

# A Case of Carbapenem Resistant Non-K1/K2 Serotype *Klebsiella pneumoniae* Liver Abscess

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## ABSTRACT

*Klebsiella pneumoniae* liver abscess (KPLA) has been described as an invasive syndrome with extrahepatic complications. The majority of KPLA is caused by capsular serotype K1 and K2 isolates. We report a case of carbapenem resistant *Klebsiella pneumoniae* liver abscess. The patient initially presented with infected right above-the-knee amputation and was later found with a large liver abscess. Initial antimicrobial susceptibility showed carbapenem resistant *K. pneumoniae* (CRKP). Further molecular workup revealed that the isolate was a less virulent non-K1/K2 serotype, and both *rmpA* and *kfu* genes were negative. The lack of outer membrane porins likely contributed to the carbapenem resistance. To our knowledge, this is a first reported case of carbapenem resistant, non-K1/K2 serotype *K. pneumoniae* liver abscess in the United States.

**Keywords:** Liver Abscess; *Klebsiella pneumoniae*; Capsular Serotype; Carbapenem Resistance

## 1. Introduction

*Klebsiella pneumoniae* liver abscess (KPLA) has been described as an invasive syndrome with extrahepatic complications via hematologic spread. The hypermucoviscosity phenotype and capsular serotype (K1 and K2) are the major virulence factors, along with capsular polysaccharide synthesis regulator gene *rmpA*, iron chelator aerobactin, and iron uptake gene *kfu*, which have been described to contribute to the virulence and invasive features of *K. pneumoniae* [1-3]. To our knowledge, we describe the first case of non-K1/K2 KPLA in the United States.

## 2. Case Report

A 64-year-old African American man presented to the Veterans Affairs New Jersey Healthcare System with an infected right above-the-knee amputation (AKA). The patient's hospital course was complicated by poor wound healing, wound debridement, revision of the AKA, and a urinary tract infection with *Pseudomonas aeruginosa* and *Klebsiella pneumoniae*, for which he was treated with doripenem. After one and a half month of hospitalization, the right AKA began to heal appropriately. On hospital

day 49, the patient developed transient right upper quadrant abdominal pain. He then developed intermittent fevers ranging from 100.7°F to 101°F, however fever workup including blood and urine cultures, as well as chest X-ray were negative. The patient did not have any further physical complaints and remained hemodynamically stable.

On hospital day 57, the patient was febrile (temperature, 102°F), with a heart rate of 121 beats per minute and blood pressure of 155/77 mm-Hg. The physical examination was unremarkable including normal thoracic and abdominal findings. The white cell count was 19,600 cell/μL with 90 percent neutrophils; the hemoglobin level was 8.5 g/dL, and the platelet count was 377,000 cell/μL. The aspartate aminotransferase and alanine aminotransferase were mildly elevated, 38 IU/L and 51 IU/L, respectively. The total bilirubin level was 0.6 mg/dL. A computerized axial tomography scan of the thorax and abdomen showed a large loculated abscess in the right lobe of the liver and intrahepatic biliary ductal dilatation (**Figure 1**). Piperacillin-tazobactam was started empirically, and the patient underwent fine-needle aspiration of the liver abscess and pigtail placement by interventional radiology. Culture of the liver abscess grew carbapenem

resistant *Klebsiella pneumoniae* (CRKP), which prompted change of antibiotic therapy to tigecycline. Follow up computerized axial tomography of the abdomen at the end of two week treatment showed significantly diminished liver abscess. However, there was communication between the abscess and the gallbladder. The patient underwent laparoscopic cholecystectomy. He remained asymptomatic and was discharged to a nursing home.

Antimicrobial susceptibility of the CRKP isolate was determined by using the broth microdilution test [4], except for that of ertapenem, which was determined by E-test strip. MICs are as follows: ceftazidime, >128 µg/mL; ceftriaxone, 128 µg/mL; cefepime, >16 µg/mL; ciprofloxacin, >2 µg/mL; gentamicin, 16 µg/mL; ertapenem, 6 µg/mL; imipenem, 1 µg/mL; and meropenem, <1 µg/mL. Capsular serotyping of the isolate was performed as previously described [5,6], and it was a non-K1/K2 serotype. Serum bactericidal assay [7], neutrophil phagocytosis [8,9], and mice lethality studies were performed. All animal care procedures and protocols were approved by the Institutional Animal Care and Use Committee of the National Health Research Institute (NHRI-IACUC-096004-A). Susceptibility to serum killing and neutrophil phagocytosis, and LD<sub>50</sub> ≥ 1 × 10<sup>7</sup> CFU were consistent with the less virulent non-K1/K2 serotype. The colonies were phenotypically mucoid but string test negative.

