	0.29939
Det. Conc. x Water Temp. x Fabric	6	0.99549
Error	48	
Total	71	

*Significant at 95% level.

TABLE IX

ANALYSIS OF VARIANCE FOR COUNT IN WASH WATER TAKEN AFTER ONE MINUTE OF AGITATION

Source of Variance	Degrees of Freedom	F Test for Significance
Detergent Concentration	3	8.45055 *
Water Temperature	2	155.73055 *
Fabric	1	0.29526
Det. Conc. x Water Temp.	6	1.67820
Det. Conc. x Fabric	3	1.37699
Water Temp. x Fabric	2	0.36541
Det. Conc. x Water Temp. x Fabric	6	1.20387
Error	48	
Total	71	

*Significant at 95% level.

TABLE X

ANALYSIS OF VARIANCE FOR COUNT IN WASH WATER TAKEN AT THE END OF WASH PERIOD

Source of Variance	Degrees of Freedom	F Test for Significance
Detergent Concentration	3	3.63860 *
Water Temperature	2	139.19664 *
Fabric	1	0.14392
Det. Conc. x Water Temp.	6	0.68225
Det. Conc. x Fabric	3	0.89674
Water Temp. x Fabric	2	0.03659
Det. Conc. x Water Temp. x Fabric	6	1.33107
Error	48	
Total	71	

*Significant at 95% level.

TABLE XI

ANALYSIS OF VARIANCE FOR COUNT IN WASH WATER TAKEN AT THE END OF RINSE PERIOD

Source of Variance	Degrees of Freedom	F Test for Significance
Detergent Concentration	3	5.10745 *
Water Temperature	2	22.11099 *
Fabric	1	2.26965
Det. Conc. x Water Temp.	6	1.66993
Det. Conc. x Fabric	3	0.35009
Water Temp. x Fabric	2	2.34341
Det. Conc. x Water Temp. x Fabric	6	2.02313
Error	48	
Total	71	

*Significant at 95% level.

TABLE XIV

ANALYSIS OF VARIANCE FOR REPRESENTATIVE COUNT REMAINING IN DRYER

Source of Variance	Degrees of Freedom	F Test for Significance
Detergent Concentration	3	2.12258
Water Temperature	2	1.07124
Fabric	1	0.55595
Det. Conc. x Water Temp.	6	0.75711
Det. Conc. x Fabric	3	0.39122
Water Temp. x Fabric	2	1.39061
Det. Conc. x Water Temp. x Fabric	6	0.69352
Error	48	
Total	71	

*Significant at 95% level.

APPENDIX C

MIL-S-12549E
3 December 1965
Pages 1 to 4.

MILITARY SPECIFICATION
SOCKS, MEN'S, NYLON AND COTTON
RIBBED, STRETCH TYPE

Class 1 - Black 94

This specification is mandatory for use by all Departments and Agencies of the Department of Defense.

3.2 Material

3.2.1 Knitting yarn.- The knitting yarn shall be made by plying or twisting one end of the cotton yarn specified in 3.2.1.1 with one end of the nylon stretch yarn specified in 3.2.1.2 using 2 to 4 turns of twists per inch when tested as specified in 4.3.1.1.

3.2.1.1 Cotton yarn.- The yarn shall be a singles 60 ± 2 count, carded and combed, mercerized cotton yarn. Testing shall be as specified in 4.3.1.1.

3.2.1.2 Nylon stretch yarn.- The yarn shall be a stretch type nylon yarn processed from two ends of 70-denier (± 5%) nylon. Testing shall be as specified in 4.3.1.1.

3.4 Color.- The color of the finished socks shall be as specified. The use of sulfur dyes and dyes containing elementary sulfur or compounds capable of oxidation to sulfuric acid is prohibited. The dyestuffs shall be chosen and applied so that the dyed socks shall show no more free or sulfide sulfur than the standard sample when tested as specified in 4.4.2.

3.4.2 Colorfastness.- The dyed socks shall show fastness to laundering and bleaching equal to or better than the standard sample. When no standard sample is available, the dyed sock shall show "good" fastness to laundering and bleaching when tested as specified in 4.3.3.

