

# Separation of an Amino Acid Mixture Using a Luna™ Omega Polar C18 Column

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# **Overview**

Amino acids are the building blocks of proteins, making them an extremely well researched compound class. With thousands of naturally occurring amino acids available, separating and quantitating a mixture of amino acids can be challenging. While HPLC is a well characterized method for amino acid analysis, LC-MS/MS analysis of amino acids poses several challenges.

In this application note, a Luna Omega 3  $\mu$ m Polar C18 column and a robust LC-MS/MS method was used to separate an amino acid mixture. The stationary phase provides a unique selectivity within a wide elution window and increased retention for both polar and non-polar analytes. The all-purpose C18 ligand provides hydrophobic interactions while a polar modified particle surface provides enhanced polar retention and aqueous stability.

# **LC Conditions**

Column: Luna Omega 3 µm Polar C18

**Dimensions:** 150 x 0.3 mm **Part No.:** 00F-4760-AC

Mobile Phase: A: 0.1 % Formic Acid in Water

B: 0.1 % Formic Acid in Methanol

Gradient: Time (min) %B
0 2
6 10
12 85
13 2
20 2

Flow Rate:  $10 \,\mu\text{L/min}$  Injection:  $1 \,\mu\text{L}$  Temperature:  $40 \,^{\circ}\text{C}$ 

Detector: QTRAP® 6500

System: NanoLC™ 425 (SCIEX®)

**Detection:** MS/MS

Table 1. Amino Acids Used and Their Respective MRM Transitions.

| Metabolite             | Q1<br>(m/z) | Q3<br>(m/z) |
|------------------------|-------------|-------------|
| Creatinine (D3)        | 117         | 47          |
| L-Phenylalanine (13C6) | 172         | 126         |
| L-Leucine (13C6)       | 138         | 91          |
| L-Tryptophan (13C11)   | 216         | 155         |
| L-Tyrosine (13C6)      | 188         | 142         |
| L-Alanine (13C3/15N)   | 94          | 47          |

# **MS/MS Conditions**

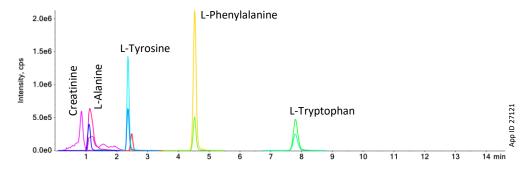
Scan Type: MRM
Polarity: Positive

Source: OptiFlow® Turbo V™

SG1: 20 SG2: 30 CUR: 30 Temperature: 150 °C

Spray Voltage: 5000 V

Figure 1. Extracted Ion Chromatogram of 6 Amino Acids on a Luna Omega 3 μm Polar C18 Column.



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