

Yiu-hong Leung, MBBS, FHKCCM;¹
 Monica Man-ha Wong, MBBS, FHKCCM;¹
 Shuk-kwan Chuang, MBBS, FHKCCM¹

Affiliations: 1. Field Epidemiology Training Programme, Centre for Health Protection, Department of Health, Hong Kong, China.

Address correspondence to Yiu-hong Leung, Room 325, 147C Argyle Street, Kowloon, Hong Kong, China (fansh@graduate.hku.hk).

Presented in part: Royal Australasian College of Medical Administrators/Hong Kong College of Community Medicine (RACMA/HKCCM) International Conference on Healthcare Reforms in Comparative Health Systems; Hong Kong; September 4–6, 2010.

Infect Control Hosp Epidemiol 2011;32(10):1048–1050

© 2011 by The Society for Healthcare Epidemiology of America. All rights reserved. 0899-823X/2011/3210-0017\$15.00. DOI: 10.1086/662019

REFERENCES

- DeLeo FR, Otto M, Kreiswirth BN, Chambers HF. Community-associated methicillin-resistant *Staphylococcus aureus*. *Lancet* 2010;375(9725):1557–1568.
- Centers for Disease Control and Prevention. Four pediatric deaths from community-acquired methicillin-resistant *Staphylococcus aureus*—Minnesota and North Dakota, 1997–1999. *MMWR Morb Mortal Wkly Rep* 1999;48(32):707–710.
- Gillet Y, Issartel B, Vanhems P, et al. Association between *Staphylococcus aureus* strains carrying gene for Panton-Valentine leukocidin and highly lethal necrotizing pneumonia in young immunocompetent patients. *Lancet* 2002;359(9308):753–759.
- Gonzalez BE, Martinez-Aguilar G, Hulten KG, et al. Severe staphylococcal sepsis in adolescents in the era of community-acquired methicillin-resistant *Staphylococcus aureus*. *Pediatrics* 2005;115(3):642–648.
- Miller LG, Perdreau-Remington F, Rieg G, et al. Necrotizing fasciitis caused by community-associated methicillin-resistant *Staphylococcus aureus* in Los Angeles. *N Engl J Med* 2005;352(14):1445–1453.
- Ammerlaan HS, Kluytmans JA, Wertheim HF, Nouwen JL, Bonten MJ. Eradication of methicillin-resistant *Staphylococcus aureus* carriage: a systematic review. *Clin Infect Dis* 2009;48(7):922–930.
- Ellis MW, Griffith ME, Dooley DP, et al. Targeted intranasal mupirocin to prevent colonization and infection by community-associated methicillin-resistant *Staphylococcus aureus* strains in soldiers: a cluster randomized controlled trial. *Antimicrob Agents Chemother* 2007;51(10):3591–3598.

confirmed as identical soon afterward.¹ To date, 9 other variants (KPC-3 to KPC-11) have been reported (<http://www.lahey.org/studies>). Among all of these carbapenemases, KPC-2, which had been discovered in many areas, was the predominant one and played a crucial role in carbapenem resistance of *K. pneumoniae*. We isolated a strain of *K. pneumoniae* carrying a novel KPC variant from an inpatient in the First Affiliated Hospital, College of Medicine, Zhejiang University, in the Hangzhou city of China.

An 81-year-old man with acute exacerbation of chronic obstructive pulmonary disease accompanied by gastrointestinal hemorrhage was admitted to our hospital on February 27, 2010. An exploratory laparotomy was performed on March 20, and colorectal polyps were found and removed. Three days later, abdominal drainage appeared, and a strain of *K. pneumoniae*, labeled zjm002, was isolated from the drainage fluid.

The isolate was identified by Vitek gram-negative identification cards (bioMérieux). Antimicrobial susceptibility tests of 9 antibiotics were performed by the microdilution method with cation-adjusted Mueller–Hinton broth (Oxoid) according to the recommendations of the Clinical and Laboratory Standards Institute (CLSI). The minimum inhibitory concentrations of 26 other antibiotics were determined by the Etest technique (bioMérieux) according to the manufacturer's instructions. The susceptibility breakpoints were interpreted as recommended by the CLSI and previous reports. The strain zjm002 was sensitive to amikacin, tigecycline, chloramphenicol, and trimethoprim-sulfamethoxazole and intermediate sensitive to tetracycline, but it was resistant to all other 27 antibiotics (Table 1).

