Transparency declarations

None to declare.

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J Antimicrob Chemother 2011
doi:10.1093/jac/dkr180
Advance Access publication 19 May 2011

Dissemination of the New Delhi metallo-β-lactamase-1 (NDM-1) among Enterobacteriaceae in a tertiary referral hospital in north India

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Keywords: carbapenemases, NDM, integrons

Sir,

Of the different classes of carbapenemases, class B enzymes are clinically the most significant; they possess the widest substrate hydrolysis range, including penicillins, cephalosporins and carbapenems, but not monobactams. In recent years, metallo-β-lactamase (MBL) genes have spread from *Pseudomonas aeruginosa* to members of the Enterobacteriaceae.[1] An alarming report by the HPA UK in 2009 warned of a rapidly proliferating novel carbapenem-hydrolysing β-lactamase (carbapenemase) designated New Delhi MBL-1 (NDM-1) among the Enterobacteriaceae family, identified in UK hospital patients.[2] Moreover, the common presence of these β-lactamase genes in transferable mobile elements means that these genes could reach virtually any Gram-negative bacterium and this provides an added risk of dissemination in the community.[3]

Our present work was undertaken with the objective of detecting the $bla_{\text{NDM-1}}$ gene among clinical isolates of the Enterobacteriaceae family in a tertiary referral hospital in north India.

A total of 780 consecutive, non-duplicate isolates of *Escherichia coli* ($n=528$), *Klebsiella pneumoniae* ($n=126$), *Citrobacter* species ($n=84$), *Enterobacter aerogenes* ($n=22$), *Proteus mirabilis* ($n=11$) and *Morganella morganii* ($n=9$) were recovered from different clinical specimens from patients who were admitted to different wards, as well as from those who attended the outpatient departments of S.S. Hospital, BHU, Varanasi, India. The study was conducted from February 2010 to July 2010. The work was approved by the Ethics Committee.

An imipenem/EDTA disc potentiation test[4] was performed for phenotypic detection of MBLs. Antimicrobial susceptibility tests were performed using the Kirby–Bauer disc diffusion method and results were interpreted according to the CLSI recommendations.[5] For partial gene PCR amplification, primers specific for the $bla_{\text{NDM-1}}$ gene were used for reaction with bacterial DNA as template. PCR was performed as described previously.[6]

Random amplified polymorphic DNA (RAPD) was performed using primer 7 (5′-GTGGATGCGA-3′) and isolates were typed according to their band patterns.

Sixty-four isolates were phenotypically found to be MBL producers. On performing PCR for all of the MBL-producing isolates the presence of the gene encoding NDM-1 was confirmed among 54 isolates, consisting of *E. coli* ($n=30$), *Citrobacter* species ($n=12$) and *K. pneumoniae* ($n=12$), with an overall occurrence of 6.9% (54/780). Similar to this study, Deshpande *et al*.[7] reported 22 NDM-1-producing Enterobacteriaceae in a short span of 3 months, while in a previous multicentre study, the occurrence was reported to be 2% from this centre.[8] Most often, NDM producers were recovered from intensive care unit patients (35.1%). The age of the patients ranged from 1 day to 85 years, with 32 male patients and 22 female patients (Table S1, available as Supplementary data at *JAC* Online).

The presence of class 1 integrons was demonstrated in all of the NDM-1-harbouring isolates. On typing the NDM-1-harbouring isolates, 18 patterns of *E. coli*, 8 patterns of *K. pneumoniae* and 5 patterns of *Citrobacter* species were found by RAPD. These results suggest horizontal transmission of the gene at both the intraspecies and interspecies level.

Disc diffusion susceptibility testing showed that 46 (85.1%) of the NDM-1-producing isolates were susceptible to polymyxin B and 25 (46.2%) were susceptible to tigecycline. For other antimicrobials, 51.8% showed susceptibility to piperacillin/tazobactam and 22.2%, 5.5% and 1.8% showed susceptibility to amikacin, gentamicin and tobramycin, respectively. Among 23 urinary tract isolates, 8 showed susceptibility to nitrofurantoin. As many as 27 (50%), 26 (48.1%) and 16 (29.6%) isolates were found to be susceptible to ertapenem, imipenem and meropenem, respectively. A previous study has demonstrated 32.4% similarity of NDM-1 to VIM-1/VIM-2-type MBL[6] and it

has also been reported that the first characterized MBL from India was VIM-2.[9] A nationwide survey also established the presence of VIM-2, -5, -6 and -11 and the evolution of a new variant, VIM-18, from this country.[10] These studies indicate the dissemination of various MBL genes in the Indian subcontinent.

Although NDM-1 hydrolyses almost all β-lactams efficiently, several of our isolates were found by disc diffusion testing to be susceptible to various carbapenems as well as to piperacillin/tazobactam, which was not discussed in any previously conducted studies. However, MICs of these carbapenems and piperacillin/tazobactam, which were not determined in the present study, could be more informative and would have added more value and clinical significance. Colistin and tigecycline were also found to be active in several cases. In our study it was observed that some of the bla_{NDM-1}-harbouring isolates were found to be phenotypically susceptible to all three carbapenems (imipenem, meropenem and ertapenem) tested. So, irrespective of antibiograms, all isolates should be routinely screened for molecular detection of the NDM-1 gene in the hospital setting. Thus, the routine susceptibility testing for carbapenems showing false susceptibility may lead to treatment failure.

Recognition of patients at risk and prevention of transmission is urgently needed in our regions. Besides stringent infection control in hospitals and good sanitation in the community, focus on the basis of the evolution of such genes is needed in order to contain their vertical and horizontal spread.

Acknowledgements

We would like to acknowledge the technical staff for their support.

Funding

This study was supported by internal funding.

Transparency declarations

None to declare.

Supplementary data

Table S1 is available as Supplementary data at *JAC* Online (http://jac.oxfordjournals.org/).

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J Antimicrob Chemother 2011
doi:10.1093/jac/dkr184
Advance Access publication 12 May 2011

Rapid detection of the bla_{NDM-1} gene by real-time PCR

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Keywords: β-lactamases, carbapenem resistance, multidrug resistance, laboratory methods, polymerase chain reaction

Sir,

New Delhi metallo-β-lactamase-1 (NDM-1) is a recently reported novel plasmid-borne metallo-β-lactamase that represents an emerging public health threat.[1] To date, the international spread of NDM-1 has already been reported to diverse locations such as the UK, the USA, Japan, Australia and most recently the Middle East.[2] With the increasing need for efficient surveillance and