ORIGINAL ARTICLE

Comparison of the Efficacy of a Hydrogen Peroxide Dry-Mist Disinfection System and Sodium Hypochlorite Solution for Eradication of *Clostridium difficile* Spores

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OBJECTIVE. To compare a hydrogen peroxide dry-mist system and a 0.5% hypochlorite solution with respect to their ability to disinfect *Clostridium difficile*–contaminated surfaces in vitro and in situ.

DESIGN. Prospective, randomized, before-after trial.

SETTING. Two French hospitals affected by *C. difficile*.

INTERVENTION. In situ efficacy of disinfectants was assessed in rooms that had housed patients with *C. difficile* infection. A prospective study was performed at 2 hospitals that involved randomization of disinfection processes. When a patient with *C. difficile* infection was discharged, environmental contamination in the patient's room was evaluated before and after disinfection. Environmental surfaces were sampled for *C. difficile* by use of moistened swabs; swab samples were cultured on selective plates and in broth. Both disinfectants were tested in vitro with a spore-carrier test; in this test, 2 types of material, vinyl polychloride (representative of the room's floor) and laminate (representative of the room's furniture), were experimentally contaminated with spores from 3 *C. difficile* strains, including the epidemic clone ribotype 027–North American pulsed-field gel electrophoresis type 1.

RESULTS. There were 748 surface samples collected (360 from rooms treated with hydrogen peroxide and 388 from rooms treated with hypochlorite). Before disinfection, 46 (24%) of 194 samples obtained in the rooms randomized to hypochlorite treatment and 34 (19%) of 180 samples obtained in the rooms randomized to hydrogen peroxide treatment showed environmental contamination. After disinfection, 23 (12%) of 194 samples from hypochlorite-treated rooms and 4 (2%) of 180 samples from hydrogen peroxide treated rooms showed environmental contamination, a decrease in contamination of 50% after hypochlorite decontamination and 91% after hydrogen peroxide decontamination ($P < .005$). The in vitro activity of 0.5% hypochlorite was time dependent. The mean ($\pm$SD) reduction in initial $\log_{10}$ bacterial count was 4.32 $\pm$ 0.35 $\log_{10}$ colony-forming units after 10 minutes of exposure to hypochlorite and 4.18 $\pm$ 0.8 $\log_{10}$ colony-forming units after 1 cycle of hydrogen peroxide decontamination.

CONCLUSION. In situ experiments indicate that the hydrogen peroxide dry-mist disinfection system is significantly more effective than 0.5% sodium hypochlorite solution at eradicating *C. difficile* spores and might represent a new alternative for disinfecting the rooms of patients with *C. difficile* infection.

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Clostridium difficile, a gram-positive, spore-forming anaerobic bacillus, is a major nosocomial enteropathogen responsible for 15%–25% of antibiotic-associated diarrhea and virtually all the cases of pseudomembranous colitis in adults.[1,2] The main established risk factors for *C. difficile* infection (CDI) are receipt of antibiotic therapy, age older than 65 years, severity of underlying disease, and length of hospital stay.[3]

Since 2003, a number of studies have reported outbreaks of CDI due to a highly virulent strain variously characterized as ribotype 027, restriction enzyme analysis type BI, and North American pulsed-field gel electrophoresis type 1 (NAP1).[4,5] This clone is responsible for more-severe infection with complications that include septic shock, toxic megacolon, digestive system perforation, and death. The emergence of this strain was associated with an increase of the incidence of CDI in North America and northern Europe.[6–8]

C. difficile infection is spread by the oral-fecal route, and strains are usually transmitted on the hands of healthcare workers. However, increasing evidence suggests that the environment might play an important role in cross-transmission. The rooms of patients with CDI are frequently contaminated with *C. difficile*, and the percentage of environmental samples from such rooms that are positive for *C. difficile* ranges from 9% to 59%.[3,9,10] Environmental contamination is more common in

