Published in final edited form as:

Transpl Infect Dis. 2009 June ; 11(3): 257–265. doi:10.1111/j.1399-3062.2009.00374.x.

Fatal cross infection by carbapenem resistant *Klebsiella* in two liver transplant recipients

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Abstract

Members of the family Enterobacteriaceae including *Klebsiella* have re-emerged as major pathogens in solid organ transplantation. The recent appearance and dissemination of carbapenemase-producing Enterobacteriaceae in Europe and the northeastern United States represents a major challenge to the treatment of enteric gram-negative bacterial infections in immunocompromised patients; however, few reports have detailed the outcomes of such infections. Here we report two cases of *Klebsiella pneumoniae* carbapenemase (KPC)-producing *Klebsiella* infections in orthotopic liver transplant recipients, which were the index case and initial secondary case for an outbreak of KPC-producing Enterobacteriaceae in our institution. In both instances, the pathogens were initially misidentified as being carbapenem sensitive, the infections recurred after cessation of directed therapy, and the patients ultimately succumbed to their infections.

MeSH Keywords

Adult; Cross infection/epidemiology; Cross Infection/microbiology*; Diseases Outbreaks*; DNA, Bacterial/genetics; Drug Resistance, Bacterial; Enterobacteriaceae Infections*/epidemiology; Enterobacteriaceae Infections*/microbiology; Enterobacteriaceae Infections*/mortality; Fatal Outcome; Humans; *Klebsiella**/isolation & purification*; *Klebsiella**/enzymology; *Klebsiella**/genetics; Liver Transplantation*; Sepsis/microbiology*; beta-Lactam Resistance*; beta-Lactamases*/genetics

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Potential financial conflicts of interest:

None declared.

Substances: Anti-Bacterial Agents; Carbapenems; Polymyxins; tigecycline

Introduction

Over the last decade, gram-negative bacilli have re-emerged as important pathogens in solid organ transplant (SOT) recipients (1). While numerous factors likely contributed to this epidemiologic shift, the resurgence of gram-negative pathogens has coincided with the dissemination of multidrug resistance in Enterobacteriaceae and nonfermentative bacilli. In Enterobacteriaceae, resistance to expanded-spectrum β -lactam antibiotics is usually conferred by plasmid-encoded extended spectrum β -lactamases (ESBLs), and ESBL-containing isolates are typically resistant to other classes of antibiotics. Since carbapenems are highly resistant to ESBL-mediated hydrolysis, these agents are preferred for the treatment of invasive infection caused by these organisms (2).

For that reason, the recent emergence of carbapenem-hydrolyzing β -lactamases (carbapenemases) in Enterobacteriaceae represents a concerning threat to the treatment of invasive enterobacterial infections. While carbapenemase-producing Enterobacteriaceae (CP-E) are rare in most parts of the world, certain geographic areas have seen significant outbreaks that may soon approach endemicity in some hospitals. In the northeastern United States and Israel, the plasmid encoded Ambler class A serine β -lactamase KPC (for *Klebsiella pneumoniae* carbapenemase) has been found in large institutional outbreaks of multidrug resistant *Klebsiella* and *Enterobacter* spp. (3–5). In southern Europe, the Ambler class B metallo- β -lactamase VIM has been associated with multiple genera outbreaks of gram-negative pathogens (6–8). Recent reports suggest that KPC-producing Enterobacteriaceae may be rapidly spreading across North America, while metallo- β -lactamase-producing Enterobacteriaceae are being reported in many locations worldwide (9–11). SOT recipients are vulnerable to infection by multidrug resistant bacteria due to their underlying end-stage organ failure combined with a major surgical procedure and the necessity of life long immunosuppression. In this report, we describe the clinical and microbiologic features of CP-E infection in two liver transplant (LT) recipients caused by KPC-producing *Klebsiella*.

