Penetration of Piperacillin and Tazobactam into Inflamed Soft Tissue of Patients with Diabetic Foot Infection

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Received 20 April 2005/Returned for modification 29 May 2005/Accepted 18 July 2005

We investigated the pharmacokinetics of piperacillin and tazobactam in the extracellular space fluid of inflamed soft tissues of six patients with diabetic foot infection using in vivo microdialysis and found similar penetration for piperacillin but not for tazobactam into inflamed and noninflamed soft tissue.

Patients with chronic diabetes mellitus are prone to the development of foot ulcers, with a lifetime risk of 10 to 15% (6). The major pathogenic factors are peripheral neuropathy and peripheral arterial occlusive disease (PAOD) (2). Diabetic foot ulcers facilitate the invasion of bacteria and the development of foot infections, which increase the risk of foot amputation and life-threatening septicemia (1). Diabetic ulcer patients frequently require minor or major amputations of the lower extremities (15 to 27%), and in more than 50% of these cases, infection is a predisposing risk factor for amputation (5, 6). Successful treatment of bacterial infections depends on choosing appropriate anti-infective agents and administering them in doses large enough to reach sufficient concentrations at the site of infection. The target site concentration of a specific anti-infective agent can be determined by means of in vivo microdialysis, which is becoming an increasingly popular means of determining the tissue penetration and pharmacokinetics of anti-infective drugs. However, despite the availability of this technique, only three antimicrobial agents (ciprofloxacin, fosfomycin, and moxifloxacin) have been studied for their tissue penetration and pharmacokinetics of anti-infective drugs. Therefore, we investigated the pharmacokinetics of piperacillin in the extracellular space fluid of inflamed soft tissues of six patients with diabetic foot infection using in vivo microdialysis and found similar penetration for piperacillin but not for tazobactam into inflamed and noninflamed soft tissue.

This was an open-label study performed at a single center. The study was approved by the ethics committee of the Medical University Graz and was performed in accordance with the Declaration of Helsinki and the Good Clinical Practice Guidelines of the European Commission. Informed consent was obtained from all patients. Six patients (four women, two men; age [means ± standard deviations {SD}]), 72.3 ± 13.1 years; weight, 80.5 ± 17.2 kg; height, 168.3 ± 16.2 cm) with diabetic foot infections were enrolled in the study. These patients had had insulin-dependent (four out of six) or non-insulin-dependent (two out of six) diabetes mellitus for 12.2 ± 6 years. Four patients had stage I to IIa PAOD (according to the Fontaine classification system). At admission to the hospital, intravenous anti-infective therapy with a commercially available combination of piperacillin and tazobactam (Tazonam, 4.5 g; Wyeth-Lederle, Vienna, Austria) was administered as described below. In addition, patients received a number of other non-anti-infective drugs (median number, 8.5; range, 4 to 12). Each patient received 4.0 g piperacillin and 0.5 g tazobactam (Tazonam; 4.5 g) in a 30-min intravenous infusion three times per day (i.e., every 8 h for a daily dose of 12 g piperacillin and 1.5 g tazobactam). This equaled three single doses of piperacillin and tazobactam totaling 154.8 ± 32.7 mg/kg body weight and 19.4 ± 4.1 mg/kg body weight, respectively. To ensure consistent experimental conditions after the initial dose, administration of piperacillin plus tazobactam was synchronized to begin at 0800 h, 1600 h, and 2400 h each day.

