ORIGINAL ARTICLE

Effectiveness of Low-Temperature Domestic Laundry on the Decontamination of Healthcare Workers' Uniforms

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OBJECTIVE. Most professionals in the healthcare environment wear uniforms. For the purpose of this study, we concentrated on nurses' uniforms. In the United Kingdom, many nurses are expected to launder their uniforms at home by using a domestic washing machine that frequently has low-temperature wash cycles. We have investigated whether the use of low-temperature wash cycles results in a microbiologically acceptable product to wear on the wards.

METHODS. We have assessed the bioburden on uniforms before and after laundry and the effectiveness of low-temperature wash cycles and ironing on removal of methicillin-resistant *Staphylococcus aureus* (MRSA) and *Acinetobacter baumannii*. We did not assess the role of tumble drying.

RESULTS. We demonstrate contamination of uniforms by gram-negative bacteria after wash, the removal of MRSA at low-temperature wash cycles in the presence of detergent, and the eradication of gram-negative bacteria after ironing.

CONCLUSIONS. Our conclusions are that laundry in a domestic situation at 60° C (140° F) for 10 minutes is sufficient to decontaminate hospital uniforms and reduces the bacterial load by more than 7-log reduction, that items left in the pockets are decontaminated to the same extent, that the addition of either a biological detergent or a nonbiological detergent is beneficial in removing MRSA from experimentally contaminated swatches, and that uniforms become recontaminated with low numbers of principally gram-negative bacteria after laundry but that these are effectively removed by ironing.

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Hospital-acquired infection (HAI) is currently an important topic both for government and the public. Multidrug-resistant organisms are of particular importance, and huge efforts are made to prevent their spread, for example, patient isolation and contact precautions. Organisms often involved in relation to HAI are methicillin-resistant *Staphylococcus aureus* (MRSA), *Acinetobacter* spp., and various coliforms.

There have been a number of publications identifying articles of clothing as potential sources of infection for patients.¹⁻⁴ By the nature of their job, healthcare workers are exposed to a wide range of microorganisms, many of which have a role to play in HAI. These microorganisms are often shed from sick patients or are frequently present in the environment and may contaminate the nurse's uniform. These uniforms may subsequently act as sources for the transmission of nosocomial pathogens to patients. A UK study of the contamination of nurses' uniforms resulted in advocating the wearing of a plastic apron to minimize the acquisition of nosocomial pathogens during close-contact nursing duties.⁵ There is some debate as to whether nurses' uniforms are a vehicle for transferring microbes from 1 patient to another,⁶ and a recent review that looked at the significance of uniforms and HAI showed that there is evidence that nurses' uniforms become contaminated with microorganisms but little evidence that nurses' uniforms are responsible for HAI.⁷

However, after changes in organization of the National Health Service in the United Kingdom, many hospitals no longer provide an in-house laundry service, and, as part of this reorganization, many nurses now launder their uniforms at home or in public laundromats.

Because clothing could be a means of transfer of organisms, we need to know how nurses' uniforms can be effectively cleaned and whether laundering at home or in a public laundry provides a uniform with a low bioburden. Commercial laundries utilize a continuous-batch (or tunnel) washer and operate according to Health Service Guideline (95)18,⁸ decontaminating the load at 65°C (149°F) for a minimum of 10 minutes or, more usually, 71°C (159°F) for a minimum of 3 minutes. Outside the United Kingdom, however, different temperatures are allowable, depending on the country. Extra mixing time of either 4 or 8 minutes is added, depending on the degree of loading.

