Isomeric Compounds From Blood Cells
Separation of UDP-Glucose, UDP-Galactose and Galactose-1-phosphate

Method Conditions

Column: Cogent Diamond Hydride™ 4µm, 100Å.
Catalog No.: 70000-15P-2
Dimensions: 2.1mm x 150mm
Solvents:
A: DI water + 0.1% formic acid
B: 90% acetonitrile/10% DI water
16.5 mM ammonium acetate
Gradient:

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<th>Time (min)</th>
<th>%B</th>
<th>Time (min)</th>
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<tr>
<td>0.00</td>
<td>95.0</td>
<td>10.00</td>
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<td>1.00</td>
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<td>3.00</td>
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<td>9.00</td>
<td>75.0</td>
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Post Time: 5 min.
Flow Rate: 0.4 mL/min.
Samples:
1. UDP-glucose: uridine 5'-diphosphateglucose, 565.0477 m/z (M-H)⁻
2. UDP-galactose: uridine 5'-diphosphategalactose, 565.0477 m/z (M-H)⁻
3. Galactose-1-phosphate, 259.0224 m/z (M-H)⁻

Stock standard solutions for analysis were prepared in DI water (1mg/mL of each sample) and were stored at -20°C. For LC-MS analysis samples were diluted 1:100 with 50% acetonitrile/50% DI water solution.


Discussion

It is essential to separate UDP-Glu and UDP-Gal even when using LC-MS detection, because they are isomeric compounds and hence they have the same molecular weight. For most physiologically relevant nucleotides, use of MS is the most practical approach for the analysis of bio samples. This method was developed using an MS detector and a MS friendly mobile phase. The samples were blood cell extracts and the separation resulted in excellent precision.

For more information visit www.MTC-USA.com

Notes:
UDP-glucose, UDP-galactose and galactose-1-phosphate determination can be used for diagnosis of galactosemia in newborn babies [1-3].

Literature: