

Microbiological Contamination of Bed Linen and Staff Uniforms in a Hospital

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Received September 4, 2013; revised October 20, 2013; accepted November 7, 2013

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ABSTRACT

Hospital linen is clearly recognized as a potential reservoir for microorganisms and could be a vector of disease transmission. The aim of this study was to isolate, count and identify fungi and bacteria from different kinds of clean and dirty linen in a hospital. Microbiological samples have been collected on clean bed linen (n = 200), dirty bed linen (n = 192) and staff uniforms (n = 192) by using contact plates. 55% of samples from clean bed linen were contaminated before contact with the patient, with a mean count of 3 cfu/25 cm² (range: 1 - 117 cfu) when contaminated. Virtually all samples from dirty bed linen carried microorganisms, with a mean count of 23 cfu/25 cm² (range 1 - 191 cfu). In addition, staff hospital uniforms showed the highest contamination rates in the study, with an average of 45 cfu/25 cm² (range: 1 - 218 cfu). Microbial species were mostly bacteria commonly found in the environment or on human skin, such as staphylococci or micrococci. Nevertheless, 57% of the identified species may be opportunistic pathogens for humans, representing a risk for people with a deficient or weakened immune system, especially in cases of superinfection. Since contamination of linen seems to occur after washing, actively antimicrobial textiles would represent a valuable measure to prevent textiles from being a vehicle for transfer of microorganisms.

Keywords: Linen; Bed Sheets; White Coat; Contamination; Hospital Acquired Infections

1. Introduction

Healthcare-associated infections (HAIs) represent a heavy social and financial burden. In the USA, it was estimated that 1.7 million cases of HAIs occurred yearly, with almost 99,000 deaths related to these cases [1]. European figures give an average prevalence of 7.1%, corresponding to 4.5 million nosocomial infections each year [2].

Direct contamination is not the sole way of acquiring HAIs; contamination through environment of the patient has been identified as a possible route. Indeed, items in contact with the patient or in its vicinity may be frequently contaminated. In two separate studies, 86% and 87% of stethoscopes were found to be contaminated with bacteria, including staphylococci [3,4]; potentially pathogenic bacteria were found on 81% of medical charts [5]. Textiles may act as reservoirs of microorganisms, since pathogens may be able to survive on such surfaces for periods ranging from a few minutes to several months [6, 7].

Various species of microorganisms, including some relevant for HAIs, have been isolated from hospital textiles. A study on white coats [8] reported 95% contamination, while others found presence of *Staphylococcus aureus* in 23% of the white coats [9]; these bacteria appear to be found more specifically around sleeves and pockets [10]. Pathogens were also found on other medical staff uniforms [11], or on bed linen in patients' rooms [12]. Textiles may thus be responsible for transmission of pathogenic microorganisms to patients, as a few studies described transmission of microorganisms from uniforms to patient and bed linen [13], or from dirty bed linen to staff uniforms [14].

The aim of the present study was to evaluate the level of microbial contamination on hospital textiles, based on samples from clean and dirty bed linen, and from staff uniforms. The analysis was both quantitative and qualitative, including identification of microbial species.

2. Materials and Methods

2.1. Interviews

Methods for handling clean and dirty linen in the hospital have been recorded. The industrial launderer responsible

for washing textiles was also interviewed.

2.2. Sampling

Two sampling campaigns were carried out in a French hospital on November and December 2011. Samples were collected exclusively by trained staff from the microbiology laboratory. Textiles were sampled according to the following plan:

- 20 non-occupied rooms were selected in various hospital departments. In each room, 10 samples were collected from the bed, which had recently been made by the staff: fitted sheet (two sampling points at the centre), draw sheet (two sampling points at the centre), upper sheet (inside and outside, at the centre), duvet (near the head), pillow (at the centre), pillowcase (centre and opening). 200 samples (referred to as "clean bed linen") were thus obtained.
- Bed sheets that had been removed after use were sampled in laundry rooms, which are actually storage rooms for dirty linen before packing and sending to the launderer. According to available pieces, fitted sheets, draw sheets, or upper sheets were sampled (respectively 25, 51, and 20 sheets), with two sample points at the centre per item, amounting to 192 samples (referred to as "dirty bed linen").
- Staff uniforms were sampled either in the room where dirty uniforms are returned before laundering, or directly on the person (for the second sampling campaign only). For each uniform, two sampling points were selected, one on the chest and one on the pocket (right by default, possibly left if the holder was present and indicated left-handedness). Job title and department of the staff member having worn the uniform were recorded. 96 uniforms were sampled, with two sampling points for each piece, making a total of 192 samples, regardless of the uniform's nature (white coat, scrub, or other).

Bed sheets were 50% polyester/50% cotton, while uniforms were 65% polyester/35% cotton.

