

# MMA from Plasma Analyzed by HPLC using Luna® Omega C18 Columns Coupled with Verex™ Filter Vials

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## Overview

Samples are prepared by a variety of methods, all of which can influence chromatographic results by affecting the sample injected into the system. Ineffective sample preparation can lead to a reduction in chromatographic separation efficiency and reproducibility, and many other problems including decreased column lifetimes. Inappropriate choice of sample preparation devices could release extractable compounds into the sample, influencing the quality and reproducibility of the results observed.

In this application, Methylmalonic Acid (MMA) samples were prepared and filtered using Verex Filter Vials. Samples were then run by HPLC-MS to determine if there was a loss of sample as well as determine if any extractables were introduced by using the Verex Filter Vials. When compared to the MMA samples without a filtration device, the filtered samples using the Verex Filter Vials showed similar results. This would indicate no adsorption to the membrane nor vial occurred and no extractables were introduced from the Verex Filter Vials.

## Sample Preparation

<b>Make:</b>	100 ng/mL of SA, MMA in 2x stripped plasma from a 1 mg/mL stock solution of each compound.
<b>Mix:</b>	100 µL with 300 µL of 0.5 % Formic Acid in methanol.
<b>Spin:</b>	10 min at 4,000 RPM
<b>Load:</b>	300 µL of supernatant into Verex Filter Vial 0.2 µm, RC <a href="#">ARO-F103-12</a>
<b>Inject:</b>	10 µL final eluate onto LC-MS

## LC-MS Conditions

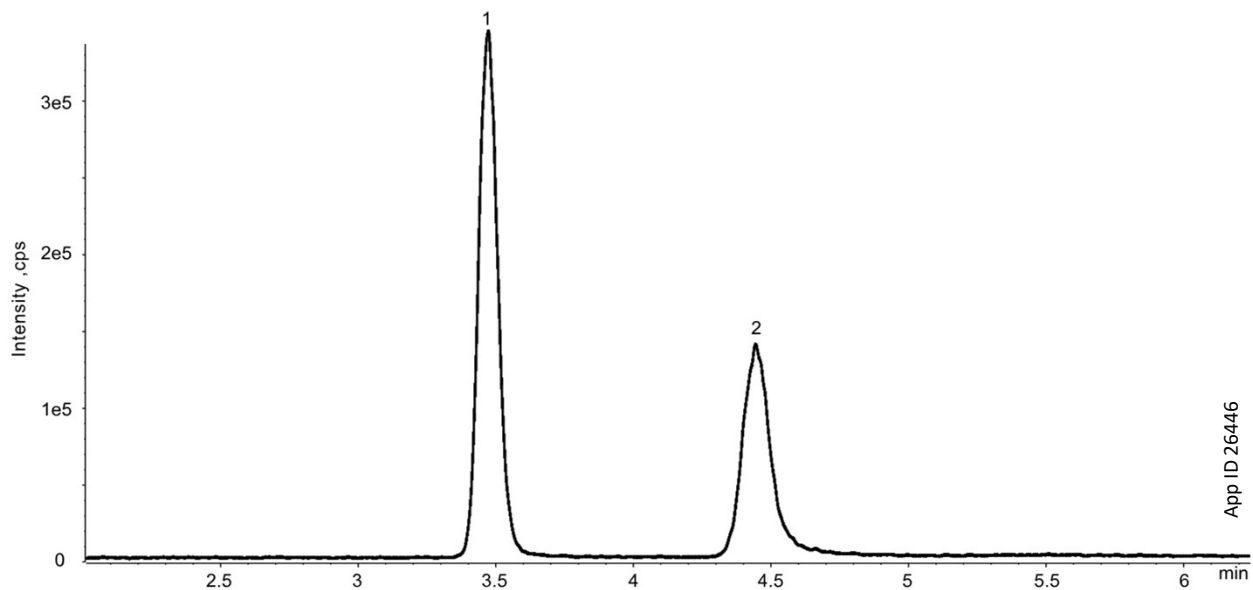
<b>Column:</b>	Luna Omega 3 µm C18	
<b>Dimension:</b>	150 x 3.0 mm	
<b>Part No.:</b>	<a href="#">00F-4784-YO</a>	
<b>Pressure (bar):</b>	350	
<b>Mobile Phase:</b>	A: 0.1 % Formic Acid in Water B: 0.1 % Formic Acid in Methanol	
<b>Gradient:</b>	<b>Time (min)</b>	<b>% B</b>
	0	5
	0.4	5
	5.4	95
	5.9	95
	6.4	5
	15	5
<b>Flow Rate:</b>	450 µL/min	
<b>Temperature:</b>	Ambient	
<b>Detection:</b>	ESI-	
<b>Detector:</b>	5500 QTRAP® (SCIEX®)	
<b>Injection Volume:</b>	10 µL	
<b>Instrument:</b>	Agilent® 1100 HPLC with Quaternary Pump	

