NDM-1 producing *Acinetobacter baumannii* isolated from a patient repatriated to the Czech Republic from Egypt, July 2011

J Hrabák (Jaroslav.Hrabak@lfp.cuni.cz)[1], M Štolbová[2], V Študentová[1], M Fridrichová[3], E Chudáčková[1], H Zemlickova[3]
1. Department of Microbiology, Faculty of Medicine in Plzeň, Charles University in Prague, Plzeň, Czech Republic
2. Department of Clinical Microbiology, Masaryk´s Hospital, Ústí nad Labem, Czech Republic
3. National Reference Laboratory for Antibiotics, National Institute of Health, Prague, Czech Republic

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We describe the isolation of an NDM-1-producing *Acinetobacter baumannii* in a Czech patient repatriated in July 2011 from Egypt. The infection spread to another patient on the same ward. Both isolates showed the same resistance pattern and were susceptible only to colistin. They had an identical PFGE pattern and belonged to the same sequence type ST 1. Sequencing of the bla_{NDM} gene identified the NDM-1 variant of the carbapenemase, surrounded by two copies of insertion sequence IS*Aba125*.

Here we describe the isolation of a New Delhi metallo-beta-lactamase-1 (NDM-1)-producing *Acinetobacter baumannii* in a Czech citizen repatriated from Egypt in July 2011. The patient was hospitalised in Egypt, and then transferred to a hospital in the Czech Republic. The patient developed ventilator-associated pneumonia caused by *A. baumannii* in addition to a primary neurological diagnosis. A carbapenem-resistant *A. baumannii* strain (V509) was isolated from bronchoalveolar lavage and an oral cavity swab. He was initially treated by meropenem and metronidazole. Due to progression of the primary disease, the patient was transferred to a long-term intensive care unit. Although the antibiotic regimen was not changed, the patient recovered according to the biochemical markers of inflammation within seven days and the antibiotic therapy was then stopped. The available data are not conclusive as to whether this patient was infected or colonised. However, the resistant isolate has been detected in low quantity in oral swab and bronchoalveolar lavage until the transfer to the long-term intensive care unit. The intensive care centre was informed about the epidemiological risk associated with this patient so that they could prepare for appropriate measures upon transfer.

A second *A. baumannii* isolate (V566) with the same resistance pattern was recovered six days later from the airways of another ventilated patient sharing the same room. The patient was treated with amoxicillin/clavulanic acid, chloramphenicol and ciprofloxacin. He died due to respiratory failure four days after the first isolation of NDM-1-producing *A. baumannii*.

Laboratory analysis

The isolates from both patients were identified as *A. baumannii* by biochemical test API ID32 GN (bioMérieux, France) and by matrix-assisted laser desorption/ionisation time-of-flight (MALDI-TOF) mass spectrometry (Bruker Daltonics, Germany). The minimum inhibitory concentrations (MICs) to 14 antibiotics were tested and the results were interpreted according to the EUCAST recommendation [1]. The isolates from both patients were resistant to all beta-lactams tested including carbapenems and other antibiotics (Table).

Typing performed by pulsed-field gel electrophoresis (PFGE) [2] showed that the isolates had indistinguishable macrorestriction patterns. Carbapenemase production was confirmed by MALDI-TOF mass spectrometry [3]. Production of metallo-beta-lactamase (MBL) activity was verified by ethylenediaminetetraacetic acid (EDTA) double-disk synergy test [4].

The bla_{NDM} gene of both isolates was amplified and sequenced as described previously [5], and revealed the NDM-1 variant of the enzyme. The bla_{NDM-1} together with other genes was located between two copies of the insertion sequence IS*Aba125* in the same orientation as found by Pfeifer et al. [6]. Because plasmid preparations from the two isolates did not yield any plasmids visible after electrophoretic separation, and no transformants were obtained after transformation experiments performed as previously described [7], it can be hypothesised that bla_{NDM-1} is located on the bacterial chromosome.

Multi-locus sequence typing (MLST) was performed [8] and the MLST database available at the website of the Pasteur Institute was used to assign the sequence type (ST). Both isolates belonged to sequence type (ST) 1 (allelic profile 1-1-1-1-5-1-1) which represents the epidemiologically successful European clone I [9].

Discussion and conclusion

Reports describing NDM-type carbapenemase producers isolated from patients previously hospitalised in high-prevalence countries have been increasing. Pfeifer et al. detected NDM-1 in *A. baumannii* isolated from a patient repatriated to Germany from Serbia in 2007 [6]. Importation of NDM-1-producing *A. baumannii* strain from Serbia has also been described by Poirel et al. [10]. Other *A. baumannii* isolates expressing NDM-1 MBL have been isolated in China and India [11,12]. It is remarkable that bla_{NDM-1} was also found on a plasmid in *A. lwoffii* in China [13]. The new NDM-2 variant was first detected in *A. baumannii* from a patient transferred from Egypt to Germany [5]. Recently, clonal spread of NDM-2-producing *A. baumannii* strains have been described in a rehabilitation ward in Israel and in the United Arab Emirates [14,15].

Until this report, no NDM-1 producing bacterium had been described in the Czech Republic, a country with a low prevalence of carbapenemase-producing bacteria [16-18]. Although routine procedures were in place in the hospital department, the strain quickly spread within one ward to another patient. After the death of the second patient and the transfer of the first patient to the long-term intensive care unit centre, the department was closed for two weeks and general cleaning including decontamination of all equipment was undertaken. No NDM-1-producing strain has been detected after the cleaning. Due to the importance of international travel in the spread of bacterial resistance, fast detection and active surveillance of bacteria producing acquired carbapenemases is needed [5-7,10,16,18,19].

We also tested the new MALDI-TOF mass spectrometry approach [3] for the detection of carbapenemase activity in the isolates. Although phenotypical detection of carbapenem-hydrolyzing enzymes in *A. baumannii* seems to be difficult by conventional methods [20], we were able to see a clear carbapenemase activity by this assay. Further validation, however, is necessary.

Table

Antimicrobial susceptibility of the NDM-1-producing *Acinetobacter baumannii* isolates, Czech Republic, July 2011 (n=2)

Antibiotic	MIC [µg/ml]
Ampicillin-sulbactam	32
Piperacillin	› 64
Piperacilin-tazobactam	› 64
Ceftazidime	› 32
Cefepime	› 32
Meropenem	› 32
Ciprofloxacin	› 32
Nalidixic acid	› 64
Gentamicin	32
Amikacin	32
Tetracycline	64
Chloramphenicol	8
Colistine	‹ 0.5
Trimethoprim-sulfamethoxazole	16

MIC: minimum inhibitory concentrations; NDM-1: New Delhi metallo-beta-lactamase-1.

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