

Trends in Antimicrobial-Drug Resistance in Japan

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Multidrug resistance in gram-positive bacteria has become common worldwide. In Japan until recently, gram-negative bacteria such as *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, and *Serratia marcescens* were controlled by carbapenems, fluoroquinolones, and aminoglycosides. However, several of these microorganisms have recently developed resistance against many antimicrobial drugs.

In Europe and the United States, methicillin-resistant *Staphylococcus aureus* (MRSA) and vancomycin-resistant enterococci (VRE) have been widely disseminated in many medical institutions; gram-negative rods, including *Klebsiella pneumoniae* and *Escherichia coli* producing extended-spectrum β -lactamases (ESBLs), are also becoming a clinical threat. In Japan, many antimicrobial drugs, such as carbapenems, fluoroquinolones, and aminoglycosides, have been freely used as first-line drugs for more than 15 years. During that time, drug-resistant bacteria have been emerging and proliferating (1-3). Isolation of VRE and ESBL-producing gram-negative rods is still rare (4), but MRSA and penicillin-intermediate-resistant and penicillin-resistant *Streptococcus pneumoniae* are widely distributed in clinical settings (5), as in western countries. In addition, several carbapenem- and fluoroquinolone-resistant gram-negative rods have been isolated from geographically separate hospitals (6-8).

Development and Use of Antimicrobial Drugs

Many clinically effective antimicrobial drugs have been developed by Japanese pharmaceuti-

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cal companies since the 1960s. These drugs are mainly broad-spectrum cepheems, fluoroquinolones, macrolides, and aminoglycosides, which are quite effective against various pathogenic bacterial species. Moreover, the official prices of these antimicrobial drugs are set at relatively high levels compared with those of earlier drugs such as penicillins, older quinolones, and kanamycin. Therefore, newer drugs have been used preferentially as first-line drugs under the Japanese health insurance system, which allows medical institutions to obtain benefit from the difference between the official price (selling price) and the actual market cost. In Japan, three carbapenems (imipenem, panipenem, and meropenem) and at least nine fluoroquinolones (enoxacin, fleroxacin, norfloxacin, ofloxacin, ciprofloxacin, lomefloxacin, tosufloxacin, sparfloxacin, and levofloxacin) are used as first-line drugs in every clinical setting, although use of these drugs is restricted in many western countries, including the United States. Arbekacin, clarithromycin, and teicoplanin are also widely used for chemotherapy.

Drug-Resistant Bacteria

The following summary is based on a preliminary survey of VRE and MRSA and a report of surveillance for antimicrobial susceptibility in Japan conducted by the Medical Information System Development Center.

Gram-Positive Cocci

VRE

Since vancomycin was approved for IV use in 1991 only for MRSA infections, isolation of VRE is still rare in Japan; however, VanA-type *Enterococcus faecium* was first isolated in Kyoto in 1996 (9). Fewer than 50 cases of *vanA*- or *vanB*-type VRE isolation have been reported throughout Japan (unpublished data). An increase in VRE isolation may be inevitable because of international travel and importation of chicken contaminated with VRE (10).

MRSA

The percentage of MRSA among nosocomial *S. aureus* strains in Japan is estimated to be 50% to 70%. Several deaths associated with MRSA infection have been reported (11,12), despite preventive measures against nosocomial infections. However, the actual number of deaths associated with MRSA infection, as well as the trend, is unknown.

Isolation of a vancomycin-homo-resistant *S. aureus* strain Mu50 from a clinical sample in Japan was reported in 1997 (13). This strain was selected from vancomycin-hetero-resistant *S. aureus* strain Mu3, which intrinsically contains a few Mu50. Identifying Mu50 by the antibiotic susceptibility testing method recommended by the National Committee for Clinical Laboratory Standards (NCCLS) (14) may be difficult. Therefore, clinicians were concerned that emergence of strains corresponding to Mu3 and Mu50 could become a serious clinical problem.

We conducted a nationwide survey for MRSA by collecting >6,600 clinical isolates from 245 medical settings. When >10⁵ CFU of bacteria were inoculated, 3% to 4% of clinical isolates grew colonies on brain-heart-infusion agar (BHI agar) plates containing 4 mg/L of vancomycin, as recommended (13). However, the assay reproducibility was poor and we were unable to confirm a stable resistant phenotype. Several vancomycin-tolerant strains (MIC, 4 mg/L by NCCLS method) were selected after repeated in vitro selection with BHI agar plates containing sub-MIC concentrations of vancomycin. No strain corresponding to Mu50, which had an MIC for vancomycin of 8 mg/L by the standard NCCLS method, could be identified in 6,625 clinical isolates. If strains with vancomycin-resistance profiles similar to those of Mu3 or Mu50 were

widely disseminated in Japan, vancomycin-homo-resistant strains such as Mu50 would be expected in clinical settings and would be isolated by the routine NCCLS antibiotic-susceptibility testing method (15). Since vancomycin has been in widespread use for nearly 9 years in Japan, several Mu50-like strains should have been selected and predominant. Possibly the Mu3 and Mu50 strains reported earlier are unusual strains that may have critical defects or mutations in the genes responsible for with synthesis or degradation of the peptidoglycan layer. The clinical importance and genetic background of these strains need to be elucidated. Clinicians should be alert for emergence of glycopeptide-resistant Staphylococci that have acquired endogenous gene mutations or exogenous genes responsible for vancomycin resistance (e.g., the *vanA* or *vanB* gene clusters found in VRE).

