

Bacterial contamination of nonsterile disposable gloves before use

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Background: After *Bacillus cereus* recovery in opened boxes of disposable gloves, the bacteriological contamination of disposable nonsterile gloves kept stored in native packages was investigated prospectively.

Methods: Thirty-six commercially available nonsterile nonpowdered disposable gloves made of latex, vinyl, or nitrile were cultured.

Results: A large variety of spore-forming and non-spore-forming bacteria was recovered, including *Bacillus cereus* and *Clostridium perfringens*.

Conclusion: This finding must be taken into consideration for care involving gloves in very immunocompromised patients. (Am J Infect Control 2006;34:128-30.)

In accordance with standard precautions, the use of disposable gloves followed immediately by hand hygiene after removal is intended to minimize cross-transmission of microorganisms in the hospital setting.¹⁻³ The use of disposable nonsterile gloves by health care workers (HCWs) is recommended for a large variety of care procedures, such as those involving contact with mucosa or secretions.

During the investigation of an outbreak of mucosal colonization with *Bacillus cereus* (eyes, throat, and stools) in neonatal intensive care unit, we isolated this bacterium in opened boxes of disposable gloves. A few months later, a cluster of digestive colonization and infection cases with *Clostridium perfringens* was recorded in the same unit without recovery of this bacterium from the hospital environment (data not shown). Because these bacteria can survive for several months in the environment and because nonsterile disposable gloves are widely used for care procedures involving mucosae, a study was conducted from April 2004 to June 2004 in the Microbiology Department of the University Hospital of Saint-Etienne to investigate the bacteriological contamination of nonsterile disposable gloves before the boxes were opened.

METHODS

The studied gloves included commercially available nonsterile nonpowdered disposable gloves currently used in our hospital and packaged in boxes of 100 units; they were made of three different materials: latex (Examinex, Thermofina, France), nitrile (Nitratex EP, Ansell Medical, Kulim, Malaysia), and polyvinyl (Line, Didactic, France). For the study, 2 different batches of each of the 3 sizes (small, medium, large) were tested for each material.

All of the experiments were performed in a class II microbiological safety cabinet to exclude airborne contamination. After treatment of the surfaces of the boxes with a low-level disinfectant (Anios TSA, Anios, Lille, France), each box was opened and the gloves were removed. The operator wore sterile gloves and used sterile forceps. For each box, two gloves were cultured: the glove on the top and one glove in the middle of the box. Each glove was immersed into 240 mL of sterile resuscitation broth containing 3% Tween 80. After stirring for 2 minutes, the solution was sonicated for 1 minute. Eighty milliliters of this solution was filtered through a 0.45- μ m filter (Millipore Corporation, Bedford, MA); the filter was placed onto a plate count agar (Biokar, Beauvais, France) and incubated at 22°C under aerobic conditions. A colony count was performed after 72 hours of incubation. The remaining 160 mL of the initial solution was warmed up to 80°C during 10 minutes. One half of the solution was filtered as described above; the filter was placed onto a meat liver glucose agar plate (Biokar) and incubated under aerobic conditions at 37°C. The other half of the solution was also filtered; the filter was placed upside down onto a meat liver glucose agar plate and incubated under anaerobic conditions at 37°C. The ability of this procedure to detect sulfite-reducing anaerobic bacteria from a large

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Table 1. Contamination of nonsterile disposable gloves before use, stratified by material and position in the box

Material	Box position	Bacteria	Bacterial cell count (CFU/80 mL) per glove					
			Small size		Medium size		Large size	
			Batch 1	Batch 2	Batch 1	Batch 2	Batch 1	Batch 2
Latex	Top	Non-spore-forming	18	37	0	6	6	12
		Aerobic spore-forming	3	6	>300	4	>300	4
		Anaerobic spore-forming	0	0	0	0	0	0
	Middle	Non-spore-forming	8	1	2	2	2	5
		Aerobic spore-forming	1	2	>300	0	0	9
		Anaerobic spore-forming	0	0	0	0	0	0
Polyvinyl	Top	Non-spore-forming	4	1	5	8	10	1
		Aerobic spore-forming	>300	1	0	2	>300	1
		Anaerobic spore-forming	0	0	0	4	0	0
	Middle	Non-spore-forming	2	0	3	1	15	1
		Aerobic spore-forming	>300	1	0	0	>300	0
		Anaerobic spore-forming	0	0	0	0	5	0
Nitrile	Top	Non-spore-forming	4	20	25	17	23	35
		Aerobic spore-forming	>300	12	0	9	0	26
		Anaerobic spore-forming	2	0	0	0	0	0
	Middle	Non-spore-forming	30	8	12	29	3	42
		Aerobic spore-forming	0	4	>300	4	>300	7
		Anaerobic spore-forming	0	0	0	0	0	0

