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ORIGINAL ARTICLE

Isolation of *Acinetobacter baumannii* Complex and Methicillin-Resistant *Staphylococcus aureus* from Hospital Rooms Following Terminal Cleaning and Disinfection: Can We Do Better?

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OBJECTIVE. To study the frequency of isolation of *Acinetobacter baumannii* complex (ABC) and methicillin-resistant *Staphylococcus aureus* (MRSA) from surfaces of rooms newly vacated by patients with multidrug-resistant (MDR) ABC following various rounds of routine terminal cleaning and disinfection (C/D) with bleach or 1 round of C/D followed by hydrogen peroxide vapor (HPV) treatment.

SETTING. A 900-bed tertiary care hospital.

METHODS. ABC and MRSA cultures were obtained from hospital rooms including 312 rooms (mean, 18.3 sites/room) following 4 rounds of C/D, 37 rooms (mean, 20 sites/room) following 1 round of C/D before and after HPV treatment, and 134 rooms (mean, 20 sites/room) following 1 round of C/D and HPV treatment.

RESULTS. Following 4 rounds of C/D, 83 (26.6%) rooms had 1 or more culture-positive sites; 102 (1.8%) sites in 51 (16.4%) rooms grew ABC, and 108 (1.9%) sites in 44 (14.1%) rooms grew MRSA. The addition of HPV treatment to 1 round of C/D resulted in a significant drop in ABC- and MRSA-positive room sites (odds ratio, 0 [95% confidence interval, 0–0.8]; $P = .04$ for both organisms). Following 1 round of C/D and HPV treatment, 6 (4.5%) rooms were culture-positive for ABC, MRSA, or both.

CONCLUSIONS. Routine terminal C/D of hospital rooms vacated by MDRABC-positive patients may be associated with a significant number of ABC- or MRSA-positive room surfaces even when up to 4 rounds of C/D are performed. The addition of HPV treatment to 1 round of C/D appears effective in reducing the number of persistently contaminated room sites in this setting.

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Acinetobacter spp. is an increasingly important cause of hospital-associated infections,^{1,2} with a propensity to persist on inanimate surfaces.³ Although aggressive or meticulous cleaning of the hospital environment has been proposed as a means of limiting the transmission of *Acinetobacter baumannii* complex (ABC),^{1,3,4} little is known about what constitutes adequate cleaning and disinfection (C/D) of ABC-contaminated rooms, particularly in nonoutbreak settings.⁵

We investigated the rate of isolation of ABC from environmental surfaces of terminally cleaned and disinfected hospital rooms vacated by patients colonized and/or infected with multidrug-resistant *Acinetobacter baumannii* complex (MDRABC). Because methicillin-resistant *Staphylococcus aureus* (MRSA) may also persist in the hospital environment³ and its risk factors for infection or colonization often overlap those of MDRABC,^{6–8} its isolation from room surfaces was simultaneously studied.

METHODS

Background

St. John's Mercy Medical Center is a suburban 900-bed community teaching medical center in St. Louis, Missouri, with a burn unit, 5 adult intensive care units (ICUs), and 2 intermediate care units (ie, step-down from ICUs). Multidrug resistance was defined as in vitro susceptibility to 2 or fewer antibiotic classes (ie, cephalosporins, aminoglycosides, monobactams, carbapenems, extended-spectrum penicillins, sulfa derivatives, tetracyclines, and quinolones),⁹ excluding colistin. The burn unit was considered an outbreak setting for MDRABC because of its zero baseline rate of healthcare-associated MDRABC infections before November 2004. Conversely, in the absence of any detectable outbreaks, MDRABC isolates from patients in other wards (eg, ICUs and general wards) and all MRSA isolates were considered endemic strains.

