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## Major article

## Are community environmental surfaces near hospitals reservoirs for gram-negative nosocomial pathogens?

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## Key Words:

Acinetobacter

Environmental contamination

**Background:** Hospital visitors and staff visit neighboring businesses, creating the potential for contamination of surfaces with hospital flora.

**Methods:** Cultures were obtained from environmental surfaces in hospital lobbies and the surrounding community of 6 hospitals in Brooklyn, NY. As a control, cultures were taken from surfaces >1.5 miles from any hospital. Screening for  $\beta$ -lactamases was done by polymerase chain reaction (PCR), and select isolates were fingerprinted by the repetitive extragenic palindromic sequence-PCR method.

**Results:** Of 493 cultures, most (70%) involved doors from local businesses. Cephalosporin-resistant *Citrobacter freundii* (n = 3), *Escherichia coli* (n = 2), and *Enterobacter* sp (n = 2) were recovered from surfaces near hospitals, but not from control sites. One isolate of *Stenotrophomonas maltophilia* harbored an integron-associated VIM-2. *Acinetobacter baumannii* was recovered in 15 samples, including 4.5% of swabs from  $\leq 0.5$  miles of the hospitals versus 0% from  $\geq 0.6$  miles ( $P = .004$ ). Eleven *A baumannii* isolates were clonally related by repetitive extragenic palindromic sequence-PCR and were also related to a known clinical isolate.

**Conclusions:** Strains of *A baumannii* and cephalosporin-resistant Enterobacteriaceae can be recovered from environmental surfaces surrounding hospitals. Finding these pathogens in the perihospital environment suggests hand cleansing should be emphasized for all people entering and leaving hospitals. The finding of integron-associated VIM-2 in our region is disconcerting, and further vigilance is warranted.

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Contaminated inanimate objects within the hospital setting can facilitate transmission of certain nosocomial pathogens. Contamination of environmental surfaces with *Staphylococcus aureus*, *Clostridium difficile*, vancomycin-resistant enterococci, and *Acinetobacter* sp have been well documented and contribute to sustained outbreaks.<sup>1,2</sup> Even with adherence to hand hygiene practices and environmental cleaning protocols, eradication of these pathogens from hospitals has been proven difficult.

Relatively little is known about contamination of environmental surfaces outside hospital departments and in the immediate vicinity of medical centers. In New Delhi, extensive environmental contamination with NDM-1 carrying bacteria has been documented, with 30% of city samples positive for this resistance gene.<sup>3</sup> For more than a decade, hospitals in the New York City region have been plagued by multidrug-resistant *Pseudomonas aeruginosa*,

*Acinetobacter baumannii*, and KPC-possessing *Klebsiella pneumoniae*.<sup>4</sup> In this report, we examine the degree of contamination with gram-negative bacilli of environmental surfaces in the communities surrounding medical centers in Brooklyn, NY.

## METHODS

Cultures were obtained from surfaces from hospital lobbies and the surrounding (<1 mile radius) community. Environmental samples from six Brooklyn hospitals were included in our study. As a control, environmental samples were obtained from a region in Brooklyn that is >1.5 miles from the nearest hospital. Environmental samples were obtained by swabbing a premoistened (in sterile saline) sterile calcium alginate swab. Preference was given to surfaces in frequent contact with human hands (eg, door handles and railings). The swab was immediately placed into a nonselective medium (Mueller-Hinton broth), and incubated overnight at 37°C. Sterility controls were included in all experiments. A sample (50  $\mu$ L) of the overnight culture was then streaked

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onto a selective medium (MacConkey agar with 2 µg/mL ceftazidime). All colonies growing on the selective medium were then identified using the API 20E and 20NE panels (bioMérieux, Inc, Durham, NC). Minimal inhibitory concentrations (MICs) of ceftazidime and imipenem were determined by the agar dilution method using Mueller-Hinton agar, according to established standards.<sup>5</sup> *P aeruginosa* ATCC 27853 was included as the control strain. Samples of the initial overnight nonselective broth, and all isolates recovered from the MacConkey selective plates, were screened by polymerase chain reaction for genes encoding the following β-lactamases: *bla*<sub>SHV</sub>, *bla*<sub>TEM</sub>, *bla*<sub>OXA-23-like</sub>, *bla*<sub>OXA-24/40-like</sub>, *bla*<sub>OXA-48</sub>, *bla*<sub>OXA-58</sub>, *bla*<sub>IMP</sub>, *bla*<sub>VIM</sub>, and *bla*<sub>KPC</sub>.<sup>6–9</sup> In addition, screening for *bla*<sub>NDM-1</sub> was performed using the following primers: NDMfor 5'-GGAACTGGCGACCAACG-3' and NDMrev 5'-ATGCGGGCCGTATGAGTGA-3'.<sup>10</sup> Positive controls were included for *bla*<sub>SHV</sub>, *bla*<sub>TEM</sub>, *bla*<sub>OXA-23-like</sub>, *bla*<sub>OXA-24/40-like</sub>, *bla*<sub>IMP</sub>, *bla*<sub>VIM</sub>, *bla*<sub>KPC</sub>, and *bla*<sub>NDM-1</sub>. For 1 isolate, the partial sequence of a type 1 integron was mapped using the forward primer for type 1 integrons and a reverse primer for *aac6'-1b*.<sup>11,12</sup> Genetic relatedness of select isolates was determined by the repetitive extragenic palindromic sequence-polymerase chain reaction method using ERIC-2 primers and Ready-To-Go RAPD Analysis Beads (GE Healthcare Biosciences, Piscataway, NJ), as previously described.<sup>4</sup> Isolates with a >1 band difference were considered unrelated strains. Statistical analysis was performed using Fisher exact test, with a 2-tailed *P* value < .05 being considered significant.