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the rooms of patients with CDI than in the rooms of asymptomatic carriers (49% vs 29% of environmental samples), which suggests that diarrhea is the key factor in strain dissemination.[3] More surprisingly, the percentage of *C. difficile*–contaminated samples recovered from the rooms of patients who are neither infected nor colonized with the bacteria can reach 8%, which probably reflects the inefficacy of detergents and/or disinfectants. Spores can persist for up to 5 months on the surface of objects.[9] Recent data have suggested that epidemic clones, including the 027/NAP1 type, have a better sporulation capacity than do other *C. difficile* strains. Additionally, the rate of sporulation might be positively influenced by subinhibitory concentrations of detergent alone or detergent and disinfectant combined.[11–13] The persistence of *C. difficile* spores in the environment creates a reservoir, the elimination of which requires a meticulous disinfection process that uses sporicidal products. The commonly available disinfectants with sporicidal properties include sodium hypochlorite, peracetic acid, glutaraldehyde, and hydrogen peroxide associated with different diffusion processes (eg, directed spray, wet spray, nebulization, or vaporization).[14–19] Recently, a new disinfection process that uses a hydrogen peroxide–based dry-mist disinfection system was developed.[15,16]

The present study compares the efficacy of a hydrogen peroxide–silver cation dry-mist system (Sterinis-Sterusil; Gloster Santé Europe) and a 0.5% sodium hypochlorite solution (5,000 ppm available chlorine) for disinfection of *C. difficile*–contaminated surfaces both in situ, in rooms previously occupied by patients with CDI, and in vitro, by use of spore carriers contaminated with different *C. difficile* strains.

METHODS

Materials

Five percent of the hydrogen peroxide–silver cation product (Sterusil) consists of hydrogen peroxide, phosphoric acid at a concentration of less than 50 ppm, silver cations at a concentration of less than 50 ppm, and gum arabic at a concentration of less than 1 ppm; the remaining 95% of the product is biosmotic water. The product is nontoxic, noncorrosive, and more than 99.99% biodegradable. The bactericidal, fungal, sporicidal, and virucidal properties of the product are the result of the oxidative action of hydrogen peroxide on the lipid membrane of microorganisms, which leads to modification in ribosomes and DNA, and the cationic effect of silver cations, which reverses membrane polarity and inhibits protein synthesis and cytoplasmic enzyme activity. The disinfection process (Sterinis) is performed with a dry-mist dispenser that produces electrically charged particles (smaller than 10 μm) that circulate freely in air as a dry aerosol disinfectant that has access to all surfaces. The system consists of a robot that can be preprogrammed to dispense the required concentration of hydrogen peroxide dry aerosol needed for full disinfection, which depends on the exact volume of the room, at a rate of 6 mL/m^3 (30 mL/minute). For the purposes of this study, the disinfection process (which requires approximately 18 minutes of diffusion time, depending on the volume of the room, and 1 hour of exposure time) was carried out in vacant rooms. The rooms' doors and windows were closed, but it was not necessary to seal the rooms. The dry-mist diffusion of the product was evaluated by use of colorimetric strips (Nocotest; Oxy'Pharm) that were placed in the room and its attached bathroom; the color of the strip changed in the presence of excess hydrogen peroxide (ie, more than 100 mg/mL).

Sodium hypochlorite solution at a concentration of 0.5% (5,000 ppm available chlorine) is known for its bactericidal, fungal, sporicidal, and virucidal properties. Both European guidelines and the French Ministry of Health's Technical Committee for Nosocomial Infections and Health Care–Related Infections recommend it for the disinfection of rooms occupied by patients with CDI.[7,14,20] Sodium hypochlorite at a concentration of 0.5% was prepared as needed using a 1 : 5 dilution of a 2.6% sodium hypochlorite solution.

In Situ Study

A prospective, randomized study was performed in 2 French university hospitals (Henri Mondor–Albert Chenevier Hospital and Saint-Antoine Hospital) from April through August 2007. The design of the study was approved by the infection control committee of each hospital. These hospitals were not affected by the epidemic-related, highly virulent 027/NAP1 strain. A CDI case patient was defined as a patient with a diarrhetic stool (ie, a stool that took the shape of the sample container) and a toxin test result positive for *C. difficile* toxin(s). At Saint-Antoine Hospital, toxin was detected by use of a stool cytotoxicity assay performed on MRC-5 cells; at Henri Mondor–Albert Chenevier Hospital, it was detected by use of the Premier Toxins A&B assay (Meridian Bioscience). When patients with CDI were discharged, their rooms were randomized to 1 of 2 decontamination arms, either decontamination with 0.5% sodium hypochlorite solution (hereafter, "hypochlorite disinfection") or decontamination with the hydrogen peroxide–silver cation dry-mist system (hereafter, "peroxide disinfection"). Prior to disinfection, the rooms (ie, their floors and other surfaces) were cleaned by use of a detergent-disinfectant (Surfanios; Anios Laboratoires) and rinsed with tap water. Hypochlorite disinfection was performed by use of a single-use cloth soaked in 0.5% hypochlorite solution, which was prepared daily in accordance with a standardized written procedure. It was stressed to the individuals who performed the disinfection process that surfaces that were frequently touched, such as bed rails, bedside tables, doorknobs, and call buttons, were to be disinfected. All disinfected surfaces were allowed to air dry except for stainless steel surfaces, which were rinsed with tap water after an exposure time of 10 minutes. The housekeeping staff in