## MRM Transitions

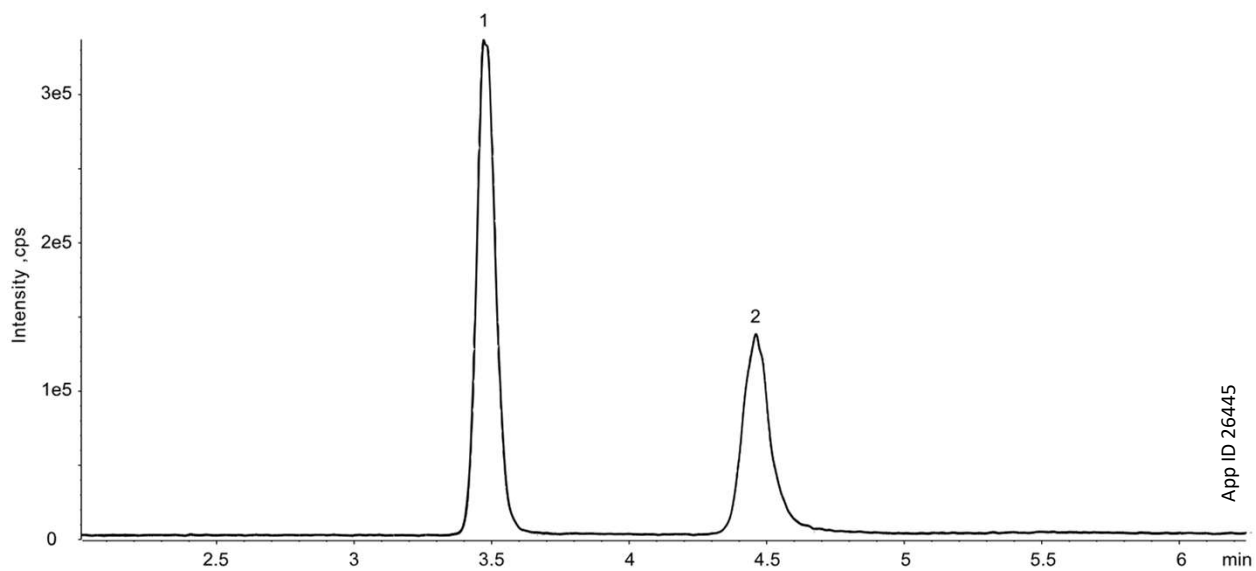
Peak No.	Analyte	Q1 (m/z)	Q3 (m/z)
1	Succinic Acid	117	73
2	Methylmalonic Acid	117	73



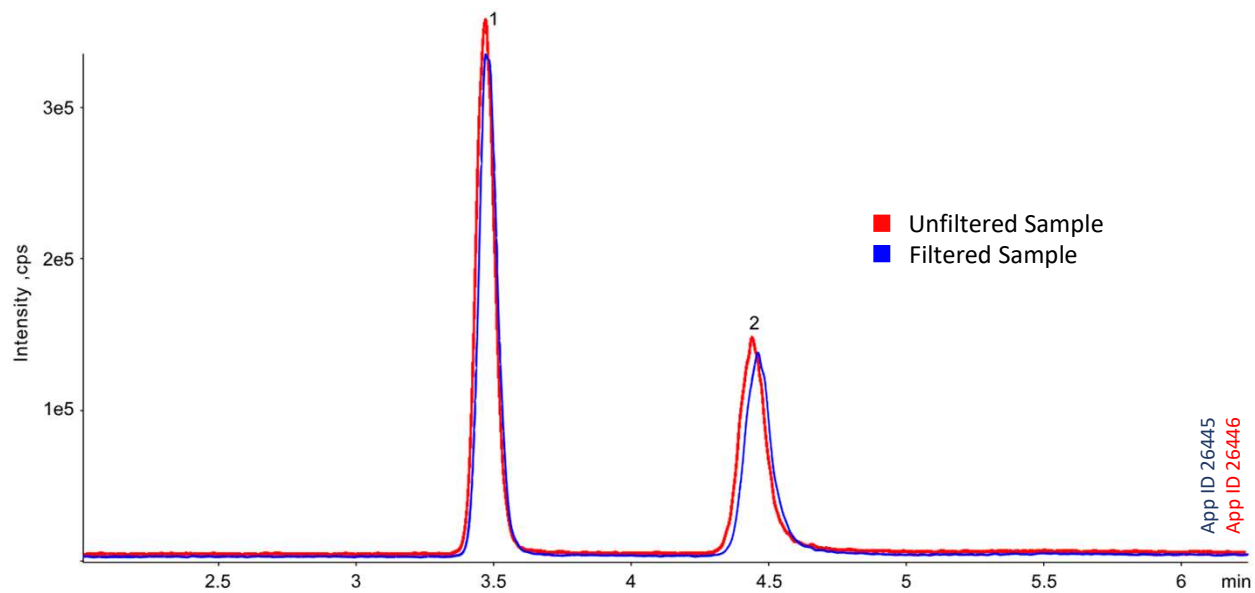
**Figure 1.** Luna® Omega C18 separation of MMA without the use of a Verex™ Filter Vial.



**Figure 2.** Luna Omega C18 separation of MMA with the use of a Verex Filter Vial.



**Figure 3.** Overlay of unfiltered and filtered samples.



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