Materials and Methods

Microbiology

Identification and antibiotic susceptibility testing was performed using the Vitek 2 system with the AST GN14 card (bioMérieux, Durham, NC). Minimum inhibitory concentrations (MICs) were determined by the microbroth dilution method in cation-adjusted Mueller-Hinton broth and the Kirby Bauer agar disk diffusion method, in accordance with the Clinical and Laboratory Standards Institute (CLSI) guidelines (12). Modified Hodge testing was performed as previously described (13). Briefly, a Mueller-Hinton agar (MHA) plate was inoculated with a one-tenth dilution of the carbapenem sensitive strain *Escherichia coli* ATCC 25922 adjusted to a McFarland 0.5 standard. A 10- μ g imipenem disk was then placed in the center of the plate and the scrutinized *Klebsiella* isolates were struck radially from the disk to the plate edge. Invagination of *E. coli* growth at interface between the *Klebsiella* streak and the imipenem zone of inhibition was read as a positive modified Hodge test (13).

DNA Techniques

Plasmid DNA was isolated from *Klebsiella* strains using the QIAGEN Plasmid Midi Kit (QIAGEN, Valencia, CA) using 60°C pre-warmed eluting buffer, as recommended by the manufacturer for large plasmid isolation. PCR amplification of the *bla*_{KPC} gene was performed using previously reported primers and condition (14). *bla*_{KPC} verification and

allele identification was performed by bidirectional DNA sequencing (Biomolecular Research Facility, University of Virginia Health System).

Southern Analysis

Plasmid DNAs were subjected to electrophoresis on a 0.6% Pulse Field Certified Agarose (Bio-Rad Laboratories, Hercules, CA) gel using the following parameters: 0.5X TBE, 70 volts, 4°C, 20 hours. The gel was stained with ethidium bromide and migration distances of the DNA bands from the clinical isolates were compared to reference plasmid DNAs from *E. coli* strain V517. Plasmid DNA was transferred to Amersham Hybond N⁺ nylon membrane (GE Healthcare, Piscataway, NJ) by capillary transfer and hybridized with a 1009-bp DNA amplicon of *bla*_{KPC-2} labeled using the Amersham ECL Direct Nucleic Acid Labeling and Detection System (GE Healthcare) in accordance with the manufacturer's instructions.

Pulse Field Gel Electrophoresis

Pulsed-field gel electrophoresis (PFGE) of *Xba*I-digested total DNA was performed using the PulseNet protocol for *E. coli* and other *Enterobacteriaceae* (15). PFGE profile analysis and dendrogram construction were performed using BioNumerics software (Applied Maths, Austin, TX).

Case Reports

Case 1

A 45-year-old man with decompensated liver disease resulting from hepatitis C virus infection and alcoholic cirrhosis underwent liver transplantation at our center in July 2007. The patient had been discharged from a Philadelphia hospital four days prior to LT after a nine day hospitalization for spontaneous bacterial peritonitis due to *E. coli*. LT using the piggyback technique with duct-to-duct biliary anastomosis was performed without complication; piperacillin-tazobactam (TZP) was given for perioperative antibiotic prophylaxis. Peritoneal fluid obtained during the transplant grew vancomycin resistant *Enterococcus faecium* (VRE). The patient was discharged on prophylactic immunosuppression (tacrolimus with trough levels of 5–10 ng/mL, mycophenolate mofetil (MMF), and a steroid taper), prophylactic trimethoprim-sulfamethoxazole (TMP-SMX), and a two week course of linezolid on postoperative day seven. Two weeks later he was readmitted to a Philadelphia hospital with increasing ascites and abdominal pain. His condition rapidly deteriorated and he was transferred to our hospital within 48 hours, where he was diagnosed with an anastomotic bile leak, peritonitis due to linezolid-resistant *E. faecium* (LRE) and *Candida glabrata*, and hepatic artery thrombosis. He underwent exploratory laparotomy and washout, ERCP with multiple biliary stent deployments, and unsuccessful attempts at hepatic artery stenting. Over the next two weeks additional complications arose including CMV hepatitis, *Aspergillus fumigatus* and ventilator-associated pneumonia, tacrolimus toxicity, acute kidney injury requiring renal replacement therapy, pancytopenia and luminal GI bleeding. Antimicrobial therapy during this period included tigecycline, ciprofloxacin, anidulafungin, voriconazole and ganciclovir; MMF was discontinued.