2.3. Microbiological Analysis

The samples were taken by placing agar contact Hygicount 4N (AES Chemunex, Bruz, France) on each sampling point (during 15 seconds). The surface of the plates is 25 cm². Plates were transported in ice to the laboratory. After incubation at 32.5 +/-2.5°C for 48 to 72 h, colonies were differentiated according to their macroscopic appearance: colour, opacity, bulging, gloss, and contour. The number of colonies of each type was counted. A copy of each colony type was then isolated on PCA (Plate Count Agar, AES Chemunex) for bacteria, or OGA (Oxytetracycline Glucose Agar, Biokar Diagnostics, Beauvais, France) for moulds. Bacteria isolated on PCA were iden-

tified to the species level by API strips (bioMérieux, Craponne, France). Moulds were analyzed by scotch-test to determine their genus.

2.4. Statistical Analysis

Statistical computations and tests were done using R statistical software [15]. Statistical differences evaluated by Wilcoxon's test were considered as significant when P-value < 0.05.

3. Results

3.1. Laundry

In the hospital, dirty bed sheets are collected by the staff in dedicated bags. These bags are stored in each aisle (corresponding to one half of a floor) before being gathered in a basement room where the launderer picks them up for washing.

Uniforms are labeled with an electronic chip: each staff member may collect up to 3 clean, personal uniforms per week. An automatic dispenser stores clean uniforms until claimed by the staff members. Dirty uniforms are returned when the person decides to, in a unique return point. They are then collected by the launderer for washing.

Washing is performed in industrial tunnel washers. Procedures include application of chemicals (bleach, antimicrobial products) and high temperatures (up to 80°C for washing, smoothing out by vapor). Clean items are packed in plastic bags. Periodic microbiological sampling of textiles leaving the laundry is performed.

In the hospital, clean bed sheets are stored in their plastic bags in a dedicated, clean room in each aisle. Beds are made by the hospital staff.

3.2. Clean Bed Linen

A summary of quantitative results on clean bed linen is presented in **Table 1**. Pillows, pillowcases, and duvets were the most frequently contaminated items. It was not possible to determine whether the department (floor/aisle) of the room had an influence on microbiological contamination, because of the incomplete and unbalanced repartition of sampled rooms.

According to species identified from samples, a summary showing most represented microbial genera is presented in **Table 2**. Almost one half of the samples contained Gram-positive cocci. Although most species correspond to common environmental or skin microflora, 53% of bacterial isolates (present in 38% of textile samples) may be opportunistic pathogens for immunocompromised patients. Examples of such identified species include various *Staphylococcus* species (91 isolates in 67 samples,

Table 1. Contamination levels on clean bed linen, sorted by decreasing order of contamination frequency; Q1, Q3: lower and upper quartiles.

Sampling	Contamination	Plate count, cfu/25 cm ^{2b}			Signif.c
point	frequency	Mean	Median [Q1, Q3]	Max	oigiiii.
Pillow	18/20	5	6 [3, 9]	30	AB
Pillowcase opening	16/20	3	3 [2, 6]	14	ACD
Pillowcase centre	15/20	6	6 [4, 12]	54	В
Duvet	15/20	5	4 [2, 10]	117	BC
Inside upper sheet	11/20	3	2 [1, 6]	24	BDE
Fitted sheet	19/40	2	1[1, 3]	12	E
Outside upper sheet	6/20	2	2[1, 3]	3	CE
Draw sheet	9/40	1	1 [1, 2]	3	E
All	109/200	3	3 [1, 7]	117	

^anumber of samples with at least one cfu/total number of samples; ^bfor samples with non-empty plates; ^crows that do not share an uppercase letter are statistically different (P-value < 0.05).

Table 2. Microbial genera found on clean bed linen.

Genus	Number (%) of samples		
Staphylococcus	69 (35%)		
Micrococcus	62 (31%)		
Bacillus	14 (7%)		
Pseudomonas	13 (7%)		
Aspergillus	13 (7%)		
Other Gram-negative bacilli	22 (11%)		
Other Gram-positive bacilli	2 (1%)		
Other Gram-positive cocci	4 (2%)		
Other moulds	11 (6%)		
Not identified	15 (8%)		

with two samples containing *S. aureus*), various *Pseudo-monas* species (14 isolates in 13 samples), *Bacillus cereus* (two samples), *Stenotrophomonas maltophilia* (one sample), or *Gardnerella vaginalis* (one sample).

3.3. Bed Sheets and Uniforms after Use

Contamination of dirty bed linen and uniforms was significantly higher than that of clean bed linen (P-value < 0.05). Results on these items are summarized in **Table 3**. On average, contamination of uniforms was twice as high as contamination of dirty bed linen, although the latter were sometimes visibly soiled by organic fluids. When

available, the time elapsed before the uniform was returned to the laundry (not necessarily connected to the time it was worn, since staff members may keep clean uniforms in their lockers) was recorded; no correlation between this time and the level of contamination was found (data not shown).

Microbial genera identified on these textiles are presented on **Table 4**, as a summary from identified species. Staphylococci and micrococci were again the most widely found genera. Opportunistic pathogens represented approximately 58% of bacterial isolates (found in 95% of the textile samples), with various *Staphylococcus* species (557 isolates in 356 samples, including six samples with

Table 3. Contamination levels on bed linen and uniforms after use, sorted by decreasing order of contamination frequency; Q1, Q3: lower and upper quartiles.

