Resistance in gram-negative bacteria: Enterobacteriaceae

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The emergence and spread of resistance in Enterobacteriaceae are complicating the treatment of serious nosocomial infections and threaten to create species resistant to all currently available agents. Approximately 20% of Klebsiella pneumoniae infections and 31% of Enterobacter spp infections in intensive care units in the United States now involve strains not susceptible to third-generation cephalosporins. Such resistance in K pneumoniae to third-generation cephalosporins is typically caused by the acquisition of plasmids containing genes that encode for extended-spectrum β-lactamases (ESBLs), and these plasmids often carry other resistance genes as well. ESBL-producing K pneumoniae and Escherichia coli are now relatively common in healthcare settings and often exhibit multidrug resistance. ESBL-producing Enterobacteriaceae have now emerged in the community as well. Salmonella and other Enterobacteriaceae that cause gastroenteritis may also be ESBL producers, which is of relevance when children require treatment for invasive infections. Resistance of Enterobacter spp to third-generation cephalosporins is most typically caused by overproduction of AmpC β-lactamases, and treatment with third-generation cephalosporins may select for AmpC-overproducing mutants. Some Enterobacter cloacae strains are now ESBL and AmpC producers, conferring resistance to both third- and fourth-generation cephalosporins. Quinolone resistance in Enterobacteriaceae is usually the result of chromosomal mutations leading to alterations in target enzymes or drug accumulation. More recently, however, plasmid-mediated quinolone resistance has been reported in K pneumoniae and E coli, associated with acquisition of the qnr gene. The vast majority of Enterobacteriaceae, including ESBL producers, remain susceptible to carbapenems, and these agents are considered preferred empiric therapy for serious Enterobacteriaceae infections. Carbapenem resistance, although rare, appears to be increasing. Particularly troublesome is the emergence of KPC-type carbapenemases in New York City. Better antibiotic stewardship and infection control are needed to prevent further spread of ESBLs and other forms of resistance in Enterobacteriaceae throughout the world. (Am J Infect Control 2006;34:S20-8.)

Gram-negative bacteria of the Enterobacteriaceae family are important causes of urinary tract infections (UTIs), bloodstream infections, hospital- and healthcare-associated pneumonias, and various intra-abdominal infections. Within this family, Escherichia coli is a frequent cause of UTIs, Klebsiella spp and Enterobacter spp are important causes of pneumonia, and all of the Enterobacteriaceae have been implicated in bloodstream infections and in peritonitis, cholangitis, and other intra-abdominal infections. Additionally, organisms such as Salmonella produce gastroenteritis and subsequently, in some patients, invasive infection. Emerging resistance in Enterobacteriaceae is a significant problem that requires immediate attention. Resistance related to production of extended-spectrum β-lactamases (ESBLs) is a particular problem in the handling of Enterobacteriaceae infections, but other mechanisms of resistance are also emerging, leading to multidrug resistance and threatening to create panresistant species.

