

Contaminated Handwashing Sinks as the Source of a Clonal Outbreak of KPC-2-Producing *Klebsiella oxytoca* on a Hematology Ward

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We investigated sinks as possible sources of a prolonged *Klebsiella pneumoniae* carbapenemase (KPC)-producing *Klebsiella oxytoca* outbreak. Seven carbapenem-resistant *K. oxytoca* isolates were identified in sink drains in 4 patient rooms and in the medication room. Investigations for resistance genes and genetic relatedness of patient and environmental isolates revealed that all the isolates harbored the *bla*_{KPC-2} and *bla*_{TEM-1} genes and were genetically indistinguishable. We describe here a clonal outbreak caused by KPC-2-producing *K. oxytoca*, and handwashing sinks were a possible reservoir.

The prevalence of carbapenem-resistant *Enterobacteriaceae* has increased over the past 10 years (1). The production of *Klebsiella pneumoniae* carbapenemases (KPCs) is one way of conferring resistance to carbapenems belonging to Ambler class A (2). Nosocomial infections due to KPC-producing *Enterobacteriaceae* are of increasing concern, especially in long-term care facilities and intensive care units (ICUs) (2). In 2011, a nosocomial outbreak of KPC-producing *Klebsiella oxytoca* occurred in a medical ICU at the Medical University of Graz in Graz, Austria. Five patients, all of whom survived, were affected (3).

From October 2011 to October 2013, KPC-producing *K. oxytoca* isolates with resistance patterns identical to that of the outbreak strain were identified from 10 more patients in the Division of Hematology ward. As *K. oxytoca* isolates were detected over a 2-year period, a reservoir in the environment was suspected. Nosocomial outbreaks with *K. oxytoca* have been reported previously (4–10). In two publications, handwashing sinks, especially sink drains, were identified as the source of the outbreaks (9, 10).

The aim of this study was to investigate sink drains as possible sources of the KPC-producing *K. oxytoca* isolates, the genetic relatedness of patient and environmental isolates, and the underlying mechanism of carbapenem resistance.

The outbreak occurred at the Division of Hematology at the Medical University of Graz, a tertiary care facility. This 28-bed ward has approximately 1,200 admissions annually, almost exclusively of patients with hematological malignancies. The index patient involved in the outbreak of KPC-producing *K. oxytoca* was in the medical ICU in December 2010 (3). The patient was considered colonized but not infected according to CDC criteria (11). The patient was then transferred to the hematology ward, where contact precautions were continued.

KPC-producing *K. oxytoca* isolates with resistance patterns identical to those of the outbreak strain were identified from 10 more patients in the Division of Hematology. Three patients were identified in late 2011 and 2012, and seven were detected from January to July 2013. Six patients developed an infection (Table 1). All the patients were admitted more than once during the outbreak period either for chemotherapy or because of side effects during chemotherapy.

Screening swabs from dry surfaces are taken and tested routinely 4 times per year by our infection control team and have never yielded *K. oxytoca*. In addition, in two publications dealing with nosocomial outbreaks of *K. oxytoca*, swabs from dry surfaces in the environment surrounding the patients did not yield any *K. oxytoca* isolates. In contrast, *K. oxytoca* was identified in sinks and drains (9, 10). Therefore, we chose to focus our investigation on sinks. Fifty-eight swabs were taken from handwashing sink drains, 23 from handwashing sink overflows (not all sinks had overflows), and 19 from shower drains in the Division of Hematology. Swabs from sink surfaces were taken from contaminated sinks in a second round of testing.

Swabs were seeded onto MacConkey agar and chromID Carba (both from bioMérieux, Marcy l'Étoile, France) and analyzed according to microbiological standards (12). The isolates were screened for the presence of resistance genes with the DNA microarray-based Check-MDR 103 kit (Check-Points, Wageningen, Netherlands) according to the manufacturer's protocol (<http://www.check-points.com/support/manuals/>). Sequencing of the detected carbapenemases and other beta-lactamase gene families was performed as previously described (13, 14). Repetitive sequence-based PCR (rep-PCR) was carried out with the DiversiLab system (bioMérieux, Nürtingen, Germany). In addition, all the isolates were tested using multilocus sequence typing (MLST), as described previously (15).

