
Lack of Mezlocillin and Piperacillin Interference in Measurement of Vancomycin in the Abbott TDx. Gerald J. Merritt, Barney H. Hunter, and W. Carey Hall (Dept. of Pharmacy, Wilford Hall USAF Medical Center, Lackland AFB, TX 78236-5300)

Our Therapeutic Drug Monitoring Service has observed several unusually low vancomycin concentrations in serum from patients receiving vancomycin concurrently with mezlocillin or piperacillin. Mezlocillin and piperacillin are both known to inactivate aminoglycoside antibiotics: a bond is formed between the beta-lactam ring and an amino group on the aminoglycoside, the product being a biologically inactive amide (1). This inactivation is dependent on concentration, temperature, and time. Vancomycin contains an amino group that also could react with beta-lactam antibiotics. A MEDLINE search back to 1966 revealed no published studies of this potential, nor were there any studies evaluating cross reactivity between mezlocillin and piperacillin with the Abbott TDx assay. We therefore conducted a study to determine if vancomycin is inactivated by either mezlocillin or piperacillin and if either drug interferes in the Abbott TDx assay for vancomycin.

Drug-free serum was supplemented with vancomycin (from Vancocin Injection; Eli Lilly & Co, Indianapolis, IN 46285) to give a vancomycin concentration of 54.5 mg/L. Piperacillin was added to give concentrations of 0, 125, 250, and 500 mg/L. Identical dilutions were prepared with mezlocillin. In addition, samples of drug-free serum were prepared which contained only piperacillin or mezlocillin in the above concentrations. All solutions were maintained at room temperature and assayed in duplicate at 0, 1, 2, 6, 12, and 24 h. All vancomycin concentrations ranged from 49 to 55 mg/L (mean 52.37, SD 1.74) at time 0, and there was no significant change in these concentrations over the 24-h period, in either blank serum or mezlocillin- or piperacillin-supplemented serum. Interference by piperacillin or mezlocillin was negligible during the 24-h period.

Reference

Comparison of Solutions for Extracting CK, AK, and LD from Brain Samples, Wayne L. Chandler, Kathleen J. Clayson, and James S. Fine (Dept. of Lab. Med., SB-10, Univ. of Washington, Seattle, WA 98195)

Anoxic injury to the brain releases enzymes—including creatine kinase BB (EC 2.7.3.2, CK-BB), mitochondrial CK (CK-mt), lactate dehydrogenase (EC 1.1.1.27, LD), and adenylate kinase (EC 2.7.4.3, AK)—into the cerebrospinal fluid (1). Previous studies have attempted to measure the total activity of these enzymes in brain tissue, to estimate the amount available for release. We compared two solutions commonly used to extract these enzymes from brain, 50 mmol/L Tris (pH 7.4 at 25 °C) and 100 mmol/L ammonium acetate (pH 9.5), with and without Triton X-100 surfactant, 10 mL/L, to determine which solution extracted the most CK, AK, and LD from samples of cerebellar cortex obtained within 60 min of death from a large normal mongrel dog. Each experiment was performed in triplicate. Samples of brain were homogenized at 4 °C in 19 mL of solution per gram wet weight of tissue, centrifuged at 15,000 × g for 20 min, and the supernates were diluted 10-fold in 50 mmol/L Tris (pH 7.4, at 25 °C) with 20 mmol/L added diethiothreitol to stabilize the enzyme activity (2). Total CK, AK, and LD activities and CK isoenzymes were measured as described previously (3).

<table>
<thead>
<tr>
<th>Solution</th>
<th>Tris +</th>
<th>NH4Ac*</th>
<th>NH4Ac</th>
<th>Triton X</th>
</tr>
</thead>
<tbody>
<tr>
<td>CK-BB</td>
<td>324 ± 16b</td>
<td>428 ± 17</td>
<td>306 ± 13</td>
<td>398 ± 11</td>
</tr>
<tr>
<td>CK-mt</td>
<td>5 ± 1</td>
<td>14 ± 4</td>
<td>27 ± 8</td>
<td>33 ± 6</td>
</tr>
<tr>
<td>LD</td>
<td>98 ± 7</td>
<td>133 ± 4</td>
<td>92 ± 4</td>
<td>123 ± 5</td>
</tr>
<tr>
<td>AK</td>
<td>33 ± 1</td>
<td>41 ± 2</td>
<td>30 ± 2</td>
<td>43 ± 1</td>
</tr>
</tbody>
</table>

*NH4Ac = ammonium acetate. All activities in U/g, mean ± SD.

Ammonium acetate alone released approximately three times more CK-mt (P <0.0005, paired t-test) than did Tris alone, and essentially equal amounts of CK-BB, LD, and AK. Addition of Triton X to the extraction solutions significantly increased (P <0.0005) the yield of all three cytoplasmic enzymes with either buffer. Previous workers who did not use surfactants in their extracting solutions may have underestimated the activity of these enzymes by as much as 30% to 40% (4). Correct estimation of CK, AK, and LD in brain is important in developing models of enzyme release into the blood and CSF after stroke or global brain ischemia.

References


Southern blot analyses of restriction fragment length polymorphisms (RFLPs) are being used to diagnose human genetic diseases (1). We recently noticed artifacts that may lead to the potential misinterpretation of banding patterns.