J Infect Chemother (2009) 15:248–251
DOI 10.1007/s10156-009-0695-7


CASE REPORT

**Yoshitaka Kohayagawa · Keiko Nakao · Misuzu Ushita
Norio Niino · Masayuki Koshizaki · Yuji Yamamori
Yusuke Tokuyasu · Hiroshi Fukushima**

# Pyogenic liver abscess caused by *Klebsiella pneumoniae* genetic serotype K1 in Japan

Received: December 1, 2008 / Accepted: April 6, 2009

**Abstract** Pyogenic liver abscess caused by *Klebsiella pneumoniae* is an emerging disease worldwide, and we know the serotype K1 strain to be the most virulent strain. We report a Japanese case of septic pyogenic liver abscess caused by *K. pneumoniae* genetic serotype K1. A 60-year old man presented at our hospital in a state of cardiopulmonary arrest. From the patient's chief complaint of chest pain, we suspected acute coronary syndrome, i.e., acute myocardial infarction. We used extracorporeal circulation and checked coronary angiography, but the 75% stenosis by itself could not adequately account for the patient's critical condition. The patient's laboratory data indicated multiple organ failure. The patient's condition did not improve while in intensive care and he died 20 h after the onset of the cardiopulmonary arrest. Pathological autopsy later showed colliquative necrosis in the deltoid and left greater pectoral muscles, as well as liver abscesses. The patient's blood, gastric juice, and stool cultures all grew a Gram-negative bacillus identified as *Klebsiella pneumoniae*. We also performed capsular polysaccharide synthesis (*cps*) genotyping by polymerase chain reaction for the detection of K serotype-specific alleles at the *wzx* and *wzy* loci. The result indicated that *wzx*_K1 and *wzy*_K1 were positive. This is the first reported Japanese case of septic pyogenic liver abscess caused by *K. pneumoniae* genetic serotype K1.

**Key words** *Klebsiella pneumoniae* · K1 · Liver abscess · PCR · *magA* · Sepsis

Y. Kohayagawa · K. Nakao · M. Ushita · N. Niino · M. Koshizaki · Y. Yamamori
Department of Critical Care and Emergency Medicine, Shimane Prefectural Central Hospital, 4-1-1 Himebara, Izumo, Shimane 693-8555, Japan
Tel. +81-853-22-5111; Fax +81-853-21-2975
e-mail: kahedon@spch.izumo.shimane.jp

Y. Tokuyasu
Department of Clinical Pathology, Shimane Prefectural Central Hospital, Shimane, Japan

H. Fukushima
Shimane Prefectural Institute of Public Health and Environmental Science, Shimane, Japan

## Introduction

*Klebsiella pneumoniae* is a Gram-negative pathogen that causes a variety of infections, especially urinary tract infections and pneumonia.[1] Primary pyogenic liver abscess caused by community-acquired *K. pneumoniae* has recently become an emerging disease. Many cases of *K. pneumoniae* liver abscess with septic metastatic infection have been reported in Taiwan, but few cases have been reported in the rest of Asia, or in North America or Europe.[2–9] Despite advances in intensive care medicine, pyogenic liver abscess is still a catastrophic illness with a high morbidity and mortality rate.[10] Capsular serotype K1 *K. pneumoniae* and *magA* (mucoviscosity-associated gene A) gene-positive *K. pneumoniae* have been reported to be extremely virulent.[4,11–13] There are few reports of pyogenic liver abscess in Japan, and the genotype and/or serotype of *K. pneumoniae* were not investigated in these reports.

In this case report, we report a Japanese case of pyogenic liver abscess with septic metastatic infection caused by *K. pneumoniae* genetic serotype K1.

## Case report

A 60-year-old man presented at a hospital in Shimane with complaints of fatigue plus left chest and shoulder pain. The patient worked at a factory that makes boiled fish paste. He had been absent from work for the previous 5 days. He lived alone and his past medical history was unknown. He walked to the emergency room (ER) and lay on a bed. The electrocardiogram monitor indicated sinus tachycardia (180 beats/min), but suddenly a flat line (asystole) occurred. An ER physician immediately started cardiopulmonary resuscitation. The patient was suspected of having cardiac disease, e.g., acute myocardial infarction, and was referred to our tertiary care medical center.

