Extended-spectrum beta-lactamases

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The definition of extended-spectrum beta-lactamases (ESBLs) has expanded rapidly, in terms of both the number and the variety of enzymes. Bacteria bearing plasmidic ESBLs are spreading widely throughout the world. In Canada, the problem has grown less rapidly than in other parts of the world; however, ESBLs still present a significant impediment to the treatment of serious infections with extended-spectrum cephalosporins and penicillins. Although SHV-derived enzymes were the primary concern in the late 1980s and early 1990s, these enzymes have been rapidly overtaken by the CTX-M family of ESBLs. There is no reason to believe that the problem of ESBLs will not expand rapidly in the face of intense antimicrobial pressure and lapses in infection control practices. Control should focus on early detection, accurate characterization, effective treatment and measures to prevent further spread.

Key Words: *Cephalosporinases; Extended-spectrum beta-lactamases; Resistance*

Les bêta-lactamases à spectre élargi

La définition de bêta-lactamases à spectre élargi (BLSÉ) a rapidement pris de l'ampleur, tant pour ce qui est du nombre que de la variété des enzymes. Les BLSÉ plasmidiques porteuses de bactéries se propagent partout dans le monde. Au Canada, le problème a moins pris d'expansion que dans les autres parties du monde, mais les BLSÉ constituent tout de même un obstacle important pour les graves infections traitées aux céphalosporines à spectre élargi et aux pénicillines. Bien que les enzymes dérivées du SHV aient constitué la principale inquiétude à la fin des années 1980 et au début des années 1990, elles ont vite été dépassées par la famille CTX-M des BLSÉ. Tout porte à croire que le problème des BLSÉ se mettra à prendre une expansion rapide en raison de l'intense pression antimicrobienne et des écueils des pratiques de contrôle des infections. Le contrôle devrait être axé sur le dépistage précoce, la caractérisation précise, le traitement efficace et les mesures visant à prévenir la poursuite de la propagation.

During the past 50 years, bacteria have rapidly evolved the mechanisms to allow them to resist antibiotics. Bacteria have become particularly adept at resisting beta-lactams. Resistance to beta-lactam antibiotics may occur as a result of permeability barriers, efflux pumps, altered penicillin binding proteins and through the production of beta-lactamases. Of these mechanisms, the production of beta-lactamases has been the subject of the greatest degree of bacterial evolution. Beta-lactamases may be encoded, on both the chromosome and the plasmid. Transferable elements, such as transposons and integrons, further enhance the ability of beta-lactamase genes to spread widely. Mutations of older 'legacy' beta-lactamases, such as TEM-1 and SHV-1, have dramatically altered their hydrolysis profiles, leading to an entirely new class of beta-lactamases now referred to as extended-spectrum beta-lactamases (ESBLs).

Currently, there are more than 130 enzymes derived from TEM-1 and another 57 derived from SHV-1 (1). However, not all of the TEM-1-derived enzymes are ESBLs; many are inhibitor-resistant beta-lactamases that retain their narrower TEM-1-like spectrum of activity (1).

To give structure to the growing body of knowledge of beta-lactamases, two organizational schemes are commonly cited. The first, by Ambler (2), is based on the molecular relatedness of the various enzymes; the second system, proposed by Bush et al (3), combines both molecular relatedness and the functional characteristics of beta-lactamases. ESBLs have been placed in Ambler class A, Bush group 2be (3). These enzymes are typically active against third-generation cephalosporins, especially ceftazidime and aztreonam (Table 1). They are generally inactive against cephamycins such as cefoxitin and carbapenems (meropenem, imipenem and ertapenem). These enzymes are inhibited in vitro by beta-lactamase inhibitors such as clavulanic acid and tazobactam. Although there are large numbers of TEM- and SHV-derived enzymes, a small number of amino acid substitutions at critical points in the hydrolysis pocket of the enzyme are responsible for changing designation.

A new and rapidly spreading group of ESBLs are those of the CTX-M family. These enzymes were likely originally derived from beta-lactamases in *Kluyvera ascorbata* and are interesting in that they hydrolyze cefotaxime more rapidly than ceftazidime and are more readily inhibited by tazobactam than clavulanic acid or sulbactam (4). Strains expressing CTX-M enzymes have been responsible for outbreaks in Canada, South America, Asia and Europe (5-8).

All of the above ESBLs are typically spread on plasmids and many outbreaks have been described, reflecting the ability of plasmids to spread among various genera and species of bacteria. Outbreaks of both *Escherichia coli* and *Klebsiella pneumoniae* have been described in a variety of health care settings. The first ESBL-obtaining strains were described two years after the introduction of the first third-generation cephalosporin (9). Within three years, the first ESBL outbreak was described in France and by 1989, the first strains were detected in the United States. ESBLs are increasingly encountered in Canadian hospitals (8). Mulvey et al (8) examined strains of *E coli* and *Klebsiella* species from 12 hospitals across Canada. One hundred twelve strains bearing ESBLs were encountered. The numbers increased significantly over the course of the study. SHV-derived enzymes were most common, representing 64% of strains; also of interest, CTX-M-derived enzymes were the second most common (23%). Pitout et al (10) found that outpatients in Calgary, Alberta, often had ESBL-bearing

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TABLE 1
Molecular and phenotypic classification of more commonly occurring or important beta-lactamases (BLAs)

Functional group (3)	Structure class (2)	Substrate preference				Inhibition clavulanate/EDTA	Representative enzyme(s)
		Penicillin	Cephaloridine	Cefotaxime	Carbapenems		
Serine BLAs							
1	C	++	+++	+	–	–/–	AmpC
2a	A	+++	+	–	–	++/–	*Staphylococcus aureus* BLAs
2b	A	+++	++	–	–	++/–	TEM-1, SHV-1
2be	A	+++	++	++	–	++/–	TEM-3, SHV-2
2br	A	+++	+	–	–	–/–	TEM-30
2c	A	++	+	–	–	+/–	PSE-1
2d	D	++	++	– (++)	–	+/–	OXA-1
2e	A	++	++	++	–	++/–	PER-1
2f		++	+	+	++	+/–	IMI-1
4	Not characterized			Variable		–/?	SAR-2
Metalloenzymes							
3	B	++	++	++	++	–/++	IMP-1

+ Some; ++ Moderate; +++ Substantial; – Minimal; ? Unknown

E coli. In their study, 70% of strains of beta-lactamases were in the CTX-M family and greater than two-thirds were from patients in the community (10). In their patient population, ciprofloxacin resistance was often associated with ESBL-mediated resistance.

