Extensively drug-resistant *Raoultella planticola* carrying multiple resistance genes including *bla*<sub>NDM-1</sub>

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**Introduction:** *Raoultella planticola* is an uncommon pathogen rarely associated with clinical infections. Previous studies showed that most *R. planticola* isolates were sensitive to cephalosporins, aminoglycosides, fluoroquinolones and carbapenems, and no extensively drug-resistant (XDR) strains have been reported.

**Case presentation:** In this study, an XDR *R. planticola* strain, RP01, was isolated from a Chinese patient with multiple chronic diseases. Antimicrobial susceptibility testing showed that RP01 was resistant to almost all clinically available antibiotics except tigecycline. PCR and sequencing revealed the presence of *bla*<sub>NDM-1</sub> encoding a metallo-β-lactamase with hydrolysing activity against carbapenems. Further genomic sequencing and ResFinder analysis identified 20 resistance genes, encoding resistance to β-lactams, aminoglycosides, sulfonamides, tetracycline, fluoroquinolones, trimethoprim and fosfomycin.

**Conclusion:** These results described the ability of the rare pathogen *R. planticola* to acquire multiple resistance genes.

**Keywords:** *Raoultella planticola; bla*<sub>NDM-1</sub>; extensively drug-resistant; tigecycline.
aminoglycosides, sulfonamides, tetracycline, fluoroquinolones, trimethoprim and fosfomycin (Table 1). The isolate was susceptible to tigecycline and colistin with MIC values of 2 and 0.125 μg ml⁻¹, respectively. After comprehensive analysis of other examination results, including normal body temperature, normal white blood cell count, and slightly elevated levels of serum procalcitonin, the responsible physician felt the clinical scenario was more consistent with colonization rather than infection. The patient received symptomatic treatment for 2 weeks, which alleviated his respiratory distress. Further specific treatment for coronary disease was then carried out and the patient was discharged after 32 days.

**Investigations**

Although the modified Hodge test for RP01 showed a negative result, it was positive by the imipenem/EDTA double-disc synergy test, indicating the presence of metallo-β-lactamase genes. Multiplex PCR detection for 11 acquired carbapenemase genes was carried out as described previously (Poirel et al., 2011) and identified the blaNDM-1 gene in RP01. The PCR product was further sequenced to confirm the blaNDM-1 gene sequence. RP01 was the first clinical isolate producing blaNDM-1 identified in FAHG, and no blaNDM-1-positive strains were detected from other hospitalized patients during the patient’s stay.

Attempts to transfer the blaNDM-1 gene to donor strain *E. coli* J53 by conjugation failed. To investigate the molecular mechanism for the XDR phenotype, high-throughput DNA sequencing was carried out by the Ion Torrent personal genome machine (Life Technologies). Total DNA of RP01 was prepared by DNA extraction kits (Qiagen) and sequenced following the manufacturer’s protocols. A total of 3,294,078 reads with a mean length of 199 bp were obtained, which represented about 79 x coverage data for the RP01 genome. *De novo* assembly was performed using the MIRA assembler (version 3.4.0.1) and 170 large contigs (>500 bp) were obtained. Antibiotic-resistant genes were analysed in the ResFinder database (Zankari et al., 2012) using the assembled contigs. A total of 20 resistance genes were identified on different contigs, including 5 for β-lactam resistance (blaCTX-M-3, NDM-1, OXA-30, SHV-12 and TEM-122), 4 for aminoglycoside resistance [aac(6')Ib-cr, aadA16, aadA5 and armA], 3 for macrolide resistance (mph(A), mph(E) and msr(E)), 2 for quinolone resistance [aac(6')Ib-cr and qnrB6], 2 for trimethoprim resistance (dfrA1 and dfrA27), 1 for fosfomycin resistance (fosA), 1 for phenicol resistance (catB3), 1 for rifampicin resistance (ARR-3), 1 for sulfonamide resistance (sulI) and 1 for tetracycline resistance (tetD). The presence of aac(6')Ib-cr contributes to both aminoglycoside and quinolone resistance (Strahilevitz et al., 2009). The blaNDM-1 gene was located on a 5.8 kb contig with 100 % sequence homology to the counterparts of pNDM-HN380, an IncX3 plasmid isolated from *Enterobacteriaceae* strains carrying blaNDM-1 in China (Ho et al., 2012). Alignment of pNDM-HN380 sequence to the sequencing reads identified all functional regions for the plasmid (data not shown), suggesting that the gene environment of blaNDM-1 in RP01 was likely identical to pNDM-HN380, a resistance plasmid distributed in *Enterobacteriaceae* isolates in China (Ho et al., 2012).

**Discussion**

Clinical isolates carrying blaNDM-1 have been associated with serious infection and high mortality, and the co-production of blaNDM-1 with other resistance genes always leads to the failure of antimicrobial treatment. Based on high throughput genome sequencing and ResFinder resistance gene analysis, the current report presents a description of a blaNDM-1 gene co-harboured with other 19 resistance genes in the rare pathogen *R. planticola*, highlighting the ability of blaNDM-1 to spread to unusual pathogens.

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**References**