3.5 Design.- The socks shall be seamless, circular ribbed knit, stretch-type with a ribbed elastic top.

3.6 Construction

3.6.1 Knitting.- The socks shall be knit seamless in one integral unit on a 200 needle circular machine having a cylinder diameter of 3½ inches.

3.6.1.2 Sole, heel, toe, and ring toe.- The sole, heel, toe, and 1-inch ring toe shall be plain knit using one end of the knitting yarn specified in 3.2.1. A minimum of 26 gore needles shall be used in knitting the heel. A minimum of 26 gore needles shall be used in knitting the toe.

TABLE XII

ANALYSIS OF VARIANCE FOR pH

Source of Variance	Degrees of Freedom	F Test for Significance
Detergent Concentration	3	9.33503 *
Water Temperature	2	4.36238 *
Fabric	1	2.37744
Det. Conc. x Water Temp.	6	4.44371 *
Det. Conc. x Fabric	3	3.48192 *
Water Temp. x Fabric	2	4.17549 *
Det. Conc. x Water Temp. x Fabric	6	4.84761 *
Error	48	
Total	71	

*Significant at 95% level.

TABLE XIII

ANALYSIS OF VARIANCE FOR REPRESENTATIVE COUNT REMAINING IN WASHER

Source of Variance	Degrees of Freedom	F Test for Significance
Detergent Concentration	3	0.50742
Water Temperature	2	33.52454 *
Fabric	1	7.72775 *
Det. Conc. x Water Temp.	6	1.72364
Det. Conc. x Fabric	3	0.92131
Water Temp. x Fabric	2	1.96608
Det. Conc. x Water Temp. x Fabric	6	1.70366
Error	48	
Total	71	

*Significant at 95% level.

MIL-S-48G
6 December 1962
Pages 1 to 5

MILITARY SPECIFICATION
SOCKS, MEN'S WOOL, CUSHION SOLE, STRETCH TYPE

Class 1 - Black - 197

3.2 Materials.-

3.2.1 Yarn-

3.2.1.1 Stretch-type knitting yarn.- The yarn for knitting the top of foot and leg portion adjacent to the high heel, and for plating the high heel, heel, sole, toe, and ring toe shall consist of a single end of the merino yarn specified in 3.2.1.1.1, twisted or plied with the nylon stretch yarn specified in 3.2.1.1.2, using knitting twist. A stretch core yarn will not be acceptable.

3.2.1.1.1 Merino yarn.- The merino yarn shall be 1/30 (worsted count) yarn, made from fleece, pulled sheep's wool, or a combination of both not lower in grade than 56's, U.S. Standard, and cotton, blended in such proportion that the finished yarn contains not less than 50 percent wool on a dry weight basis when tested as specified in 4.3.2. Cotton core yarn will not be acceptable. The merino yarn shall be spun on either the cotton or worsted system.

3.2.1.1.2 Nylon stretch yarn.- The yarn shall be a 140 denier ±5%, 2 ply, nylon stretch yarn.

3.2.1.3 Terry stitch yarn.- The yarn for the terry stitch on the inside of the high heel, heel, sole, toe, and ring toe shall be made from wool not lower in grade than 50's, U.S. Standard. The yarn shall be spun on the worsted system. Not finer than 1/16s, 1/18s, and 1/20s or equivalent yarn count shall be used for 108 to 114, 116 to 122, and 124 to 136 needle machines respectively.

3.2.1.4 Looping yarn.- The yarn for looping the toe of the sock shall be as specified in 3.2.1.1.

3.3 Color.- The color of the finished socks shall be as specified. The use of sulfur dyes and dyes containing elementary sulfur or compounds capable of oxidation to sulfuric acid is prohibited. The dyestuffs shall be chosen and applied so that the dyed socks shall show no more free or sulfide sulfur than the standard samples when tested as specified in 4.3.3.

3.3.2 Colorfastness.- The dyed socks shall show fastness to perspiration, laundering and crocking equal to or better than the standard sample. In comparing the colorfastness of the standard sample with that of the material under test, specific care will be taken to insure that the same area of both the standard and the test material are taken for testing by any specific test method. When no standard sample is available, the dyed black-197 socks shall show "good" fastness to perspiration and "fair" fastness to laundering and crocking. Testing shall be as specified in 4.3.3.