OVERVIEW OF RESISTANCE TRENDS AND OUTCOMES

Third-generation cephalosporins were originally developed as β-lactams able to overcome resistance caused by common β-lactamases. When first introduced, third-generation agents like ceftriaxone, cefotaxime, and ceftazidime were stable in the presence of common β-lactamases. However, within a few years, hospital-acquired gram-negative bacilli like Klebsiella pneumoniae and others began producing mutated versions of these β-lactamases that made them resistant to third-generation cephalosporins and to the monobactam aztreonam. According to the National Nosocomial Infections Surveillance (NNIS) System report, in 2003, 20.6% of all K pneumoniae isolates from patients in intensive care units (ICUs) in the United States were nonsusceptible to third-generation cephalosporins. This represented a 47% increase compared with resistance rates for 1998 to 2002. Nonsusceptibility to third-generation cephalosporins was also observed in 31.1% of Enterobacter spp and 5.8% of E coli isolated from patients in ICUs in 2003. Those rates were approximately the same as for 1998 to 2002. International studies report even higher rates of nonsusceptible K pneumoniae in hospitals and particularly in ICU settings.
Enterobacteriaceae resistance to third-generation cephalosporins is typically caused by production of β-lactamases. An example are the ESBLs that hydrolyze broad- and extended-spectrum cephalosporins, monobactams, and penicillins. The genes that encode ESBLs are frequently found on the same plasmids as genes that encode resistance to aminoglycosides and sulfonamides, and many Enterobacteriaceae possess changes that confer high-level resistance to quinolones. This means that ESBL-producing Enterobacteriaceae species are common multidrug resistant, which poses a particular challenge for the treatment of nosocomial infections, especially in critically ill patients. Inappropriate empiric antimicrobial treatment for nosocomial- or community-acquired infections has been reported to contribute to significantly greater mortality rates in the ICU, and inadequate antimicrobial treatment of infection was the most important independent determinant of hospital mortality. Other studies have reported that inappropriate initial antibiotic treatment for nosocomial bacteremia caused by ESBL-producing *K pneumoniae* or *E coli* is associated with a significantly higher mortality rate than is initial therapy involving an agent with activity against these ESBL-producing bacteria.

### EXTENDED-SPECTRUM β-LACTAMASE–PRODUCING ENTEROBACTERIACEAE

#### General issues and nomenclature

Infections caused by ESBL-producing Enterobacteriaceae are serious concerns in the current environment. Many ESBLs represent enzymes that have evolved from class A β-lactamases—namely, TEM-1, TEM-2, and SHV-1, which are frequently expressed in gram-negative bacteria and which confer resistance to ampicillin, amoxicillin, and other penicillins, as well as to early- but not later-generation cephalosporins. ESBLs arose when mutations of the genes encoding TEM-1, TEM-2, or SHV-1 gave rise to new β-lactamases that became able to hydrolyze third-generation cephalosporins and aztreonam. TEM- or SHV-type ESBLs are typically not active against cefamycins (eg, cefotetan, cefoxitin, or cefmetazole) or carbapenems (imipenem, ertapenem, and meropenem), and can generally be inhibited by β-lactamase inhibitors such as clavulinate, sulbactam, or tazobactam. Enterobacteriaceae may also express ESBLs that are not closely related to TEM- or SHV-related species, includingCTX-M- and OXA-type ESBLs, among others. CTX-M-type ESBLs typically hydrolyze cefotaxime more efficiently than ceftazidime. Unlike most ESBLs that have been found in *E coli, K pneumoniae*, and other Enterobacteriaceae, OXA-type ESBLs have been found mainly in *Pseudomonas aeruginosa* and only rarely in Enterobacteriaceae. Another important fact about ESBLs is that they are typically plasmid mediated rather than chromosomally mediated β-lactamases.

ESBLs should be distinguished from other β-lactamases capable of hydrolyzing extended-spectrum cephalosporins. Examples include AmpC and carbapenemases. Carbapenemases may be further grouped as either metallo-β-lactamases (class B) or serine carbapenemases (classes A and D). Like ESBLs, AmpC β-lactamases hydrolyze third-generation or expanded-spectrum cephalosporins, but unlike ESBLs, they are also active against cefamycins and are resistant to inhibition by clavulanate or other β-lactamase inhibitors. Carbapenemases have broader-range activity, covering carbapenems as well as expanded-spectrum cephalosporins. Carbapenemase-producing Enterobacteriaceae are currently relatively rare, but there are concerns about the emergence and spread of these strains. Table 1 presents a brief summary of different β-lactamases produced by Enterobacteriaceae or other gram-negative bacteria.

### In vitro susceptibility profiles and clinical outcomes

In vitro susceptibility profiles for ESBL-producing Enterobacteriaceae for cephalosporins can be misleading because they suggest that an isolated strain is susceptible to a given cephalosporin when in fact the drug may not be effective when used to treat a serious infection caused by such an organism. This phenomenon is sometimes known as hidden resistance. Susceptibility break points from the National Committee for Clinical Laboratory Standards (NCCLS), now known as the Clinical and Laboratory Standards Institute (CLSI), for third-generation cephalosporins in Enterobacteriaceae were created in the early 1980s, at a time when ESBL production was not a common occurrence. Thus, there is a problem in that ESBL-producing strains sometimes may be incorrectly recorded by the microbiology laboratory as susceptible to third-generation cephalosporins.

