The Spread of *Klebsiella pneumoniae* Carbapenemase–Producing *K. pneumoniae* to Upstate New York

Ben M. Lomaestro, Ellis H. Tobin, Wenchi Shang, and Thomas Gootz

Clinical Pharmacology, Albany Medical Center Hospital, and Upstate Infectious Diseases Associates, Albany, New York; and Department of Antibiotics, Immunology and Cancer, Pfizer Global Research and Development, Groton, Connecticut

*K. pneumoniae* carbapenemases (KPCs) have previously been identified in distinct geographic locations. We report the spread of KPC-2 to upstate New York. Our intention is to alert clinicians to problems encountered in identifying KPC-containing isolates. Possible errors as a result of inferring susceptibility of untested carbapenems from the routine antibiogram using agar-based methodology or microdilution testing are discussed.

Enzyme-mediated carbapenem resistance in Enterobacteriaceae is relatively rare. However, *Klebsiella pneumoniae* carbapenemase (KPC) type β-lactamases capable of hydrolyzing penicillins, cephalosporins, aztreonam, and carbapenems are becoming increasingly common in the northeastern United States [1]. They are categorized as Ambler class A and Bush functional group 2f, and they differ from metallo-β-lactamases, which require divalent cations as metal cofactors for activity [2, 3].

There has been a well described outbreak of KPC infection among patients residing in the New York City area [1, 4–6]. We now report the identification of KPC-2 infection in 3 patients residing in upstate New York. Although we cannot determine whether our cases were sporadic in nature or attribute them to the existence of an outbreak, this enzyme has not been previously identified in this geographic area. Routine laboratory testing was unable to identify these isolates. In addition, our assumption that ertapenem activity could be inferred from meropenem susceptibility results in an extended-spectrum β-lactamase (ESBL)–containing organism may have contributed to treatment failure.

**Case reports.** Patient 1 was a 44-year-old man recovering from a kidney transplantation in New York City in January 2003. He was admitted to Albany Medical Center Hospital on 2 March 2005 for *K. pneumoniae* bacteremia that appeared susceptible to meropenem, as determined by disk diffusion testing. He received 2 days of meropenem treatment, which was switched to ertapenem for ease of administration. Recurrent *K. pneumoniae* bacteremia was identified 7 days later while the patient was receiving ertapenem. Resistance to ertapenem was subsequently demonstrated by Etest (AB Biodisk), with an MIC of >8.0 mg/L. The Etest result for meropenem was 3.0 mg/L, which is indicative of susceptibility. The isolate was sent to an outside laboratory for analysis, where it was subjected to PCR amplification of *bla* and *ompK* genes and DNA sequencing. The primers used for amplification of the *bla* and *ompK* genes are presented in table 1. Amplification conditions were essentially those previously reported [6]. Exact gene identification was accomplished by sequencing amplicons using standard methods, through the use of gene terminator kits obtained from Applied Biosystems and primers for sequencing were synthesized by Operon/Qiagen. PCR products from 2 independent reactions were analyzed for each gene. Analysis confirmed the presence of KPC-2, TEM-1, and SHV-12 enzymes. The DNA sequence analysis was consistent with an inactive Ompp35, and functional Ompp36 and Ompp37 porins.

Patient 2 was a 39-year-old man who experienced multiple trauma from a motor vehicle accident on 19 April 2005. He developed sepsis and ventilator-associated pneumonia. A non-ESBL *K. pneumoniae* strain was isolated from a sputum culture performed on 4 May. On 10 May, he developed *K. pneumoniae* bacteremia; an Etest demonstrated MICs of 3.0 mg/L to meropenem and >8 mg/L to ertapenem. This isolate was submitted to a reference laboratory and was subsequently shown to possess KPC-2 by sequencing of the PCR amplicon. Testing for other enzymes or porins changes was not performed.

Patient 3 was a 41-year-old woman with end-stage renal disease requiring hemodialysis, in whom *K. pneumoniae* bacteremia was identified 30 days after hospital admission. This isolate contained an ESBL initially determined to be meropenem indeterminate by Kirby-Bauer testing. A subsequent Etest revealed an MIC of 3.0 mg/L for meropenem and an MIC >8.0 mg/L for ertapenem. A reference laboratory confirmed the presence of KPC-2 by PCR analysis but did not test for other resistance mechanisms.

Patients 2 and 3 did not make recent health care–related
visits to the New York city area. All 3 patients survived their infections and were discharged after lengthy hospital stays.

