Could Acinetobacter pittii act as an NDM-1 reservoir for Enterobacteriaceae?

Pierre Bogaerts*, Te-Din Huang, Roberta Rezende de Castro, Warda Bouchahrouf and Youri Glupczynski

National Reference Centre for Antibiotic Resistance in Pseudomonas and Acinetobacter spp., CHU UCL Mont-Godinne-Dinant, Yvoir, Belgium

*Corresponding author. E-mail: pierre.bogaerts@uclouvain.be

Keywords: carbapenemases, non-fermenters, plasmids, type IV secretion systems

Sir,

Carbapenemases are the most common mechanism of carbapenem resistance and include β-lactamases of Ambler classes A, B (metallo-β-lactamases) and D. Among the carbapenemases, the New Delhi metallo-β-lactamase-1 (NDM-1) has been recovered worldwide in Gram-negative bacteria, especially in Enterobacteriaceae, but also in Pseudomonas aeruginosa and in several Acinetobacter species, including Acinetobacter baumannii and other Acinetobacter species isolated from clinical samples, environmental samples and food animals.1–4 Several reports showed that blaNDM-1 is frequently located on mobile genetic elements inserted into the chromosome or on plasmids that often carry several additional resistance genes.1 Here, we report a case of infection due to an NDM-1-producing Acinetobacter pittii recovered in Belgium from an immunocompromised patient who had previously travelled to Egypt and to north-west India (Rajasthan).

In 2012, a patient was admitted to hospital for an episode of acute alcoholic pancreatitis. This patient had received successive courses of treatment with various antimicrobial broad-spectrum antibiotics, including amoxicillin/clavulanate and amikacin, piperacillin/tazobactam and meropenem. A rectal swab obtained upon admission for the screening of asymptomatic carriage of carbapenemase producers was cultured on a selective Brilliance CRE agar plate (Oxoid, Cambridge, UK), but yielded no growth after 24 h.

A few days later, the patient developed severe abdominal pain and high-grade fever with elevated serum amylase and lipase levels. An abdominal CT scan image showed the presence of multiple pancreatic pseudocysts, which led to surgical drainage of the intra-abdominal collections. Perioperative abdominal fluid and liquid from pancreatic cyst punctures grew an A. pittii isolate, 2012276, identified by matrix-assisted laser desorption/ionization–time of flight mass spectrometry (MALDI-TOF MS; Bruker Daltonik, Bremen, Germany), which displayed resistance to piperacillin/tazobactam (MIC >64/4 mg/L), ceftazidime (MIC >256 mg/L), cefepime (MIC >256 mg/L), meropenem, doripenem and imipenem (MIC >32 mg/L) and susceptibility to ciprofloxacin (MIC 0.5 mg/L), gentamicin (MIC 2 mg/L), tobramycin (MIC ≤1 mg/L), amikacin (MIC 16 mg/L) and colistin (MIC 0.5 mg/L) by the broth microdilution method using CLSI interpretative criteria.5 The production of a metallo-β-lactamase was confirmed by the Neo-Sensitabs® combined disc test (Rosco Diagnostica, Taastrup, Denmark) using meropenem and dipicolinic acid as an inhibitor of metallo-β-lactamases (results not shown).

The presence of blaNDM-1 was detected using a Check-Points array (CT102, Wageningen, the Netherlands) and further analysis of the presence of β-lactamase-coding genes by endpoint multiplex PCRs targeting minor extended-spectrum β-lactamases (blaTEM, blaSHV, blaGES and blaPER), carbapenemases (blaOXA-48, blaOXA-58, blaOXA-24 and blaOXA-58), and OXA carbapenemases (blaOXA-23, blaOXA-24 and blaOXA-58)6 confirmed that A. pittii was positive for blaNDM-1 only.

Plasmid extraction revealed the presence of a 47 kb plasmid, which was easily transferred by conjugation to azide-resistant Escherichia coli J53, conferring on the recipient strain nonsusceptibility to ertapenem (MIC 16 mg/L), imipenem (MIC 4 mg/L), meropenem (MIC 2 mg/L) and doripenem (MIC 2 mg/L) (results not shown).