A modified Hodge test (MHT), which might assist in confirming the presence of carbapenemase, was carried out according to Endimiani et al.³ As a result, the positivity of MHT for this isolate indicated that it carried carbapenemase. Since MHT could not exclusively detect the KPC-type carbapenemase, we detected *bla*_{KPC} and an additional 39 β -lactamases genes, including 13 class A carbapenemases genes (*bla*_{TEM}, *bla*_{SHV}, *bla*_{CTX-M1}, *bla*_{CTX-M2}, *bla*_{CTX-M8}, *bla*_{CTX-M9}, *bla*_{CTX-M25}, *bla*_{PER}, *bla*_{VEB}, *bla*_{GES}, *bla*_{CARB}, *bla*_{RTG}, *bla*_{LAF}), 10 class B carbapenemases genes (*bla*_{IMP}, *bla*_{VIM}, *bla*_{SIM}, *bla*_{SPM}, *bla*_{GIM}, *bla*_{AIM}, *bla*_{NDM}, *bla*_{KHM}, *bla*_{TMB}, *bla*_{DIM}), 8 class C carbapenemases genes (*bla*_{LEN}, *bla*_{OKP}, *bla*_{DHA}, *bla*_{ACT/MIR}, *bla*_{LAT/CMY}, *bla*_{MOX/CMY}, *bla*_{FOX}, *bla*_{ACC}), and 8 class D carbapenemases genes (*bla*_{OXA-1}, *bla*_{OXA-2}, *bla*_{OXA-10}, *bla*_{OXA-23}, *bla*_{OXA-24}, *bla*_{OXA-48}, *bla*_{OXA-51}, *bla*_{OXA-58}). Primer pairs for *bla*_{KPC} and *bla*_{TEM} polymerase chain reaction detection were 5'-ATGTCCTGATCGCCGTCTA-3' and 5'-TTACTGCCCCGTTGACGCCCAA-3' for *bla*_{KPC} and 5'-AGGAAGAGTATGATTCAACA-3' and 5'-CTCGTCGTTTGGTATGGC-3' for *bla*_{TEM}. The amplicons were sequenced on an ABI PRISM3730 sequencer analyzer (Applied Biosystems). As a result, *bla*_{KPC} and *bla*_{TEM} were detected, whereas the remaining 38 genes were not. The presence of *bla*_{TEM} confirmed as *bla*_{TEM-1} was determined by sequencing and Blast analysis. The amino acid sequence of KPC showed an amino acid change

Novel KPC Variant from a Carbapenem-Resistant Strain of *Klebsiella pneumoniae* in a Chinese Hospital

To the Editor—*Klebsiella pneumoniae* carbapenemase (KPC) was first reported to be the carbapenem-hydrolyzing β -lactamase from a carbapenem-resistant strain of *K. pneumoniae* in 1996¹ and was termed KPC-1. Two years later, KPC-2 was discovered by the same group.² However, a *bla*_{KPC-1} sequence error was found, and the *bla*_{KPC-1} and *bla*_{KPC-2} sequences were

TABLE 1. Antimicrobial Susceptibilities of the Clinical Isolate Labeled zjm002

Antimicrobial agent	Etest	Agar dilution	Clinical laboratory report
Ampicillin	≥256		R
Ampicillin/sulbactam	≥256		R
Amoxicillin/clavulanic acid	≥256		R
Cefalotin	≥256		R
Cefoxitin	≥256		R
Cefuroxime	≥256		R
Ceftazidime	≥256		R
Ceftriaxone	≥256		R
Cefotaxime	≥256		R
Cefepime	≥256		R
Aztreonam	≥256		R
Imipenem	≥32		R
Meropenem	≥32		R
Amikacin	12		S
Tobramycin	16		R
Netilmicin	32		R
Norfloxacin	≥32		R
Ofloxacin	≥32		R
Ciprofloxacin	≥32		R
Levofloxacin	≥32		R
Tetracycline	8		I
Tigecycline	1.5		S
Chloramphenicol	6		S
Trimethoprim-sulfamethoxazole	1.5		S
Ticarcillin/clavulanic acid	≥256		R
Cefoperazone/sulbactam	≥256		R
Piperacillin,		≥128	R
Piperacillin/tazobactam		≥4	R
Cefoperazone		≥64	R
Cefotetan		≥64	R
Ertapenem		≥8	R
Gentamycin		2	S
Polymyxin E		0.5	S
Polymyxin B		0.5	S
Fosfomycin		≥256	R

NOTE. I, intermediate sensitive; R, resistant; S, sensitive.