charge of disinfection was periodically trained in how to perform the disinfection process.

Samples were collected before cleaning and after disinfection (ie, after drying for hypochlorite or after 1 hour of exposure time for peroxide). In each room, either 12 or 13 100-cm^2 environmental surface areas (13 if all were present, otherwise 12) were sampled using swabs premoistened in Schaedler broth (bioMérieux). These surfaces included the toilet seat, bathroom sink, bathroom floor, bedside table, care table, telephone, door handle, chair arm, floor of the main room, windowsill, remote control for the bed, bed barrier, and alcohol-based hand gel dispensers. Swab samples were directly inoculated on brain-heart infusion agar plates supplemented with 5% defibrinated horse blood, 0.1% taurocholate, 8 μg/mL cefoxitin, 250 μg/mL cycloserine (TCCA plates) and in Schaedler broth supplemented with 0.1% taurocholate, 8 μg/mL cefoxitin, and 250 μg/mL cycloserine. TCCA plates and broth were incubated at 37°C in an anaerobic atmosphere for 48 hours. Each broth culture with visible growth was plated on TCCA plates. Colonies of *C. difficile* were initially identified by their macroscopic appearance and odor; suspect colonies were conclusively identified with a commercial identification system (API Rapid ID32A; bioMérieux). For each sample that was positive for *C. difficile* by direct plating, the colonies of *C. difficile* were counted (as colony-forming units per plate).

In Vitro Study

The sporicidal property of both disinfectants was tested in vitro by use of the spore-carrier test.[21] The spore carriers used were small (2 cm^2) pieces of vinyl polychloride (representative of the room's floor) or laminate (representative of the room's furniture). Each spore carrier was experimentally contaminated with 1 of the following 3 toxigenic strains of *C. difficile:* the reference strain VPI 10463 (toxinotype 0), the CD196 strain (CIP107932 from the Pasteur Institute Collection; 027 historical strain), and the 1067 strain (027/NAP1 American epidemic strain). For each assay, the 3 strains were simultaneously but separately tested on the 2 different spore carriers. Each assay was repeated 4 times.

C. difficile spores were obtained by use of the method described by Wullt et al.[22] Strains were incubated in Wilkins-Chalgren broth (bioMérieux) at 37°C in an anaerobic atmosphere for 7 days. We mixed 8 mL of absolute ethanol with 8 mL of contaminated Wilkins-Chalgren broth, which was then left to sit for 30 minutes at room temperature. The mixture was then centrifuged at 3,000 *g* for 20 minutes, at 4°C. We checked for the presence of spores under a microscope after Gram staining. The pellet containing the spores was suspended in 1 mL of absolute ethanol. Spore carriers were soaked overnight in a detergent solution (Hexanios; Anios Laboratoires), washed in tap water, and sterilized (at 121°C for 15 minutes). Each spore carrier was experimentally contaminated with 100 μL of spore suspension and then dried for 15 minutes at 37°C.

The test spore carriers were placed in a vacant room in a predefined arrangement near the bed and in the bathroom (Figure 1) and subjected to the peroxide-based disinfection process. The control spore carriers (controls) were left on the laboratory bench at room temperature and were not subjected to the disinfection process. After disinfection, to evaluate the efficacy of the peroxide-based process, test and control spore carriers were placed in 3 mL of a neutralizing solution (30% polysorbate 80, 1% histidine, 3% egg lecithin, and 0.5% sodium thiosulfate [DNP solution; AES Laboratories]), mixed with a vortex mixer for 30 seconds, sonicated for 5 minutes, and mixed with a vortex mixer again for 30 seconds.