In vitro studies show that the minimum inhibitory concentration (MIC) of third- and fourth-generation cephalosporins for ESBL-producing Enterobacteriaceae is elevated severalfold when high versus standard inocula are used in susceptibility testing. This can lead to a situation in which in vitro testing indicates the isolate is susceptible to late-generation cephalosporins, but clinical failure occurs when these agents are used. In a study of patients with ESBL-producing *K pneumoniae* bacteremia, 54% of patients receiving treatment with a susceptible cephalosporin, as determined by in vitro methods, experienced clinical failure. The
data suggested that an antibiotic MIC of 8 μg/mL was universally associated with clinical failure and that high rates of failure were also observed with a MIC of 4 μg/mL (Table 2). These results are consistent with those from a variety of observational studies that show rates of clinical failure >90%, approximately 67%, and <30% with MICs of 8, 4, and ≤2 μg/mL, respectively. Whether the susceptibility break points of Enterobacteriaceae for cephalosporins should be changed is currently under consideration. The 2005 CLSI guidelines recommend that laboratories report ESBL-producing isolates as resistant to all penicillins, cephalosporins, and aztreonam irrespective of in vitro tests results. The presence of inadequately identified ESBL producers has important implications for both antibiotic therapy and infection control.

### Treatment of ESBL producers

The presence of ESBL-producing Enterobacteriaceae complicates therapy, especially because these organisms are often multidrug resistant. When isolates from a patient indicate an ESBL-producing organism, the first thing to consider is whether the patient has a true infection. Patients with positive isolates from urine or perhaps the respiratory tract may be only colonized, and clearly, there is no indication for treatment in those situations. Assuming the patient has a serious infection due to ESBL-producing Enterobacteriaceae, the choice of empiric therapy is made difficult by the likelihood of multidrug resistance and the fact that there are no large, randomized, controlled trials designed to compare one antibiotic therapy with another for infections caused by ESBL-producing organisms. Moreover, for a variety of reasons, it is unlikely that such a study will ever be performed. Nonetheless, data from a number of studies strongly point to carbapenems as the drugs of choice for empiric treatment of serious infections involving ESBL-producing Enterobacteriaceae.

Subgroup analysis from a randomized, evaluator-blind trial comparing cefepime with imipenem in patients with nosocomial pneumonia showed that 100% of patients (10 of 10) receiving imipenem for pneumonia caused by an ESBL producer experienced a positive clinical response compared with only 69% of patients (9 of 13) treated with cefepime. Similarly,