(Leu(168)→Met) when compared with the sequence of KPC-2, so this carbapenemase was confirmed to be a novel variant designated KPC-2-like (GenBank accession no. HQ258934).

As an important subgroup of class A carbapenemases, KPC has spread among Enterobacteriaceae throughout the world.⁴ The *bla*_{KPC} gene is often located on a Tn3-like transposon, Tn4401, which might mediate the rapid gene spread. Tn4401 was inserted on different-sized plasmids that belonged to different incompatibility groups.⁵

Though KPC played a major role in high-level resistance to carbapenems and other β -lactams, several other β -lactamases, such as TEM, SHV, and CTX-M enzymes, also might have existed in *K. pneumoniae* simultaneously.^{6,7} However, in our study, only *bla*_{TEM} and *bla*_{KPC} were discovered in zjm002. Considering that the TEM enzyme contributed nothing directly to carbapenem resistance, the KPC enzyme might be the result of it.

There were some other mechanisms, for instance, alterations in the outer membrane proteins of OmpK35/OmpK36 and overexpressions of efflux pumps, involving high levels of resistance to carbapenems.⁷ To zjm002, these mechanisms were unclear.

So far, 11 types of KPC enzymes were discovered all over the world. KPC-3 was found to differ from KPC-2 by a single amino acid substitution (His(272)→Tyr).⁸ Similarly, compared with KPC-2, in KPC-5 there was a single amino acid substitution⁹ (KPC-6 [EU555534], KPC-11 [HM066995]). Furthermore, two amino acid substitutions were found in KPC-4⁵ (KPC-7 [EU729727], KPC-8 [FJ234412], KPC-9 [FJ624872], KPC-10 [GQ140348]), compared with KPC-2. Our research confirmed that the KPC enzyme carried by zjm002 was a novel variant. As this variant shared 99% homology with KPC-2 from Genbank, it was termed KPC-2-like carbapenemases.

In clinic, owing to the carbapenem resistance and sensitivity to 2 aminoglycosides (amikacin and gentamycin), carbapenems and other β -lactams were abandoned, and etimicin, a type of aminoglycoside, was utilized to control the abdominal infection. To our great relief, 6 days later, the therapy was successful in clearing the abdominal drainage.

Accompanied by the dissemination of KPC-2 throughout the world, novel KPC variants emerged continuously. In our study, a novel KPC variant termed KPC-2-like was discovered in a *K. pneumoniae* isolate from the abdominal drainage of an 81-year-old patient. This KPC-2-like carbapenemase shared 99% homology with KPC-2. However, an attempt to transfer carbapenem resistance or to present the biochemical characterization of this new variant should be further performed. All β -lactams including carbapenems were virtually useless; etimicin was chosen so that the abdominal infection was controlled. KPC carbapenemases posed serious challenges to clinical therapy and the health of patients. Surveillance of the spread of KPC-producing *K. pneumoniae* should be urgently undertaken.

ACKNOWLEDGMENTS

Potential conflicts of interest. All authors report no conflicts of interest relevant to this article.

Jian Ming Zhu, BS;¹ Ru Jin Jiang, BS;¹
Zu Huang Mi, BS;² Hai Sheng Kong, BS;³
Feng Zhang, MD⁴

Affiliations: 1. Microbiology Laboratory, Hangzhou Yuhang Hospital of Traditional Chinese Medicine, Hangzhou, China; 2. Wuxi Clone Gen-Tech Institute, Wuxi, China; 3. Clinical Laboratory Department of the First Affiliated Hospital, College of Medicine, Zhejiang University, Hangzhou, China; 4. Clinical Laboratory Department of the Third Affiliated Hospital, Nantong University, Wuxi, China.

