ing responsible for nosocomial infection. Items used during the patient’s ocular surgery were confirmed to be disposable and nonreused.

Topical drops of corticosteroids commonly applied during cataract surgery for intraocular lens implantation penetrate ocular structures. An alternative hypothesis is that corticosteroids applied during ocular surgery reactivate a latent ocular infection. Our review indicated that 13 of 19 patients with documented T. whippelii uveitis had received topical or systemic corticosteroids before the diagnosis (Table) (7). Worsening of Whipple disease has been reported in patients receiving corticoid therapy for arthralgia (10). We speculate that our patient had an asymptomatic ocular infection before surgery.

This case shows that ocular surgery and use of topical corticosteroids that penetrate ocular structures could reactivate a latent T. whippelii ocular infection. We suggest that patients with postoperative panendophthalmitis be tested for T. whippelii by PCR.

M.D., F.F., and D.R. were supported by Unité de Recherche sur les Maladies Infectieuses et Tropicales Emergentes; D.D. was supported by Assistance Publique–Hôpitaux de Marseille.

Michel Drancourt,
Florence Fenollar,
Danièle Denis,
and Didier Raoult

Author affiliations: Université de la Méditerranée, Marseille, France; and Assistance Publique–Hôpitaux de Marseille, Marseille

DOI: 10.3201/eid1505.081209

References

1. Raoult D, Birg ML, La Scola B, Fournier PE, Enea M, Lepidi H, et al. Cultivation of the bacillus of Whipple’s disease. N Engl J Med. 2000;342:620–5. DOI: 10.1056/NEJM200003023420903
2. Fenollar F, Puechhal X, Raoult D. Whipple’s disease. N Engl J Med. 2007;356:55–66. DOI: 10.1056/NEJMra062477
3. Font RL, Rao NA, Issaescu S, McEntee WJ. Ocular involvement in Whipple’s disease: light and electron microscopic observations. Arch Ophthalmol. 1978;96:1431–6.
4. Durant WJ, Flood T, Goldberg MF, Tso MO, Pasquali LA, Peyman GA. Vitreom- tony and Whipple’s disease. Arch Ophthalmol. 1984;102:848–51.
5. Selsky EJ, Knox DL, Maumenee AE, Green WR. Ocular involvement in Whipple’s disease. Retina. 1984;4:103–6. DOI: 10.1097/00006982-198404020-00006
6. Williams JG, Edward DP, Tessler HH, Persing DH, Mitchell PS, Goldstein DA. Ocular manifestations of Whipple disease: an atypical presentation. Arch Ophthal- mol. 1998;116:1232–4.
7. Drancourt M, Berger P, Terrada C, Bodaghi B, Conrath J, Raoult D, et al. High preva- lence of fastidious bacteria in 1,520 cases of uveitis of unknown etiology. Medicine (Baltimore). 2008;87:167–76.
8. Davis JL, Miller DM, Ruiz P. Diagnostic testing of vitrectomy specimens. Am J Ophthalmol. 2005;140:822–9. DOI: 10.1016/j.ajo.2005.05.032
9. Knox DL, Green WR, Troncoso JC, Yardley JH, Hsu J, Zee DS. Cerebral ocular Whipple’s disease: a 62-year odyssey from death to diagnosis. Neurology. 1995;45:617–25.
10. Mahnel R, Kalt A, Ring S, Stallmach A, Strober W, Marth T. Immunosuppressive therapy in Whipple’s disease patients is associated with the appearance of gastro- intestinal manifestations. Am J Gastroen- terol. 2005;100:1167–73. DOI: 10.1111/ j.1572-0241.2005.40128.x

Address for correspondence: Didier Raoult, Unité des Rickettsies, Faculté de Médecine, Université de la Méditerranée, 27 Blvd Jean Moulin, 13385 Marseille Cedex 5, France; email: didier.raoult@gmail.com

Klebsiella pneumoniae Carbapenemase, Canada

To the Editor: Carbapenems are used to treat life-threatening infections caused by extremely drug-resistant gram-negative pathogens; these drugs represent the last line of defense in the antimicrobial drug armamentarium against serious or invasive infection (1). The rapid global spread of Klebsiella pneumoniae that produces K. pneumoniae carbapenemase (KPC), especially in the northeastern United States (e.g., New York state), is of major concern (2,3). KPC β-lactamases belong to the family of serine car- bapenemases and are usually found in K. pneumoniae and Escherichia coli. KPC hydrolyzes β-lactam agents, thereby reducing their action. KPC activity has been reported, albeit less frequently, in other family Enterobacteriaceae (K. oxytoca, Enterobacter spp., Salmonella spp., Citrobacter fre- undii, and Serratia spp.) as well as in Pseudomonas aeruginosa (1).

