

Untargeted Metabolomics

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OSPHERE

Traditionally, metabolomics is a difficult analysis with conventional chromatographic tools in the lab. MAC-MOD worked in collaboration with Dr. Tim Garrett at the University of Florida to develop an application with the goal of improving quantitative and qualitative analytical capabilities.

Here we have an untargeted metabolomic screen of a plasma extract via a dried blood spot analysis. We are utilizing monodisperse fully porous particle technology to achieve increased performance and efficiency.

The Evosphere® C18/PFP column was chosen for this due to its combination of regioisomer and hydrophobic selectivity as well as polar retention capacity.

Experimental Conditions

| Instrument: | Thermo Q-Exactive with Dionex Ultimate UHPLC | | |
|------------------------|---|--|--|
| Column: | Evosphere C18/PFP 100 Å , 3 μm , 2.1 x 100 mm Monodisperse Particle Column | | |
| PN#: | EVO18FP020503 | | |
| Mobile Phase A: | 0.1% Formic Acid in H_2O | | |
| Mobile Phase B: | Acetonitrile | | |
| Sample: | Plasma Extract | | |
| Temp: | 25 °C | | |
| Injection Volume: 2 µl | | | |
| | | | |

| Time (min) | % B | Flow Rate |
|---------------|------------|-----------|
| 1 | 0 | 0.35 |
| 13 | 80 | 0.35 |
| 16 | 80 | 0.35 |
| 16.5 | 0 | 0.35 |
| 16.8 | 0 | 0.60 |
| 20 | 0 | 0.60 |
| 20.5 | 0 | 0.60 |
| | | |

8.48

8.15

15.03

13.34 13.97

16.33

н

12.33

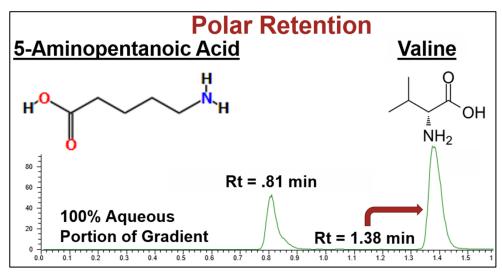
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Application Note

Untargeted Metabolomics

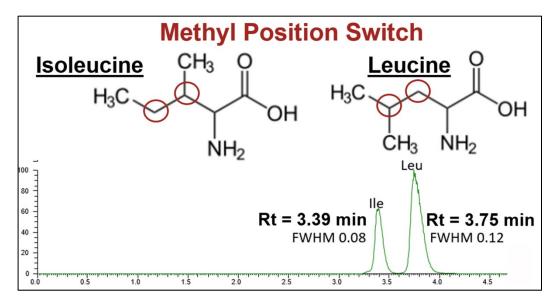
A- 5-Aminopentoic Acid, B- Valine

5-Aminopentoic Acid and Valine are difficult to retain in reversed-phase on a standard C18 column, whereas these analytes are easily retained in the 100% aqueous portion of the gradient with C18/PFP. This leverages the 100% aqueous compatibility and polar retention capability of the C18/PFP phase chemistry.



C- Isoleucine, D- Leucine

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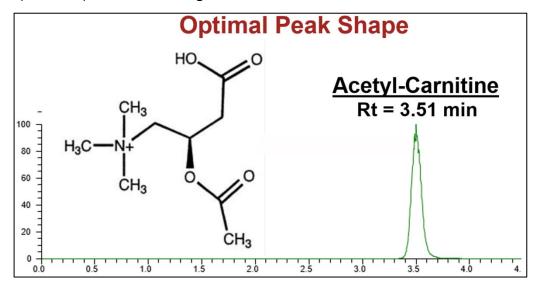




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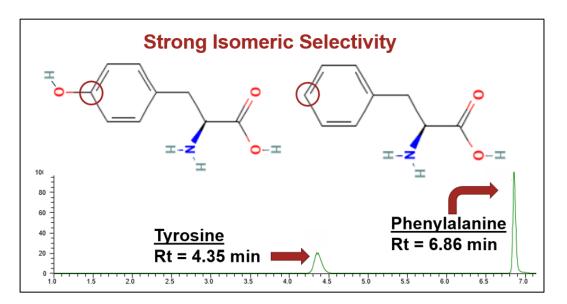
E- Acetyl-Carnitine

In separations utilizing standard C18 columns, Acetyl-Carnitine exhibits tailing due to its positively charged quaternary amine chemical structure. This makes it difficult to get excellent peak shape with low ionic strength mobile phases, whereas the C18/PFP provides Gaussian peak shape with no tailing.



F- Tyrosine, G- Phenylalanine

The Evosphere C18/PFP is able to separate two closely related molecules in Tyrosine and Phenylalanine due to the combination of PFP functionality and C18 hydrophobicity in the bonding chemistry.

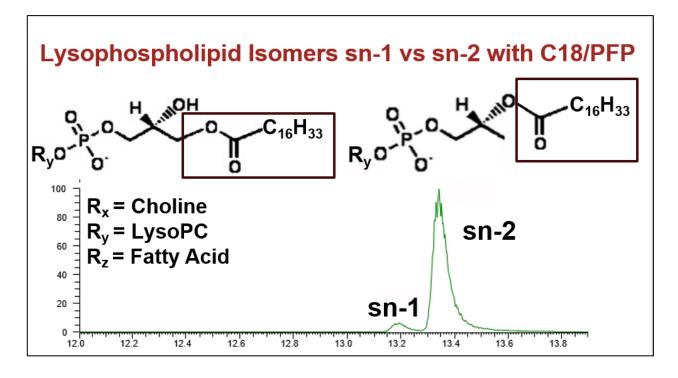




Application Note

H- Lysophospholipid sn-1, I- Lysophospholipid sn-2

Traditionally, sn-1 and sn-2 Lysophospholipid isomers are difficult to separate with standard C18 columns, whereas C18/PFP is able to baseline resolve these isomers through its unique separating capability.



For technical support or applications contact:

info@mac-mod.com

For more information visit:

www.mac-mod.com

For all supply in North America and Canada please contact MAC-MOD Analytical

Tel: 800-441-7508

info@mac-mod.com



Fortis ••••• Technologies Ltd.

mac-mod.com

info@mac-mod.com

800-441-7508