To evaluate the efficacy of hypochlorite disinfection, test spore carriers were softly shaken in 0.5% sodium hypochlorite solution, and control spore carriers were softly shaken in water. The following exposure times were tested: 10 seconds, 5 minutes, 10 minutes, and 20 minutes. After the exposure time had elapsed, the liquid was drained, and the spore carriers were placed in a sterile vial. We added 3 mL of 0.9% sodium chloride 0.5% sodium thiosulfate to neutralize the action of chlorine. The vials were then mixed with a vortex mixer for 30 seconds, sonicated for 5 minutes, and mixed with a vortex mixer again for 30 seconds.

The spores from test and control spore carriers were counted by inoculating 100 μL of 10-fold serial dilutions of each suspension on TCCA plates. For the test carriers, 1 mL of pure suspension was inoculated on 2 TCCA plates to obtain a threshold of sensitivity of 3 spores/spore carrier. The TCCA

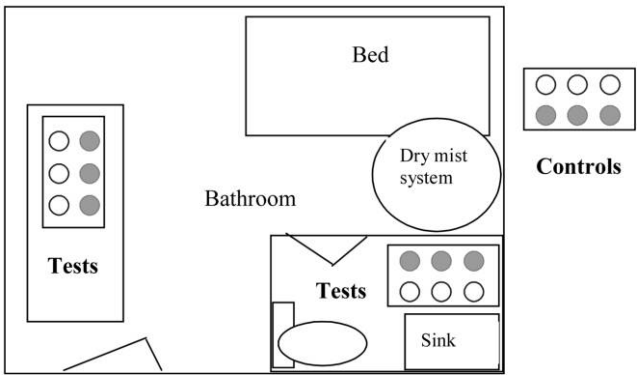

FIGURE 1. Diagram of a hospital room and its attached bathroom showing the arrangement of pieces of laminate *(white circles)* and vinyl polychloride *(gray circles)* contaminated with *Clostridium difficile* spores. These pieces of material (spore carriers) were used for in vitro studies of the efficacy of 2 decontamination processes (0.5% sodium hypochlorite solution and a hydrogen peroxide–silver cation dry-mist system). Spore carriers were experimentally contaminated with 1 of 3 strains of *C. difficile* (CD196, VPI 10463, or 1067) and placed as shown. Control spore carriers (which were not subjected to the decontamination processes) were left on the laboratory bench at room temperature.

plates were incubated at 37°C in anaerobic atmosphere for 48 hours. Spores were then counted and results were expressed as $\log_{10}$ colony-forming units. The reduction in initial contamination was calculated as the difference between the number of spores on control carriers and the number on test carriers after each disinfection process.

To ensure that the disinfectant-neutralization procedure itself was not detrimental to spore viability, spore carriers were treated with either saline alone or saline containing the neutralizers (for the peroxide-based process, the solution containing 30% polysorbate 80, 1% histidine, 3% egg lecithin, and 0.5% sodium thiosulfate; for the hypochlorite process, 0.9% sodium chloride containing 0.5% sodium thiosulfate). After an exposure time of 10 minutes, the spores were eluted and counted on selective media.

Statistical Analysis

For the in situ study, the percentage of *C. difficile*–positive samples obtained before and after each disinfection process was compared by use of the χ^2 test (Epi Info, version 6.01; Centers for Disease Control and Prevention). The percentage of rooms from which *C. difficile* was recovered before and after each disinfection process was compared by use of the Fisher 2-tailed exact test. Significance was set at $P < .05$. For the in vitro study, the reduction in initial contamination was compared by use of 1-way analysis of variance (Prism 4; GraphPad Software).

RESULTS

In Situ Evaluation of Efficacy

When patients with CDI were discharged, their rooms ($n = 31$) were cleaned and randomized into 1 of 2 disinfection groups; there were 15 rooms treated with peroxide and 16 rooms treated with hypochlorite. A total of 748 surface samples (360 from peroxide-treated rooms and 388 from hypochlorite-treated rooms) were collected; for each disinfection method, half of the samples were collected before treatment and half were collected after treatment. The surfaces sampled included the toilet seat ($n = 58$), bathroom sink ($n = 58$), bathroom floor ($n = 58$), bedside table ($n = 62$), care table ($n = 62$), telephone ($n = 54$), door handle ($n = 60$), chair arm ($n = 58$), main room floor ($n = 62$), windowsill ($n = 60$), remote control for the bed ($n = 62$), bed barrier ($n = 62$), and alcohol-based hand gel dispensers ($n = 32$).