On his arrival at the medical center, he was still in asystole. The patient received percutaneous cardiopulmonary support and intraaortic balloon pumping. Cardioangiogra-

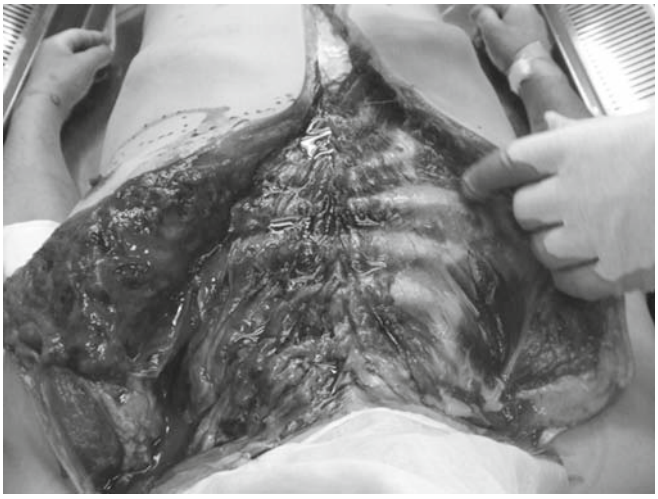

**Fig. 1.** Colliquative necrosis in the left chest (at autopsy)

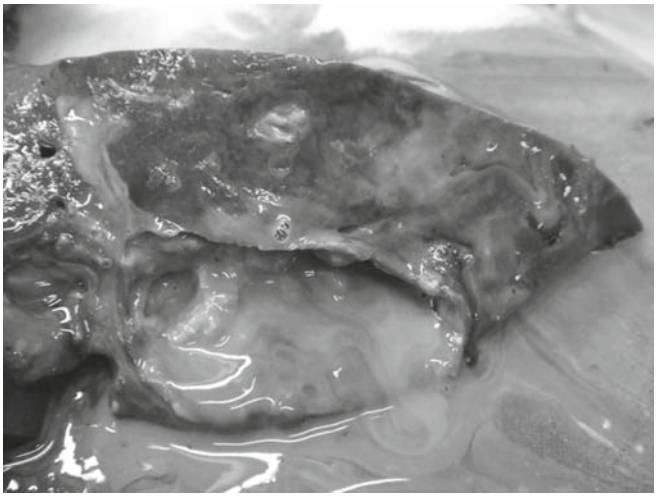

**Fig. 2.** Liver abscesses (at autopsy)

phy revealed stenosis covering 75% of the anterior descending artery.

Laboratory evaluation indicated a highly elevated white blood cell count (18600/μl), aspartate aminotransferase (AST), 1165 IU/l; alanine aminotransferase (ALT), 306I U/l; blood urea nitrogen (BUN), 77.4 mg/dl; creatinine, 2.29 mg/dl; myoglobin, 1971 ng/ml; creatine kinase (CK), 322 IU/l; creatine kinase muskle-brain fraction (CK-MB), 39 IU/l; C-reactive protein, 22.4 mg/dl; and hemoglobin A1c, 11.9%; as well as disturbed mineral balance (Na, 118.5 mmol/l; K, 8.5 mmol/l; Cl, 83.6 mmol/l). Blood, gastric juice, and stool samples were collected for bacterial culture. Chest X-ray demonstrated subcutaneous emphysema. Computed tomography could not be performed. After coronary angiography, the patient was treated in the intensive care unit. He did not respond at all, and his relatives requested conservative treatment. So, we continued with conservative treatment, and he died 20 h after the onset of the cardiopulmonary arrest.

Pathological autopsy was performed and showed macroscopically colliquative necrosis in the left greater pectoral muscle and deltoid muscle, as well as liver abscesses (Figs. 1, 2). These abscesses included gas. There were also microabscesses in cardiac muscle, bilateral adrenal glands, kidneys, muscle, lung, and throughout the whole body. We observed Gram-negative bacilli in the intravascular space of the heart and in the kidneys, muscles, and lung. In the gastrointestinal tract, we observed intramucosal inflammation with fibrosis in the small intestine and colon. We did not perform a cranial autopsy. The liver abscess, blood, gastric juice, and stool cultures from the patient all grew a Gram-negative bacillus identified as *Klebsiella pneumoniae*. All these *K. pneumoniae* strains showed the same tendency in drug susceptibility tests. They were susceptible to most antibacterial agents, but resistant to ampicillin, piperacillin, and fosfomycin. We performed capsular polysaccharide synthesis (*cps*) genotyping of SPCH-K0001 (a *K. pneumoniae* strain derived from liver abscess) by polymerase chain reaction for the detection of K serotype-specific alleles at the *wzx* and *wzy* loci, as previously described.[12] We used the following primers for *cps* genotyping :

