**Analysis of Beta Blockers Using a Core Enhanced Technology Accucore HPLC Column**

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**Abstract**

This application note demonstrates the use of the Thermo Scientific Accucore RP-MS HPLC column for the fast analysis of six beta-blockers.

**Introduction**

Accucore™ HPLC columns use Core Enhanced Technology to facilitate fast and high efficiency separations. The 2.6 µm diameter particles are not totally porous, but rather have a solid core and a porous outer layer. The optimised phase bonding creates a series of high coverage, robust phases. Accucore RP-MS uses an optimized alkyl chain length for more effective coverage of the silica surface. This coverage results in a significant reduction in secondary interactions and thus highly efficient peaks with very low tailing. The tightly controlled 2.6 µm diameter of Accucore particles provides much lower backpressures than typically seen with sub-2µm materials.

Beta blockers (or beta antagonists) are a category of drugs used to treat a number of medical complaints, such as hypertension, angina, heart failure and heart attacks. Beta blockers are designed to stop the functioning of a naturally occurring compound, noradrenaline. Noradrenaline is a chemical released in the body which can cause the arteries to narrow and the heart beat to increase.

The separation of these six compounds and more significantly the speed of analysis and the reduced back pressure are demonstrated in this application.

**Sample Preparation**

Primary standard of metoprolol, propranolol and alprenolol at a concentration of 1 mg/mL in water.

Primary standard of atenolol, nadolol and pindolol at a concentration of 1 mg/mL in methanol.

Working standard contained the following in mobile phase:

- 200 µg/mL of pindolol
- 100 µg/mL of atenolol, metoprolol and alprenolol
- 30 µg/mL of nadolol and propranolol

**Thermo Scientific Column**

<table>
<thead>
<tr>
<th>Part Number</th>
<th>Thermo Scientific Column</th>
<th>Measured pressure: 250 bar</th>
</tr>
</thead>
<tbody>
<tr>
<td>17626-052130</td>
<td>Accucore RP-MS 2.6 µm 50 x 2.1 mm</td>
<td>17626-052130</td>
</tr>
</tbody>
</table>

**Thermo Scientific Accela**

<table>
<thead>
<tr>
<th>Part Number</th>
<th>Thermo Scientific Accela</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Column temperature 45 ºC</td>
</tr>
<tr>
<td></td>
<td>Injection volume 1 µL</td>
</tr>
<tr>
<td></td>
<td>Flow rate 0.7 mL/min</td>
</tr>
<tr>
<td></td>
<td>UV detection 220 nm</td>
</tr>
</tbody>
</table>

**Mobile Phase**

Mobile phase A: 0.1% TFA in water
Mobile phase B: 0.1% TFA in methanol
Gradient: 15-65%B in 2.5 minutes

**Consumables**

<table>
<thead>
<tr>
<th>Part Number</th>
<th>Consumables</th>
</tr>
</thead>
<tbody>
<tr>
<td>W/0106/17</td>
<td>Fisher Scientific HPLC grade water</td>
</tr>
<tr>
<td>M/4056/17</td>
<td>Fisher Scientific HPLC grade methanol</td>
</tr>
<tr>
<td>T/3268/PB05</td>
<td>Fisher Scientific Analytical grade TFA</td>
</tr>
<tr>
<td>MSCERT4000-34W</td>
<td>NSC Mass Spec Certified 2 mL clear vial with blue bonded PTFE silicone cap</td>
</tr>
</tbody>
</table>
Results

The analysis was carried out on an Accucore RP-MS 2.6 µm 50 x 2.1 mm column. As shown on Figure 1, the elution of six beta-blockers occurred in less than 2.5 minutes. The backpressure generated using Accucore RP-MS was approximately 250 bar. This application would therefore be suitable for use on a conventional HPLC system.

The peak splitting associated to nadolol (peak 3) was due to the presence of different diastereoisomers of that compound.

Conclusions

Accucore RP-MS columns are an excellent choice for the fast analysis of beta-blockers, allowing high sample throughput at lower back pressures.

![Chromatogram](image)

Figure 1: Chromatogram for atenolol (1), pindolol (2), nadolol (3), metoprolol (4), propranolol (5) and alprenolol (6) separated on an Accucore RP-MS 2.6 µm 50 x 2.1 mm column

<table>
<thead>
<tr>
<th>Analyte</th>
<th>Analyte name</th>
<th>tᵢ (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Atenolol</td>
<td>0.38</td>
</tr>
<tr>
<td>2</td>
<td>Pindolol</td>
<td>0.85</td>
</tr>
<tr>
<td>3*</td>
<td>Nadolol</td>
<td>1.11</td>
</tr>
<tr>
<td>4</td>
<td>Metoprolol</td>
<td>1.58</td>
</tr>
<tr>
<td>5</td>
<td>Propranolol</td>
<td>2.19</td>
</tr>
<tr>
<td>6</td>
<td>Alprenolol</td>
<td>2.28</td>
</tr>
</tbody>
</table>

Table 1: Results obtained from Accucore RP-MS