volume of broth was ascertained in preliminary experiments by comparing the growth rates of serial dilutions of a *C. perfringens* strain tested either by filtration as described above or by direct incorporation of the broth into prewarmed agar medium as recommended in the NF T 90-415 AFNOR French standard procedure. For both aerobic and anaerobic cultures, a final colony count was performed after 48 hours of incubation. Bacteria and fungi were subcultured onto selective or nonselective media before identification by using API galleries (bioMerieux, Marcy l'Etoile, France) or BBL Crystal Identification System (Becton Dickinson, Cockeysville, MD). The sterility of culture media and of the surfaces of the microbiological safety cabinet was assessed periodically during the study.

The SPSS 10.1 software (SPSS Inc, Chicago, IL) was used for data analysis and comparison. The Wilcoxon nonparametric test was used for mean comparison; *P* values below the 5% level were considered significant.

RESULTS

A total of 36 gloves were tested as described above. As shown in Table 1, spore-forming and/or non-spore-forming bacteria were recovered from all of the tested gloves. Gloves made of nitrile were significantly more contaminated with non-spore-forming bacteria than those made of latex or polyvinyl, with a mean of 20.7, 8.25, and 4.25 CFU per 80 mL (median, 21.5, 5.5, and 2.5) for each material respectively (*P* = .001). Considering the contamination by non-spore-forming bacteria, the glove on the top of the box tended

to harbor more germs than the glove in the middle of the box, with a mean of 9 CFU/80 mL (median, 6) for latex (*P* = .02) and of 3.5 CFU/80 mL (median, 2) for vinyl (*P* = .08); in contrast, the bacterial load was not different for nitrile gloves according to the position of gloves in the box.

When considering glove contamination by spore-forming bacteria, it seemed that all of the types of gloves were contaminated mainly with *Bacillus subtilis* but also with other aerobic bacteria (notably *B. cereus*). By contrast, anaerobic spore-forming bacteria were only recovered in two glove boxes (*Clostridium acetobutylicum* and *C. perfringens*). In addition, *Aspergillus versicolor* was recovered once in a box of nitrile gloves.

DISCUSSION

To our knowledge, this study is the first to have investigated the bacterial contamination of nonsterile disposable gloves before use in the hospital setting. To reach this goal, ultrasound and resuscitation broth solution were used to maximize the recovery of bacteria from gloves; culture conditions were adapted from a microbiological method used for the study of hand contamination.⁴

We had previously observed two episodes of colonization/infection in the neonatal intensive care unit of our hospital, respectively with *B. cereus* and *C. perfringens*, two agents known to be responsible for severe infections in immunosuppressed infants.^{5,6} Because *B. cereus* had been recovered in opened glove boxes and although it could not be excluded that this

contamination was attributable to an external source during glove removal from the packaging, it was speculated that contaminated gloves inside the box could have played a role in the colonization of neonates.

To investigate this hypothesis, the prospective study described herein was conducted to evaluate the bacterial contamination of stored nonsterile disposable gloves. The results confirm that a large variety of spore-forming and non-spore-forming bacteria can be isolated from nonsterile gloves, including *B. cereus* and *C. perfringens*. This finding is not surprising because the European manufacturing standards for nonsterile gloves only require physical testing (EN 455-1, 2). The *B. cereus* was recovered on the top of the gloves but also in the middle of the box, suggesting a contamination during the manufacturing process, whereas *C. perfringens* was recovered on the top of the glove box, rather suggesting a contamination during storage. Another interesting observation is the higher mean bacterial load found in nitrile gloves as compared with those made of other materials. However, further studies are needed to verify whether this finding is related to the material or to the manufacturing conditions.

CONCLUSIONS

The hands of HCWs can be highly contaminated, notably during routine care procedures, when exposed to moist body substances.⁷ In addition to hand hygiene, the use of gloves is important to minimize the cross-transmission of microorganisms^{1,2} and can help to control nosocomial outbreaks.⁸ However, HCWs must be aware that nonsterile disposable gloves could be contaminated with a wide range of bacteria, including

spore-forming agents. These findings plead for introducing additional controls regarding the absence of spore-forming bacteria in nonsterile disposable gloves and for improving the absence of leaks of glove packaging. They also suggest that recommendations must be given to HCWs to store glove boxes in clean areas and to use preferentially sterile disposable gloves for care procedures involving mucosa in very immunocompromised patients.

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