The blaKPC genes have been identified on conjugative plasmids and pose an infection control problem because plasmids could theoretically be transmitted from one species to another (4). The few therapeutic options for treating infections caused by organisms containing these β-lactamases are aminoglycosides, glycyclioclines, pol- ymyxins, or combinations (1). A ma- jor concern is that routine susceptibil- ity testing methods based on existing breakpoints can falsely identify KPC producers as susceptible to carbapenems. Such results pose the potential risk for increased illness and death, longer hospital stays, and nosocomial spread of infection.

In 2008, the Public Health Labo- ratory in Toronto received clinical iso- lates of K. pneumoniae from urine and sputum of 1 patient. The hospital labor- atory had forwarded the isolates to the
Public Health Laboratory because they were possible KPC producers. The patient was a 73-year-old man with a history of emphysema and hypertension, seen at a tertiary care hospital in the Toronto area, 80 miles from the New York state border, for a laparoscopic right radical nephrectomy because of hypernephroma. He had no risk factors for acquisition of KPC producers, e.g., travel to the United States or prior carbapenem exposure.

Susceptibility testing of *K. pneumoniae* was performed by the agar dilution method, using breakpoints set by the Clinical and Laboratory Standards Institute (5,6). The sputum isolate (7315) was susceptible to meropenem (MIC 4 μg/mL), and the urine isolate (7184) was intermediately susceptible (MIC 8 μg/mL). The *K. pneumoniae* isolates were screened for extended-spectrum β-lactamases (ESBLs) and AmpC production according to Ontario guidelines (7).

Briefly, to screen for ESBL enzymatic activity, a double-disk diffusion method was used: a clavulanic acid-containing disk was placed adjacent to a disk containing one of several cephalosporins such as ceftazidime and cefotaxime. Enhanced killing of the organism in the area between the drug with and without clavulanate indicates ESBL. Cefoxitin resistance (zone ≤17 mm) indicates AmpC-like β-lactamase activity. In addition, testing for ESBL/AmpC was performed according to Clinical and Laboratory Standards Institute guidelines (6). When the screening result for ESBL or AmpC is positive, the clinical laboratory issues a warning that no β-lactam except carbapenems can effectively treat this infection. The Table summarizes results of initial susceptibility testing and supplementary laboratory testing for KPC.

The initial result was consistent with a possible AmpC/ESBL producer for the sputum and urine isolates (6,7). However, because the patient responded poorly to empiric vancomycin and imipenem therapy and because of the elevated MIC to meropenem for isolate 7184, further laboratory testing was conducted to rule out the possibility of carbapenemase activity.

The modified Hodge test is a phenotypic test proposed to confirm the presence of carbapenemase activity such as KPC in *K. pneumoniae* and *E. coli* (8). Universal primers for *bla*KPC family, Uni-KPC-F (5′-ATGTC ACTGTATCGCCGTCT-3′) and -R (5′-TTACTGGCCGT TGAGC GCCC-3′), were used for the entire 882-bp coding sequence. Amplicons were bidirectionally sequenced by using the BigDye Terminators method and a 3130xl Genetic Analyzer (Applied Biosystems, Foster City, CA, USA) and primers Uni-KPC-F and -R. Multiple nucleotide and protein sequence alignments were performed with the ClustalW2 software (www.ebi.ac.uk/Tools/clustalw2/index.html). To aid the clinician, an Etest method was used to measure the MIC of this KPC-producing *K. pneumoniae* isolate to colistin (0.5 μg/mL) and tigecycline (2.0 μg/mL). However, before this information could be used, the patient had died of respiratory failure, presumably caused by *K. pneumoniae*. Infection control measures and laboratory screening were undertaken in the hospital to limit transmission to other patients.