*wzx*_K1-F: 5′-GTAGGTATTGCAAGCCATGC-3′, and *wzx*_K1-R: 5′-GCCCAGGTTAATGAATCCGT-3′; *wzy*_K1-F: 5′-GGTGCTCTTTACATCATTGC-3′, and *wzy*_K1-R: 5′-GCAATGGCCATTTGCGTTAG-3′; *wzx*_K2-F: 5′-GGAGCCATTTGAATTCGGTG-3′, and *wzx*_K2-R: 5′-TCCCTAGCACTGGCTTAAGT-3′; *wzy*_K2-F: 5′-GGATTATGACAGCCTCTCCT-3′, and *wzy*_K2-R: 5′-CGACTTGGTCCCAACAGTTT-3′).

For the genotyping procedure, we extracted genomic DNA from the tested strains as templates. Initial denaturation at 96°C for 3 min was followed by denaturation at 96°C for 30 s, annealing at 56°C for 15 s, and extension at 74°C for 1 min for 30 cycles. There was a final 10-min hold at 72°C. *wzx*_K1 and *wzy*_K1 were positive (Fig. 3).

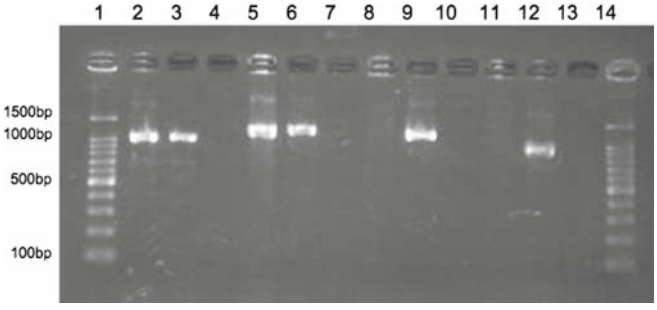


**Fig. 3.** Polymerase chain reaction (PCR) for detection of K serotype-specific alleles at *wzx* and *wzy* loci: *lanes 1 and 14*, size marker; *lanes 2–4*, PCR for *wzx*_K1; *lanes 5–7*, PCR for *wzy*_K1; *lanes 8–10*, PCR for *wzx*_K2; *lanes 11–13*, PCR for *wzy*_K2. *Lanes 2, 5, 8, and 11* are SPCH-K0001; *lanes 3 and 6* are K1 strain (NTUH-K2044) as a K1 positive control; *lanes 4, 7, 10, and 13* are pure water as a negative control; *lanes 9 and 12* are SPCH-K0002 (detected previously in our hospital) as a K2 positive control

## Discussion

To the best of our knowledge, this is the first case of septic metastatic infection caused by *K. pneumoniae* genetic serotype K1 in Japan. We were overwhelmed by the rapidly progressive state of the disease. The principal virulence factor that has been described for *K. pneumoniae* is its polysaccharide capsule, which occurs in more than 70 antigenic varieties.[14] The *K. pneumoniae cps* gene cluster has a conserved organization similar to the *Escherichia coli* group 1 capsule biosynthesis gene cluster.[11,15,16,17] The *wzx* and *wzy* alleles are the serotype-specific alleles that produce the integral inner membrane proteins WZX and WZY. *magA* is the serotype K1 *wzy* allele (*wzy*_K1).[12] Chuang et al.[11] reported that all *magA*-positive strains were capsular serotype K1, whereas none of the *magA*-negative strains were serotype K1. Antisera Seiken (Denka Seiken, Tokyo, Japan) was unavailable for sale in Japan, so we could not directly investigate the serotype. Therefore, we determined our strain to be serotype K1 by investigating only the genotype, but not the serotype of *K. pneumoniae* (genetic serotyping).

