Postantibiotic Effects of Daptomycin against 14 Staphylococcal and Pneumococcal Clinical Isolates

G. A. Pankuch, M. R. Jacobs, and P. C. Appelbaum

Department of Pathology, Hershey Medical Center, Hershey, Pennsylvania 17033, and Department of Pathology, Case Western Reserve University, Cleveland, Ohio 44106

Received 27 February 2003/Returned for modification 25 April 2003/Accepted 2 June 2003

Daptomycin mean staphylococcal postantibiotic effects (PAEs) were 1.1 to 6.2 h, with a mean of 2.5 h. The mean pneumococcal PAEs were 1.7 h, ranging between 1.0 and 2.5 h. The staphylococcal and pneumococcal postantibiotic sub-MIC effects at 0.4 times the MIC ranged from 3.0 to >12.0 h and 1.9 to >12.0 h, respectively.

The postantibiotic effect (PAE) is a pharmacodynamic parameter contributing to the choosing of antibiotic dosing regimens. It is defined as the length of time that bacterial growth is suppressed following brief exposure to an antibiotic (4, 5, 10). Odenholt-Tornqvist and coworkers have suggested that during intermittent dosage regimens, supra-MIC levels of antibiotic are followed by subinhibitory levels that persist between doses, and those authors have hypothesized that persistent subinhibitory levels could extend the PAE (3, 13–15). The effect of sub-MICs on growth during the PAE period has been defined as the postantibiotic sub-MIC effect (PA-SME), representing the time interval that includes the PAE plus the additional time during which growth is suppressed by sub-MICs. In contrast to the PA-SME, the SME measures the direct effect of subinhibitory levels on cultures which have not been previously exposed to antibiotics (13–15).

We examined the PAE, PA-SME, and SME of daptomycin, a lipopeptide active against gram-positive organisms (1, 2, 6, 8, 16–18). We studied two clinical strains each of methicillin-susceptible Staphylococcus aureus, methicillin-resistant S. aureus, methicillin-susceptible coagulase-negative staphylococci, methicillin-resistant coagulase-negative staphylococci, and penicillin-susceptible, -intermediate, and -resistant Streptococcus pneumoniae. Organisms were identified by standard methods (11). No attempt was made to identify coagulase-negative staphylococci by species. Daptomycin MICs were determined by macrodilution procedures (12). Mueller-Hinton broth was adjusted to contain 50 mg of calcium per liter for testing daptomycin, as recommended by the NCCLS (12).

The PAE was determined by the viable plate count method (5), using Mueller-Hinton broth supplemented with calcium as described above and 5% lysed horse blood when testing pneumococci. The PAE was induced by exposure to 10 times the MIC of daptomycin for 1 h. Because the protein binding of daptomycin is 90 to 92%, this corresponds to total serum drug concentrations of approximately 100 times the MIC and approximates serum free drug levels at peak concentrations only (8, 18).

For PAE testing, tubes containing 5 ml of broth with antibiotic were inoculated with approximately 5 × 10^6 CFU/ml. Inocula were prepared by suspending growth from an overnight blood agar plate in broth. Growth controls with inoculum but no antibiotic were included with each experiment. Inoculated test tubes were placed in a shaking water bath at 35°C for an exposure period of 1 h. At the end of the exposure period, cultures were diluted 1:1,000 in prewarmed broth to remove the antibiotic by dilution. Antibiotic removal was confirmed by comparing growth curves of a control culture containing no antibiotic to another containing daptomycin at 0.01 times the exposure concentration.

Viability counts were determined before exposure and immediately after dilution (0 h) and then every 2 h for up to 12 h or until turbidity of the tube reached 1 McFarland standard. The PAE was defined as PAE = T – C; T is the time required for viability counts of an antibiotic-exposed culture to increase by 1 log_{10} above counts immediately after dilution, and C is the corresponding time for growth control (5).

In cultures designated for PA-SME determinations, the PAE was induced as described above after exposure to 6 times or 10 times the MIC (see above). Following 1:1,000 dilution, cultures were divided into four tubes. To three tubes, daptomycin was added to produce final subinhibitory concentrations of 0.2, 0.3, and 0.4 times the MIC. The fourth tube did not receive antibiotic. Viability counts were determined before exposure, immediately after dilution, and then every 2 h for up to 12 h or until their culture turbidity reached 1 McFarland standard. SME experiments were performed as for PA-SME experiments; however, cultures designated for SME determinations were treated the same as for PA-SME testing except that PAE was not induced. SME cultures were not exposed during a PAE phase and were simply exposed after each culture dilution and were under the constant influence of 0.2, 0.3, or 0.4 times the corresponding sub-MIC.