**Discussion.** KPC-1 was named and identified from an isolate of *K. pneumoniae* collected in North Carolina in 2001 [7] and occurred in combination with reduced expression of porins OmpK35 and OmpK37. KPC-2 was described in 2003 from a stool specimen of *Salmonella enterica* from a patient hospitalized in Maryland [8]. *K. pneumoniae* isolates containing KPC-2 have been reported in New York City, Boston, and Maryland [2, 8, 9]. KPC-2 has also been identified in *Enterobacter* species [10]. These isolates produced multiple β-lactamases, which hindered identification via antibiogram. In 2004, KPC-3 was identified in isolates of *K. pneumoniae* collected in New York City and was present in a strain of *Escherichia coli* from Hackensack, New Jersey, in 2005 [11, 12]. The New York City *Klebsiella* isolates of KPC-3 were encoded on a plasmid and lacked the OmpK35 porin [12].

To date, KPC-2 has occurred most frequently and has been best analyzed. One isolate of *Klebsiella oxytoca* and 18 additional isolates of *K. pneumoniae* from 7 different New York City hospitals containing KPC-2 and inhibitor-resistant TEM-30 were described in 2003 [1, 4]. Most of these isolates also contained ESBLs, and 14 isolates possessed SHV-12, similar to the strain in our first case. Bacterial isolates containing KPC enzymes confer greater resistance to cephapslorins than to penicillins; it is postulated that cephalosporin use may be selective for KPC enzymes [12]. KPC enzymes are often reported to be susceptible to carbapenems when automated systems are used, thereby hindering identification of the isolates [13].

The degree to which carbapenemics confer resistance may be linked to additional resistance mechanisms, such as increased permeability barriers [14, 15]. Alteration of 3 porin proteins has been described for *K. pneumoniae* (OmpK35, OmpK36, and OmpK37), and resulted in an increased MIC for carbapenems through decreased entry into bacteria [7]. Two additional surveillance studies from New York City noted that KPC-containing *K. pneumoniae* isolates were frequently resistant to virtually all commonly used antimicrobials and also noted that identification of KPC-mediated resistance was often missed by initial routine microbiologic testing [5, 6]. Although 95% of isolates were correctly identified as carbapenem resistant using agar-based diffusion testing, many isolates were considered to be susceptible or intermediate to imipenem on the basis of broth-based test results [5].

Reduced susceptibility to carbapenems (MIC >1 mg/L) may be an early identifier of KPC-producing strains [2]. Standard broth microdilution techniques are highly inoculum dependent and may fail to identify carbapenem resistance, especially if imipenem is the tested carbapenem [6]. An inoculum effect was not observed with ertapenem, which suggests that ertapenem may be preferred in carbapenem susceptibility testing of KPC-containing isolates [5]. Our institution does not perform broth microdilution techniques because they may misidentify these agents as a result of inoculum-dependent factors. The presence of multiple β-lactamases often renders isolates resistant to β-lactamase inhibitors [1]. In addition, some isolates may have acquired an AmpC β-lactamase and may have a negative confirmatory test result for ESBL production, but they may produce a KPC enzyme [9]. This complex phenotype makes detection of a specific carbapenemase (such as the KPC group) very difficult without a reliance on molecular analysis.

Our first case illustrates potential deficiencies when using meropenem susceptibility test results to predict ertapenem susceptibility and should serve to alert clinicians to possible discrepancies with this practice. Although it has been suggested that susceptibility to ertapenem in Enterobacteriaceae can be predicted reliably from imipenem susceptibility [16], others have also found this to be a poor surrogate for ertapenem susceptibility in ESBL-producing organisms [17, 18]. Our 3 cases also confirm the difficulty in identifying new resistance phenotypes by conventional microbiology testing. We currently screen all ESBL-positive strains for ertapenem susceptibility as a marker for KPC enzymes.

In summary, dissemination of KPC enzymes has expanded beyond New York City. In addition to our results at Albany Medical Center Hospital, we have been alerted to the presence of KPC-containing strains from several hospitals and a nursing home in upstate New York (personal communication).

On 29 August 2005, the State of New York Department of Health (Albany, New York) issued an advisory regarding the spread of multidrug-resistant *K. pneumoniae* throughout the state. Verification of KPC enzymes is now performed in submitted isolates by the New York state laboratory in Albany. Merck also administers a program to determine the presence of KPC enzymes in Enterobacteriaceae. Their program was used to confirm the presence of KPC-2 in the isolates identified in cases 2 and 3.
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References