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Routine Terminal C/D of Hospital Rooms

Routine terminal C/D of rooms newly vacated by MDRABC-positive patients included wiping of visibly soiled surfaces with a quaternary ammonium compound followed by disinfection with 0.525% sodium hypochlorite solution (1:10 dilution of household bleach; henceforth referred to as bleach). Briefly, a dedicated cart with buckets was set up outside of the room, with separate buckets used for disinfection of the floor. Multiple wiping cotton cloths were used to avoid double dipping of clean cloths into the buckets containing bleach. Soiled curtains were replaced. Total C/D time commonly ranged between 30 and 60 minutes per room. The importance of meticulous C/D was stressed to the environmental service staff upon hire, daily during infection control ward rounds, and through weekly meetings of the infection control with the environmental service staff. While occupied by MDRABC-positive patients, such hospital rooms also underwent daily C/D with bleach (twice daily in the burn unit).

Hydrogen Peroxide Vapor (HPV) Treatment

HPV (Bioquell) treatment of selected hospital rooms was initiated at St. John's Mercy Medical Center on December 15, 2008. Subsequently, all rooms newly vacated by MDRABC-positive patients underwent 1 round of C/D with bleach followed by HPV treatment. The procedure for HPV treatment of rooms was as previously described.¹⁰ Biological indicators (more than 1.0×10^6 *Geobacillus stearothermophilus* spores in Tyvek pouches; Apex) were routinely utilized (4 per room) during each cycle for quality control purposes. The entire process usually took 3–4 hours/room.

Microbiologic Sampling of the Environment

Premoistened culture swabs (BactiSwab, Remel) were used to culture room sites.¹¹ Cultures were obtained when rooms were considered clean and dry by visual inspection. Swabs were simultaneously used for isolation of ABC and MRSA. Each swab was streaked on 5% sheep blood and MacConkey agar plates. Standard methodology was used to identify *S. aureus* (latex test, Staphaurex, Remel); screening for MRSA was based on cefoxitin diffusion testing.¹² For ABC, an automated identification and drug susceptibility testing was performed (VITEK 2 system, bioMérieux).

Table 1 shows the selection of rooms for microbiological sampling by study period, number of cultured sites and rooms, and the timing of cultures relative to the C/D status of the room with or without HPV treatment. Except for period A, the majority of rooms cultured were located outside of the burn unit. During periods A and B, the number of sites cultured per room ranged from 9 to 30, in part related to the variation in the number of existing items and equipment in the room at the time cultures were obtained and the uncertainty of the number of cultures necessary before a room could be declared decontaminated. Subsequently, however, a fixed number of 20 sites per room were cultured, with priority

given to surfaces likely to have been touched by patients or healthcare workers (periods C and D). During period C, to assess the efficacy of HPV treatment, 37 newly vacated rooms underwent serial cultures following 1 round of C/D before and after HPV treatment; for 35 rooms, pre-clean cultures were also available from the same sites. During period D, 136 rooms were vacated by MDRABC-positive patients, of which 134 were cultured; 58 (43.3%) rooms were vacated by patients with concurrent MRSA infection and/or colonization.

Persistently ABC-positive rooms underwent another 4 rounds of C/D before the availability of HPV or a repeat round of C/D followed by HPV treatment thereafter. Rooms were released only if all cultures were negative for ABC. The relatedness of environmental and clinical MDRABC isolates was studied by pulsed-field gel electrophoresis (PFGE) during 3 separate months (March 2005, January 2006, and December 2006). No attempt was made to study the relatedness of environmental and clinical MRSA isolates.

Statistical Analysis

Statistica (StatSoft) and InStat (GraphPad) were used for data analysis. Fisher exact test of significance was used for comparison of categorical data; χ^2 McNemar test of significance was used for comparison of paired data related to serial sampling of the same room sites. $P < .05$ was considered statistically significant.

RESULTS

Isolation of ABC and MRSA from Burn Unit Rooms Following 2 Rounds of C/D

During period A (Table 1), 17 room sites (12.1%) grew ABC and 5 (3.6%) grew MRSA. Four (44.4%) and 5 (55.6%) rooms had 1 or more persistently culture positive sites for ABC and MRSA, respectively. ABC-positive room sites included the television, patient chair/recliner, contaminated needle container, patient table, cardiac monitor, sink, and IV pump, while MRSA-positive room sites included the in-room computer keyboard and mouse, television, countertop, lift, bed rails, and floor.