### Table 1. Selected β-lactamases of gram-negative bacteria

<table>
<thead>
<tr>
<th>β-Lactamase</th>
<th>Examples</th>
<th>Substrates</th>
<th>Inhibition by clavulanate*</th>
<th>Molecular class</th>
</tr>
</thead>
<tbody>
<tr>
<td>Broad-spectrum</td>
<td>TEM-1, TEM-2, SHV-1</td>
<td>Penicillin G, aminopenicillins, carboxypenicillins, piperacillin, narrow-spectrum cephalosporins</td>
<td>+++</td>
<td>A</td>
</tr>
<tr>
<td></td>
<td>OXA family</td>
<td>Substrates of the broad-spectrum group plus clavulanic acid, methicillin, and oxacillin</td>
<td>+</td>
<td>D</td>
</tr>
<tr>
<td>Extended-spectrum</td>
<td>TEM family, SHV family</td>
<td>Substrates of the broad-spectrum group plus oximino-cephalosporins, and monobactam (aztreonam)</td>
<td>+++</td>
<td>A</td>
</tr>
<tr>
<td></td>
<td>CTX-M family</td>
<td>Substrates of the expanded-spectrum group plus, for some enzymes, cefepime</td>
<td>+++</td>
<td>A</td>
</tr>
<tr>
<td></td>
<td>OXA family</td>
<td>Same as for CTX-M family</td>
<td>+</td>
<td>D</td>
</tr>
<tr>
<td></td>
<td>Others (PER-1, PER-2, BES-1, GES/IBC family, SFO-1, TLA-1, VEB-1, VEB-2)</td>
<td>Same as for TEM family and SHV family</td>
<td>+++</td>
<td>A</td>
</tr>
<tr>
<td>AmpC</td>
<td>ACC-1, ACT-1, CFE-1, CMY family, DHA-2, FOX family, LAT family, MIR-1, MOX-1, MOX-2</td>
<td>Substrates of expanded-spectrum group plus cephemycins</td>
<td>0</td>
<td>C</td>
</tr>
<tr>
<td>Carbapenem</td>
<td>IMP family, VIM family, GIM-1, SPM-1 (metallo-β-enzymes)</td>
<td>Substrates of the expanded-spectrum group plus cephemycins and carbapenems</td>
<td>0</td>
<td>B</td>
</tr>
<tr>
<td></td>
<td>KPC-1, KPC-2, KPC-3</td>
<td>Same as for IMP family, VIM family, GIM-1, and SPM-1</td>
<td>+++</td>
<td>A</td>
</tr>
<tr>
<td></td>
<td>OXA-23, OXA-24, OXA-25, OXA-26, OXA-27, OXA-40, OXA-48</td>
<td>Same as for IMP family, VIM family, GIM-1, and SPM-1</td>
<td>+</td>
<td>D</td>
</tr>
</tbody>
</table>

Adapted from N Engl J Med. 10
*+, ++, and +++ denote relative sensitivity to inhibition.

### Table 2. Outcome of cephalosporin treatment of serious infections due to extended-spectrum β-lactamase–producing organisms

<table>
<thead>
<tr>
<th>MIC (μg/mL)</th>
<th>Experienced failure of cephalosporin therapy</th>
<th>Died of bacteremia within 14 days</th>
</tr>
</thead>
<tbody>
<tr>
<td>8</td>
<td>100 (6/6)</td>
<td>33 (2/6)</td>
</tr>
<tr>
<td>4</td>
<td>67 (2/3)</td>
<td>0 (0/3)</td>
</tr>
<tr>
<td>2</td>
<td>33 (1/3)</td>
<td>0 (0/3)</td>
</tr>
<tr>
<td>≤1</td>
<td>27 (3/11)</td>
<td>18 (2/11)</td>
</tr>
<tr>
<td>Total*</td>
<td>54 (15/28)</td>
<td></td>
</tr>
</tbody>
</table>

MIC, minimum inhibitory concentration.
Adapted from J Clin Microbio. 14
*Includes 5 patients with isolates for which MICs were recorded simply as 0.5 to 4 μg/mL.
a prospective, observational, international study of patients with *K. pneumoniae* bacteremia reported an all-cause 14-day mortality rate of 5.7% (1 of 27) with carbapenem alone, compared with rates of 36.3% and 44.4% with quinolone and noncarbapenem β-lactam monotherapy, respectively.7 For patients infected with ESBL-producing *K. pneumoniae*, the corresponding 14-day mortality rates were 4.8% (2 of 42) among patients receiving carbapenem monotherapy or combination therapy and 27.6% (8 of 29) among those receiving treatment with a noncarbapenem antibiotic.