Address correspondence to Feng Zhang, MD, Clinical Laboratory Department of the Third Affiliated Hospital, Nantong University, 585 Xing Yuan North Road, Wuxi 214041, China (zf9958@163.com).

Infect Control Hosp Epidemiol 2011;32(10):1050-1052

© 2011 by The Society for Healthcare Epidemiology of America. All rights reserved. 0899-823X/2011/3210-0018\$15.00. DOI: 10.1086/662021

REFERENCES

1. Yigit H, Queenan AM, Anderson GJ, et al. Novel carbapenem-hydrolyzing beta-lactamase, KPC-1, from a carbapenem-resistant strain of *Klebsiella pneumoniae*. *Antimicrob Agents Chemother* 2001;45:1151–1161.
2. Yigit H, Queenan AM, Rasheed JK, et al. Carbapenem-resistant strain of *Klebsiella oxytoca* harboring carbapenem-hydrolyzing beta-lactamase KPC-2. *Antimicrob Agents Chemother* 2003;47:3881–3889.
3. Endimiani A, Perez F, Bajaksouzian S, et al. Evaluation of updated interpretative criteria for categorizing *Klebsiella pneumoniae* with reduced carbapenem susceptibility. *J Clin Microbiol* 2010;48:4417–4425.
4. Nordmann P, Cuzon G, Naas T. The real threat of *Klebsiella*

pneumoniae carbapenemase-producing bacteria. *Lancet Infect Dis* 2009;9:228–236.

5. Cuzon G, Naas T, Truong H, et al. Worldwide diversity of *Klebsiella pneumoniae* that produce beta-lactamase *bla*_{KPC-2} gene. *Emerg Infect Dis* 2010;16:1349–1356.
6. Zhang R, Wang XD, Cai JC, et al. Outbreak of KPC-2-producing *Klebsiella pneumoniae* with high *qnr* prevalence in a Chinese hospital. *J Med Microbiol* 2011;60(7):977–982.
7. Mataseje LF, Boyd DA, Willey BM, et al. Plasmid comparison and molecular analysis of *Klebsiella pneumoniae* harbouring *bla*_{KPC} from New York City and Toronto. *J Antimicrob Chemother* 2011;66:1273–1277.
8. Woodford N, Tierno PM Jr, Young K, et al. Outbreak of *Klebsiella pneumoniae* producing a new carbapenem-hydrolyzing class a beta-lactamase, KPC-3, in a New York medical center. *Antimicrob Agents Chemother* 2004;48:4793–4799.
9. Wolter DJ, Kurpiel PM, Woodford N, Palepou MF, Goering RV, Hanson ND. Phenotypic and enzymatic comparative analysis of the novel KPC variant KPC-5 and its evolutionary variants, KPC-2 and KPC-4. *Antimicrob Agents Chemother* 2009;53:557–562.

Investigation and Control of a Nosocomial Norovirus Outbreak in a Long-Term Care Facility

To the Editor—We report the investigation and control of an important nosocomial outbreak of norovirus infections that occurred in a long-term care facility (240 beds on 3 floors) affiliated with the university hospital of Brest, France, during the winter of 2008.

Norovirus is an RNA virus of the *Caliciviridae* family and the agent that causes most nonbacterial gastroenteritis.¹ Transmission is essentially fecal-oral, either direct through

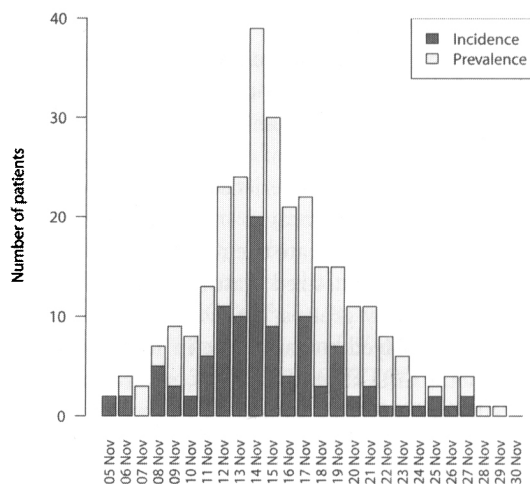


FIGURE 1. Winter 2008 norovirus outbreak in René Fortin long-term care facility: incident and prevalent cases, as daily observed.