On hospital day 21, the patient became septic and *Klebsiella pneumoniae* grew from blood (strain Kpn1014), urine, abdominal drainage, and bronchial cultures. Antibiotic susceptibilities by Vitek 2 were notable for a meropenem MIC of ≤ 0.25 μ g/mL (Table 1). LRE was also isolated from the blood. Meropenem and daptomycin were started and tigecycline was stopped. Despite initial improvement, the patient again developed septic shock and multiorgan dysfunction twelve days later while still receiving meropenem. *K.*

pneumoniae (strain Kpn1016) was cultured from abdominal drain cultures, while *Klebsiella oxytoca* was isolated from both abdominal drain cultures (strain Kox1015) and respiratory secretions. The *K. oxytoca* isolate had a Vitek meropenem MIC of 1 µg/mL, and the *K. pneumoniae* isolate had a Vitek meropenem MIC of ≥ 16 µg/mL (Table 1). Imaging studies and a second exploratory laparotomy failed to identify any undrained purulent collections. Kox1015 was initially interpreted as meropenem susceptible; however, the elevated meropenem MIC in strain Kpn1016 raised concern for the presence of carbapenemase, which previously had not been observed in any clinical Enterobacteriaceae isolates at our medical center. Molecular studies confirmed the presence of *bla*_{KPC-2} in both Kpn1016 and Kox1015 (see below). Meropenem was discontinued and a two week course of colistimethate sodium (colistin) and tigecycline was given. Isolation of carbapenemase-producing *Klebsiella* triggered a hospital-wide epidemiology surveillance program and updated infection control training programs for transplant and ICU healthcare staff. The patient remained critically ill and the bile leak was not amenable to repair by interventional radiology or surgery. Over the next several months, he developed a large liver abscess and had several episodes of bacteremia with KPC-producing *K. pneumoniae* as well as *Stenotrophomonas maltophilia*, *Enterococcus faecalis*, and *E. faecium*. Multiple courses of culture-directed antibiotics, including combinations of colistin, tigecycline, amikacin, high dose TMP-SMX, and daptomycin, were given. On hospital day 115 (post-LT day 138), his clinical condition was deemed to be sufficiently stable to attempt re-LT as a final attempt for cure. The original graft was necrotic with several large abscesses. Intraoperatively the patient developed a severe coagulopathy and refractory hypotension despite massive resuscitation efforts, and he died several hours after graft reperfusion.

Case 2

A 64-year-old woman underwent LT for end-stage liver disease due to nonalcoholic steatohepatitis and hepatocellular carcinoma at our center in August 2007. LT using the piggyback technique with duct-to-duct biliary anastomosis was performed without complications, and she was admitted to the same surgical intensive unit as patient 1. TZP was given perioperatively. Prophylactic immunosuppression included tacrolimus (trough levels, 5–10 ng/mL), MMF and corticosteroids. Initial graft function was poor and she developed steroid-resistant acute cellular rejection, which responded to antithymocyte globulin. On postoperative day fourteen she developed a large pulmonary embolism with hemodynamic instability. Subsequent problems over the next month included GI bleeding, CMV viremia, *Clostridium difficile* colitis, VRE bacteremia, and acute kidney injury requiring renal replacement therapy. Antimicrobial therapy during this period included linezolid, cefepime, metronidazole, ganciclovir, and prophylactic TMP-SMX; she did not receive a carbapenem.

She slowly improved until post-LT day 45, when she developed septic shock and respiratory failure. *K. pneumoniae* was isolated from blood (strain Kpn1017) and urine cultures. The meropenem MIC was 4 µg/mL by Vitek 2, which was interpreted as susceptible. Meropenem was started and subsequent blood cultures were negative; however, she remained febrile and hypotensive requiring inotropic support. Five days later, Kpn1017 was confirmed to be modified Hodge test and *bla*_{KPC} PCR positive, and meropenem was changed to colistin. She responded well to a two week course of colistin; unfortunately, graft function further deteriorated when *K. pneumoniae* bacteremia and septic shock recurred several weeks later. Due to her multiple medical problems, re-LT was not an option and she died within days of withdrawal of intensive medical care.