Our patient had been living alone and we had no information about his medical history. The condition at onset was not clear. Our laboratory data indicated at least a possibility of diabetes mellitus, because hemoglobin A1c was 11.9%. Fung et al.[13] reported *K. pneumoniae* serotype K1 to be significantly associated with liver abscess and the complication of endophthalmitis, especially in diabetic patients. Lin et al.[18] have reported that poor glycemic control plays a role in impairing the neutrophil-mediated phagocytosis of K1/K2 *K. pneumoniae*, but does not significantly affect the phagocytosis of non-K1/K2 *K. pneumoniae*. However, Fang et al.[12] reported that in cases of septic ocular or central nervous system complications from pyogenic liver abscess, genotype K1 was the only significant predictor of these complications. Chen et al.[10] reported that the clinical outcome of patients with pyogenic liver abscess admitted to the intensive care unit was not significantly different between patients with diabetes mellitus and patients without. Thus, the importance of diabetes mellitus in this regard remains controversial.

Why is infection with *K. pneumoniae* serotype K1 so catastrophic? The mechanism by which serotype K1 *Klebsiella pneumoniae* increases pathogenicity is not well understood. One reason is its hypermucoviscous form, which is characterized by the formation of elongated (>5 mm) mucoviscous strings from a colony. The *mag A*-positive strains were found to form a capsule-associated mucopolysaccharide web, and increased serum resistance as well as resistance to phagocytosis was shown.[4] Wu et al.[19] recently reported that Toll-like receptor 4 (TLR4) recognition of *magA*-positive *K. pneumoniae* was interrupted by the mucoviscosity of the *magA*-positive *K. pneumoniae* capsular polysaccharides (CPS). The role of TLR4 is important for the recognition of lipopolysaccharide (LPS).[20] The *magA*-positive *K. pneumoniae* CPS hinders the underlying bacterial component LPS, and heat-treatment disruption of CPS structure results in more tumor necrosis factor (TNF)-α production via interaction with TLR4 and underlying LPS. TNF-α is essential for the prevention of bacterial attacks. Thus, *magA*-positive *K. pneumoniae* causes TNF-α production to be reduced, thereby causing the host to be immunocompromised.

Susceptibility tests for the strain isolated from our patient indicated good susceptibility to antimicrobial drugs. However, as extended-spectrum beta-lactamase (ESBL)-producing *K. pneumoniae* is increasing in Japan,[21] we should choose appropriate antimicrobial drugs according to the situation.

At present, the *K. pneumoniae* serotype does not affect the choice of treatment, but it is important to investigate the *K. pneumoniae* serotype because in the future treatments may be based on the serotype. Recently, Wu et al.[19] reported that monoclonal antibodies against CPS of *magA*-positive *K. pneumoniae* protected mice from *magA*-positive *K. pneumoniae*-induced liver abscess formation and lethality. With a rapid-diagnosis system (i.e, real-time polymerase chain reaction [PCR]) for the detection of the *magA* gene, the administration of monoclonal antibodies would become the main therapy for serotype K1 *K. pneumoniae*-induced liver abscess. In Japan, even medical professionals are often unaware of pyogenic liver abscess caused by *K. pneumoniae* serotype K1 and its catastrophic consequences. Further research about this disease is necessary.

**Acknowledgments** The authors thank Professor Jin-Town Wang (Department of Microbiology, National Taiwan University College of Medicine) for the gift of chromosomal DNA (NTUH-K2044) and his excellent advice.