Eleven *K. oxytoca* isolates were found in the sink drains. Seven

Received 15 September 2014 Returned for modification 13 October 2014
Accepted 23 October 2014

Accepted manuscript posted online 27 October 2014

Citation Leitner E, Zarfel G, Luxner J, Herzog K, Pekard-Amenitsch S, Hoenigl M, Valentin T, Feierl G, Grisold AJ, Högenauer C, Sill H, Krause R, Zollner-Schwetz I. 2015. Contaminated handwashing sinks as the source of a clonal outbreak of KPC-2-producing *Klebsiella oxytoca* on a hematology ward. *Antimicrob Agents Chemother* 59:714–716. doi:10.1128/AAC.04306-14.

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doi:10.1128/AAC.04306-14

isolates showed multidrug resistance, including resistance to carbapenems, one was an extended-spectrum beta-lactamase (ESBL)-producing *K. oxytoca* isolate, and three isolates showed wild-type resistance. The 7 KPC-producing *K. oxytoca* isolates were found in rooms 23, 25, and 29 (double rooms housing 2 patients each with two sinks), room 37 (a single room), and 32A (a room in which medication is prepared by the nursing staff). Five KPC-producing *K. oxytoca* isolates were detected in sink drains (in rooms 23, 25, 29, 37, and 32A). One isolate was found in the sink overflow (room 37), and one was found in the shower drain (room 25). Swabs taken from sink surfaces in a second round did not yield any additional KPC-producing *K. oxytoca* isolates.

All the isolates from the patients and the seven isolates derived from sinks were multidrug resistant, exhibited susceptibility to amikacin, colistin, and fosfomycin only, and were found to be KPC producers. When microarray technology was used, *bla*_{KPC} and *bla*_{TEM} were detected in all the isolates and were identified as *bla*_{KPC-2} and *bla*_{TEM-1} by sequencing. rep-PCR revealed that all strains tested were indistinguishable with a similarity index of >97.5%. MLST yielded only one sequence type, ST4, for all the KPC-2-producing *K. oxytoca* isolates.

The starting point for this outbreak was a colonized patient from the ICU who later was transferred to the hematology ward. We hypothesize that in the case of this patient, KPC-2-producing *K. oxytoca* got into the sink most likely during personal hygiene activities or by the disposal of contaminated body fluids where it persisted. In the Division of Hematology, the water from the sink faucets directly hits the mesh that covers the sink drain. We hypothesize that some patients were colonized by contaminated aerosols when using the sinks for personal hygiene; 6 of 10 affected patients stayed in rooms with contaminated sinks, and 2 patients shared a room with a patient who later proved to be infected or colonized. We speculate that cross-contamination took place either by direct contact between the patients or through the hands of health care workers. The potential involvement of the health care workers is indicated by the fact that KPC-2-producing *K. oxytoca* was found in the sink of the room where medication is prepared by the health care staff.

As a consequence of the prolonged outbreak, the existing infection control measures (isolating colonized patients, enforcing hand hygiene measures, and cleaning the ward, particularly the sinks and equipment) were reinforced. An exchange of all the colonized sinks is under way. Since October 2013, no more KPC-producing *K. oxytoca* isolates have been identified.

In conclusion, a clonal relationship between environmental isolates and patient strains was determined and pointed to hand-washing sinks as a possible reservoir for the prolonged nosocomial outbreak of KPC-2-producing *K. oxytoca*.