Microbiology

Carbapenem susceptibilities and genetic analysis of the *Klebsiella* isolates are presented in Table 1. Kpn1014, the initial *K. pneumoniae* isolate from patient 1, had a meropenem MIC ≤ 0.25 $\mu\text{g/mL}$ by Vitek 2 and microbroth dilution and a zone diameter of 28 mm by disk diffusion. The isolate had a negative modified Hodge test and the *bla*_{KPC} gene was not detected by PCR amplification (Fig. 1, Table 1). Kox1015, isolated from patient 1 after ten days of meropenem therapy, was found to have an MIC to meropenem of 1 $\mu\text{g/mL}$ (susceptible) by Vitek 2 but 8 $\mu\text{g/mL}$ (intermediate) by microbroth dilution (Table 1). Kpn1016 had an MIC to meropenem of ≥ 16 $\mu\text{g/mL}$ (resistant) by Vitek 2 and 4 $\mu\text{g/mL}$ (susceptible) by microbroth dilution (Table 1). Kpn1017 isolated from patient 2 thirty-five days after isolating Kpn1016 from patient 1, had an MIC to meropenem of 4 $\mu\text{g/mL}$ (susceptible) by Vitek 2 and 16 $\mu\text{g/mL}$ (resistant) by microbroth dilution (Table 1). On disk diffusion testing, the zones of inhibition for Kox1015, Kpn1016, and Kpn1017 (zone diameters 16–17 mm) were smaller than that for Kpn1014 but remained in the susceptible range. Recently, detection of CP-E by automated susceptibility testing systems has been shown to be greatly improved by using ertapenem rather than meropenem or imipenem as the class agent for carbapenems (16). Kox1015, Kpn1016, and Kpn1017 had an MIC to ertapenem of 4 $\mu\text{g/mL}$ or greater (non-susceptible) by Vitek 2 and microbroth dilution (Table 1). Kox1015, Kpn1016, and Kpn1017 also had positive modified Hodge and *bla*_{KPC} PCR tests (Fig. 1, Table 1). DNA sequencing confirmed that all isolates carried the *bla*_{KPC-2} allele.

Plasmids were extracted from the *Klebsiella* isolates and separated by gel electrophoresis. As shown in Figure 2A, the plasmid profiles of Kpn1016 and Kpn1017 were identical, while those of Kpn1014 and Kox1015 differed by several bands. Southern blot analysis using a *bla*_{KPC} specific probe confirmed the presence of *bla*_{KPC} plasmids in strains Kox1015, Kpn1016, and Kpn1017 but not Kpn1014 (Fig. 2B). Kpn1016 and Kpn1017 had a single plasmid of 54-kb that hybridized to the *bla*_{KPC} probe, while Kox1015 had two *bla*_{KPC}-positive plasmid bands of approximately 80- and 110-kb.

PFGE of the *K. pneumoniae* isolates revealed that Kpn1014 was closely related to Kpn1016, differing by a total of three bands, and that Kpn1016 and Kpn1017 belong to the same lineage (87% Dice similarity coefficient) (Fig. 3). Kpn1016 and Kpn1017 were not related (<60% Dice similarity coefficient) to other KPC-producing *K. pneumoniae* isolates in the CDC database, including archived strains obtained in 2005 from the Philadelphia hospital where patient 1 was hospitalized early in his course (data not shown).

Discussion

This report highlights several challenges regarding the diagnosis and treatment of CP-E infections. First, it demonstrates the difficulty in recognizing and verifying carbapenemase-production in Enterobacteriaceae by clinical microbiology laboratories. In the United States, the KPC enzyme has emerged in diverse array of Enterobacteriaceae, including *K. pneumoniae*, *K. oxytoca*, *Enterobacter* spp., *E. coli*, *Citrobacter freundii*, and *Salmonella enterica* (17). As seen in these cases, automated susceptibility testing can incorrectly report a KPC-producing organism as carbapenem sensitive (4,16,18). In hospitals with no prior experience with CP-E, initial recognition of an enteric carbapenemase producer may only occur due to the consideration of an astute clinician or microbiologist. Second, the cases confirm that the treatment of CP-E infections with carbapenemases is associated with clinical and/or microbiological failure. Erroneous assignment as carbapenem susceptible may have grave clinical consequences. Third, unrecognized, undrained or inaccessible collections may serve as a nidus for recurrent infection. Although not observed in these

cases, re-treatment of CP-E infections has the potential to further select for resistance in these isolates.