In the United Kingdom, domestic washing machines usually operate at lower temperatures (40°C [104°F] or 60°C

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[140°F]) or have a high-temperature wash at 90°C (194°F). The current trend in washing machine manufacture in the United Kingdom is the use of lower temperatures and less water in order to save energy as part of an increased awareness of environmental issues. Current A-rated washing machines use 0.56 kWh/cycle at 40°C (104°F), 0.94 kWh/cycle at 60°C (140°F), and 1.22 kWh/cycle at 90°C (194°F). As part of the Energy-Using Products Directive (2009/125/EC) the benchmark is, for example, 0.85 kWh/cycle at 60°C (140°F) for a 5-kg rated-capacity washing machine (EU 1015/2010). There is thus a strong pressure to use lower-temperature/lowerwater-usage washing machines, with a machine lifetime expected savings of £65 (\$106) and a reduction of 400 kg of CO₂ into the atmosphere.^{9,10} In the United States, washing machines are covered by a single federal standard that does not limit water consumption, although the standard was due for review in January 2011. A more stringent standard issued by the US Environmental Protection Agency called "Energy Star" was reviewed in January 2011, and Energy Star washing machines use 37% less energy and 50% less water than noncertified machines. Many washing machines in the United States already exceed these standards in relation to reduction in water consumption and energy efficiency. Overall, the Energy Star program has saved \$18 million on utility bills in 2010.

All these changing factors of lower temperature and lower water consumption may influence the risk of nurses' uniforms being inadequately laundered under the home circumstances and thus acting as a potential vehicle for infection to patients.

There is no stipulation on how these uniforms should be laundered within a domestic environment. How these uniforms will be cleaned depends on the facilities available to the individual and his or her own methods. There are no means to check whether this clothing has been effectively decontaminated. Therefore, because nurses often take their laundry home to wash it in a domestic washing machine or in a public laundry, we investigated the bioburden on the uniform immediately after wearing it and compared it to the bioburden after laundry. Additionally, we determined the removal of multidrug-resistant bacteria from artificially contaminated fabric by washing in a front-loading washing machine at different time and temperature combinations to simulate the domestic environment. We also investigated the effects of ironing on the fabric bioburden.

METHODS

We undertook 4 related studies. First, we assessed the bacterial load on nurses' uniforms immediately after wearing them on a shift and the effect of home laundering on the bioburden (study 1). Second, we assessed the effect of varying the times and temperatures during the laundry process on decontamination of artificially contaminated swatches of a polyester/ cotton mix with multidrug-resistant bacteria known to cause HAI (study 2). The third study was an assessment of effectiveness of the washing process at decontaminating items left in the pockets (study 3). Because the most frequent type of organism detected after laundering was gram-negative bacilli, we elected to assess the effect of ironing on the survival of *Acinetobacter* (study 4).

Study 1: Method

Nurses' uniforms (either scrubs or tunics, depending on the location; both top and trousers were analyzed together) from 5 areas (intensive care unit, accident and emergency unit, infectious diseases unit, hematology ward, and gynecology ward) of the hospital were investigated. Two nurses participated from each area. Each uniform was worn for a shift and given to the microbiology department in a sterile container. The total viable count of organisms on the uniform was determined by soaking the uniform, with agitation, in 1 L of sterile phosphate-buffered saline (PBS) in a sterile container.

Decimal dilutions of the wash were plated on 5% Columbia blood agar, McConkey medium, and chromogenic MRSA plates (Oxoid), and after incubation at 37°C (98°F) for 48 hours, the colonies were counted.

On a separate occasion, the individuals wore another uniform for a shift and laundered the uniform themselves. The uniform was laundered either in the nurses' homes in Maytag machines at 40°C (104°F) or in public self-service laundries in Electrolux machines at 40°C (104°F). The nurses brought the washed items to the microbiology department in a sterile plastic container, and the microbial load was determined as above. Each nurse repeated the exercise twice.

Study 2: Method

The organisms chosen for this study were MRSA and *Acinetobacter baumannii* (AB). A front-loading Electrolux FOM 71CLS scientific washing machine with a Claris control system allowing independent programmable cycles and providing a printout of the parameters for each cycle was used in the study. All washes were undertaken with colored sterile 30-cm (1-ft) squares of polyester/cotton (ballast) at a total weight of 5 kg.