Although TEM- and SHV-type ESBLs do not effectively hydrolyze cephamycins (such as cefoxitin or cefotetan). Enterobacteriaceae may exhibit resistance to those agents due to plasmid-mediated expression19 or overexpression19 of AmpC β-lactamases. Additionally, resistance to β-lactam–β-lactamase inhibitor combinations (such as piperacillin-tazobactam) may occur due to the coexistence of AmpC-type β-lactamases and ESBLs. The development of porin-deficient mutants may also contribute to resistance to cephamycins and β-lactam–β-lactamase inhibitor combinations.20 Such occurrences are not infrequent and argue against the use of cephamycins or β-lactam–β-lactamase inhibitor combinations in patients with serious infections due to ESBL-producing Enterobacteriaceae. As mentioned earlier, the CLSI recommends that isolates found to be ESBL producing be considered resistant to all penicillins, all cephalosporins, and aztreonam. Similarly, quinolones, aminoglycosides, and trimethoprim-sulfamethoxazole (TMP-SMX) are generally not appropriate initial therapeutic choices for serious infections caused by ESBL-producing Enterobacteriaceae because ESBL-producing Enterobacteriaceae are often resistant to these drugs as well.21-23 Moreover, multidrug resistance among ESBL-producing *K. pneumoniae* and *E. coli* species appears to be increasing.23 With quinolones, even in the presence of apparent susceptibility, there may be a substantial failure rate. In our international study discussed earlier, 35.4% of patients who received treatment with a quinolone for bacteria caused by ESBL-producing *K. pneumoniae* died within 14 days.7 Quinolone resistance in Enterobacteriaceae is described in greater detail below.

**Community-acquired ESBLs**

ESBL-producing Enterobacteriaceae are prevalent in the hospital setting, and there is now evidence that they are emerging and spreading in the community as well.24 Most cases of ESBL-producing organisms in the community have been reported internationally; the situation in the United States is not yet known. Most commonly, the cases of community-acquired ESBLs (CA-ESBLs) involve urinary tract infections (UTIs), although gastrointestinal infections in the community may also be important. A population-based laboratory surveillance study of ESBL-producing *E. coli* infections in the Calgary Health Region of Canada reported that 71% of patients had community-onset disease.25 The study did not address whether the ESBL-producing *E. coli* were necessarily acquired in the community, but the data do speak to the high prevalence of infections associated with ESBL-producing species in the community, and cautions that many clinical laboratories may not be aware of the importance of screening for ESBL-producing organisms when dealing with infections originating in the community.24 Failure to do so may lead to inappropriate treatment and adverse outcomes.

The surveillance study also reported that 70% of the ESBL in *E. coli* isolated from patients with community-onset infections were of the CTX-M-type.25 Other studies, too, have reported a high prevalence of CTX-M-type ESBLs in community-onset infections.24 This contrasts with the general preponderance of TEM- and SHV-type ESBLs in isolates from hospitalized patients with *K. pneumoniae* or *E. coli* infections both in the United States and worldwide. In fact, CTX-M-type ESBLs have only very recently been identified in patients with nosocomial *E. coli* infections in the United States.26 In our international study of nosocomial bloodstream infections, CTX-M-type ESBLs were identified in isolates from patients in all countries except the United States.27 Outside the United States, among nonhospitalized patients, ESBL-producing *E. coli* have been identified in various countries—including Spain, Israel, Canada, and the United Kingdom—and frequently, the ESBLs have been of the CTX-M type.25,28-31 As with TEM- or SHV-type ESBLs, CTX-M-type ESBLs are often multidrug resistant.24 This underscores the importance of screening for ESBL-producing pathogens in certain groups in the community.

