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Brief Report

The relevance of sink proximity to toilets on the detection of *Klebsiella pneumoniae* carbapenemase inside sink drains

Blake W. Buchan PhD, D(ABMM) [a,*], Mary Beth Graham MD [b], Jill Lindmair-Snell RN, MSN, CIC [d], Jennifer Arvan BSN, RN [d], Nathan A. Ledeboer PhD, D(ABMM) [a], Rahul Nanchal MD [c], L. Silvia Munoz-Price MD, PhD [b]

[a] *Department, of Pathology, The Medical College of Wisconsin, Milwaukee, WI*
[b] *Division of Infectious Diseases, The Medical College of Wisconsin, Milwaukee, WI*
[c] *Division of Pulmonary and Critical Care Medicine, The Medical College of Wisconsin, Milwaukee, WI*
[d] *Froedtert Memorial Lutheran Hospital, Milwaukee, WI*

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We report a higher prevalence of bla_{KPC} in patient room sink drains located next to toilets (87.0%) when compared with sink drains located farther away from toilets (21.7%) using direct polymerase chain reaction assay. However, culture methods were only able to recover bla_{KPC}-positive isolates from 16% of polymerase chain reaction–positive drains.

BACKGROUND

Recent findings demonstrate that sink drains can be highly contaminated with carbapenemase-producing organisms,[1,2] potentially splashing to adjacent counter surfaces and patient care equipment.[3,4] The duration of colonization or persistence of these organisms in sink drains is unknown. We conducted a surveillance study of sink drains focusing on a medical intensive care unit (MICU) with no documented infections with *Klebsiella pneumoniae* carbapenemase (KPC)–producing organism in the past year to determine the prevalence of bla_{KPC} and KPC-producing organisms in sink drains. We present the results of molecular and culture-based tests used for MICU sink surveillance and compare findings based on the proximity of the sinks to toilets.

METHODS

Specimen collection

This study was performed in a 600-bed hospital in Milwaukee, Wisconsin. The MICU is a 26-bed unit, in which each room has a patient bed, 2 sinks, and a toilet without physical barriers in between (Fig 1). Each room is surface-cleaned (including sink bowls but not drains) with hydrogen peroxide/peracetic acid on a daily basis. Separate cloths are used to clean each of the 2 sinks in patient rooms to avoid cross-contamination.

Two swab types were used to collect specimens from each sink drain: a wound Dacron swab (BBL Dual CultureSwab; Becton Dickinson, Sparks, MD) and ESwab (Copan, Brescia, Italy). Swabs were inserted into the sink drain and rotated to collect specimen from the inner walls for a minimum of 3 insertions or until the swabs were visibly soiled. Laboratory testing was initiated within 4 hours of collection.

Molecular detection of bla_{KPC}

All specimens were tested for the presence of bla_{KPC} using the research use only Becton Dickinson MAX CRE test kit. One of 2 BBL Dual CultureSwabs was removed from the transport tube and broken off into a Becton Dickinson MAX Sample Buffer Tube (SBT). Similarly, 100 μL of ESwab broth was transferred into a Becton Dickinson MAX SBT. SBTs were vortexed and analyzed with the Becton Dickinson MAX System.

Culture methods

The remaining BBL Dual CultureSwab and 100μL of ESwab medium were used in parallel to directly inoculate CHROM CRE chromogenic agar plates (Hardy, Santa Maria, CA). Cultures were

* Address correspondence to Blake W. Buchan, The Medical College of Wisconsin, Department of Pathology, 9200 W Wisconsin Ave, Milwaukee, WI 53226.
E-mail address: bbuchan@mcw.edu (B.W. Buchan).
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Fig 1. Image illustrating configuration of the medical intensive care unit room surveyed in this study. The location of the toilet, sink adjacent to the toilet, and sink far from the toilet are labeled.

incubated 18-24 hours at 35°C before examination for bacterial growth and colonies characteristic of Enterobacteriaceae. All characteristically colored colonies were isolated to blood agar to obtain pure cultures. Isolates were identified with MALDI-ToF MS (Bruker Daltonik, Bremen, Germany) and Etest (bioMerieux, Marcy l' Etoile, France) used for susceptibilities. All isolates were tested for the presence of bla_{KPC} using a real-time polymerase chain reaction (PCR) assay.[5] Difference in proportions of bla_{KPC} positivity were determined with χ^2 tests.

RESULTS

The bla_{KPC} gene was detected in 25 of 46 (54.3%) of sink drain specimens tested directly by PCR (Table 1). Direct PCR was positive in 20 of 23 (87.0%) sinks located near the toilet compared with only 5 of 23 (21.7%) sinks located near the room entry door ($P < .00001$; Table 1). In 4 of 5 (80.0%) rooms with a positive testing entry door sink, the sink near the toilet was also positive, suggesting a potential source for cross-contamination within the same room. Both ESwab and wound fiber swab specimens tested positive for 17 of 25 (68.0%) sink drains. Among the remaining 8 sink drains, 5 (62.5%) were positive by wound fiber swab alone, and 3 (37.5%) were positive by ESwab alone.

Bacterial culture recovered bla_{KPC}-positive isolates from 3 of 20 (15.0%) sinks near the toilet and 1 of 5 (20.0%) entry door sinks. All positive cultures were obtained with the ESwab device only. Isolates recovered included *K pneumoniae*, *Enterobacter cloacae*, *Raoultella planticola*, *Raoultella ornitinolytica*, and *Aeromonas hydrophila*. All isolates displayed nonsusceptible MICs to either ertapenem, meropenem, or both (Table 1).