Twelve cases of invasive CP-E infection, including the 2 presented herein, have been reported in the English language literature and are summarized in Table 2 (8,19–23). The cases included 7 liver, 3 kidney, and one heart transplant recipient; the organ transplanted was not reported in one case. Thirteen Enterobacteriaceae were cultured: 9 *K. pneumoniae*, 2 *K. oxytoca*, and 2 *Enterobacter cloacae*. Nine of the twelve cases involved bloodstream infection. In North America all isolates (n=7) carried *bla*_{KPC}, while in Europe the isolates (n=6), most from a single Italian hospital outbreak, carried *bla*_{VIM-1}. This pattern generally reflects the current distribution of carbapenemases in Enterobacteriaceae, with a preponderance of KPC (serine) carbapenemases in North America and VIM metallo-β-lactamases in Europe. The crude mortality rate was 58%; six of the seven LT patients died. No clear relationship between the chosen antimicrobial regimen and survival could be determined.

Several factors place SOT recipients at high risk for infection by CP-E. First, SOT recipients often receive prolonged courses of broad spectrum antibiotics, an enormous selective pressure for the acquisition of antimicrobial-resistant organisms. Determinants of resistance, including KPC and VIM carbapenemases, are often encoded on transposons or plasmids that carry suites of other resistance genes. Therefore, selective pressure occurs even with the use of unrelated classes of antibiotics. In this regard, it is instructive that patient 2 developed *bla*_{KPC-2}-positive *K. pneumoniae* bacteremia without antecedent carbapenem therapy. Second, transplant recipients undergo numerous invasive therapeutic and diagnostic procedures. Technical complications, including early graft injury, as well as the intensity of immunosuppression, poor allograft function and rejection are particularly important risks for the development of nosocomial infections in the early post-transplant period (24,25). Third, SOT patients have high rates of hospitalization, which increase the risk of becoming colonized with healthcare-associated multidrug resistant organisms like CP-E. Intra- and interhospital transfers are common amongst organ transplant recipients and may geographically expand opportunities for cross-colonization by drug-resistant organisms. We speculate that the index patient may have become colonized with *bla*_{KPC-2}-containing *Klebsiella* during a hospitalization in Philadelphia, although PFGE failed to show genetic relatedness between his isolates and tested Philadelphia KPC-containing *Klebsiella* isolates. Alternatively, he may have acquired a strain during an earlier hospitalization that later served as a *bla*_{KPC-2}-plasmid or transposon donor.

Infections in SOT patients and other immunocompromised hosts can provide sentinel warning of changes in pathogen trafficking and dissemination, both in the hospital and the community (26,27). In this instance, the reported patients were the first cases of KPC-producing Enterobacteriaceae infection identified at our institution. A subsequent hospital-wide outbreak involving two dozen additional patients has been recognized and epidemiologically and/or microbiologically linked to the index cases, despite reinforced infection control education and monitoring efforts (A.M., H.C., K.C.H. and C.D.S., unpublished data). Similarly, in an Italian report describing an outbreak of VIM-1 producing *Klebsiella pneumoniae* bloodstream infections, the index case occurred in a kidney transplant recipient (8). These reports illustrate that SOT recipients can act not only as sentinels for new pathogens but may be conduits for their spread to new locations or facilities.

Unfortunately, the risk for continued CP-E dissemination will remain high due to suboptimal clinical laboratory recognition of carbapenem-resistance in enteric gram-negative bacilli (as demonstrated by the initial antibiotic susceptibility results in these cases), unrecognized

“silent” fecal carriage of CP-E, and environmental contamination. While susceptibility testing systems that use ertapenem as the class agent for carbapenems have been shown to be vastly improved at detecting CP-E, they are not infallible (16). Other detection methods under investigation include the use of CHROMagar optimized for CP-E detection and the assessment of potentiation of carbapenem resistance by boronic acid compounds (28,29). The optimal method for detection and screening for carbapenemase production in Enterobacteriaceae remains to be defined.