Before cleaning, *C. difficile* spores were detected in 80 (21%) of 374 surface samples and in 23 (74%) of 31 rooms. There was not a significantly different percentage of *C. difficile*–positive samples obtained from rooms randomized to be treated with hypochlorite (46 [24%] of 194 samples) and rooms randomized to be treated with peroxide (34 [19%] of 180 samples) ($\chi^2 = 1.29$; $P = .256$). After disinfection, the percentage of samples that showed environmental contamination significantly decreased in both arms. A total of 23 (12%) of 194 samples obtained after disinfection from the hypochlorite-treated rooms were positive for *C. difficile* ($\chi^2 = 9.3$; $P < .002$), 4 (2%) of 180 postdisinfection samples from the peroxide-treated rooms were positive for *C. difficile* ($\chi 2 = 28.9$; $P < .001$). The decrease in the percentage of contaminated samples was significantly greater in the peroxide group (91%) than in the hypochlorite group (50%) ($\chi^2 = 17.4$; $P < .005$). In rooms treated with hypochlorite disinfection, surface samples were positive for *C. difficile* by direct plating in 31 (16%) of 194 rooms before treatment and 8 (4%) of 194 rooms after treatment; in rooms treated with peroxide disinfection, surface samples were positive by direct plating in 19 (11%) of 180 rooms before treatment and 3 (2%) of 180 rooms after treatment. In the hypochlorite disinfection arm, the percentage of rooms with at least 1 sample positive for *C. difficile* was 69% (11 of 16) before treatment and 50% (8 of 16) after treatment ($\chi^2 = 1.17$; $P = .28$); in the peroxide disinfection arm, the percentage of rooms with at least one sample positive for *C. difficile* was 80% (12 of 15) before treatment and 20% (3 of 15) after treatment ($\chi^2 = 8.53$; $P = .003$).

The distribution of contaminated samples from different environmental sites both before and after the hypochlorite and peroxide disinfection processes is presented in Figure 2. The sites most commonly contaminated with *C. difficile* before disinfection included the bathroom floor (11 [38%] of 29 samples), toilet seat (10 [34%] of 29), bathroom sink (9 [31%] of 29), telephone (8 [30%] of 27), bedside table (7 [23%] of 31), and windowsill (6 [20%] of 30). Regardless of the disinfection process used, the sites that remained contaminated after disinfection were the bathroom floor (5 [17%] of 29), the arm of the chair (4 [14%] of 29), and the toilet seat (3 [10%] of 29).

In Vitro Evaluation of Efficacy

The neutralizers did not significantly reduce the spore titer in fluid from the test spore carriers, compared with fluid from saline-treated control carriers, and the neutralizers had no deleterious effect on the viability of spores (data not shown). The mean ($\pm$SD) initial contamination of spore carriers was 5.54 ± 0.67 $\log_{10}$ cfu in the hypochlorite group and 5.51 ± 0.76 $\log_{10}$ cfu in the peroxide group ($P = .65$). For peroxide-treated carriers, the mean reduction of initial contamination was 4.18 ± 0.80 $\log_{10}$ cfu. For hypochlorite-treated carriers, a time-dependent decrease in spore contamination was observed, and reduction ranged from 1.76 ± 0.96 $\log_{10}$ cfu after 10 seconds of exposure to 4.33 ± 0.37 $\log_{10}$ cfu after 20 minutes of exposure (Figure 3). The differences observed between the 2 surfaces (ie, laminate and vinyl polychloride) with respect to reduction were not statistically significant for either hypochlorite (pooling the data obtained from 10 and 20 minutes of exposure) or peroxide (Table 1).

