Activity of Temocillin against KPC-Producing Klebsiella pneumoniae and Escherichia coli

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Temocillin, a 6-a-methoxy derivative of ticarcillin, is currently approved for treatment of infections due to *Enterobacteriaceae* in Belgium and the UK. It is stable against hydrolysis by most β-lactamases including extended-spectrum β-lactamases (ESBLs) and AmpC-type β-lactamases with studies reporting MIC₉₀s between 16 and 32 µg/ml (3, 4, 8). Temocillin is thus drawing attention as a potential alternative to carbapenems in treatment of infections caused by *Enterobacteriaceae* producing these broad-spectrum β-lactamases.

Carbapenem-resistant *Klesbiella pneumoniae* producing KPC-type β-lactamase has emerged in recent years and caused hospital outbreaks of serious infections in the United States and other parts of the world (7). Furthermore, KPC-type β-lactamase is increasingly identified in other species of *Enterobacteriaceae* as well, including *Escherichia coli*. One concerning recent phenomenon is the occurrence of urinary tract infections due to KPC-producing organisms at nursing homes (10). Currently, the limited treatment options for infections due to KPC-producing organisms include colistin and tigecycline. Concern for nephrotoxicity due to colistin limits its use outside closely monitored settings, whereas tigecycline does not achieve therapeutic urinary
concentration (2). Furthermore, emergence of resistance to these agents has been recently recorded in *Enterobacteriaceae* (5, 6).

The present study was conducted to evaluate the *in vitro* activity of temocillin against clinical isolates of *K. pneumoniae* and *E. coli* producing KPC-type β-lactamase. A total of 33 KPC-producing clinical isolates (30 *K. pneumoniae* and 3 *E. coli*) were used. KPC production was confirmed by ertapenem resistance phenotype, positive modified Hodge test and positive PCR for the KPC structural gene. They were collected from hospitals in 3 states in the United States. The minimum inhibitory concentration (MIC) of temocillin was determined by the standard agar dilution method (1). Temocillin was provided by Eumedica (Brussels, Belgium). In addition, MICs of temocillin against an *E. coli* isogenic clone producing KPC-3 was tested to determine the direct effect of KPC production on temocillin MIC. *E. coli* ATCC25922 was used as the control strain.

Table 1 summarizes the results. For *K. pneumoniae*, the MICs ranged between 16 µg/ml and 64 µg/ml (MIC$_{50}$=32 µg/ml, MIC$_{90}$=32 µg/ml). The *E. coli* clinical isolates had MICs between 8 and 16 µg/ml. Both *E. coli* DH10B with and without cloning vector pBCSK- (Stratagene, La Jolla, CA) encoding *bla*$_{KPC-3}$ had an MIC of 8 µg/ml. Inoculum
The effect was not observed at $10^5$ CFUs, whereas mild inoculum effect averaging within two-fold MIC difference was seen with *K. pneumoniae* when $10^6$ CFUs were inoculated (Table 1). This result was in line with a previous study documenting modest inoculum effect of temocillin in non-KPC-producing isolates (9). The frequency of mutants of representative clinical isolates that grew at their MICs and 2×MICs was calculated to be approximately $1\times10^{-10}$ and 0 for *K. pneumoniae* and $3\times10^{-10}$ and $1\times10^{-10}$ for *E. coli*, respectively.

Currently, the British Society for Antimicrobial Chemotherapy (BSAC) is the only organization that defines temocillin MIC breakpoints for *Enterobacteriaceae*. The BSAC defines temocillin susceptibility at $\leq 8$ and $\leq 32$ µg/ml in systemic and urinary tract infections, respectively (http://www.bsac.org.uk/). One gram of temocillin is known to achieve a peak serum concentration of approximately 160 µg/ml with serum binding of 85% and a half life of 4 to 5 hours (9). The urinary concentration after a 500-mg dose is approximately 500 µg/ml (9). These pharmacokinetic properties of temocillin make it a potential alternative treatment option for mild to moderate urinary tract infections caused by KPC-producing *Enterobacteriaceae*. 
Acknowledgements

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References


Table 1. Susceptibility of KPC-producing *K. pneumoniae* and *E. coli* against temocillin.

<table>
<thead>
<tr>
<th>Innoculum (CFU)</th>
<th>Species</th>
<th>MIC of temocillin (µg/ml)</th>
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<tbody>
<tr>
<td></td>
<td></td>
<td>1</td>
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<tr>
<td>1×10⁴</td>
<td><em>K. pneumoniae</em> (30)</td>
<td>12</td>
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<tr>
<td></td>
<td><em>E. coli</em> (3)</td>
<td>1</td>
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<tr>
<td>1×10³</td>
<td><em>K. pneumoniae</em> (30)</td>
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<td><em>E. coli</em> (3)</td>
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108a BSAC breakpoint for systemic infections.
109b BSAC breakpoint for urinary tract infections.