C1iit	Contamination frequency ^a	Plate count, cfu/25 cm ^{2b}			C::CC
Sampling point		Mean	Median [Q1, Q3]	Max	-Signif. ^c
Uniform pocket	96/96	49	55 [32, 87]	218	A
Uniform chest	96/96	42	39 [27, 82]	153	A
Upper sheet	40/40	24	23 [11, 61]	191	В
Draw sheet	102/102	23	35 [10, 69]	152	В
Fitted sheet	49/50	22	22 [13, 31]	132	В
All	383/384	32	37 [18, 73]	218	

^anumber of samples with at least one cfu/total number of samples; ^bfor samples with non-empty plates; ^crows that do not share an uppercase letter are statistically different (P-value < 0.05).

Table 4. Microbial genera found on bed linen and uniforms after use.

Genus	Number (%) of samples	
Staphylococcus	357 (93%)	
Micrococcus	340 (89%)	
Pseudomonas	90 (23%)	
Bacillus	66 (17%)	
Kocuria	22 (6%)	
Pasteurella	20 (5%)	
Ralstonia	20 (5%)	
Other gram-negative bacilli	68 (18%)	
Other gram-positive bacilli	35 (9%)	
Other gram-negative cocci	11 (3%)	
Other gram-positive cocci	11 (3%)	
Moulds or yeasts	8 (2%)	
Not identified	25 (7%)	

S. aureus), Pseudomonas species (99 isolates in 90 samples), Bacillus species (34 isolates in 33 samples), Burkholderia cepacia (eight samples), Stenotrophomonas maltophilia (two samples), Aerococcus urinae (two samples), or Gardnerella vaginalis (one sample).

4. Discussion

The contamination level of dirty bed linen and uniforms evidenced in the present study is not uncommon. In a study, the authors found a microbial load on white coats after use at an average level of 24 cfu/25 cm² [16], which is similar to the average level of 45 cfu/25 cm² on uniforms in the present study. Other researchers sampled pajamas and bed sheets after one-night use, and found contamination levels ranging from 10² to more than 10⁵ cfu/10 cm², which were larger than the average value of 23 cfu/25 cm² on dirty bed linen in the present study, but it was using a different sampling method, namely swabbing [17]. While bed linen contamination obviously originates mostly from the patient, uniforms were most likely contaminated both by skin microflora of the wearer and by contact with contaminated surfaces during the work shift.

Contamination of clean bed linen may appear more unexpected. In a number of studies, bed sheets sampled before use did not appear to carry microbial loads [17,18]. Another one found that counts on clean white coats were, on average, lower than 2 cfu/25 cm² [16]. Washing procedures seem efficient for a removal of microorganisms [19]. However, contamination may occur after washing, but before contact with the patient: 14% of clean clothes in staff dressing rooms were found to be contaminated by S. aureus, and 10% by Clostridium sp. [20]. In the present study, since textiles leaving the laundry appear to be free from contamination (according to the launderer), although they are not claimed to be sterile, most bacteria found on "clean" bed linen likely originated from contact between clean bed sheets and uniforms, while the bed was made by the hospital staff. Since all samples from uniforms were positive for presence of microorganisms, regardless of the job of the wearer, these uniforms represent the most likely source of contamination of bed linen. This is in agreement with studies showing a transfer of microor- ganisms between various textiles [14]. It should be noted that the relatively highly contaminated pillows and du- vets are not changed as frequently as other items.

Most species identified from the samples correspond to usual members of skin microflora or to bacteria from the environment. Gram-positive cocci were the most prevalent species. These findings are coherent with other reports [8,21,22]. A number of studies specifically focused on searching HAI-relevant pathogens [2], mostly *S. aureus* or enterococci [9,11,23]. In the present study, only eight samples contained *S. aureus* (two clean bed items, three dirty bed sheets, three uniforms), and a single dirty

bed sheet was contaminated with a non-pathogenic *Ente-rococcus* strain. However, a majority of isolated species may act as opportunistic pathogens, given the fragility and possibly weakened immune system of patients staying in a hospital. Bacterial isolates were not tested for antimicrobial susceptibility, since our final objective is to reduce the microbial load overall.

Contamination of staff uniforms may be considered as the most relevant hazard identified from this study. Indeed, although rules on hand hygiene have received wide attention and nowadays are largely complied with, benefits from clean hands may be partially lost by the fact that contaminated uniforms are worn from one room to another. The ubiquity of staff in a hospital thus represents a likely route of infection spread possibly involving textiles. Therefore, the use of treated textiles having an antimicrobial activity may be beneficial for limiting HAIs [24,25]. As a follow-up to the present results, similar tests will be performed by using bed sheets and uniforms with an antimicrobial activity.

5. Acknowledgements

This work was supported by Région Nord-Pas-de-Calais and European regional development fund in the framework of CROSSTEXNET ERA-NET Project Transnational Call 2010 (Antibacterial flexible barrier to fight nosocomial diseases in hospital and for medical applications-BACNOS) from the European commission. Partners of the project BACNOS are gratefully acknowledged: Devan Chemicals NV, Colortex byba, Centexbel, and Subrenat.

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