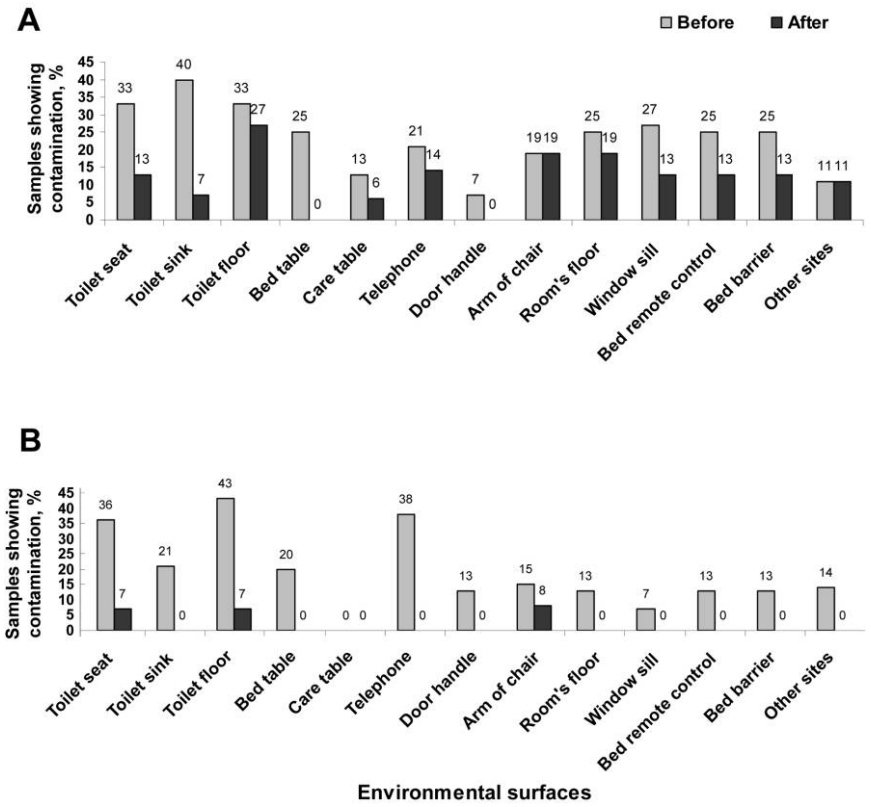

FIGURE 2. Percentage of samples that showed environmental contamination with *Clostridium difficile* before and after disinfection with 0.5% sodium hypochlorite solution *(A)* or a hydrogen peroxide–silver cation dry-mist system *(B)*.

DISCUSSION

The environment of patients with CDI is heavily contaminated with *C. difficile*. In the present study, *C. difficile* spores were detected in 21% of surface samples (80 of 374) and in 74% of rooms (23 of 31) that had housed patients with CDI. This finding is in agreement with previous reports showing that the percentage of contaminated sites in the rooms of infected patients ranges from 9.3% to 59%.[3,9,10,23] Consistent with the results obtained by others,[23,24] the surfaces most frequently found to be contaminated in this study included the floors of the main room and bathroom and the toilet, which emphasizes the need to clean these areas thoroughly. Even in studies in which an association between environmental contamination and the incidence of CDI has been observed,[24] the role of this contamination in cross-transmission is difficult to interpret. It might be the consequence of strain dissemination by an infected patient or the source of the infection. However, the role of environmental contamination in *C. difficile* cross-transmission is suggested by both the control of outbreaks after the modification of cleaning procedures[25–27] and the significant decrease in the incidence of *C. difficile* infection after the use of a sporicidal disinfectant.[19,28]

In many countries (including France), hypochlorite-based products are currently recommended for disinfecting the rooms of patients with CDI.[7,14,20] The use of hypochlorite has been shown to decrease the incidence of CDI, particularly in departments with high endemic levels of CDI.[13,19,29] Nevertheless, the use of hypochlorite-based disinfectants has several drawbacks, including the following: (1) the disinfection procedure must be performed manually, it is time consuming, and the quality of disinfection is strongly dependent on the practices of the housekeeping staff who perform disinfection[25]; (2) hypochlorite-based products can be corrosive to various materials; and (3) the odor of chlorine may irritate healthcare workers' and patients' eyes and respiratory tracts. There is a need to develop a safe, rapid, and highly effective procedure to disinfect the rooms of patients with CDI. The hydrogen peroxide associated with different diffusion processes (spray and vaporization) has been previously reported to be effective against *C. difficile* spores.[16,30–32] Moreover, hydrogen peroxide–based formulations have been shown to have much higher materials compatibility while being less toxic to human beings and the environment.[33]