Swatches (white) of a typical hospital-quality uniform fabric (supplied by Carrington Career and Work Wear) made from a 67% polyester/33% cotton blend with a weight of 195 g M^{-2} were soaked with a 10-mL concentrate of microorganisms (approximately 2 × 10⁸/mL) prepared in a sterile 3.5% bovine serum albumin (BSA) solution. BSA was used to represent body fluids and also to aid in the adherence of microbes to the fabric.

Three 30-cm² test swatches were prepared per wash and air-dried. These swatches were then attached to a clean load of fabric and processed in the washing machine. In addition, 2 clean swatches were also placed in the load to determine whether there was any cross-contamination within the washing machine.

Location Unif		Before wash, mean CFU/L (range)	Organism	After wash, mean CFU/L (range)	Organism	
Intensive care unit	Scrubs	2.0×10^{5} (1.4 × 10 ⁴ -5.6 × 10 ⁵)	Staphylococci, micrococci, streptococci	4.6×10^4 (0-6.0 × 10 ⁶)	GNR	
Accident and emergency unit	Tunics	$\begin{array}{rrrr} 1.1 \ \times \ 10^7 \\ (6.0 \ \times \ 10^4 - 5.2 \ \times \ 10^7) \end{array}$	Staphylococci, micrococci, streptococci	5.1×10^{2} $(1.3 \times 10^{1} - 1.0 \times 10^{3})$	GNR	
Infectious diseases unit	Tunics	$5.5 \times 10^{6} (8.4 \times 10^{4} - 1.7 \times 10^{7})$	Staphylococci, micrococci, streptococci	1.0×10^2 (0-4.0 × 10 ³)	GNR Staphylococcus	
Hematology ward	Tunics	1.5×10^{5} (1.7 × 10 ⁶ -2.0 × 10 ⁴)	Staphylococci, micrococci, streptococci	1.5×10^2 (0-5.3 × 10 ³)	GNR Bacillus	
Gynecology ward	Tunics	1.3×10^{7} (1.7 × 10 ⁴ -4.4 × 10 ⁷)	Staphylococci, micrococci, streptococci GNR	1.1×10^2 (0-3.0 × 10 ²)	GNR Bacillus	

TABLE 1. Contamination of Nurses' Uniforms before and after Wash

NOTE. Nurses' uniforms assessed after work and after laundry by serial dilution from a total volume of 1 L after immersion of the whole garment. CFU, colony-forming unit; GNR, gram-negative rod.

We simulated typical laundry conditions as follows. A temperature range was chosen on the basis of the washing temperatures most popularly selected by domestic users. The machine was standardized to deliver the same volume of water per run and to deliver 3 rinses at ambient temperature (~20°C [68°F]). This program was used for all the studies.

Two of the variables were the washing temperature and wash cycle time. The temperatures and times selected were as follows: 30° C (86° F), 40° C (104° F), and 60° C (140° F) for 10 and 20 minutes per main wash cycle and 90° C (194° F) for 3 minutes.

A third variable we assessed was the presence of a detergent. We compared 2 commercially available detergents: a biological detergent (containing enzymes such as lipases and proteases) and a nonbiological detergent. These were designated D1 and D2, respectively.

First, we assessed the mechanical action of the wash cycle without detergent but at different temperatures. The washes were then repeated using each of the detergents separately at the same temperature/time relationship.

After the wash was completed, the swatches were detached and processed for 15 minutes in a stomacher containing 20 mL of PBS, and the total viable count was performed by serial dilution and plated onto 5% horse blood agar and Oxoid's chromogenic MRSA medium. Plates were incubated at 37°C for 48–72 hours and colonies counted. This count was then compared to the controls, which were identically contaminated swatches that had not been laundered but otherwise processed in the same way.

Study 3

Three swatches per organism (MRSA and AB) were attached to the inside of a pocket (1 swatch per pocket) and 3 swatches

per organism were attached to the ballast (1 swatch per ballast) and placed in the washing machine. Two clean swatches were also placed in the washing machine to check for carryover/cross-contamination effects. The washing machine was run at 2 different time/temperature cycles: 40° C (104° F) for 20 minutes and 60° C (140° F) for 10 minutes with D1 and D2 separately. After the wash, the swatches were removed and placed individually into separate stomacher bags with 20 mL of PBS and stomached for 15 minutes. Total viable counts were performed by decimal dilution, and the plates were incubated at 37° C (98° F) for 48 hours.