3.5 Shrink resistant treatment.- All of the wool for the finished sock shall be treated for resistance to felting shrinkage in stock, top, yarn or sock form by a controlled oxidation process approved by the contracting agency. The shrink resistant treatment shall not be identified by name or trademark on the socks or on the package.

3.6 Design.- The socks shall be seamless, stretch-type, with a true rib-knit top and a plain knit leg and foot with a terry or tuft stitch on the inside of the high heel, heel, sole, toe and toe ring.

3.7 Construction.-

3.71 Knitting.- The socks shall be knit seamless on a circular machine of not less than 3½ nor more than 4 inches in cylinder diameter with not less than 108 nor more than 136 needles. A minimum of 15 gore needles shall be used in knitting the heel, and a minimum of 15 gore needles shall be used in knitting the toe. The socks shall be knit so that they will finish to the proper size and length without undue stretching during boarding.

3.7.1.2 High heel, remaining portion of leg and foot.- The high heel, the remaining portion of the leg adjacent to the high heel, and the foot, shall be plain knit with one end of the stretch-type knitting yarn specified in 3.2.1.1. The high heel, heel, sole, toe and ring toe whall be reinforced with a terry stitch thrown to the inside, made with the wool terry yarn specified in 3.2.1.3, and every knitting course of these areas. The terry yarn, for the high heel and sole, shall be laid in at a point not less than 3 needles after the last short butt needle in the heel gore. The knitting on all of these needles shall be terried. The terry stitch may be omitted from not more than two courses before the looping course, provided the terry yarn is knit with the stretch-type knitting yarn into the knitting of the looper rounds. The two yarns shall be knit together for at least two courses beyond the looping or loose course.

APPENDIX D

ARTIFICIAL SOIL USED IN RESEARCH*

Gold Metal Flour	15 g.
Argo Corn Starch	15 g.
Domino Cane Sugar, granulated	15 g.
Powdered Carbon	1 g.
Wesson Oil	15 ml.
Mineral Oil	15 ml.
Carnation Evaporated Milk	100 ml.
Water	250 ml.

All ingredients were mixed in a Waring Blender for five minutes to form a relatively stable emulsion. A mold inhibitor, Anti-dine, was added to the soil in a ratio of 1:10,000. The resultant pH of the soil was 6.2. Fifteen to twenty cc. of the soil were used per 8" x 12" swatch of fabric.

*Ridenour, p. 95 (Herein Designated Soil #1).

THE SURVIVAL OF STAPHYLOCOCCUS AUREUS ON MILITARY SOCK FABRIC LAUNDERED AT VARIOUS WATER TEMPERATURES AND DETERGENT CONCENTRATIONS

by

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The prevention of disease and the transmission of pathogenic microorganisms through the laundry has received increased concern. Obtaining sufficiently high water temperatures to destroy bacteria during washing may be difficult in the home. Neither soaps nor synthetic detergents alone remove bacteria. The objectives of the study were to determine the effect of water temperature and detergent concentration upon the survival of Staphylococcus aureus in a home laundry situation and to determine the transference of the organism during laundering.

Two knitted fabrics meeting military specifications for U. S. Air Force socks were soiled and inoculated with Staphylococcus aureus, washed at three water temperatures (140° F., 100° F., and 60° F.) with detergent concentrations of 0%, .1%, .2%, and .4% by weight in an automatic washer, and tumble dryed. The survival after washing and drying, the redeposition count, bacterial count in the wash and rinse waters, pH of the wash water, and a representative count remaining in the washer and dryer were analyzed statistically.

Water temperature had the greatest effect on survival but increasing detergent concentrations increased staphylococcial removal. Test organisms remained on the fabric and were transferred with 140° F. wash water at all detergent concentrations. Bacterial removal from the nylon and cotton fabric was higher than from the wool blend fabric. Results indicated drying also decreased the survival. Bacterial redeposition during washing showed similar trends to bacterial survival on inoculated swatches at the end of the wash. Test organisms remained in the washer and dryer after fabric removal. Since only 100% removal of the test organism was satisfactory, none of the combinations of water temperatures and detergent concentrations were found adequate in providing a sanitary wash.