## RESULTS

A total of 493 environmental cultures were obtained. Of the cultures involving the regions around (<1 mile radius) 6 medical centers, 95 were obtained from hospital A (including 65 from doors and 9 from hand railings), 99 from hospital B (66 from doors and 7 from hand railings), 90 from hospital C (65 from doors and 9 from hand railings), 73 from hospital D (56 from doors and 1 from a hand railing), and 86 from hospital complex E-F (55 from doors, 11 from hand railings). As a control, 50 cultures (including 31 doors and 4 hand railings) were obtained from a region in Brooklyn with the nearest hospital outside a 1.5-mile radius.

A total of 70 samples yielded gram-negative bacilli. Most were bacteria of modest clinical importance, including *Pseudomonas fluorescens* (n = 16), *Burkholderia cepacia* (n = 15), and *Alcaligenes faecalis* (n = 14). Three isolates of *Citrobacter freundii* (1 each from a hospital lobby bathroom, satellite outpatient clinic bathroom, and standing water in a street) and 2 isolates of *Escherichia coli* (both from standing water in street) were recovered. One isolate each of *Enterobacter cloacae* and *Pseudomonas putida* (both from standing street water), and 1 isolate of *Pantoea agglomerans* (from a door to a deli), were also identified. All but the *Pantoea agglomerans* were highly resistant to ceftazidime (MIC ≥32 µg/mL). Two isolates of *Stenotrophomonas maltophilia* were recovered; 1 isolate was from a hospital lobby bathroom from hospital complex E-F and found to harbor *bla*<sub>VIM-2</sub>. This metallo-β-lactamase was flanked by sequences encoding a type 1 integron and an *aac6'-1b*-type aminoglycoside-modifying enzyme (GenBank accession KF471098). None of the other isolates were found to harbor any of the screened β-lactamases.

Fifteen isolates of *A baumannii* were recovered from the environmental samples (Table 1). Ten were recovered from doors located in the community surrounding the hospitals. *A baumannii* was recovered from 15 of 336 (4.5%) cultures from surfaces within a 0.5-mile radius from the hospitals, compared with none of the 157 cultures from a distance >0.5 miles (*P* = .004). Eleven of the isolates were from the community around 1 hospital. All had ceftazidime MICs ≥4 µg/mL, and 1 isolate was carbapenem-resistant. Genetic fingerprinting demonstrated 11 of the isolates were closely related,

**Table 1**

Sources, susceptibilities, and repetitive extragenic palindromic sequence-polymerase chain reaction fingerprints of isolates of *Acinetobacter baumannii* recovered from environmental surfaces

Hospital	Isolate	Source	Ceftazidime	Imipenem	Random group
			MIC (µg/mL)		
A	30	Restaurant door	8	0.25	3
B	26	Emergency room bathroom	16	0.5	3
B	50	Internet café bathroom	8	0.5	2a
B	60	Clinic door	4	0.25	2a
B	64	Bakery door	4	0.25	2a
B	66	Diner door	4	0.25	2a
B	67	Pizza parlor door	16	0.5	2a
B	68	Donut shop door	4	0.25	2a
B	71	Subway door	4	0.25	2b
B	76	Subway hand railing	4	0.25	2b
B	78	Internet café door	4	0.25	2c
B	89	Deli door	4	0.25	2b
C	57	Sidewalk water	16	2	4
E-F	12	Restaurant bathroom	>32	>16	1
E-F	61	Grocery door	8	0.25	2a

MIC, minimal inhibitory concentration.

and similar to a clinical isolate recovered from 1 of the participating hospitals (data not shown). However, none of the environmental isolates belonged to the previously recognized multidrug-resistant strains endemic to our region.

## DISCUSSION

There are 2 noteworthy observations regarding our investigation of community environmental surfaces surrounding the hospitals in Brooklyn, NY. The first is the finding of a single strain of *A baumannii* that contaminated community surfaces, especially around 1 of our area hospitals. A clinical isolate was also identified as this strain, suggesting transmission between the community and hospital. This strain did not appear related to multidrug-resistant isolates previously identified in our region. *A baumannii* is well equipped to survive on environmental surfaces; survival from 26 days to 4 months has been described on dry surfaces.<sup>13,14</sup> Contamination of inanimate surfaces, particularly in intensive care areas, commonly occurs in rooms occupied by patients infected or colonized with *A baumannii*. Admission to a hospital room previously occupied by a patient with *A baumannii* has been identified as a risk factor for infection or colonization, suggesting the environment plays an important role in transmission of this pathogen.<sup>15</sup> Approximately 39%–48% of rooms occupied by a patient with *A baumannii* have at least 1 positive environmental culture, typically with the same strain as that recovered from the patient.<sup>16–19</sup> Even with aggressive environmental cleaning protocols with recommended disinfectants, as many as one-fourth of rooms remain culture positive.<sup>20,21</sup>

Because as many of 29% of hands of health care workers in intensive care areas can be colonized,<sup>17</sup> it is certainly plausible that carriage of this pathogen can occur in and out of the hospital and contaminate environmental surfaces in the community. Given the dismal failure in controlling the spread of *A baumannii* in most medical centers in Brooklyn,<sup>4</sup> further investigation of the roles of environmental surfaces both within and outside the hospital should be undertaken. An emphasis on hand hygiene for all persons entering and leaving hospital units should also be considered.

Second, the finding of an isolate with an integron-associated *bla*<sub>VIM-2</sub> is disconcerting. Sporadic cases have occurred in the United States,<sup>22,23</sup> but to our knowledge, this is the first report of this enzyme in New York City. This metallo-β-lactamase is frequently found in *P aeruginosa*,<sup>24</sup> but spread to Enterobacteriaceae has also been documented.<sup>25</sup> Given the fact that

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