Isolation of ABC and MRSA from Hospital Rooms Following 4 Rounds of C/D

During period B (table 1), 102 (1.8%) and 108 (1.9%) room sites grew ABC or MRSA, respectively. Overall, ABC or MRSA grew from 1 or more sites of 51 (16.4%) and 44 (14.1%) rooms, respectively; 83 (26.6%) rooms grew one or both of these organisms. In the burn unit, ABC or MRSA grew from 1 or more sites of 12 (21.4%) and 16 (28.6%) rooms, respectively, with 22 (39.3%) rooms growing one or both of these organisms. Outside of the burn unit, ABC or MRSA was recovered from 1 or more sites of 61 (23.8%) rooms; 39 (15.2%) rooms grew ABC, and 28 (11%) rooms grew MRSA.

ABC or MRSA grew from several room sites with a high

TABLE 1. Isolation of *Acinetobacter baumannii* Complex and Methicillin-Resistant *Staphylococcus aureus* in 4 Study Periods

Study period (dates)	Rooms, no.		Room sites, no.		Range	Mean/room	C/D status
	Total	Burn unit	Total				
A (March 1–30, 2005)	9	9	140		9–20	15.6	After 2 × C/D ^a
B (January 1, 2006–December 14, 2008)	312	56	5,705		16–30	18.3	After 4 × C/D ^a
C (December 15, 2008–March 25, 2009)	37	2	740		...	20 ^b	After C/D, before and after HPV
	35	2	700		...	20 ^b	Before and after C/D
D (March 27, 2009–April 23, 2010)	134	3	2,680		...	20 ^c	After C/D followed by HPV

NOTE. C/D, cleaning and disinfection; HPV, hydrogen peroxide vapor.

^a Disinfection with bleach; 2 × C/D, 2 complete rounds; 4 × C/D, 4 complete rounds.

^b 20 paired sites/room.

^c 20 sites/room.

likelihood of patient contact (eg, beds, wheelchairs, pillows), as well as from sites with a lower likelihood of direct patient contact but a high likelihood of contact with hands of personnel (eg, interior of cabinets and medication drawers; table 2). Medication drawers and cabinet interiors had a greater than 3% contamination rate for both ABC and MRSA. Cabinet interiors were more likely to grow ABC than cabinet exteriors (6.3% vs 1.3%; odds ratio [OR], 5.2 [95% confidence interval {CI}, 1.0–27.6]).

Isolation of ABC and MRSA from Hospital Rooms before and after 1 Round of C/D

During period C (table 1), 12 previously ABC-positive room sites in 8 precleaned rooms became culture negative, while 3 ABC-negative sites in 3 rooms became culture positive. Overall, there was a significant reduction in the number of ABC-positive room sites (OR, 0.25 [95% CI, 0.045–0.93]; $P = .04$) but not rooms ($P = .23$). For MRSA, 10 culture-positive room sites in 5 precleaned rooms became culture negative, while 6 culture-negative room sites in 4 rooms became culture positive. Overall, there was no significant reduction in the number of MRSA-positive room sites or rooms ($P = .45$ and $P = 1.0$, respectively).

Isolation of ABC and MRSA from Hospital Rooms Following 1 Round of C/D before and after Hydrogen Peroxide Treatment

Six previously ABC-positive sites in 5 rooms became culture negative following HPV treatment; no previously culture-negative room sites grew ABC. Overall, there was a significant reduction in the number of ABC-positive room sites (OR, 0 [95% CI, 0–0.8]; $P = .04$) and near significant reduction in the number of ABC-positive rooms following HPV treatment (OR, 0 [95% CI, 0–1.09]; $P = .07$). For MRSA, 6 culture-positive sites in 4 rooms became culture negative; no previously culture-negative room sites grew MRSA. Overall, there was a significant reduction in the number of MRSA-positive room sites (OR, 0 [95% CI, 0–0.85]; $P = .04$), but not in the number of MRSA-positive rooms (OR, 0 [95% CI, 0–1.5]; $P = .13$), following HPV treatment.

