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High Frequency of IMP-6 among Clinical Isolates of Metallo-β-Lactamase-Producing *Escherichia coli* in Japan

Carbapenems are the most potent agents for treating infections by Gram-negative bacteria due to their stability with respect to the majority of β-lactamases and their high rate of penetration through the bacterial outer membranes. However, there have been increasing reports of Gram-negative bacteria carrying transferable carbapenem resistance genes such as bla_{IMP}, especially in Japan (4, 6, 7). Most of the metallo-β-lactamases confer resistance not only to carbapenems but also to other β-lactams. Therefore, there is a possibility of treatment failure in patients with metallo-β-lactamase-producing strains and the spread of these strains in hospitals is a serious medical problem. Here we report the high frequency of IMP-6 among clinical isolates of metallo-β-lactamase-producing *Escherichia coli* in Japan.

From February to July 2011, 54 consecutive and nonduplicate clinical isolates of *E. coli* were collected throughout Japan that showed a positive result in the sodium mercaptoacetic acid (SMA; Eiken Chemical Co. Ltd., Tokyo, Japan) test. Antimicrobial susceptibility testing was performed by the broth microdilution method (2). PCR was done to detect β-lactamase genes (IMP-1, IMP-2, VIM-1, VIM-2, CTX-M, TEM, and SHV types) (5, 8, 9), and the PCR products of the metallo-β-lactamase gene were sequenced.

All 54 isolates were positive for the IMP-1 type but negative for IMP-2, VIM-1, and VIM-2 types by PCR. Sequencing revealed that 49 of the 54 *E. coli* isolates carried IMP-6 and five carried IMP-1. Of the 49 IMP-6-positive isolates, all were positive for CTX-M, 23 were positive for TEM, and none were positive for SHV by PCR. All IMP-6-positive isolates showed 100% susceptibility to imipenem and 71.4% susceptibility to meropenem according to the criteria of the Clinical and Laboratory Standards Institute (Table 1) (3).

We previously reported the first isolate of IMP-6-producing *Serratia marcescens*, which was found in the urine of a Japanese patient with urinary tract infection (12). We mentioned that the strain carrying the IMP-6 gene, which is encoded by a transferable plasmid, was more resistant to meropenem than to imipenem. Recently, Shigemoto et al. reported that five strains of IMP-6-producing *Klebsiella pneumoniae* were isolated in Japan and that those strains were also resistant to meropenem but susceptible to imipenem (10). In our study, the susceptibility rate of IMP-6-positive *E. coli* was higher for imipenem than meropenem (100% versus 71.4%). In Japan, imipenem is often used as a representative carbapenem for susceptibility testing (10), so IMP-6-producing isolates may be falsely categorized as susceptible if imipenem is tested. Weisenberg et al. reported that KPC-producing *K. pneumoniae* strains seem susceptible to carbapenems according to routine testing, although clinical and microbiological failure are frequent when these agents are chosen (11). Therefore, when IMP-6-producing isolates are falsely categorized as susceptible and treated with carbapenems, clinical and microbiological failure may occur, as happens with KPC producers. Meropenem is widely used for the treatment of severe Gram-negative infections, because it is slightly more active than imipenem against Gram-negative bacteria (1). Accordingly, IMP-6-producing isolates may be selected by use of carbapenems, especially meropenem, becoming a major problem for antimicrobial therapy in Japan. Therefore, it is necessary to establish a laboratory screening method for these isolates, and further study, including typing of integrons and analysis using multilocus sequence typing, is needed.

TABLE 1 Activity of various antimicrobial agents against 49 IMP-6-positive isolates of *Escherichia coli*

Drug	MIC (μg/ml)			% susceptible
	Range	50%	90%	
Meropenem	0.125 to 8	1	2	71.4
Imipenem	0.125 to 0.5	0.25	0.5	100
Piperacillin	32 to ≥256	≥256	≥256	0
Tazobactam/piperacillin	1 to ≥256	4	8	98.0
Cefazolin	≥256	≥256	≥256	0
Ceftazidime	4 to ≥256	16	64	1.9
Cefotaxime	8 to ≥256	128	≥256	0
Cefepime	4 to ≥256	16	128	18.4
Levofloxacin	≤0.06 to 64	16	32	10.2

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Published ahead of print 4 June 2012

Address correspondence to Hisakazu Yano, yanohisa@med.tohoku.ac.jp.

H. Yano and M. Ogawa contributed equally to this article.

doi:10.1128/AAC.00617-12

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Hisakazu Yano
Miho Ogawa
Shiro Endo
Department of Infection Control and Laboratory Diagnostics
Internal Medicine
Tohoku University Graduate School of Medicine
Miyagi, Japan

Risako Kakuta
Department of Otolaryngology
Head and Neck Surgery
Tohoku University Graduate School of Medicine
Miyagi, Japan

Hajime Kanamori
Shinya Inomata
Noriomi Ishibashi
Tetsuji Aoyagi
Masumitsu Hatta
Department of Infection Control and Laboratory Diagnostics
Internal Medicine
Tohoku University Graduate School of Medicine
Miyagi, Japan

Yoshiaki Gu
Mitsuhiro Yamada
Department of Regional Cooperation for Infectious Diseases
Tohoku University Graduate School of Medicine
Miyagi, Japan

Koichi Tokuda
Department of Infection Control and Laboratory Diagnostics
Internal Medicine
Tohoku University Graduate School of Medicine
Miyagi, Japan

Hiroyuki Kunishima
Miho Kitagawa
Yoichi Hirakata
Mitsuo Kaku
Department of Infection Control and Laboratory Diagnostics
Internal Medicine
Tohoku University Graduate School of Medicine
Miyagi, Japan