On the third day of anti-infective treatment, concentrations of piperacillin and tazobactam in the interstitial space fluid of soft tissues were determined by means of in vivo microdialysis as previously described (8, 15). Microdialysis probes (CMA 10; molecular cutoff, 20 kDa; outer diameter, 0.5 mm; membrane length, 16 mm; Microdialysis AB, Stockholm, Sweden) were inserted into the upper subcutaneous layer of the thigh (non-inflamed soft tissue) and into the upper subcutaneous layer within the area of inflammation (inflamed soft tissue). Piperacillin and tazobactam in both plasma and microdialysates were quantified by high-performance liquid chromatography and UV detection as previously described (11). The limit of detection was 1.0 mg/liter. Kineta 3.0 software (InnaPhase Corp., Philadelphia, PA) was used for analysis of pharmacokinetic, noncompartmental data. The time to maximal concentration (Tmax) and the maximal concentration of drug (Cmax) were calculated for plasma and soft tissue. The area under the concentration-versus-time curve from 0 to 8 h (AUC0–8) was determined for plasma (AUC0–8, plasma), inflamed soft tissue (AUC0–8,inflamed), and noninflamed soft tissue (AUC0–8,noninflamed) by the trapezoid rule. As measures
zobactam are presented in Fig. 1 and Table 1, respectively. The peak concentration of piperacillin was similar in inflamed and noninflamed soft tissues (Fig. 1a and Table 1). The $C_{\text{max}}$ of tazobactam in plasma was significantly higher than in noninflamed soft tissue but not significantly higher than in inflamed soft tissue (Fig. 1b and Table 1). The piperacillin concentration at 8 h ($C_8$) was 7.8 ± 6.2 mg/liter in plasma, 4.1 ± 1.9 mg/liter in inflamed soft tissue, and 3.1 ± 1 mg/liter in noninflamed soft tissue (Fig. 1a and Table 1). The $C_R$ for tazobactam was below the limit of detection (1 mg/liter) in plasma, inflamed soft tissue, and noninflamed soft tissue (Fig. 1b and Table 1). For piperacillin, the $\text{AUC}_{0-8, \text{plasma}}$ was significantly larger than the $\text{AUC}_{0-8, \text{inflamed}}$ and $\text{AUC}_{0-8, \text{noninflamed}}$; however, the $\text{AUC}_{0-8, \text{inflamed}}$ and $\text{AUC}_{0-8, \text{noninflamed}}$ values were similar (Table 1). The mean $\text{AUC}_{0-8, \text{inflamed}}/\text{AUC}_{0-8, \text{plasma}}$ and $\text{AUC}_{0-8, \text{noninflamed}}/\text{AUC}_{0-8, \text{plasma}}$ ratios (means ± SD), which served as measures of drug penetration into inflamed and noninflamed soft tissue, were 0.45 ± 0.22 and 0.42 ± 0.36, respectively, for piperacillin and 1.36 ± 0.75 and 0.53 ± 0.17, respectively, for tazobactam. The mean $\text{AUC}_{0-8, \text{inflamed}}/\text{AUC}_{0-8, \text{noninflamed}}$ ratio was 1.47 ± 0.82 for piperacillin and 2.39 ± 1.73 for tazobactam.

In the present study, the β-lactam antibiotic piperacillin easily penetrated the interstitial space fluid of both inflamed and noninflamed soft tissues of patients with diabetic foot infections. This was demonstrated by the observation that the $C_{\text{max}}$ and $\text{AUC}_{0-\infty}$ after intravenous administration of piperacillin and the piperacillin concentration-versus-time curves during the 8-h dosing interval were similar for both inflamed and noninflamed soft tissue. These data indicated that the local inflammation accompanying diabetic foot infection did not reduce the penetration of piperacillin into the interstitial space. Moreover, this ability of piperacillin to penetrate equally into inflamed and noninflamed soft tissue was not shared by tazobactam, as demonstrated by two pieces of evidence. First, tazobactam concentrations in plasma, inflamed soft tissue, and noninflamed soft tissue were considerably lower than piperacillin concentrations in the same places. Second, tazobactam concentrations became undetectable (i.e., fell below 1 mg/liter) within 4 h after intravenous administration began.

These findings appear to agree with previous findings by others. In a rat model, Dalla Costa et al. (4) showed that 2 h after intravenous administration of a bolus containing 120 mg/kg piperacillin plus 15 mg/kg tazobactam, the plasma concentration of tazobactam was about 4 mg/liter. The dose per kilogram of body weight that they used was about 2.4 times the dose that we administered to our patients and would have equaled single doses of 9.6 g of piperacillin plus 1.2 g of tazobactam for our patients. In a study of human patients with pneumonia and metapneumonic pleural empyema, Tomaselli et al. (14) administered piperacillin and tazobactam at the same doses that we used (i.e., 4.0 g and 0.5 g, respectively) and reported subsequently low concentrations of tazobactam in both lung and muscle tissues. Thus, our inability to detect tazobactam after so short a time was not unexpected. However, this does not necessarily mean that the combination of piperacillin and tazobactam was a poor anti-infective therapy, since in vitro tazobactam concentrations of 0.05 to 0.5 mg/liter are capable of inhibiting β-lactamases by 50% (12). The large

FIG. 1. Concentration-versus-time profiles for piperacillin (a) and tazobactam (b) in plasma (filled squares) as well as interstitial space fluid of inflamed (downward-pointing filled triangles) and noninflamed (upward-pointing open triangles) subcutaneous soft tissue following the application of piperacillin plus tazobactam in patients with diabetic foot infections. Piperacillin (4.0 g) plus tazobactam (0.5 g) (Tazonam; 4.5 g) were intravenously applied during a 30-min period starting at time point zero ($t = 0$ min). Results are given as means ± SD; $n = 6$ for panel a and 3 to 6 for panel b.

of drug penetration into inflamed and noninflamed soft tissues, $\text{AUC}_{0-8, \text{inflamed}}/\text{AUC}_{0-8, \text{plasma}}$ and $\text{AUC}_{0-8, \text{noninflamed}}/\text{AUC}_{0-8, \text{plasma}}$ ratios were determined. Drug concentrations in interstitial space fluid were calculated from dialysates as described previously (10). For comparisons between pharmacokinetic parameters, the Wilcoxon signed rank test was used (StatView 5.0; SAS Institute Inc., Cary, NC). $P < 0.05$ was set as the level of significance.