The typical clinical picture for community-associated infection involving ESBLs is UTI (sometimes associated with bacteremia) due to CTX-M-producing *E. coli*, with elderly women being most commonly affected. Isolates are resistant to typical first-line agents for UTI, such as ciprofloxacin, TMP-SMX, gentamicin, and ceftriaxone. So there is now the very real risk that treatment of community-acquired infections with *E. coli* may be compromised because of multidrug resistance. Data for January 1998 through June 2004 from the US-based Intensive Care Antimicrobial Resistance Epidemiology (ICARE) project indicated that only 0.6% of *E. coli* isolates and 1.8% of *K. pneumoniae* isolates from so-called outpatient areas were nonsusceptible to third-generation cephalosporins.1 At present, the existence of CA-ESBL-producing Enterobacteriaceae appears to be very limited in the United States.
Nonetheless, the healthcare community needs to be aware of the potential problem that CA-ESBL producers may present in the United States in the future, especially given what has been observed in the United Kingdom and Canada. It suggests that attention be focused on potential risk factors for community development of infection with ESBL-producing bacteria. In a Spanish study, those risk factors included diabetes mellitus, previous quinolone use, recurrent UTIs, a previous hospital admission, and older age in male patients. An Israeli study identified risk factors as previous hospitalization within the past 3 months, antibiotic treatment within the past 3 months, age >60 years, male sex, *K pneumoniae* infection, previous use of third-generation cephalosporins, previous use of second-generation cephalosporins, previous use of quinolones, and previous use of penicillin. Although both studies identified previous hospitalization as a risk factor, it should be noted that cases of infection with an ESBL-producing organism in the community have been reported in patients without recent hospitalization. So, cases of true CA-ESBL producers have been documented. As mentioned previously, ESBL-producing pathogens may also be involved in gastrointestinal infections acquired in the community. Bacterial species that have been reported to produce ESBLs leading to drug-resistant gastroenteritis include *Salmonella* spp, *Shigella*, *Vibrio cholerae*, and Shiga toxin-producing *E coli*. The possible emergence and spread of *Salmonella* strains resistant to antibiotics commonly used as treatment are concerns, because these infections can be invasive. Outside the United States, TEM-, SHV-, and CTX-M-type ESBLs, as well as AmpC β-lactamases, have been identified in infection-causing *Salmonella*. Within the United States, the mechanism of *Salmonella* resistance to third-generation cephalosporins has been linked to production of AmpC β-lactamases. In particular, resistance has been associated with the plasmid-mediated AmpC β-lactamase known as CMY-2. *Salmonella* strains resistant to third-generation cephalosporins are of concern because (1) ceftriaxone and, secondarily, quinolones are the drugs of choice for invasive salmonella disease, and (2) quinolones are not indicated for use in children. Fortunately, ceftriaxone-resistant *Salmonella* are currently rare in the United States, but they represent an area that bears further watching.

**ANTIBIOTIC RESISTANCE IN ENTEROBACTER SPECIES**

*Enterobacter* spp are significant causes of nosocomial infection and are intrinsically resistant to ampicillin, cefazolin, and cefoxitin due to production of constitutive chromosomal AmpC β-lactamases. Moreover, β-lactam exposure is capable of inducing expression of AmpC β-lactamases in *Enterobacter* spp—with consequent resistance to third-generation cephalosporins—and mutations may result in permanent hyperproduction and persistent resistance. Treatment of *Enterobacter* infections with third-generation cephalosporins may select for mutant strains associated with hyperproduction of AmpC β-lactamase. The prevalence of *Enterobacter* spp resistant to third-generation cephalosporins has increased since the introduction and common use of these antibiotics. For example, in 1 study, resistance to third-generation cephalosporins emerged in approximately 20% of patients during treatment for *Enterobacter* bacteremia. Multidrug-resistant *Enterobacter* spp in initial positive blood cultures were significantly more prevalent (*P < .001*) among patients who had previously received third-generation cephalosporins than among patients who had previously received other antibiotic treatments, and they were associated with higher mortality rates.

In summary, third-generation cephalosporins should be avoided as treatment for serious infection with *Enterobacter* spp because their use in such situations results in selection of the small number of AmpC-overproducing mutants present in any collection of *Enterobacter* spp isolates and because the typical clinical scenario is characterized by initial response followed by recurrence of infection. In contrast, cefepime is comparatively stable to AmpC β-lactamases, and therefore has been regarded as a suitable option for treatment of *Enterobacter* infections. However, ESBL-producing *Enterobacter* spp, particularly *Enterobacter cloacae*, have been identified in the United States (Table 1). At our medical center in Pittsburgh, approximately 33% of bloodstream isolates have been shown to contain *E cloacae* that produce ESBL as well as AmpC β-lactamases. The MIC of cefepime may be within that danger zone of 4–8 mg/mL, where cefepime activity may be compromised. Hence, not all resistance to later-generation cephalosporins in *E cloacae* may be the result of hyperproduction of AmpC β-lactamases, and ESBL-producing strains of *E cloacae* may be resistant to fourth- as well as third-generation cephalosporins. The advent of ESBLs in AmpC-producing *E cloacae* is certainly something to be aware of and is potentially clinically important.

