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Title: Bla NDM-1-positive Klebsiella pneumoniae from environment, Vietnam

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bla<sub>NDM-1</sub>-positive Klebsiella pneumoniae from Environment, Vietnam

To the Editor: The bla<sub>NDM-1</sub> gene, which produces the New Delhi metallo-β-lactamase (NDM-1) enzyme, confers resistance to the carbapenem class of antimicrobial drugs and can be transferred among different types of bacteria. NDM-1 was identified in 2008 in Sweden from a patient from India who had been hospitalized in New Delhi (1). Since that report, bla<sub>NDM-1</sub>-positive bacteria have been identified from patients in several countries; most of these patients had a direct link with the Indian subcontinent (2). The spread of bla<sub>NDM-1</sub> among bacterial pathogens is of concern not only because of resistance to carbapenems but also because such pathogens typically are resistant to multiple antimicrobial drug classes, which leaves few treatment choices available (3–5).

In 2011, spread of bla<sub>NDM-1</sub>-positive bacteria in an environmental setting in New Delhi was reported (6).

The possible appearance of bacteria harboring bla<sub>NDM-1</sub> in Vietnam is of concern because cultural and economic links between Vietnam and India are strongly established, including extensive person-to-person exchanges that could enable easy exchange of pathogens. In addition, Vietnam faces a serious problem of antimicrobial drug resistance because drugs are freely available and used in an indiscriminate fashion. Thus, once bla<sub>NDM-1</sub>-positive bacteria colonize persons in Vietnam, they would be able to spread easily and pose a serious public health threat.

During September 2011, we collected paired swab samples (1 for PCR, 1 for culture) of seepage water from 20 sites (rivers, lakes, and water pools in streets) within a 10-km radius of central Hanoi, Vietnam. Samples were transported in Transystem (COPAN Italia S.p.A., Brescia, Italy) to preserve bacteria and DNA. The 20 PCR swab specimens were squeezed out into 0.5-mL volumes of sterile water and centrifuged at 3,000 × g for 30 seconds; 1 μL of the resulting suspension was then used as PCR template to detect bla<sub>NDM-1</sub>, as described (7). Two samples were positive for bla<sub>NDM-1</sub>; these 2 samples were collected from the same river (Kim Ngua River) but at sites 3 km apart.

To isolate and identify the phenotype and genotype of bla<sub>NDM-1</sub>-positive bacteria, we repeatedly spread the 20 culture swab specimens onto Muller-Hinton agar (Nissui, Tokyo, Japan) containing 100 mg/L vancomycin (Nakalai, Kyoto, Japan) plus 0.5 mg/L meropenem (LKT Laboratories, St. Paul, MN, USA) until single colonies were obtained. Each colony was then subcultured by plating onto MacConkey agar (Nihon Seiyaku, Tokyo, Japan) containing 0.5 mg/L meropenem to ensure culture purity; colonies were identified by using API 20E strips (bioMérieux, Basingstoke, UK). MICs of these isolates for 13 antimicrobial drugs were calculated by using Etest (bioMérieux), and susceptibility data were interpreted by using Clinical and Laboratory Standards Institute guidelines (www.clsi.org).

We harvested several species of bacteria from the 2 seepage samples positive for bla<sub>NDM-1</sub>: Acinetobacter baumannii, Klebsiella pneumoniae, Pseudomonas aeruginosa, P. fluorescens/putida, and P. luteola. These isolates were placed onto media containing 0.5 mg/L meropenem, and bacterial DNA was extracted and used for the template for PCR analysis to detect bla<sub>NDM-1</sub>, as described (7). bla<sub>NDM-1</sub> was detected in 3 K. pneumoniae isolates from each of the 2 positive samples (6 isolates total); this result was confirmed by sequencing. All 6 isolates were highly resistant to all β-lactam antimicrobial drugs, including carbapenems (Table). To
Our results show that bla<sub>NDM-1</sub>-positive K. pneumoniae sequence type 283 is present in the Kim Ngoo River, which flows through the central part of Hanoi at 2 sites. The isolates we obtained were also positive for 2 other β-lactamases, bla<sub>TEM</sub>-1 and bla<sub>CTX-M-3</sub>, which showed high elevation of MIC against ciprofloxacin up to 1.5 mg/L. Wide-scale surveillance of environmental and clinical samples in Vietnam and establishment of a strategy to prevent further spread of bla<sub>NDM-1</sub> are urgently needed.

**References**


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