R360
Publication Only
Diagnostics, other than Molecular: Diagnostic/laboratory methods (other than molecular)

Feasibility of the disc diffusion methodology and the MIC evaluator for susceptibility testing of ceftaroline against *Staphylococcus aureus* and ESBL-negative Enterobacteriaceae

M. Kresken[1], B. Körber-Irrgang[1]

[1]Antiinfectives Intelligence GmbH, Campus of the University of Applied Science, Rheinbach, Germany

Objective: Ceftaroline (CPT), the active form of the parenteral pro-drug CPT fosamil, has shown in vitro activity against Gram-positive pathogens, including methicillin-resistant *Staphylococcus aureus* (MRSA). It is also potent against common Gram-negative pathogens, but not against extended-spectrum-β-lactamase (ESBL)- or carbapenemase-producing Enterobacteriaceae and non-fermenters. CPT fosamil has been approved by the European Medicines Agency for treatment of adults with complicated skin and soft tissue infections (cSSSI) and community-acquired pneumonia. The present study was performed to verify EUCAST zone diameter breakpoints and evaluate the utility of the M.I.C. evaluator device (M.I.C.E, Oxoid, Wesel, Germany), an MIC gradient endpoint methodology similar to that of Etest, for testing CPT against a set of *S. aureus* and ESBL-negative Enterobacteriaceae strains recovered from patients with cSSSIs.

Methods: A total of 214 isolates were collected from 19 medical microbiology laboratories during a resistance surveillance study conducted in Germany, 2011-2012. Organisms tested were methicillin-susceptible *S. aureus* (MSSA, n=40), MRSA (n=80), and ceftriaxone (CRO)-susceptible (considered as ESBL-negative) strains of five Enterobacteriaceae species: *Escherichia coli* (ECO, n=30), *Enterobacter cloacae* (ECL, n=15), *Klebsiella oxytoca* (KOX, n=15), *Klebsiella pneumoniae* (KPN, n=19), and *Proteus mirabilis* (PMI, n=15). Susceptibility testing was performed in a central laboratory. Isolates were tested for susceptibility to CPT by reference broth microdilution method (BMD) as described by the standard ISO 20776-1:2006. M.I.C.E strips were applied according to the manufacturers' instructions. Disk diffusion tests (DD) were performed according to the EUCAST guideline. Interpretive criteria were those published by EUCAST. Standard performance criteria (categorical and essential [within one 2-fold dilution] agreement) were applied for comparisons of the results achieved by M.I.C.E strips and DD with those achieved by the reference BMD method.

Results: Using the reference BMD method, the percentages of CPT-susceptible isolates were 100% each for MSSA and MRSA, and 86.7%, 100%, 100%, 84.2 and 100% for CRO-susceptible ECO, ECL, KOX, KPN, and PMI, respectively. For all organisms, M.I.C.E MICs were comparable with BMD MICs (within one 2-fold dilution). The overall level of essential and categorical agreement was 100% in each case. Comparing DD results with BMD MICs, the overall categorical agreement was 91.6%, but varied between 66.7% for PMI and 100% for MSSA, ECL, and KOX. Error rates are given in the Table. All MRSA classified as resistant by the DD had zone diameters of 19 mm, which is one mm below the breakpoint.

Conclusion: The results suggest that the M.I.C.E represents an alternative to the reference BMD method for susceptibility testing of CPT. DD provided acceptable results for most organisms, but not for MRSA and PMI. MRSA displaying zone diameters around the breakpoint should be re-tested using the reference BMD method, as recommended by EUCAST.

Table: Error rates of DD compared to BMD*

Organism (n)	Number (%) of errors	
	Major	Very major
MSSA (40)	0	n.a.
MRSA (80)	11 (13.8)	n.a.
E. coli (30)	1 (3.3)	0
E. cloacae (15)	0	n.a.
K. oxytoca (15)	0	n.a.
K. pneumoniae (19)	1 (5.3)	0
P. mirabilis (15)	5 (33.3)	n.a.
Total (214)	18 (8.4)	0

*Very major errors were defined as resistant by the BMD method (reference) and categorized as (false)-susceptible by DD. Major errors were determined for organisms that were susceptible by BMD and (false)-resistant by DD. Minor errors did not apply as CPT breakpoints to not include an intermediate category.

n.a., not applicable as CPT-resistant strains were not detected