**EMERGING QUINOLONE AND CARBAPENEM RESISTANCE**

**Quinolone resistance**

Quinolones are used widely for the treatment of serious *E coli* UTIs and may also be used to treat other
infections caused by other members of the Enterobacteriaceae family. Hence, quinolone resistance in Enterobacteriaceae may lead to treatment failures and is a significant concern, as is the recent emergence of plasmid-mediated resistance to quinolones. According to the 2004 NNIS report, means of 7.3% and 8.2% of E. coli isolates from US patients in both ICUs and non-ICU areas of hospitals, respectively, and a mean of 5.6% from US outpatients exhibited quinolone resistance. Higher rates of quinolone resistance may be found in ESBL-producing strains of E. coli and K. pneumoniae. For example, 55.8% of infections in the University of Pennsylvania Health System caused by ESBL-producing E. coli or K. pneumoniae were fluoroquinolone resistant. In Shanghai, China, 86.1% of ESBL-producing E. coli and 45.6% of ESBL-producing K. pneumoniae were reported to be resistant to levofloxacin. We studied K. pneumoniae bacteremia from 12 hospitals in 7 countries and found that overall, 18% of ESBL-producing isolates were ciprofloxacin resistant and 60% of ciprofloxacin-resistant isolates produced ESBLs. Prior receipt of a quinolone has been shown to be an independent risk factor for quinolone resistance.

Quinolone resistance in Enterobacteriaceae is usually due to alterations in target enzymes (DNA gyrase and/or topoisomerase IV) or to impaired access to the target enzymes, occurring either because of changes in porin expression or because of efflux mechanisms. Both of these principal means of resistance are caused by chromosomal mutations. More recently, plasmid-mediated quinolone resistance has emerged in K. pneumoniae and E. coli. The first case of plasmid-mediated resistance to quinolones in K. pneumoniae was reported in the United States in 1998 and was from a strain isolated at the University of Alabama in 1994. The plasmid, pMG252, confers multidrug resistance and was shown to greatly increase quinolone resistance when transferred to strains of K. pneumoniae deficient in outer-membrane porins. The gene associated with that resistance has been designated qnr. Quinolone resistance associated with qnr-containing plasmids has now emerged in E. coli and K. pneumoniae strains. A recent study in the United States reported that 11.1% of K. pneumoniae strains from 6 states exhibited plasmid-mediated quinolone resistance associated with the qnr gene, although none of the E. coli strains examined contained qnr. Some of the strains contained the original pMG252 plasmid, but qnr was carried on different plasmids for others. The mechanism of quinolone resistance associated with qnr-containing plasmids appears to involve inhibition of quinolone binding with DNA gyrase.

The emergence of this new plasmid-mediated mechanism of quinolone resistance is particularly worrisome because it provides a mechanism for the rapid development and spread of quinolone and multidrug resistance to important members of the Enterobacteriaceae family.

Carbapenem resistance

As mentioned earlier, carbapenems are currently considered to be the preferred agents for treatment of serious infections caused by ESBL-producing Enterobacteriaceae. Carbapenems are highly stable to β-lactamase hydrolysis, and porin penetration is facilitated by their general size and structure. Their susceptibility to most strains of Enterobacteriaceae makes them generally useful as treatment for multidrug-resistant organisms. Carbapenem resistance is currently rare among Enterobacteriaceae, but some worrisome signs have appeared in recent years.