PCR mapping and sequencing of the genetic context of blaNDM-1 revealed a Tn125 composite transposon bracketed by two copies of ISAba125 as previously reported in A. baumannii Ab11314 reported in Belgium (Figure 1).7 This structure is also identical to the sequence described in non-baumannii Acinetobacter spp. recovered in China.8 In particular, this transposon includes an ISCR27, which could be at the origin of the acquisition of the blaNDM-1 gene by a rolling-circle mechanism from its progenitor.9 The genetic context of the transposon was determined by PCR mapping and partial sequencing using a set of 12 primer pairs described by Hu et al.9 and showed a plasmid scaffold identical to the type IV secretion system (T4SS) plasmid pNDM-BJ01 recovered from Acinetobacter lwoffii in China (results not shown). T4SS is an extremely versatile secretion system involved in conjugation, in translocation of protein effectors, such as virulence proteins, and in DNA uptake.10 A combined therapy, including 400 mg of ciprofloxacin intravenously twice daily and 3 000 000 IU of colistin intravenously four-times daily, was administered for 21 days. The patient was eventually discharged following a control abdominal CT scan that showed a significant decrease in the collections.

Non-baumannii Acinetobacter spp. expressing NDM-1 have mostly been reported in China and have not yet been described in European countries. In the present case the patient had a history...
of travel in Egypt and in India. These data further highlight the risk of worldwide dissemination of NDM-1. The fact that NDM-1 was detected on a transposon already observed in the Belgian NDM-1-producing A. baumannii, but on a different plasmid (T4SS), harbouring multiple virulence factors, which was easily transferred to E. coli, underlines the potential of A. pittii as a resistance reservoir for the dissemination of NDM-1 towards Enterobacteriaceae.

Figure 1. Close genetic context and general features of the Tn125 transposon carrying blaNDM-1 in A. pittii 2012276 recovered in Belgium. Genes and their transcription orientation are indicated by arrows (not to scale). The origin of replication of ISCR27 (ori) is indicated by a filled circle. blaNDM-1 is located on a transposon bracketed by two copies of ISAba125 inserted on a T4SS plasmid (not shown) between an aphA6 gene encoding a 3’-aminoglycoside phosphotransferase of type VI and a resolvase-coding gene. The whole plasmid is similar to the pNDM-BJ01 NDM-1-harbouring plasmid described in A. lwoffii recovered in China.9

Transparency declarations
None to declare.

References

J Antimicrob Chemother 2013
doi:10.1093/jac/dkt190
Advance Access publication 16 May 2013

Complete sequencing of an IncFII NDM-1 plasmid in Klebsiella pneumoniae shows structural features shared with other multider resistance plasmids

Akira Hishinuma1*, Atsushi Yoshida1, Hiromichi Suzuki2, Katsuko Okuzumi3 and Takeshi Ishida4

1Department of Infection Control and Clinical Laboratory Medicine, Dokkyo Medical University, Mibu, 321-0293, Tochigi, Japan; 2Clinical Laboratory Center, Dokkyo Medical University, Mibu, 321-0293, Tochigi, Japan; 3Division of Infection Control, Dokkyo Medical University, Mibu, 321-0293, Tochigi, Japan; 4Saitama Citizens Medical Center, Saitama, 331-0054, Japan

*Corresponding author. Tel: +81-282-87-2139; Fax: +81-282-86-6212; E-mail: a-hishi@dokkyomed.ac.jp

Keywords: community-acquired blaNDM-1 plasmid, horizontal transfer of blaNDM-1 gene, recombination of multidrug resistance region, metallo-β-lactamases

Sir,
Since the first report of New Delhi metallo-β-lactamase 1 (NDM-1) in 2009, it has spread worldwide in Europe, America, Africa, Asia and Australia.1 The first case was a Swedish patient who had previously been treated in India.2 Retrospective analysis of carbapenemase-producing Enterobacteriaceae traced the blaNDM-1 gene to Escherichia coli collected from an Indian hospital in 2006. Recently, blaNDM-1-producing bacteria were identified from the environment in New Delhi3 and Vietnam. Also, the blaNDM-1 gene has been found on a mobile plasmid in an Acinetobacter lwoffii isolate from chicken meat.4

We reported the first case of NDM-1-producing E. coli in Japan from a patient who had been treated in India in 2009.5 Subsequently, we determined the complete sequence of the IncFIIA blaNDM-1-positive plasmid (pNDM-1_Dok01), which suggested a possible