Glycopeptide-Resistant Coagulase-Negative Staphylococci

Sporadic isolation of teicoplanin-resistant coagulase-negative Staphylococci such as *Staphylococcus haemolyticus* has been reported to several medical, clinical, or microbiological societies in Japan; however, no report has been published in English.

Penicillin-Resistant Pneumococci

The overall isolation frequency of penicillin-intermediate *S. pneumoniae* and penicillin-resistant *S. pneumoniae* is estimated to be approximately 50%, according to reports from individual hospitals and health districts in Japan (16,17). Several clinically isolated pneumococci are resistant to cepheems, new macrolides, and fluoroquinolones (16,18). Nosocomial penicillin-resistant *S. pneumoniae* isolates usually belong to serotypes 19, 6, and 23 (19), and substitution at Thr371 was associated with penicillin resistance in many such isolates (20).

Gram-Negative Rods

Pseudomonas aeruginosa and Related Microorganisms

P. aeruginosa is resistant to a wide range of antimicrobial drugs. Carbapenems, fluoroquinolones, and aminoglycosides such as amikacin are the last resort for treatment. However, several clinical isolates that have acquired resistance to

these drugs have been identified in Japan. Of special concern are highly carbapenem-resistant strains producing plasmid-dependent IMP-1-type metallo- β -lactamase, which are proliferating rapidly (1). According to a preliminary survey of 2,533 *P. aeruginosa* isolates, 88 (1.3%) strains had the *bla*_{IMP} gene cassette responsible for IMP-1 production. Moreover, the *bla*_{IMP} gene cassette has been dispersing into various glucose-nonfermenting gram-negative bacteria such as *Pseudomonas putida*, *P. fluorescens*, and *Burkholderia cepacia* (7).

Furthermore, approximately 20% and 5% of clinically isolated *P. aeruginosa* have acquired resistance to fluoroquinolones and amikacin, respectively. *Alcaligenes* and *Acinetobacter* spp. have also acquired a wide range of drug resistance.

Enterobacteriaceae

Two nosocomial outbreaks of *Serratia marcescens* were recently reported in Japan. This bacterium is consistently resistant to penicillins and classic cephalosporins (e.g., cephalothin and cephaloridine), but not to carbapenems. However, IMP-1 producers have recently been isolated in geographically separate clinical settings, and they often show high-level resistance to β -lactams, including cephamycins and carbapenems (21). According to a preliminary survey of 3,222 clinically isolated *S. marcescens* strains, at least 141 (4.4%) had IMP-1 productivity. Some of these strains also have acquired resistance to fluoroquinolones or amikacin.

E. coli and *K. pneumoniae* are still susceptible to oxy-imino-cephalosporins such as cefotaxime and ceftazidime. *K. pneumoniae* and *E. coli* strains that produce TEM- or SHV-derived extended-spectrum β -lactamase and are resistant to these drugs are still rare in Japan, while those producing CTX-M-2 type β -lactamase predominate (22,23). This disproportion may be related to the widespread use of cephamycins and carbapenems. However, as a result of recent restricted use of these broad-spectrum drugs, several *K. pneumoniae* and *E. coli* strains producing SHV-derived ESBLs, such as SHV-12, are emerging (4,24).

Several multidrug-resistant *Salmonella* spp. have been reported in Japan, and some of them were recently identified as *S. Typhimurium* DT104 (25). However, isolation frequency of DT104 is thought to be lower than in western countries.

Fluoroquinolone-resistant gonococci are also increasing in Japan, as in south Asian countries (26). *Haemophilus influenzae* clinical isolates that show ampicillin resistance, some of which are penicillinase nonproducers, have also been increasing (27). More than 90% of *Moraxella catarrhalis* have acquired penicillinase productivity.

Isolation frequency of multidrug-resistant *Mycobacterium tuberculosis* is estimated to be low (28) but may be increasing, as indicated by recent reports of several outbreaks (29).

Drug-Resistant Bacteria in the 21st Century

Multidrug-resistant gram-positive cocci such as MRSA and VRE have emerged and spread worldwide in the 20th century. Gram-negative rods such as *P. aeruginosa*, *K. pneumoniae*, and *S. marcescens* have until recently been controlled by carbapenems, fluoroquinolones, and aminoglycosides. However, several of these microorganisms have become resistant to many antimicrobial drugs. Extraordinary efforts will be needed to prevent global dissemination of multidrug-resistant gram-negative bacteria in the next millennium (30).

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