Study 4

Strips of polycotton fabric (5 cm × 15 cm [2 in × 6 in]) were seeded with a 2-mL suspension of AB (inoculum, 7.5 × 10^3 – $1.3 × 10^4$ mean colony-forming units [CFU]) in PBS with 3.5% BSA and allowed to air-dry for 4 hours in a sterile container. After drying, they were ironed with a Tefal UltraSmooth Glide domestic nonsteam iron set at the maximum temperature (150°C [302°F]). The iron was held on the fabric for set times ranging from 5 to 11 seconds and covered the whole fabric strip. After ironing, the strips were placed into nutrient broth and incubated at 37°C (98°F) for 24 hours and then subcultured onto solid agar to check for the presence of growth. Three replicates were performed at each time point.

Calculation of Log Reduction

The viable counts for the inoculum and postlaundry data were converted to logarithms $(LD^{I} \text{ and } LD^{L}, \text{ respectively})$, and the mean (MLD) and standard deviation (SLD) were calculated. The log reduction (LR) was calculated as

	Log reduction in colony counts							
	Water	r only	Bio	logical D1	Nonbiological D2			
Temperature, °C (°F)/ time, minutes	MRSA	AB	MRSA	AB	MRSA	AB		
30 (86)/10	2.68 ± 0.35	2.37 ± 0.18	>7	2.39 ± 0.94	>7	2.11 ± 0.36		
30 (86)/20	$3.60~\pm~0.28$	$2.66~\pm~0.20$	>7	$3.66~\pm~0.90$	>7	2.26 ± 0.39		
40 (104)/10	$3.10~\pm~0.28$	$2.62~\pm~0.18$	>7	$2.61~\pm~0.47$	>7	2.70 ± 0.35		
40 (104)/20	$4.92~\pm~0.31$	$3.39~\pm~0.17$	>7	$2.48~\pm~0.60$	>7	$2.74~\pm~0.35$		
60 (140)/10	>7	>7	>7	>7	>7	>7		
90 (194)/3	>7	>7	>7	>7	>7	>7		

TABLE 2. Effect of Temperature and Exposure Time on the Eradication of the Different Test Organisms

NOTE. Artificially contaminated uniform fabric with methicillin-resistant *Staphylococcus aureus* (MRSA) or *Acinetobacter baumannii* (AB) and laundered at differing time/temperature relations, with or without a detergent, in a scientific front-loading washing machine. AB mean final inocula (water only, 3.5×10^7 ; range, 7.0×10^6 – 5.7×10^7 ; biological detergent [D1], 7.2×10^6 ; range, 2.7×10^7 – 1.8×10^6 ; non-biological detergent [D2], 1.3×10^7 ; range, 1.0×10^7 – 7.9×10^6). MRSA mean final inocula (water only, 3.8×10^7 ; range, 7.2×10^7 – 4.2×10^7 ; D1, 2.8×10^7 ; range, 1.1×10^7 – 7.4×10^7 ; D2, 3.7×10^7 ; range, 2.3×10^6 – 1.2×10^8). >7, no growth (reduction equal to or greater than the inoculum).

$$LR = MLD^{I} - MLD^{L}$$
,

and the standard deviation of LR (SLR) was calculated as

SLR =
$$\left[\frac{(\text{SDL}^{1})^{2}}{n^{T}} + \frac{(\text{SDL}^{L})^{2}}{n^{L}}\right]^{1/2}$$
,

where n^{I} is the number of data points for the inoculum and n^{L} is the number of data points for the laundered fabrics.