The expression of AmpC or class A (TEM- or SHV-type) ESBLs plus loss of outer-membrane proteins have been associated with carbapenem resistance in K. pneumoniae. Resistance to carbapenems has also been reported in K. pneumoniae–producing class B β-lactamases (metallo-β-lactamases) in various countries outside the United States, including Brazil, Greece, China, and Singapore. A metallo-β-lactamase–producing strain of E. cloacae with reduced susceptibility to carbapenems has also recently been observed in Greece. Standard susceptibility testing may categorize metallo-β-lactamase–producing Enterobacteriaceae as susceptible to carbapenems, but an inoculum effect has been observed, suggesting that the susceptibility testing may falsely predict the susceptibility of particular Enterobacteriaceae to carbapenems in the clinical environment.

In the United States, carbapenem resistance has been observed in strains of K. pneumoniae–producing class A carbapenemases, namely, KPC-1, KPC-2, and KPC-3. The genes encoding these enzymes are apparently obtained via plasmid conjugation; the enzymes are capable of hydrolyzing and inactivating the carbapenems. KPC-producing strains have generally been shown to exhibit multidrug resistance that includes piperacillin/tazobactam, third- and fourth-generation cephalosporins, fluoroquinolones, and aminoglycosides, as well as carbapenems. KPC-1 expression itself was apparently associated with moderate- to high-level carbapenem resistance, while loss of outer-membrane proteins appeared to be a required cofactor for high-level resistance in KPC-2– and KPC-3–producing strains. In 96 isolates obtained from 10 New York hospitals, >80% of the KPC-producing organisms belonged to a single ribotype. This is worrisome because it suggests that these isolates are not just being selected by antibiotic use, but rather are being passed from person to person as a result of breakdown in infection
control. As with metallo-β-lactamase-producing Enterobacteriaceae, susceptibility testing may falsely indicate the clinical susceptibility of KPC-producing K. pneumoniae due to an inoculum effect. In vitro testing suggests that tigecycline and polymyxins may exhibit the most consistent activity against KPC-producing strains of K. pneumoniae, but this has yet to be demonstrated clinically. KPC-producing strains of Enterobacter and Salmonella spp. have also been identified in the United States.

Strains of clinically important Enterobacteriaceae have now emerged with broader multidrug resistance than has ever before been observed. As the list of antibiotics with potential activity against these strains continues to shrink, measures that prevent and slow the spread of multidrug-resistant Enterobacteriaceae strains throughout the world must be put into action.

SUMMARY

Enterobacteriaceae are significant causes of serious infections, and many of the most important members of this family are becoming increasingly resistant to currently available antibiotics. This is a troubling trend, and one that requires vigilance and intensified measures to control the further spread of resistance by these important gram-negative pathogens. Although improvements in antibiotic stewardship and infection control are discussed in greater detail by others in this supplement, it should be emphasized that such improvements are necessary if the steady rise in ESBL-producing Enterobacteriaceae and in other forms of resistance in these species is to be slowed or stopped. The widespread use of third-generation cephalosporins as the driving force behind the emergence of ESBL-producing organisms has been shown in many studies. Because of that risk, third-generation cephalosporins are no longer appropriate as workhorse antibiotics in our hospitals, and the restriction of their use in hospitals and other healthcare facilities has been shown to reduce the incidence of ESBL-producing Enterobacteriaceae like K. pneumoniae. Quinolones seem to have replaced third-generation cephalosporins in hospitals, yet their overuse, particularly in light of plasmid-mediated quinolone resistance in Enterobacteriaceae, may be of concern.

Infection control is the second aspect in restriction of the emergence and spread of resistant Enterobacteriaceae. With regard to ESBL producers, there is ample evidence of person-to-person spread. Combining reductions in third-generation cephalosporin use with traditional infection control measures—such as the use of gloves, gowns, and hand hygiene in the care of colonized or infected patients—has been reported to control the hospital spread of multidrug-resistant K. pneumoniae. Both improvement in antibiotic stewardship and infection control strategies are needed before it is too late—before we have to deal with infections caused by multidrug-resistant Enterobacteriaceae for which there are no clinically effective drugs.

References