The majority of sink drains yielded bacterial growth on the chromogenic culture media; however, most colonies did not demonstrate coloration consistent with carbapenem-resistant Enterobacteriaceae and were identified as *Pseudomonas aeruginosa*. A number of isolates did demonstrate coloration consistent with carbapenem-resistant Enterobacteriaceae, including *Elizabethkingia meningoseptica*, *Stenotrophomonas maltophilia*, *Comamonas sp*, and *Acinetobacter sp*. These species often harbor intrinsic or acquired resistance to β-lactam antibiotics, including carbapenems, but rarely carry bla_{KPC}. To investigate whether any of these non-Enterobacteriaceae were harboring bla_{KPC}, "sweep" PCR was conducted on 4 culture plates. All 4 reactions were negative for bla_{KPC}.

DISCUSSION

We found a high prevalence of KPC positivity in sink drains, especially next to toilets, when interrogated using a direct PCR method. Culture for KPC-producing organisms was low yield, with a recovery rate of only 16% (4/25) among PCR-positive drains. Both swab types used for specimen collection demonstrated similar yield when used for direct PCR; however, KPC-producing isolates were only recovered from specimens collected with ESwab, suggesting that this might be a better tool for collection, which is consistent with previous reports.[6-8]

To date, no studies have examined the relevance of sink proximity to toilets in patient rooms. Based on recent data by Mathers and colleagues,[3] it could be plausible that contamination of sinks next to toilets is occurring through biofilms growing in communal pipes between toilets and sinks. Alternatively, toilets are known to generate contaminated droplets during flushing; thus contamination of sink drains via droplets would not be unreasonable. Finally, the seeding of sinks may result from independent events such as the conduct of routine hand hygiene by patients or health care workers. Further studies examining the genetic clonality of bacterial isolates and plasmids are under way to test these hypotheses.

The low yield of culture recovery of KPC-producing organisms from sinks testing positive for bla_{KPC} by direct PCR is puzzling and could be the result of (1) poor sampling and collection technique, (2) suboptimal culture methods, (3) presence of bla_{KPC} in

Table 1
Direct PCR and culture results for 46 sinks surveyed

Room no.	Sink near toilet (direct PCR)		Sink near entry door (direct PCR)		
	Wound swab	ESwab	Wound swab	ESwab	Culture result*
1	Positive	Positive	Positive	—	*K pneumoniae:* Erta >32, Mero >32, bla_{KPC} positive *E. cloacae*: Erta 8, Mero 8, bla_{KPC} positive
2	Positive	Positive	—	—	
3	Positive	Positive	—	—	
4	Positive	Positive	—	—	
5	Positive	Positive	—	—	
6	Positive	Positive	—	—	
7	Positive	Positive	—	—	
8	Positive	Positive	—	—	
9	Positive	—	—	—	
10	Positive	Positive	—	—	
11	Positive	Positive	—	—	
12	—	Positive	—	—	
13	—	—	—	—	
14	Positive	Positive	—	—	*R. planticola*: Erta 1, Mero 1, bla_{KPC} positive
15	Positive	Positive	—	—	
16	Positive	Positive	Positive	Positive	
17	Positive	Positive	—	—	*K. pneumoniae*†: Erta 8, Mero 1, bla_{KPC} negative
18	Positive	—	—	—	
19	Positive	—	—	—	
20	Positive	Positive	Positive	—	*R ornithinolytica*: Erta 1, Mero 2, bla_{KPC} positive
21	—	—	Positive	Positive	*A. hydrophila*: Erta 2, Mero 1, bla_{KPC} positive
22	—	—	—	—	
23	—	Positive	—	Positive	

bla_{KPC}, result of bla_{KPC} PCR on isolate; *Erta*, ertapenem *MIC*; *Mero*, meropenem MIC; *PCR*, polymerase chain reaction.
*All positive cultures were obtained using ESwab. None of the specimens collected using wound fiber swab was culture positive.
†Isolate recovered from sink near entry door.

environmental bacteria uncommonly associated with KPC carriage, or (4) carriage of bla_{KPC} by viable but nonculturable organisms persisting as biofilm in sink drains.[9] The ultimate source of these genes and organisms is unknown and should be further explored by future studies. Of additional interest is the finding of multiple genera (*Klebsiella, Enterobacter, Raoultella, Aeromonas*) harboring bla_{KPC} in these sink drains. It is not clear whether this is the result of horizontal transfer of genetic material between genera or independent seeding events over time; however, 2 bla_{KPC}-harboring isolates were isolated from a single sink drain that provides the possibility of horizontal gene transfer.

In conclusion, a high prevalence of bla_{KPC} positivity was found in sink drains of a unit with no known recent history of KPC-producing organisms. The significance of these findings and the potential risk of transmission of KPC-producing organisms in absence of culturable bacterial strains is still unclear in terms of infection control. Our findings should be validated in other settings and institutions because the infection control implications are major. If sinks next to toilets are indeed a reservoir for bla_{KPC}, then additional interventions such as modified hand hygiene practices (eg, dedicated sinks), optimization of sink disinfection protocols (eg, increased frequency, optimal disinfectants), and use of engineering controls (eg, splash shields) may be needed to further mitigate the risk of transmission of KPC-producing organisms among health care providers and patients.

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