Isolation of ABC and MRSA from Rooms Following 1 Round of C/D Followed by HPV Treatment

During period D (table 1), 5 (0.19%) sites grew ABC in 5 (3.7%) rooms, while 2 (0.75%) sites grew MRSA in 2 (1.5%) rooms. Overall, 6 (4.5%) rooms were culture positive for ABC, MRSA, or both. ABC-positive room surfaces consisted of a curtain, cables, bed, in-room computer, sink, and supply drawer, while MRSA-positive sites included a patient call light and a bed. All 3 of the ABC room isolates tested were MDRABC. All culture-positive rooms had negative subsequent cultures for ABC and MRSA following a repeat round of C/D and HPV treatment.

Relatedness of Environmental and Clinical ABC Isolates

Nineteen ABC isolates from surfaces of rooms vacated by MDRABC-positive patients underwent susceptibility and PFGE testing; all were confirmed MDRABC, with 12 (63%) having >95% similarity index as those of 17 unique clinical isolates (8 blood, 6 wound, and 3 perianal isolates) from patients occupying the same room or ward during the same period.

DISCUSSION

To the best of our knowledge, this is the largest study to date to systematically investigate the frequency of isolation of ABC and MRSA from hospital rooms newly vacated by MDRABC-positive patients following terminal cleaning. We found that elimination of ABC and MRSA from hospital room surfaces is often challenging, with approximately 1 of 4 rooms remaining contaminated with either one of these organisms even after 4 rounds of C/D with bleach. The relatively high number of room sites cultured undoubtedly increased our ability to identify contaminated rooms. Nevertheless, because we did not culture all potential room sites and did not use the enrichment broth technique,^{11,13} it is likely that our results still underestimated the actual rate of persistently contaminated rooms. It is noteworthy that our findings were not limited to the burn unit (ie, outbreak setting), where extensive environmental contamination with ABC or MRSA may be expected.^{14,15}

We hypothesize that the failure to eliminate ABC and

TABLE 2. Isolation of *Acinetobacter baumannii* Complex (ABC) and Methicillin-Resistant *Staphylococcus aureus* (MRSA) from 312 Rooms Newly Vacated by Patients Colonized and/or Infected with Multidrug-Resistant ABC by Room Site Following 4 Rounds of Terminal Cleaning and Disinfection with Bleach

Room site	Total	ABC positive, no. (%)	MRSA positive, no. (%)
High likelihood of direct patient contact			
Bedside table	402	6 (1.5)	10 (2.5)
Chair/recliner	336	15 (4.5)	20 (6)
Television	279	2 (0.7)	4 (1.4)
Paper towel dispenser	258	1 (0.4)	2 (0.8)
Sink/faucet	240	0	2 (0.8)
Door (room entrance)	238	2 (0.8)	7 (2.9)
Light switch	211	4 (1.9)	0
Bed (including rails)	198	12 (6)	9 (4.5)
Blood pressure equipment	187	1 (0.5)	1 (0.5)
Telephone	171	2 (1.2)	3 (1.8)
Television remote control	148	3 (2.0)	4 (2.7)
Mattress	123	2 (1.6)	1 (0.8)
VCR	97	0	4 (4.1)
Dresser	61	2 (3.3)	1 (1.6)
Glucose meter	56	2 (3.6)	1 (1.8)
Radio/CD player	50	0	1 (2)
Pillow	45	6 (13)	3 (6.7)
Thermometer	44	0	1 (2.7)
Lift	20	0	1 (5)
Wheel chair	19	3 (15.8)	2 (10.5)
Door (bathroom)	13	1 (7.7)	0
Gait belt	4	2 (50)	0
Low likelihood of direct patient contact			
In-room computer	291	9 (3.1)	3 (1)
Suction equipment	226	1 (0.4)	2 (0.8)
Thermostat	219	0	1 (0.5)
Oxygen flow meter	212	7 (3.3)	3 (1.4)
Cardiac monitor	194	6 (3.1)	3 (1.5)
Cabinet (exterior)	157	2 (1.3)	2 (1.3)
Countertop	155	0	5 (3.2)
Closet	82	1 (1.2)	0
Cabinet (interior)	79	5 (6.3)	3 (3.8)
Curtain	52	2 (3.8)	0
Dressing cart	51	3 (5.9)	1 (2)
Contaminated needle container	51	0	1 (2)
Enteral feeding pump	46	1 (2.1)	0
Compression device pump	29	0	1 (3.4)
IV pump	29	0	1 (3.4)
Medication drawer	26	2 (7.7)	1 (3.8)
Window sill	26	0	2 (7.7)
Pill crusher	11	0	1 (9.1)