During the in vitro experiments, time-dependent sporicidal activity was observed for hypochlorite; the reduction of the initial *C. difficile* contamination ranged from 1.76 $\log_{10}$ cfu after 10 seconds of exposure to 4.33 ± 0.37 cfu after 20 minutes of exposure. After 10 minutes of exposure, which is

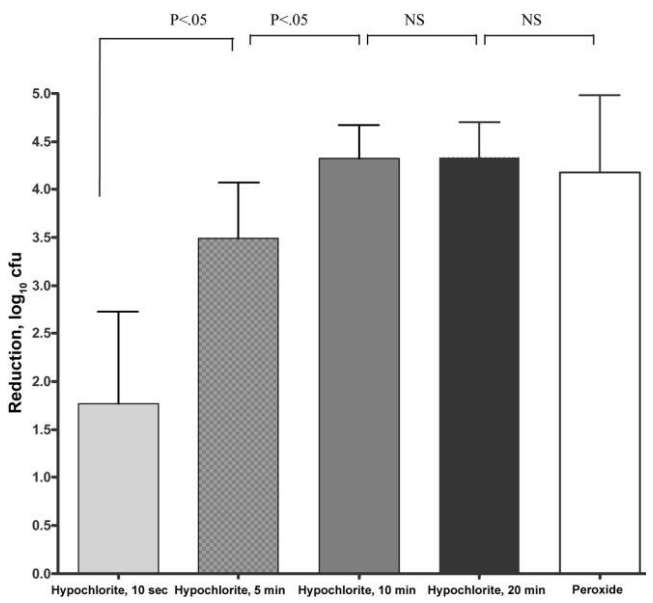

FIGURE 3. Reduction of initial contamination with *Clostridium difficile* spores during in vitro experiments. Spores were counted before and after disinfection with 0.5% sodium hypochlorite solution (hypochlorite) (10 seconds, 5 minutes, 10 minutes, and 20 minutes of exposure time) or disinfection with a hydrogen peroxide–silver cation dry-mist system (peroxide). Data are mean ± SD. For details, see Results.

the contact time usually recommended for disinfection,[14,20] the reduction reached 4.32 ± 0.35 $\log_{10}$ cfu and was not significantly different from that obtained with peroxide ($P = .42$). However, in these in vitro experiments, the reduction of spore count comes only from the disinfectant properties of hypochlorite and not from the mechanical act of cleaning. This result is consistent with previous reports that showed a 1.2–6 $\log_{10}$ cfu decrease in *C. difficile* contamination when hypochlorite was used alone or in combination with a detergent.[12,34] However, heterogeneous results have been observed in various studies. These discrepancies can be explained by the different concentrations of sodium hypochlorite solution tested (from 500 to 5,500 ppm available chlorine), the use of different formulations (such as sodium hypochlorite and sodium dichloroisocyanurate), the choice of bacterial strains (epidemic vs nonepidemic), and the types of material used for spore carriers.

With regard to the activity of peroxide, we observed a reduction of 4.18 $\log_{10}$ cfu after a single cycle of disinfection. In a recent study that used spore carriers that included 0.3% bovine serum albumin to simulate biological soiling, *C. difficile* spores were inactivated after 30 minutes of exposure to hydrogen peroxide vapor.[17] Other in vitro studies evaluated the efficacy of liquid (not dry-mist) hydrogen peroxide. Perez et al.[34] showed a 6 $\log_{10}$ cfu reduction of *C. difficile* spores after 9 minutes of exposure to 7% hydrogen peroxide (Virox STF; Virox). In contrast, Fawley et al.[12] reported that another product based on a liquid form of hydrogen peroxide (G-Force; JohnsonDiversey) had no activity against *C. difficile* spores, but the hydrogen peroxide concentration was not mentioned. During the in situ experiments, in which we tried to simulate the real conditions of use, the efficacy of peroxide was significantly greater than that of hypochlorite.