Polymerase chain reaction (PCR) for *rmpA*, *kfu*, aerobactin, SHV β-lactamase, and *K. pneumoniae* carbapenemase (KPC) genes were performed as previously described [10]. *rmpA*, aerobactin, and *kfu* genes were not present in the isolate. SHV-11 and SHV-12 genes were detected. The KPC gene was not detected. The outer membrane porins (OMPs) were isolated according to the rapid procedure of Chen *et al.* [11]. OMPs were extracted with sodium lauroylsarcosinate (Sigma) and recovered by ultracentrifugation as described previously [12]. The OMP profiles were determined by sodium dodecyl sul-

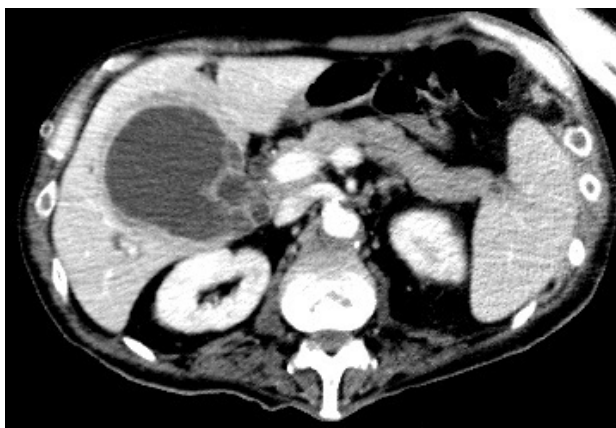


Figure 1. Computerized axial tomography of the abdomen showed a loculated abscess in the right lobe of the liver.

phate-polyacrylamide gel electrophoresis (SDS-PAGE) using 12% SDS gels and were visualized by Coomassie blue staining (Figure 2). Neither OmpK35 nor OmpK36 were present.

Pulsed-field gel electrophoresis (PFGE) and multilocus sequence typing (MLST) of the CRKP isolate and two other known KPC positive CRKP isolates from the same institution were done as previously described [13-17] (Figure 3). Sequence type (ST) 258 was identified in all three isolates.

### 3. Discussion

In the past two decades, *Klebsiella pneumoniae* has become the primary cause of liver abscess in Southeast Asia, including South Korea and Taiwan [18]. There is also increasing incidence in North America with the majority of patients being of Asian descent [19,20]. KPLA has been described as an invasive syndrome as it can cause extrahepatic complications via hematologic spread.

The major virulence factors in KPLA are the hypermucoviscosity phenotype and capsular serotype (K1 and K2). The presence of hypermucoviscosity is confirmed by a positive string test. Isolates of capsular serotype K1/K2 are the most prevalent serotypes found in KPLA, accounting for more than seventy percent of liver abscess strains reported [1-3]. There have been several case reports of confirmed K1 and K2 serotype liver abscesses in the United States [21,22]. Due to impaired neutrophil phagocytosis, K1/K2 serotypes are found exclusively in disseminated cases [18]. Other virulence factors, such as capsular polysaccharide synthesis regulator gene *rmpA*, iron chelator aerobactin, and iron uptake gene *kfu*, have been described to contribute to the virulence and invasive features of *K. pneumoniae*. Even though isolates of non-K1/K2 serotypes are less virulent, the presence of hypermucoviscosity phenotype, *rmpA*, aerobactin, and *kfu* genes in these isolates make them capable of causing