RESULTS

Study 1

The results from our first study indicated that nurses' uniforms are, unsurprisingly, contaminated with high numbers (10^4-10^7 CFU) of skin flora contaminating the whole uniform (coagulase-negative *Staphylococcus* spp., *Micrococcus* spp.) after a shift. After being washed at low temperatures, although the bioburden is reduced and the skin flora mainly eliminated, the uniforms become contaminated with gram-negative bacteria of the *Klebsiella/Enterobacter/Serratia* group of Enterobacteriaceae or *Bacillus* spp. (Table 1).

Study 2

The results of our second study indicated that the mechanical action of washing alone, without detergent, reduced the test organisms by a factor of 10^2 – 10^3 . At 40° C (104° F), the organisms were generally reduced by a factor of 10^3 – 10^4 ; at 60° C (140° F) and 90° C (194° F), the organisms were reduced by a factor of $10^{>7}$, and no organisms were recovered (Table 2).

We noticed that in the presence of either a biological detergent or a nonbiological detergent, even at a low-temperature wash (30°C [86°F] for 10 minutes), MRSA was eliminated from the material, but the gram-negative bacteria (*Acinetobacter*) were more difficult to remove (Table 2).

Study 3

From this experiment, it can be seen that at 40°C (104°F) for 20 minutes, the removal of *Acinetobacter* from the ballasts as compared with the pockets is comparable. With MRSA, however, there is an approximate 10-fold better removal of organisms from the ballasts as compared with the pockets. Addition of detergent is efficient at completely removing MRSA from the ballast and the pocket but has no effect on *Acinetobacter*. A temperature of 60°C (140°F) for 10 minutes is effective at obtaining decontamination of laundry for both MRSA and *Acinetobacter* from the pockets and ballasts (Table 3).

Study 4

When we used AB at concentrations typically found with gram-negative bacteria recontaminating items after laundry, the results (Table 4) show that a 7-second exposure to a hot domestic iron renders the item free of the organism. At 5 and 6 seconds of exposure, the organism was cultured from the broth.

DISCUSSION

This study was undertaken to assess (*a*) the contamination of nurses' uniforms after a shift and the efficacy of laundering on reducing the bioburden on the uniforms; (*b*) the effect of low-temperature washes on artificially contaminated swatches made from a polyester/cotton mix with common multidrugresistant bacteria that cause HAI; (*c*) whether laundry conditions can decontaminate items left in pockets; and (*d*) the effect of ironing on eradication of gram-negative bacteria from hospital fabric. A previous study investigated the effectiveness of low-temperature laundry on artificially contaminated fabric with an inoculum of 10^8-10^{10} CFU/mL of *S. aureus.*¹¹ The sensitivity profile of the organism was not noted. The 2 temperatures used were 40° C (104° F) and 60° C (140° F);

	Log reduction in viable count									
		Water					Detergent			
	MI	RSA	А	B MRSA			AB			
Temperature, °C (°F)/ time, minutes	Т	Р	Т	Р	Т	Р	Т	Р		
40 (104)/20 60 (140)/10	3.03 ± 0.48 >7	2.07 ± 0.25 >7	2.37 ± 0.26 >7	$2.19 \pm 0.25 > 7$		>7 >7	2.74 ± 0.13 >7	2.02 ± 0.21 >7		

TABLE 3. Effect of Laundry Condition on Items in Uniform Pockets

NOTE. Artificially contaminated fabric with methicillin-resistant *Staphylococcus aureus* (MRSA) or *Acinetobacter baumannii* (AB) placed in the pocket of a nurse's uniform and laundered at differing time/temperature relations, with or without a detergent, in a scientific front-loading washing machine. Inoculum AB, 2.8×10^7 ; inoculum MRSA, 1.9×10^7 . T, swatch free in laundry tub; P, swatch in uniform pocket. >7, no growth (reduction equal to or greater than the inoculum).

a nonbiological detergent was added and the experiment carried out once only. The results indicated that at both 40°C (104°F) and 60°C (140°F) the inoculated organism was not isolated immediately after laundry but there was contamination by environmental gram-negative bacteria (derived from the washing machine), inactivated by tumble drying/ ironing.