The SME was defined as SME = T_{pa} – C; T_{pa} is the time for cultures previously exposed to antibiotic and then reexposed to different sub-MICs to increase by 1 log_{10} above counts immediately after dilution, and C is the corresponding time for the unexposed control (13–15). The SME was defined as SME = T_{pa} – C; T_{pa} is the time for the cultures exposed only to sub-MICs to increase 1 log_{10} above counts immediately after dilution, and C is the corresponding time for the unexposed control. The PA-SME and SME (13–15) were measured in two
PA-SMEs obtained from three of the four S. aureus strains follow: staphylococci, 0.25 to 0.5; pneumococci, 0.12 to 0.5. The mechanism underlying this SME at 0.2 times the MIC, as well as that underlying the long-lasting SME at 0.2 times the MIC, has not been elucidated.

For the 14 strains, the mean PAE was 1.7 h, the mean SME was 0.9 h, and the mean PAE-SME was 0.8 h. Mean PAEs of 2.4 h and 4.1 h were found for methicillin-sensitive and -resistant S. aureus strains, respectively. The mean SME of 2.7 h for the two methicillin-sensitive S. aureus strains was shorter than the mean SME of 3.8 h for the two methicillin-resistant strains.

For the six pneumococcal isolates, the mean PAE was 1.7 h, the mean SME was 0.9 h, and the mean PAE-SME was 0.8 h. Mean PAEs of 1.1 to 6.2 h, with a mean of 2.5 h, were found for the pneumococcal isolates. Mean SMEs of 3.0 to 4.6 h, with a mean of 3.5 h, were found for the pneumococcal isolates. The mean PAE-SME was 1.5 h.

Table 1. PAE of daptomycin against 14 strains

<table>
<thead>
<tr>
<th>Strain</th>
<th>PAE (h)</th>
<th>SME (h)</th>
<th>PAE-SME (h)</th>
</tr>
</thead>
<tbody>
<tr>
<td>S. aureus (SA1)</td>
<td>2.7</td>
<td>1.2</td>
<td>1.5</td>
</tr>
<tr>
<td>S. aureus (SA2)</td>
<td>2.0</td>
<td>1.2</td>
<td>0.8</td>
</tr>
<tr>
<td>S. aureus (SA3)</td>
<td>2.7</td>
<td>1.3</td>
<td>1.4</td>
</tr>
<tr>
<td>S. aureus (SA4)</td>
<td>3.0</td>
<td>1.5</td>
<td>1.5</td>
</tr>
<tr>
<td>S. aureus (SA5)</td>
<td>2.4</td>
<td>1.2</td>
<td>1.2</td>
</tr>
<tr>
<td>S. aureus (SA6)</td>
<td>3.6</td>
<td>1.8</td>
<td>1.8</td>
</tr>
<tr>
<td>S. aureus (SA7)</td>
<td>2.9</td>
<td>1.5</td>
<td>1.4</td>
</tr>
<tr>
<td>S. aureus (SA8)</td>
<td>2.7</td>
<td>1.3</td>
<td>1.4</td>
</tr>
<tr>
<td>S. aureus (SA9)</td>
<td>2.4</td>
<td>1.2</td>
<td>1.2</td>
</tr>
<tr>
<td>S. aureus (SA10)</td>
<td>2.0</td>
<td>1.2</td>
<td>0.8</td>
</tr>
<tr>
<td>S. aureus (SA11)</td>
<td>1.8</td>
<td>1.1</td>
<td>0.7</td>
</tr>
<tr>
<td>S. aureus (SA12)</td>
<td>2.7</td>
<td>1.3</td>
<td>1.4</td>
</tr>
<tr>
<td>S. aureus (SA13)</td>
<td>2.4</td>
<td>1.2</td>
<td>1.2</td>
</tr>
<tr>
<td>S. aureus (SA14)</td>
<td>3.6</td>
<td>1.8</td>
<td>1.8</td>
</tr>
</tbody>
</table>

Duration of time (h) for PAE, SME, and PAE-SME was calculated from the MIC to the end of growth. The PAE and SME were determined from the end of growth to the end of the post-exposure period. The PAE-SME was determined from the end of growth to the end of the post-exposure period to the MIC.
compared to the other strains in this study. This same degree of variability has been reported before for levofloxacin PAEs, PA-SMEs, and SMEs using S. aureus (9).

Longer intervals between doses may be possible when an antibiotic has a long half-life as well as a prolonged PAE and PA-SME, because regrowth continues to be prevented when serum and tissue drug levels fall below the MICs (3, 5, 14, 15). Previous studies have reported daptomycin PAEs for S. aureus ranging from 2.4 to 6.3 h (2, 8). Our mean PAE results for S. aureus (2.0 to 6.2 h) were similar. In this study we tested the PAE and PA-SMEs by using daptomycin within clinically achievable free peak serum drug levels. The PA-SMEs were generally longer than the PAEs for all of the strains tested, indicating that sub-MIC levels of daptomycin extend the PAE. Therefore, a longer PAE can be achieved by sub-MIC daptomycin concentrations when they follow a suprainhibitory level (14, 15). The half-life of daptomycin in plasma is approximately 7 h (18). This together with the long PAEs and PA-SMEs found in this study supports once-daily dosing of daptomycin.

This study was supported by a grant from Cubist Pharmaceuticals, Lexington, Mass.

REFERENCES