## References

1. Michel SD. Enterobacteriaceae (chapter 215). In: Mandell, Douglas, and Bennett's principles and practice of infectious diseases. 6th ed. Philadelphia, Pennsylvania: Elsevier; 2005. p. 2567–86.
2. Chang SC, Fang CT, Hsueh PR, Chen YC, Luh KT. *Klebsiella pneumoniae* isolates causing liver abscess in Taiwan. Diagn Microbiol Infect Dis 2000;37:279–84.
3. Cheng DL, Liu YC, Yen MY, Liu CY, Wang RS. Septic metastatic lesions of pyogenic liver abscess. Their association with *Klebsiella pneumoniae* bacteremia in diabetic patients. Arch Intern Med 1991;151:1557–9.
4. Fang CT, Chuang YP, Shun CT, Chang SC, Wang JT. A novel virulence gene in *Klebsiella pneumoniae* strains causing primary liver abscess and septic metastatic complications. J Exp Med 2004;199:697–705.
5. Fang FC, Sandler N, Libby SJ. Liver abscess caused by *magA*+ *Klebsiella pneumoniae* in North America. J Clin Microbiol 2005;43:991–2.
6. Ko WC, Paterson DL, Sagnimeni AJ, Hansen DS, Von Gottberg A, Mohapatra S, et al. Community-acquired *Klebsiella pneumoniae* bacteremia: global differences in clinical patterns. Emerg Infect Dis 2002;8:160–6.
7. Lee SS, Chen YS, Tsai HC, Wann SR, Lin HH, Huang CK, et al. Predictors of septic metastatic infection and mortality among patients with *Klebsiella pneumoniae* liver abscess. Clin Infect Dis 2008;47:642–50.
8. Wang JH, Liu YC, Lee SS, Yen MY, Chen YS, Wang JH. Primary liver abscess due to *Klebsiella pneumoniae* in Taiwan. Clin Infect Dis 1998:26:1434–8.

9. Yang CC, Yen CH, Ho MW, Wang JH. Comparison of pyogenic liver abscess caused by non-*Klebsiella pneumoniae* and *Klebsiella pneumoniae*. J Microbiol Immunol Infect 2004;37:176–84.
10. Chen W, Chen CH, Chiu KL, Lai HC, Liao KF, Ho YJ, et al. Clinical outcome and prognostic factors of patients with pyogenic liver abscess requiring intensive care. Crit Care Med 2008;36: 1184–8.
11. Chuang YP, Fang CT, Lai SY, Chang SC, Wang JT. Genetic determinants of capsular serotype K1 of *Klebsiella pneumoniae* causing primary pyogenic liver abscess. J Infect Dis 2006;193:645–54.
12. Fang CT, Lai SY, Yi WC, Hsueh PR, Liu KL, Chang SC. *Klebsiella pneumoniae* genotype K1: an emerging pathogen that causes septic ocular or central nervous system complications from pyogenic liver abscess. Clin Infect Dis 2007;45:284–93.
13. Fung CP, Chang FY, Lee SC, Hu BS, Kuo BI, Liu CY, et al. A global emerging disease of *Klebsiella pneumoniae* liver abscess: is serotype K1 an important factor for complicated endophthalmitis? Gut 2002;50:420–4.
14. Sahly H, Podschun R, Ullmann U. Klebsiella infections in the immunocompromised host. Adv Exp Med Biol 2000;479:237–49.
15. Dong C, Beis K, Nesper J, Brunkan-Lamontagne AL, Clarke BR, Whitfield C, et al. Wza the translocon for *E. coli* capsular polysaccharides defines a new class of membrane protein. Nature 2006; 444:226–9.
16. Rahn A, Drummelsmith J, Whitfield C. Conserved organization in the cps gene clusters for expression of *Escherichia coli* group 1 K antigens: relationship to the colanic acid biosynthesis locus and the cps genes from *Klebsiella pneumoniae*. J Bacteriol 1999;181: 307–13.
17. Whitfield C. Biosynthesis and assembly of capsular polysaccharides in *Escherichia coli*. Annu Rev Biochem 2006;75:39–68.
18. Lin JC, Siu LK, Fung CP, Tsou HH, Wang JJ, Chen CT, et al. Impaired phagocytosis of capsular serotypes K1 or K2 *Klebsiella pneumoniae* in type 2 diabetes mellitus patients with poor glycemic control. J Clin Endocrinol Metab 2006;91:3084–7.
19. Wu MF, Yang CY, Lin TL, Wang JT, Yang FL, Wu SH, et al. Humoral immunity against capsule polysaccharide protects the host from *magA*+ *Klebsiella pneumoniae*-induced lethal disease by evading Toll-like receptor 4 signaling. Infect Immun 2009;77: 615–21.
20. Poltrak A, He X, Smirnova I, Liu MY, Van HC, Du X, et al. Defective LPS signaling in C3H/HeJ and C57BL/10ScCr mice: mutations in *Tlr4* gene. Science. 1998;282:2085–8.
21. Hawkey PM. Prevalence and clonality of extended-spectrum beta-lactamases in Asia. Clin Microbiol Infect 2008;14 Suppl 1:159–65.