In this study that extends that of Patel et al,¹¹ naturally contaminated uniforms from different ward locations were initially assessed for bacterial bioburden (study 1), and swatches prepared from the same material used to make nurses' uniforms were artificially contaminated by 2 multidrug-resistant organisms, MRSA and AB (study 2). The swatches were laundered at 4 temperatures, 30°C (86°F), 40°C (104°F), 60°C (140°F), and 90°C (194°F), using either a biological detergent or a nonbiological detergent in separate experiments. Each experiment was repeated 3 times. Additionally, we assessed the efficacy of the laundry process on fabric items left in the pockets of the uniforms (study 3). Our inoculum on the artificially contaminated swatches reflected that found on naturally contaminated clothing after a shift and was lower than that used by Patel et al^{11} (10⁶-10⁷) compared with $10^8 - 10^{12}$).

Although clothing was contaminated with high levels of mainly gram-positive skin flora after working a shift, after laundry at 40°C (104°F) in a domestic washing machine, the predominant organisms were gram-negative bacteria at much lower levels and presumably derived from the washing machine. It is well recognized that various gram-negative bacteria contaminate automated endoscope washer/disinfectors and that they are found in the biofilm present in the machine. It is not unreasonable to assume that the same will occur with a domestic washing machine. Occasional coagulase-negative staphylococci were noted after laundry, probably representing contamination during the retrieval of the uniform after wash. There was no significant difference between the bioburden before laundry and the location of working.

In the case of the swatches artificially contaminated with MRSA and AB, the laundry process eliminated MRSA even at the lowest temperature (30°C [86°F]), as long as a detergent was added, compared with a wash without detergent. Despite

the addition of a detergent, the gram-negative organism was not removed. There was no difference between a biological detergent and a nonbiological detergent in effect on both bacteria. All bacteria were eliminated even in the absence of the detergent by 60°C (140°F) for 10 minutes. This is consistent with Health Service Guideline (95)18,⁸ which advocates 65°C (149°F) for 10 minutes. The results also demonstrate that items left in the pockets of the uniforms are effectively decontaminated and that postlaundry ironing kills AB.

The shortcomings of this study are the relatively limited number of trials of each experiment and the restricted species of bacteria. However, it would be impractical to assess the effect of differing laundry conditions on all extant bacteria, and indeed it would also be impractical to increase the number of repetitions because any choice of number would be arbitrary. In any case, process control is regarded as more effective than product control. It should be emphasized that most of this work was performed on a scientific washing machine with accurate and adjustable control of cycle parameters, whereas home laundry of uniforms would be undertaken on many different brands and models of washing machines, with variable accuracy of cycle parameters. In order to accurately quantify the effects on specific organisms, most of this work utilized artificially contaminated fabric, which may not represent naturally contaminated materials, and the relationship between the reduction in numbers of bacteria under these rather artificial conditions may not accurately represent that seen in the home environment.

 TABLE 4.
 Effect of Ironing on the Survival of Acinetobacter baumannii–Contaminated Fabric

		Time, seconds						
	5	6	7	8	9	10	11	
Acinetobacter	+	+	—	_	_	—	_	

NOTE. Artificially contaminated fabric with *A. baumannii* at an inoculum of 10^3 – 10^4 and subsequently exposed to a hot iron for varying times. A plus sign indicates growth in nutrient broth after overnight incubation at 37°C (98°F); a minus sign indicates no growth.

These results raise a number of points worthy of further study: first, to determine whether any hospital-acquired contaminants became established in the biofilm of washing machines; second, to assess the effect of a wider range of detergents on decontamination of laundry, particularly on gram-negative bacteria; third, to assess other hospital fabrics; and, finally, to assess a wider range of bacteria species, including spores. We are currently undertaking some of these studies.

The results of this study suggest that a detergent should be included when laundering nurses' uniforms, and, also, as lower temperatures and lower water use is likely to increase, particular attention should be paid to the organisms colonizing washing machines after laundering hospital uniforms.

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