To our knowledge, only 2 recent studies have assessed the impact of hydrogen peroxide room disinfection on *C. difficile* environmental contamination.[31] The first study used a dry-mist hydrogen peroxide decontamination system (Sterinis) to treat *C. difficile* environmental contamination. In 10 rooms that housed high-risk elderly patients, 48 (24%) of 203 samples were positive for *C. difficile* before hydrogen peroxide decontamination. After a single cycle of hydrogen peroxide decontamination, only 7 (3%) of 203 samples were positive for *C. difficile,* a 94% reduction ($P < .001$). These findings are comparable to the 91% reduction observed in the present study, in which environmental contamination was assessed using pure sodium taurocholate–containing media to promote spore germination. A second prospective trial evaluated the efficacy of room decontamination with hydrogen peroxide vapor (Bioquell) in a university-affiliated hospital affected by the epidemic NAP1 strain of *C. difficile*.[31] For the process used in that study, all the room openings were sealed and special generators converted 30% liquid hydrogen peroxide into hydrogen peroxide vapor in the environment. Premoistened sterile cellulose sponges were used to collect environmental surface samples before and after disinfection with hydrogen peroxide vapor. Eleven (25.6%) of 43 cultures of samples obtained before disinfection yielded *C. difficile,* compared with 0 of 37 cultures of samples obtained after disinfection ($P < .001$).

In our study, residual contamination was observed at only 3 sites after disinfection with peroxide. The presence of *C. difficile* on the bathroom floor, toilet seat, and arm of the chair might be explained by the deterioration of these surfaces (lacerations were observed in the leather of the armchair and the porcelain of the toilet seat was damaged) and/or the presence of a biofilm that decreased the peroxide's access to the

TABLE 1. Comparison of the In Vitro Activity of Sodium Hypochlorite and Hydrogen Peroxide Against *Clostridium difficile* Spores, According to the Material Used as a Spore Carrier

Disinfection method	Reduction in initial contamination, mean (± SD), $\log_{10}$ cfu	
	Vinyl polychloride	Laminate
0.5% Sodium hypochlorite solution	4.18 ± 0.33	4.47 ± 0.32
Hydrogen peroxide–silver cation dry-mist	4.19 ± 0.86	4.17 ± 0.74

NOTE. For sodium hypochlorite solution, data from the 10-minute and 20-minute exposures were pooled. All *P* values were nonsignificant. cfu, colony-forming units.

spores. In contrast, residual contamination was more frequent in rooms treated with hypochlorite and was found at many different sites. This residual contamination might be explained by an inadequate preparation of the hypochlorite solution and/or the difficulty of cleaning all surfaces because the disinfection process was performed manually. One of the limitations of the present study is that neither the dilution nor the quality of the disinfection process was controlled during the study. One aim of the study was to assess the efficacy of the disinfection performed as part of the daily housekeeping practices. However, a quick audit performed in Saint-Antoine Hospital in September 2007 indicated that for housekeeping staff, the overall rate of compliance with the stated disinfection procedure was 83.3% (25 of 30), and 93.3% (28 of 30) fully obeyed the instructions for dilution of 0.5% hypochlorite (unpublished data).

Compared with hypochlorite disinfection, the dry-mist disinfection system is easy to use. Medical equipment that is difficult to disinfect manually and frequently is not fully disinfected can be effectively decontaminated by the hydrogen peroxide dry-mist disinfection process. It requires only a cleaning with detergent prior to disinfection and generates a standardized diffusion of the product in the environment. In this study, the hydrogen peroxide dry-mist disinfection process was carried out in a vacant room, but the process did not require that the room be sealed, in contrast with hydrogen peroxide vapor disinfection.

In conclusion, the results of our in situ experiments indicate a significantly higher efficacy for the hydrogen peroxide dry-mist system than for the use of 0.5% sodium hypochlorite, which is presently recommended for disinfecting the environment of patients with CDI. This evaluation of efficacy is made on the basis of the reduction in spore counts and not on clinical outcome (ie, decreased incidence of CDI). The results of our in vitro experiments suggest that the sporicidal activity of hypochlorite is time dependent. The hydrogen peroxide dry-mist system might represent an appropriate alternative to sodium hypochlorite for the eradication of *C. difficile* spores in patient environments. Further studies will be necessary to compare the impact of the disinfection processes on the incidence of CDI and to evaluate the costs and benefits of each process.

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(See the commentary by Boyce on pages 515–517.)

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