

Comparing the Separation of Water-Soluble Vitamins using Core-Shell and Fully Porous Polar C18 Columns

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Overview

Vitamins are nutrients that the body needs in small amounts to stay healthy and function properly. Watersoluble vitamins, such as vitamin C and some vitamin B compounds, are carried to the body's tissues but are not stored. Being able to successfully separate these water-soluble vitamins is a useful tool for analytical and clinical labs. In this application note, we present an LC-MS/MS method to separate water-soluble vitamins by utilizing both the Luna® Omega 3 µm Polar C18 column and the Kinetex® 2.6 µm Polar C18 column.

In this method, a panel of water-soluble vitamins was separated using both a fully porous and core-shell polar surface particle chemistry. The all-purpose C18 ligand provides hydrophobic interactions while a polar modified particle surface provides enhanced polar retention and aqueous stability. These attributes make the Polar C18 matrix an excellent choice for a balanced retention of polar and hydrophobic compounds as well as to solely enhance retention of highly polar compounds.

Here we see the Luna Omega 3 μ m Polar C18 column has a slightly longer run time for peaks to elute, but there is a larger selection of water-soluble vitamins that elute. In comparison, the Kinetex 2.6 μ m Polar C18 column has a shorter run time, but with less peaks being resolved. This would suggest that the Kinetex 2.6 μ m Polar C18 column would be a better selection for a smaller, more specific panel of water-soluble vitamins with faster separation. Taken together, this method offers users options when developing a method to separate water-soluble vitamins.

LC-MS/MS Conditions

Column: Luna Omega 3 µm Polar C18

Kinetex 2.6 μm Polar C18

Dimension: 100 x 2.1 mm (Luna Omega)

100 x 3.0 mm (Kinetex)

Part No.: <u>00D-4760-AN</u>

00D-4759-Y0

Mobile Phase: A: 10 mM Ammonium Formate in

0.1 % Formic Acid in Water B: 0.1 % Formic Acid in Acetonitrile

 Gradient: Time (min)
 %B

 0.0
 1

 0.5
 1

 3.5
 20

 4.5
 30

 4.7
 1

 5.7
 1

Flow Rate: 0.5 mL/min (Luna Omega)

0.7 mL/min (Kinetex)

Injection Volume: $10 \mu L$ Temperature: $20 \, ^{\circ}C$

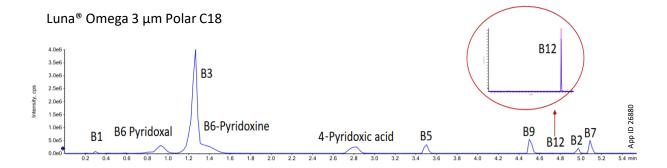
LC System: Agilent® 1260 Infinity

Detection: LC-MS/MS

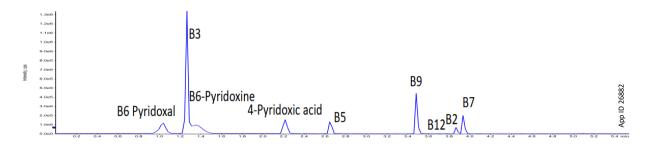
Detector: 4000 QTRAP® (SCIEX®)

MRM Transitions

Analyte	Q1 (m/z)	Q3 (m/z)	Analyte	Q1 (m/z)	Q3 (m/z)
B1 Thiamine - 1	265.2	122	B5 Pantothenic Acid - 1	220.1	72.1
B1 Thiamine - 2	2652	144	B5 Pantothenic Acid - 2	220.1	90.1
B6 Pyridoxal - 1	168	150	B5 Pantothenic Acid - 3	220.1	202.1
B6 Pyridoxal - 2	168	94	B9 Folic Acid - 1	442.2	295.1
B3 Nicotinic Acid - 1	123.98	78.1	B9 Folic Acid - 2	442.2	176
B3 Nicotinic Acid - 2	123.98	80.1	B12 Cyanocobalamin - 1	678.6	147.1
B3 Nicotinic Acid - 3	123.98	53	B12 Cyanocobalamin - 2	678.6	359.2
B6 Pyridoxine - 1	170	152	B2 Riboflavin - 1	377	243.1
B6 Pyridoxine - 2	170	134	B2 Riboflavin - 2	377	172.1
4-Pyridoxic Acid - 1	184.2	148	B7 Biotin - 1	245.1	226.9
4-Pyridoxic Acid - 2	184.2	165.9	B7 Biotin - 2	245.1	96.9



Kinetex[®] 2.6 μm Polar C18



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