REFERENCES

- Nordmann P, Naas T, Poirel L. 2011. Global spread of carbapenemase-producing *Enterobacteriaceae*. *Emerg Infect Dis* 17:1791–1798. <http://dx.doi.org/10.3201/eid1710.110655>.
- Levy Hara G, Gould I, Endimiani A, Pardo PR, Daikos G, Hsueh PR, Mehtar S, Petrikos G, Casellas JM, Daciuk L, Paciel D, Novelli A, Saginur R, Pryluka D, Medina J, Savio E. 2013. Detection, treatment, and prevention of carbapenemase-producing *Enterobacteriaceae*: recommendations from an International Working Group. *J Chemother* 25:129–140. <http://dx.doi.org/10.1179/1973947812Y.0000000062>.
- Hoenigl M, Valentin T, Zarfel G, Wuerstl B, Leitner E, Salzer HJ, Posch J, Krause R, Grisold AJ. 2012. Nosocomial outbreak of *Klebsiella pneumoniae* carbapenemase-producing *Klebsiella oxytoca* in Austria. *Antimi-*

TABLE 1 Clinical data and epidemiological information

Patient no.	Patient age (yr) and gender ^a	Comorbidity	Date of first detection (day/mo/yr)	Site(s) of detection	Infection or colonization	Outcome	Patient stayed in a room with <i>K. oxytoca</i> in sink (room no.)
1	39, m	AML ^b	25/10/2011	Anal swab	Colonization	Alive	No
2	65, m	AML	28/12/2011	BAL ^c	Infection	Death due to KO ^d pneumonia	No
3	50, f	AML	13/11/2012	Blood, skin, stool, urine, abscess	Infection	Death due to KO abdominal wall abscess	Yes (23, 25)
4	51, f	AML	8/1/2013	Stool, BAL	Infection	Death due to AML relapse	No, but shared room with patient 5
5	60, f	AML	15/1/2013	Stool	Colonization	Alive	Yes (25)
6	61, m	B-cell non-Hodgkin lymphoma	9/4/2013	Anal swab, throat, BAL	Infection	Death due to KO pneumonia	Yes (25)
7	72, m	Myelodysplastic syndrome, secondary AML	14/05/2013	Stool	Colonization	Alive	Yes (23)
8	54, m	AML	17/5/2013	BAL, throat, anal swab	Infection	Death due to KO pneumonia	Yes (25)
9	89, m	B-cell non-Hodgkin lymphoma	31/7/2013	Catheter urine, throat	Colonization	Alive	No, but shared room with patient 8
10	56, m	AML	31/10/2013	Blood, throat, anal swab, skin, stool	Infection	Alive	Yes (25)

^a m, male; f, female.

^b AML, acute myeloid leukemia.

^c BAL, bronchoalveolar lavage.

^d KO, KPC-2-producing *K. oxytoca*.