This report shows that KPC-producing organisms such as *K. pneumoniae* may pose a major risk for clinical disease and a challenge for infection control if they were to spread to other hospitals in Canada. Current testing algorithms focus on ESBL- and AmpC-producing gram-negative bacteria, which may not detect KPC-producer strains. We suggest that reference laboratories validate a screening method coupled with confirmatory phenotypic assay for carbapenemase activity for suspected organisms, especially *K. pneumoniae* and *E. coli*. Our in-house validation studies confirm that use of the ertapenem disk followed by the modified Hodge test to confirm carbapenemase activity may be effective (D.R. Pillai et al., unpub. data). Public health officials should be aware that this report further expands the international distribution of KPC-producing *K. pneumoniae*.

The research component of this report was funded by the Ontario Agency for Health Protection and Promotion.

**Dylan R. Pillai, Roberto Melano, Prasad Rawte, Stephen Lo, Nathalie Tijet, Milan Fuksa, Nancy Roda, David J. Farrell, and Sigmund Krajden**

Author affiliations: Ontario Agency for Health Protection and Promotion, Toronto, Ontario, Canada (D.R. Pillai, R. Melano, P. Rawte, S. Lo, N. Tijet, D.J. Farrell); University of Toronto, Toronto (D.R. Pillai, R. Melano, D.J. Farrell, S. Krajden); University Health Protection and Promotion, Toronto, Ontario, Canada

---

Table. Results of initial susceptibility and supplementary testing for *Klebsiella pneumoniae* carbapenemase in urine and sputum samples from 73-year-old man, Canada*

| Isolate | MIC, μg/mL† | Disk diffusion results, mm |
|---------|-------------|---------------------------|
|         | AMP         | FOX | CIP | GEN | CTRX | MEM | FOX | CAZ | CAC | CTX | CTC | Initial report | Final report|
| 7184    | >16         | >16 | >2  | 8   | >32  | 8   | 16  | 0   | 14  | 13  | 15  | AmpC/ESBL      | KPC          |
| 7315    | >16         | >16 | >2  | 8   | >32  | 8   | 0   | 0   | 8   | 13  | 15  | AmpC/ESBL      | KPC          |

*AMP, ampicillin; FOX, cefoxitin; CIP, ciprofloxacin; GEN, gentamicin; CTRX, ceftriaxone; MEM, meropenem; CAZ, ceftazidime; CAC, ceftazidime-clavulanic acid; CTX, cefotaxime; CTC, cefotaxime-clavulanic acid; ESBL, extended-spectrum β-lactamase; KPC, Klebsiella pneumoniae carbapenemase.

†MIC values for clinical isolates 7184 (urine) and 7315 (sputum) were obtained by using agar macrodilution.

‡Initial screening for ESBL or AmpC β-lactamase activity, performed by Kirby Bauer disk diffusion according to Clinical Laboratory Standards Institute guidelines (6,7), suggested ESBL or AmpC β-lactamase activity.

§Supplementary modified Hodge test; PCR (specific for *bla*KPC family), and DNA sequencing confirmed the presence of KPC activity due to *bla*KPC-2.
Cryptosporidium sp. Rabbit Genotype, a Newly Identified Human Pathogen

To the Editor: Most human cases of cryptosporidiosis are caused by Cryptosporidium parvum or C. hominis, but pathogenicity of some unusual Cryptosporidium species/genotypes is uncertain (1). In July 2008, an outbreak caused by Cryptosporidium sp. rabbit genotype was linked to consumption of tap water in Northamptonshire, England (2). On June 23 and 24, Cryptosporidium oocysts were detected by operational monitoring of treated water at a surface water treatment works. A precautionary boil-water notice was implemented on June 25.

Enhanced surveillance for cases was established by the health protection team on June 25 in the affected area. Eight single-well immunofluorescent microscopy slides, on which oocysts were detected by water company sampling of the distribution system, were sent to the UK Cryptosporidium Reference Unit, Swansea, for typing. Slides contained 49–259 oocysts. Coverslips were removed after softening the seal with nail polish remover. Fixed material was resuspended after thaw cycles, and DNA was extracted using the QiAamp DNA Mini Kit (QIAGEN), which involved digestion with proteinase K in lysis buffer AL at 56°C for 30 min, purification in a spin column, elution in 50 µL buffer AE, and storage at –20°C (3).