In our patients, both gram-positive bacteria (Staphylococcus aureus, streptococci) and gram-negative bacteria (Pseudomonas aeruginosa, Proteus vulgaris, and Escherichia coli) were isolated from skin lesions by deep swabbing. S. aureus was detected in 50% of cases. The concentration-versus-time profiles and the pharmacokinetic parameters of piperacillin and tazobactam are presented in Fig. 1 and Table 1, respectively. The $C_{\text{max}}$ of piperacillin was significantly higher in plasma than it was in the interstitial space fluid of inflamed or noninflamed soft tissue. However, the peak concentration of piperacillin was similar in inflamed and noninflamed soft tissues (Fig. 1a and Table 1).
TABLE 1. Pharmacokinetic parameters for plasma, inflamed soft tissue, and noninflamed soft tissue after administration of piperacillin and tazobactam in patients with diabetic foot infection

<table>
<thead>
<tr>
<th>Substance</th>
<th>Plasma</th>
<th>Inflamed soft tissue</th>
<th>Noninflamed soft tissue</th>
</tr>
</thead>
<tbody>
<tr>
<td>AUC0–8 (mg · h/liter)</td>
<td>671.8</td>
<td>283.3</td>
<td>283.3</td>
</tr>
<tr>
<td>T_{max} (h)</td>
<td>0.50</td>
<td>0.25</td>
<td>0.25</td>
</tr>
<tr>
<td>C_{max} (mg/liter)</td>
<td>334.5 (338.5–1288.9)</td>
<td>169.4 (84.0–535.3)</td>
<td>63.5 (50.8–203.2)</td>
</tr>
<tr>
<td>T_{max} (h)</td>
<td>52.2 (275.9–392.3)</td>
<td>83.0 (38.5–254.3)</td>
<td>111.0 (59.8–203.2)</td>
</tr>
<tr>
<td>C_{max} (mg/liter)</td>
<td>0.18 (0.33–0.67)</td>
<td>0.25 (0.83–1.50)</td>
<td>0.25 (0.83–1.50)</td>
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</tbody>
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Data are means ± SD (and range) for six patients. See the text for dosage and administration schedules. C_8 was not detectable for tazobactam.

Piperacillin (4.0 g) intravenously in combination three times a day, we found good and equal penetration of piperacillin into interstitial space fluid of inflamed and noninflamed soft tissue of patients with diabetic foot infections. The concentrations of tazobactam in inflamed and noninflamed tissue as well as in plasma would support success against bacterial organisms with MICs of up to 16 mg/liter for most patients. However, against organisms with MICs above 16 mg/liter, e.g., P. aeruginosa with a CLSI breakpoint of susceptibility for piperacillin-tazobactam of 64 mg/liter, the detected in vivo concentrations in inflamed and noninflamed tissues of our patients may not be sufficiently effective.

The in vivo microdialysis experiments that we performed were done at a time on day 3 of treatment when the concentration of piperacillin, but not tazobactam, in interstitial space fluid would have been low (i.e., before the second of three doses scheduled that day). We cannot exclude the possibility that this may have influenced our in vivo microdialysis findings by affecting our calculations of the absolute concentrations of piperacillin in interstitial space fluid. Theoretically, it could have led us to underestimate the in vivo recovery, which in turn would have led us to slightly overestimate the concentrations of piperacillin in interstitial space fluid. We do not believe, however, that this potential error would have been large enough to affect the conclusions of this study. On the other hand, we assume that our calculations of the in vivo recovery and in vivo concentrations of tazobactam were correct, because the tazobactam concentration at the time of in vivo recovery was below the drug’s limit of detection.

In summary, after administering the β-lactam antibiotic piperacillin (4.0 g) and the β-lactamase inhibitor tazobactam (0.5 g) intravenously in combination three times a day, we found good and equal penetration of piperacillin into interstitial space fluid of inflamed and noninflamed soft tissue of patients with diabetic foot infections. The concentrations of tazobactam in inflamed and noninflamed tissue as well as in plasma, however, was lower than expected by the dose of tazobactam intravenously administered to the patients. From the concentrations of piperacillin-tazobactam in inflamed and noninflamed tissues of patients with diabetic foot infection in this study, we conclude that organisms for which the drug MICs were 16 mg/liter and below should be successfully eradicated with the employed antibacterial regimen, while organisms for which the drug MICs are higher may not be sufficiently affected.

This study was supported in part by Wyeth-Lederle, Vienna, Austria.

REFERENCES