- croB Agents Chemother 56:2158–2161. <http://dx.doi.org/10.1128/AAC.05440-11>.
4. Sardan YC, Zarakolu P, Altun B, Yildirim A, Yildirim G, Hascelik G, Uzun O. 2004. A cluster of nosocomial *Klebsiella oxytoca* bloodstream infections in a university hospital. *Infect Control Hosp Epidemiol* 25: 878–882. <http://dx.doi.org/10.1086/502313>.
 5. Jeong SH, Kim WM, Chang CL, Kim JM, Lee K, Chong Y, Hwang HY, Baek YW, Chung HK, Woo IG, Ku JY. 2001. Neonatal intensive care unit outbreak caused by a strain of *Klebsiella oxytoca* resistant to aztreonam due to overproduction of chromosomal beta-lactamase. *J Hosp Infect* 48:281–288. <http://dx.doi.org/10.1053/jhin.2001.1018>.
 6. Zarate MS, Gales AC, Picao RC, Pujol GS, Lanza A, Smayevsky J. 2008. Outbreak of OXY-2-producing *Klebsiella oxytoca* in a renal transplant unit. *J Clin Microbiol* 46:2099–2101. <http://dx.doi.org/10.1128/JCM.00194-08>.
 7. Schulz-Stubner S, Kniehl E. 2011. Transmission of extended-spectrum beta-lactamase *Klebsiella oxytoca* via the breathing circuit of a transport ventilator: root cause analysis and infection control recommendations. *Infect Control Hosp Epidemiol* 32:828–829. <http://dx.doi.org/10.1086/661225>.
 8. Ruiz E, Rezusta A, Saenz Y, Rocha-Gracia R, Vinue L, Vindel A, Villuendas C, Azanedo ML, Monforte ML, Revillo MJ, Torres C. 2011. New genetic environments of *aac(6′)-Ib-cr* gene in a multiresistant *Klebsiella oxytoca* strain causing an outbreak in a pediatric intensive care unit. *Diagn Microbiol Infect Dis* 69:236–238. <http://dx.doi.org/10.1016/j.diagmicrobio.2010.09.004>.
 9. Lowe C, Willey B, O’Shaughnessy A, Lee W, Lum M, Pike K, Larocque C, Dedier H, Dales L, Moore C, McGeer A, Mount Sinai Hospital Infection Control Team. 2012. Outbreak of extended-spectrum beta-lactamase-producing *Klebsiella oxytoca* infections associated with contaminated handwashing sinks. *Emerg Infect Dis* 18:1242–1247. <http://dx.doi.org/10.3201/eid1808.111268>.
 10. Vergara-Lopez S, Dominguez MC, Conejo MC, Pascual A, Rodriguez-Bano J. 2013. Wastewater drainage system as an occult reservoir in a protracted clonal outbreak due to metallo-beta-lactamase-producing *Klebsiella oxytoca*. *Clin Microbiol Infect* 19:E490–E498. <http://dx.doi.org/10.1111/1469-0691.12288>.
 11. Horan TC, Andrus M, Dudeck MA. 2008. CDC/NHSN surveillance definition of health care-associated infection and criteria for specific types of infections in the acute care setting. *Am J Infect Control* 36:309–332. <http://dx.doi.org/10.1016/j.ajic.2008.03.002>.
 12. Lee K, Lim YS, Yong D, Yum JH, Chong Y. 2003. Evaluation of the Hodge test and the imipenem-EDTA double-disk synergy test for differentiating metallo-beta-lactamase-producing isolates of *Pseudomonas* spp. and *Acinetobacter* spp. *J Clin Microbiol* 41:4623–4629. <http://dx.doi.org/10.1128/JCM.41.10.4623-4629.2003>.
 13. Bradford PA, Bratu S, Urban C, Visalli M, Mariano N, Landman D, Rahal JJ, Brooks S, Cebular S, Quale J. 2004. Emergence of carbapenem-resistant *Klebsiella* species possessing the class A carbapenem-hydrolyzing KPC-2 and inhibitor-resistant TEM-30 beta-lactamases in New York City. *Clin Infect Dis* 39:55–60. <http://dx.doi.org/10.1086/421495>.
 14. Zarfel G, Galler H, Feierl G, Haas D, Kittinger C, Leitner E, Grisold AJ, Mascher F, Posch J, Pertschy B, Marth E, Reinthaler FF. 2013. Comparison of extended-spectrum-beta-lactamase (ESBL) carrying *Escherichia coli* from sewage sludge and human urinary tract infection. *Environ Pollut* 173:192–199. <http://dx.doi.org/10.1016/j.envpol.2012.09.019>.
 15. Herzog KA, Schneditz G, Leitner E, Feierl G, Hoffmann KM, Zollner-Schwetz I, Krause R, Gorkiewicz G, Zechner EL, Hogenauer C. 2014. Genotypes of *Klebsiella oxytoca* isolates from patients with nosocomial pneumonia are distinct from those of isolates from patients with antibiotic-associated hemorrhagic colitis. *J Clin Microbiol* 52:1607–1616. <http://dx.doi.org/10.1128/JCM.03373-13>.