Cryptosporidium oocysts were also detected by direct immunofluorescent antibody test (IFAT) (Cryptocel; TCS Biosciences, Buckingham, UK) in large bowel contents from a rabbit carcass removed by the water company from a tank at the water treatment works. Oocysts were separated from fecal debris by flotation, resuspended in reverse osmosis water (4), and processed as above.

Cryptosporidium species were identified by bidirectional sequencing of PCR products generated by nested PCR for the small subunit (SSU) rRNA gene (5) from 4 DNA aliquots of each sample. SSU rRNA sequences from 7 water samples, containing 49–197 oocysts, and the rabbit isolate were homologous with isolates from rabbits in the People’s Republic of China (6) and the Czech Republic (7)(GenBank accession nos. AY120901 and AY273771, respectively) (online Appendix Table, available from www.cdc.gov/EID/content/15/5/829-appT.pdf). One sample from 1,391 L of water contained 259 oocysts but was not amplified. Other cryptosporidia were not identified.

Human stool samples from 34 local laboratory-identified cases of cryptosporidiosis in the affected area were sent to the UK Cryptosporidium Reference Unit for typing. To differentiate rabbit genotype from C. hominis (1), enhanced typing by SSU rRNA nested PCR–restriction fragment length polymorphism analysis with SpI (1,5) was used for all isolates submitted to the UK Cryptosporidium Reference Unit during July and August. Samples from 23 cases (22 primary and 1 secondary) with rabbit genotype profiles were identified by visualization of 472-, 267-, and 109-bp bands generated by digestion with SpI (1). All case-patients lived in the area affected by the water supply incident and had onset dates consistent with exposure by drinking water consumption or by person-to-person spread. All 23 samples were homologous to AY120901 and AY273771 (online Appendix Table). Of the other 11 samples, 6 were not confirmed by IFAT or PCR, 2 were C. hominis, 1 was C. parvum, and 2 were not typeable.

Address for correspondence: Dylan R. Pillai, Ontario Public Health Laboratories, Medical Microbiology, Rm 243, 81 Resources Rd, Toronto, Ontario M9P 3T1, Canada; email: dylan.pillai@oahpp.ca

References
1. Walther-Rasmussen J, Hily N. Class A carbapenemases. J Antimicrob Chemother. 2007;60:470–82. DOI: 10.1093/jac/dkm226
2. Bradford PA, Bratu S, Urban C, Visalli M, Mariano N, Landman D, et al. Emergence of carbapenem-resistant Klebsiella species possessing the class A carbapenem-hydrolyzing KPC-2 and inhibitor-resistant TEM-30 β-lactamases in New York City. Clin Infect Dis. 2004;39:55–60. DOI: 10.1086/421495
3. Woodford N, Tierno PM Jr, Young K, Tysall L, Palepou MF, Ward E, et al. Outbreak of Klebsiella pneumoniae producing a new carbapenem-hydrolyzing class A β-lactamase, KPC-3, in a New York medical center. Antimicrob Agents Chemother. 2004;48:4793–9. DOI: 10.1128/AAC.48.12.4793-4799.2004
4. Naas T, Cuzon G, Villegas MV, Lartigue MF, Quinn JP, Nordmann P. Genetic structures at the origin of acquisition of the β-lactamase blaKPC gene. Antimicrob Agents Chemother. 2008;52:1257–63. DOI: 10.1128/AAC.01451-07
5. Clinical and Laboratory Standards Institute. Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically; approved standard (document M7-A7 Edn). Wayne (PA): The Institute; 2006.
6. Clinical and Laboratory Standards Institute Performance standards for antimicrobial susceptibility testing; eighteenth informational supplement (document M100-S18). Wayne (PA): The Institute; 2006.
7. Quality Management Program, Laboratory Services. Extended spectrum β-lactamase and AmpC in gram-negative bacilli. In: Rutherford C, editor. Broadsheet; May 15, 2006. p. 1–18.
8. Anderson KF, Lonsway DR, Rasheed JK, Biddle J, Jensen B, McDougall LK, et al. Evaluation of methods to identify the Klebsiella pneumoniae carbapenemase in Enterobacteriaceae. J Clin Microbiol. 2007;45:2723–5. DOI: 10.1128/JCM.00015-07

DOI: 10.3201/eid1505.081536