The optimal therapy for serious CP-E infection is also uncertain, and treatment of disease in SOT patients poses several additional challenges. Surgical debridement of devitalized tissue and drainage of collections remains a cornerstone for effective treatment of local infections. Reducing the level of immunosuppression may also offer the best opportunity for control of these infections, especially in cases where overimmunosuppression may have been an important contributing factor, such as the cases presented here. CP-E isolates are routinely resistant to many classes of antibiotics, often leaving polymyxins and tigecycline as the only remaining antimicrobials with *in vitro* activity against these organisms (30). In recent years, the polymyxins (colistin and polymyxin B) have been resurrected due to the emergence of carbapenem resistance in enteric and nonfermentative gram-negative bacilli. In the modern era, polymyxin-induced nephrotoxicity and neurotoxicity appears to be less common and severe compared to the historic experience (31). However, polymyxin toxicities remain important clinical considerations, and whether these toxicities are more frequent or severe in transplant patients receiving calcineurin inhibitors (with their associated nephrotoxicity and neurotoxicity) remains to be determined.

Another concern is the emergence of polymyxin resistance in CP-E. Rates of polymyxin resistance in KPC-producing *K. pneumoniae* reached 9–27% during recent outbreaks in New York, and an outbreak of colistin-resistant, metallo- β -lactamase-producing *K. pneumoniae* has been reported in a Greek intensive care unit (18,32,33). We believe that use of polymyxins for selective bowel decontamination prior to LT should be abandoned; not only is it doubtful that the practice reduces post-operative infections, but it is unambiguously linked to the development of colistin resistance (34–36).

Tigecycline is a new semisynthetic glycylcycline antibiotic that has *in vitro* activity against a broad array of nosocomial pathogens, including most Enterobacteriaceae, and is approved for the treatment of complicated skin and skin structure and complicated intra-abdominal infections (37). A recent survey of 104 CP-E isolates collected from medical centers located in North America, Europe and Latin America failed to identify a single strain that was tigecycline resistant (38). By contrast, 11.9% of the isolates were resistant to polymyxins. However, the efficacy of tigecycline to treat CP-E infection (especially endovascular infection) has been called into question, and treatment-associated increases in tigecycline MICs have been observed during therapy (22,39). Furthermore, tigecycline is not active against *Pseudomonas* species and has reduced activity against genera of the tribe Proteaceae (*Proteus*, *Providencia*, and *Morganella*).

In conclusion, carbapenem-resistant Enterobacteriaceae are perfectly suited to become important pathogens in SOT recipients. Once largely confined to certain geographic areas, these organisms are rapidly spreading to new institutions and locations around the world. Unfortunately, few new agents are in the pipeline that may have activity against these pan-resistant organisms. In what has been described as a “microbial perfect storm” of exploding drug resistance and neglected antibiotic development, pan-resistant CP-E and the specter of XDR (extreme drug resistance) Enterobacteriaceae represents an enormous threat to solid organ transplantation (40,41). Strict infection control measures, aggressive antibiotic stewardship, and improved screening strategies will be needed to curtail their spread.

Further research is urgently needed to determine the clinical impact and optimal therapeutic and prevention strategies for CP-E infection in organ transplantation.

Acknowledgments

We thank Joanne Carrol and Frankie Brewster for technical assistance and David C. Hooper (Massachusetts General Hospital) for providing us reference strain *E coli* V517.

Financial support:

Work in the Sifri lab is supported by a Howard Hughes Medical Institute Early Career Award.

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Fig. 1. Modified Hodge tests of *K. pneumoniae* isolates from patient 1. A 10- μ g imipenem disk was placed in the center of a MHA plate shortly after being inoculated with the carbapenem-sensitive strain *E. coli* ATCC 25922, and isolates Kpn1014 and Kpn1016 were struck radially from the disk to the plate edge. Kpn1014 is *bla*_{KPC-2}-negative and shows a concentric zone of growth inhibition at its intersection with ATCC 25922, whereas invagination of ATCC 25922 growth into the imipenem zone of inhibition adjacent to Kpn1016 is indicative of carbapenemase production.

Fig. 2.

(A) Plasmid DNA profiles of *Klebsiella* isolates from patient 1 (Kpn1014, Kpn1016, and Kox1015) and patient 2 (Kpn1017) resolved on a 0.6% agarose gel. *E. coli* V517 supercoiled plasmid standards (kb). C, chromosomal DNA. (B) Southern blot of the gel shown in panel A after hybridization with an 1009-bp *bla*_{KPC-2}-specific probe. DNA sequence-confirmed *bla*_{KPC-2} PCR product (right-most lane) loaded one hour before completion of the electrophoresis served as the positive control.