NOTE. Total number of sites cultured from each room often varied because of variation in the mix of items present in the room at the time cultures were obtained. Bulletin board ($n = 32$), fan ($n = 47$), soap dispenser ($n = 211$), and alcohol rub dispenser ($n = 17$) yielded no positive cultures for ABC or MRSA.

MRSA from hospital rooms was due to suboptimal environmental cleaning of hospital rooms as found by others¹⁶ rather than the failure of the disinfectants per se to destroy these pathogens. Of interest, the need for multiple rounds of C/D to render hospital rooms free of another common pathogen,

vancomycin-resistant *Enterococcus faecium*, has also been reported.¹⁷ Furthermore, we found that contaminated room sites involved not only those highly likely to be in contact with the patient (eg, chairs, pillows) but also those likely to be in contact with personnel hands only (eg, medication

drawers and cabinet interiors), underscoring the importance of thorough cleaning of all high-touch items and equipment.

We found that 1 round of C/D significantly reduced the number of ABC- but not MRSA-positive room sites. However, the addition of HPV treatment was associated with a significant reduction in number of ABC- and MRSA-positive sites; the efficacy of HPV treatment in reducing MRSA contamination of hospital rooms has been previously reported.¹⁰ Of note, several culture-negative room sites became culture positive, particularly in the case of MRSA, following 1 round of C/D. Although the variation in the size of sampled surfaces may be a possible reason for this finding, recontamination of previously culture-negative sites during the C/D process itself might be another explanation. Supporting the latter hypothesis is the finding that no previously culture-negative room site became culture positive after HPV treatment, which exposes all room surfaces to its antibacterial activity simultaneously, eliminating the possibility of recontamination of previously culture-negative sites. Our longitudinal study of 134 rooms (nearly one-half of which were vacated by patients concurrently colonized and/or infected with MRSA) further confirmed the efficacy of 1 round of C/D followed by HPV by reducing the number of ABC- or MRSA-positive rooms to less than 5%.

Several limitations of our study are worthy of discussion. First, because of their frequent MDR nature,¹⁸ ABC isolates from room surfaces underwent susceptibility testing only periodically. Nevertheless, the relatedness of many environmental and clinical isolates was inferred on the basis of the PFGE studies and MDR susceptibility pattern of the environmental isolates tested. Second, our results were based on the experience at a single institution and may not necessarily be generalizable to healthcare facilities with a different patient mix or terminal cleaning practices. Last, although HPV treatment appeared effective in reducing the number of contaminated room sites, the additional cost and time incurred to complete 1 treatment cycle may limit its widespread adoption.

We conclude that even when performed up to 4 rounds, routine terminal C/D with bleach of hospital rooms vacated by MDRABC-positive patients may not eliminate ABC or MRSA from many inanimate surfaces. HPV treatment of rooms following 1 round of C/D appears effective in reducing the number of ABC- or MRSA-contaminated room sites and may obviate the need for multiple rounds of C/D.

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