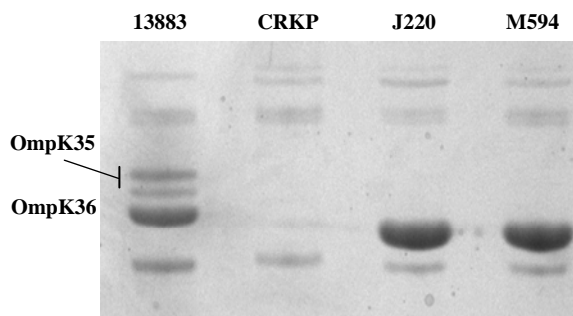
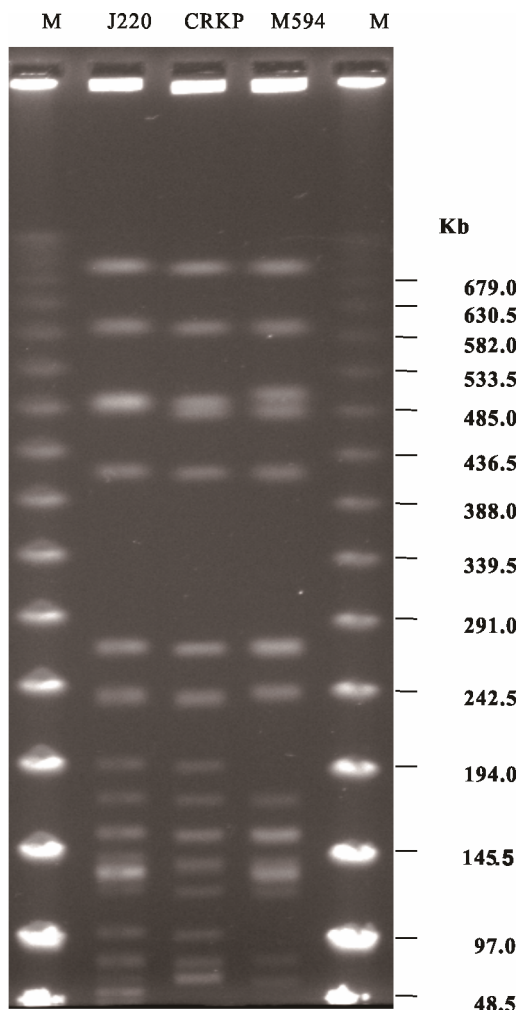


Figure 2. Outer membrane profiles of the case CRKP isolate and two other known CRKP isolates from the same institution, J220 and M594. Lane 1, ATCC13883, is the control strain with only intrinsic resistance to ampicillin. Neither OmpK35 nor OmpK36 are presented in the case isolate.



**Figure 3. Pulsed-field gel electrophoresis of the case CRKP isolate and two other known CRKP isolates. All three isolates are clonally related.**

liver abscesses [1]. However, these virulent factors were not present in the case isolate. To our knowledge, there has not been a case of nosocomial liver abscess caused by a non-K1/K2 serotype *K. pneumoniae* reported in the United States.

Cephalosporin or combination therapy has been used to treat KPLA. Liver abscesses caused by extended spectrum  $\beta$ -lactamase (ESBL)-producing *K. pneumoniae* are uncommon [18]. Carbapenems are the antibiotic mainstay of treatment for organisms harboring ESBLs.

However, with the emergence of CRKP and particularly the epidemic in the northeastern United States, treatment options are limited to colistin and tigecycline [23]. Both  $\beta$ -lactamase genes, SHV-11 and SHV-12, were identified in the case isolate. PFGE and MLST of the case isolate and two known KPC positive CRKP isolates from the same institution showed that the isolates are clonally related, with all three isolates displaying sequence type 258. The KPC gene however, was not de-

tected in the case isolate. We suspect that it may have been lost due to plasmid instability during storage. Our patient received two weeks of tigecycline, however his uneventful recovery was largely due to mechanical removal of the liver abscess via percutaneous drainage.

In addition to the production of carbapenemase, deficiency in outer membrane porins has been proposed to increase carbapenem resistance [24]. Most ESBL-producing *K. pneumoniae* strains have only OmpK36, whereas the majority of non-ESBL-producing *K. pneumoniae* have both OmpK35 and OmpK36. As with our case isolate, mutant strains with double deletion of OmpK35 and OmpK36 have been found to have increased antimicrobial resistance but decreased virulence due to weakened metabolic fitness and slowed growth rate [24]. This most likely was attributed to the case isolate's high ertapenem MIC despite being KPC negative.

Despite the low virulence of non-K1/K2 serotypes of *K. pneumoniae*, they are capable of causing liver abscess due to presence of hypermucoviscosity phenotype, *rmpA*, aerobactin, and *kfu* genes. To our knowledge, this is the first reported case of non-K1/K2 serotype KPLA in the United States. Commonly associated virulence factors were also absent in this isolate. With increasing antimicrobial resistance, health care providers should be aware of the emergence of CRKP liver abscesses and use appropriate antimicrobial agents when multidrug resistant organisms are suspected.

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