Fig. 3. Pulsed-field gel electrophoresis profiles and dendrogram of *Xba*I macrorestricted chromosomal DNA fragments of the *K. pneumoniae* isolates from patient 1 (Kpn1014, Kpn1016), patient 2 (Kpn1017), and 22 additional KPC-producing *K. pneumoniae* isolates.

Carbapenem susceptibilities and carbapenemase characterization of the clinical *Klebsiella* isolates

Table 1

Patient	Isolate	Species	Modified Hodge test	bla _{KPC} PCR	Source	Day of isolation ¹	Duration of carbapenem therapy ²	meropenem MIC (µg/ml) ³		ertapenem MIC (µg/ml) ³		imipenem disk diffusion (mm)
								Vitek 2	Microbroth dilution	Vitek 2	Microbroth dilution	
Case 1												
	Kpn1014	<i>K. pneumoniae</i>	-	-	Blood	0	0	≤ 0.25	≤ 0.25	n/d	≤ 0.5	28
	Kox1015	<i>K. oxytoca</i>	+	+	Drain	12	10	1	8	≥ 8	16	16
	Kpn1016	<i>K. pneumoniae</i>	+	+	Drain	12	10	≥ 16	4	≥ 8	8	17
Case 2												
	Kpn1017	<i>K. pneumoniae</i>	+	+	Blood	47	0	4	16	4	8	17

¹ Compared to the day of isolation of *K. pneumoniae* Kpn1014 from patient 1

² Days of carbapenem therapy prior to culturing the isolate

³ Boldface values indicate intermediate or resistant susceptibilities
n/d, not determined

Table 2

Summary of clinical and microbiological characteristics of carbapenemase-producing Enterobacteriaceae infections in solid organ transplant recipients

Patient	Ref No.	Age (y)	Locale	Transplant	Time after transplant	CP-E organism(s) isolated	Carbapenemase	Source	Treatment	Outcome
1	8	NR	Italy	Kidney	NR	<i>K. pneumoniae</i>	VIM-1	Blood	TZP	Survived
2	8	NR	Italy	Kidney	NR	<i>K. pneumoniae</i>	VIM-1	Blood, urine	MER	Survived
3	8	NR	Italy	Liver	NR	<i>K. pneumoniae</i>	VIM-1	Blood	MER	Died
4	8	NR	Italy	Liver	NR	<i>K. pneumoniae</i>	VIM-1	Blood, wound	AMK CIP LEV MER SXT TZP	Survived
5	8	NR	Italy	Liver	NR	<i>K. pneumoniae</i>	VIM-1	Blood	AMK MER	Died
6	19	44	New York City	Kidney	26 m	<i>K. pneumoniae</i>	KPC-2	Blood	NR	Survived
7	20	55	Italy	Liver	46 d	<i>Enterobacter cloacae</i>	VIM-1	Pleural fluid, BAL, bile	AMK COL ¹ IMP RIF	Died
8	21	NR	Michigan	Heart	NR	<i>K. oxytoca</i>	KPC-2	Sputum	COL ² GEN TGC	Survived
9	22	57	Pennsylvania	NR	NR	<i>K. pneumoniae</i>	KPC	Pleural fluid, BAL	GEN TGC	Died
10	23	54	Texas	Liver	45 d	<i>E. cloacae</i> ³	KPC-2	Blood, wound	AMK CIP	Died
11	PR	45	Virginia	Liver	55 d	<i>K. pneumoniae</i> , <i>K. oxytoca</i>	Both KPC-2	Blood, sputum, abdominal drain, urine	AMK COL ¹ TGC	Died
12	PR	64	Virginia	Liver	46 d	<i>K. pneumoniae</i>	KPC-2	Blood	COL ¹	Died

Abbreviations: BAL, bronchoalveolar lavage; d, days; m, months; NR, not reported; PR, present report; y, years

Antimicrobial abbreviations: AMK, amikacin; CIP, ciprofloxacin; COL¹, colistin parenteral; COL², colistin inhaled; GEN, gentamicin; IMP, imipenem; LEV, levofloxacin; MER, meropenem; RIF, rifampin; SXT, trimethoprim-sulfamethoxazole; TGC, tigecycline; TZP, piperacillin-tazobactam

³ Also bacteremic with KPC-2 producing *Pseudomonas putida*