Sulfa Drugs in Serum

HPLC Method

Column: Symmetry® C18, 3.9 x 150 mm, 5 µm
Part number: WAT046980
Mobile phase: 1% glacial acetic acid and 4% methanol in water
Flow rate: 1.0 mL/min
Injection volume: 10 µL of reconstituted porcine serum extract
Temperature: 25°C
Detection: UV @ 254 nm

Compounds:
1. Sulfadiazine
2. Sulfathiazole (I.S.)
3. Sulfamerazine

Oasis® HLB Extraction Method

Oasis® HLB 1 cc/30 mg Extraction Cartridge
Part Number WAT094225
Condition: 1 mL methanol/1 mL water
Load: 1 mL spiked porcine serum with 10 µg/mL sulfathiazole (I.S.), 20 µL phosphoric acid
Wash: 1 mL 5% methanol in water
Elute: 1 mL methanol
Evaporate and Reconstitute: 40 °C under nitrogen stream
1 mL mobile phase

Chromatogram of Serum Extracts: A) Blank B) Spiked Sample

<table>
<thead>
<tr>
<th>Compound</th>
<th>Concentration µg/mL</th>
<th>% Recovery</th>
<th>%RSD (n=6)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sulfadiazine</td>
<td>2.00</td>
<td>98.4%</td>
<td>2.7%</td>
</tr>
<tr>
<td></td>
<td>10.0</td>
<td>96.9%</td>
<td>0.65%</td>
</tr>
<tr>
<td>Sulfamerazine</td>
<td>2.00</td>
<td>93.5%</td>
<td>1.9%</td>
</tr>
<tr>
<td></td>
<td>10.0</td>
<td>96.4%</td>
<td>0.74%</td>
</tr>
</tbody>
</table>