In the United States, ESBL-mediated resistance has increased slightly more rapidly than in Canada. In a SENTRY study (11) of third-generation cephalosporin-resistant *Klebsiella* species, the prevalence in the United States was higher than that in Canada but less than that in other countries (Latin America [45%], the western Pacific region [25%], Europe [23%], the United States [8%] and Canada [5%]).

Also in Canada, outbreaks of multiresistant *E coli* in long-term care facilities have been well documented by Muller et al (12). In one long-term care facility, 93 of 149 residents were colonized with an outbreak strain. In another investigation (12) in the Durham region north of Toronto, Ontario, more than 200 individuals were colonized with an ESBL-bearing *E coli*. A number of risk factors for ESBL infection colonization have been identified. The duration of both hospital and intensive care unit stay, prior use of a third-generation cephalosporin antibiotic and severe illnesses increase patient susceptibility to infection in the acute care setting (13-15) (Table 2).

When hospital outbreaks occur, they may be difficult to control. Lucet et al (16) described their experience in a Paris, France hospital. By flagging colonized patients and by reinforcing handwashing and other infection control measures, they were able to reduce the rate of acquisition of ESBL-producing Enterobacteriaceae from 117 to only 19 cases per year. This corresponded to a nosocomial rate reduction of 0.56 to 0.06 per 100 admissions. Also of interest, in this setting, is that they did not apply any antibiotic restrictions, while at the same time, they achieved a considerable reduction in transmission.

In another study, Rahal et al (17) responded to an ESBL-bearing *Klebsiella* outbreak with a strategy that included restriction of third-generation cephalosporins. They restricted cephalosporin use to certain pediatric infections, single-dose surgical prophylaxis, acute bacterial meningitis and spontaneous bacterial peritonitis. They were able to achieve an 80% reduction in the amount of cephalosporin use and, correspondingly, a 44% reduction in the number of cases of ceftazidime-resistant *Klebsiella*. Correspondingly, there was a 140% increase in the amount of imipenem used and a 69% increase in the incidence

TABLE 2
Risk factors for infection or colonization with extended-spectrum beta-lactamase-producing organisms

Intensity of care factors
Arterial lines
Central venous catheters
Jejunostomy/gastrostomy tubes
Longer intensive care unit stay
Longer hospital stay
Urinary catheters
Ventilatory systems
Prior antibiotic use
Prior aztreonam/ceftazidime use

of imipenem-resistant *Pseudomonas aeruginosa*. Lee et al (18) found that prior exposure to third-generation cephalosporins was an independent risk factor for acquisition of ESBL-producing *K pneumoniae*, and that reducing the use of this class of antibiotics appeared to lower the acquisition rate (18). For others, formulary intervention did not appear to have a significant effect (19). At this time, the relative importance of tight infection control measures and improved antibiotic stewardship is not known. The optimal strategy presumably involves both measures undertaking the concurrent implementation of both strategies.

There is no doubt that infection with ESBL-bearing bacteria adversely affects patients' outcomes. Paterson et al (20) studied 445 patients with *K pneumoniae* bacteria; 85 of these infections were due to ESBL-producing strains (20). Eight of 23 patients not receiving antibiotics other than a carbapenem died within 10 days. Only one of 27 patients receiving a carbapenem died in the same period. Kim et al (21) examined the outcomes of children with bacteremia due to *E coli* and *K pneumoniae* with and without ESBLs; the case fatality rate for the ESBL group was significantly higher (12 of 45; P=0.001) than that for the non-ESBL group (five of 87).

The antibiotic class of choice for the treatment of ESBL-bearing Enterobacteriaceae appears to be a carbapenem (22). Although in vitro susceptibility suggests that cefepime should be active in vivo, there is a clear inoculum effect (ie, a large concentration of organisms can overwhelm the antibacterial affect of this antibiotic) (23-25). A rat model of *P aeruginosa* pneumonia responded poorly to cefepime (26). At this point in time, it cannot

be considered a first-line agent. Piperacillin-tazobactam also shows an inoculum effect, and several animal studies have suggested that it is less efficacious than carbapenems (26,27). However, the importance of the inoculum effect has recently been questioned; further studies are required to clarify the degree to which it influences in vivo response to antibiotics, particularly in settings where the bacterial burden is high (28). For many ESBL strains, aminoglycosides, fluoroquinolones and trimethoprim-sulfamethoxazole retain activity. However, several studies have shown high levels of coresistance to these antibiotics in ESBL-bearing strains (29-34).

While carbapenems are currently considered to be the drug of choice, bacterial strains with porin mutations may infer resistance (35). There is also concern that the spread of carbapenemase enzymes (of which there are two classes) will threaten the usefulness of this class of antibiotics in the future (36-37).

DISCLOSURES: In the past year, Dr Forward has served on advisory boards for both Wyeth-Ayerst Canada Inc and Merck Frosst Canada Ltd, and has developed educational materials for both. He has also received research funding from sanofi-aventis Canada.

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