

APPLICATIONS

Extraction of 25-OH Vitamin D2/D3 From Plasma Using Strata[®] DE Supported Liquid Extraction (SLE)

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Overview

In this application we provide a supported liquid extraction (SLE) method for the cleanup of 25-OH Vitamin D2/D3 metabolites from plasma. This method provides recoveries of greater than 80 % and RSD values less than 10 % for both 25-OH Vitamin D2 and D3. The results presented in this application note show an acceptable method for extracting Vitamin D metabolites from plasma by utilizing Strata DE, a diatomaceous earth SLE product.

Pre-treatment

Dilute 100 µL of spiked plasma (100 ng/mL) with 100 µL of 1:1 (IPA/Water).

SLE Protocol

96-Well Plate:	Strata DE 200 µL 96-Well Plate
Part No.:	8E-S325-FGB
Load:	200 µL Pre-treated sample onto plate (apply vacuum or positive pressure to pull/push sample into sorbent)
Wait:	5 minutes
Elute:	2x 600 µL Heptane
Apply:	Vacuum or apply positive pressure at 5-10" Hg for 10 seconds
Dry:	Sample under slow stream of Nitrogen at 30 °C
Reconstitute:	100 µL Methanol/Water (80:20) with internal standard

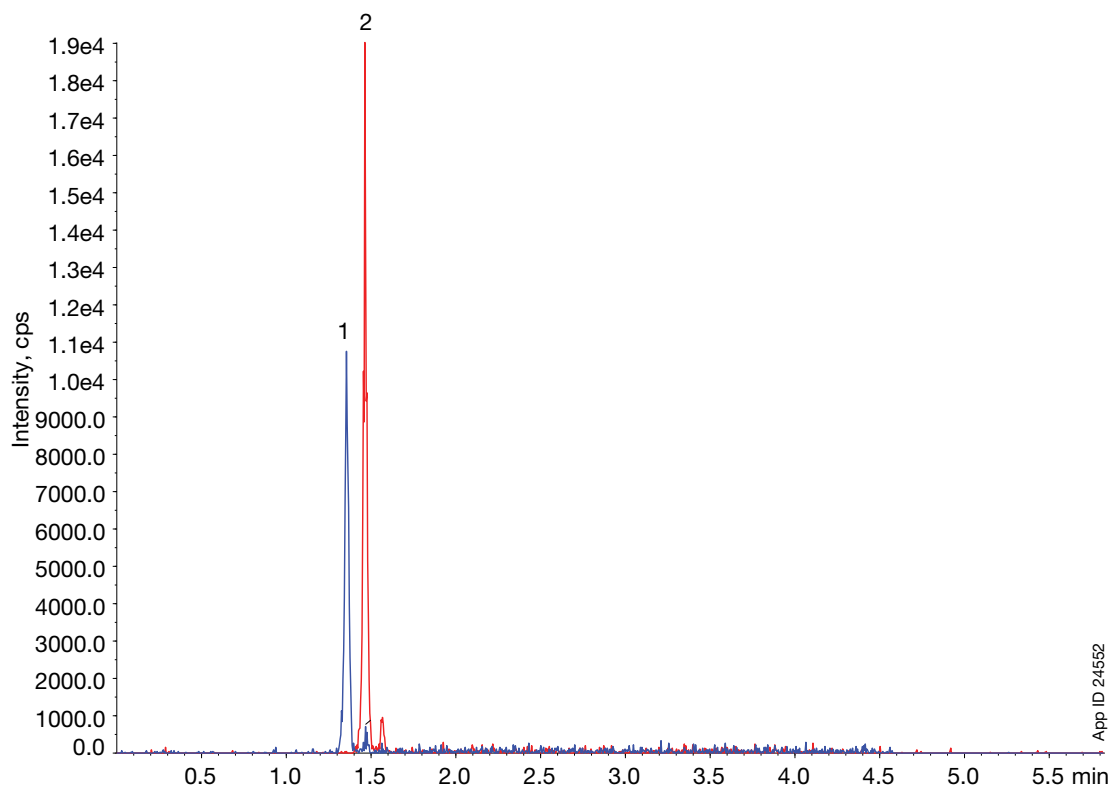
LC-MS/MS Conditions

Column: Kinetex[®] 2.6 µm EVO C18
Dimensions: 50 x 2.1 mm
Part No.: 00B-4725-AN
Guard Cartridge: SecurityGuard[™] ULTRA EVO C18
Guard Part No.: AJ0-9298
Mobile Phase: A: 0.1 % Formic acid in Water
 B: 0.1 % Formic acid in Acetonitrile

Gradient: Time (min)	% B
0	60
0.83	95
3.33	95
3.34	60
5.83	60

Injection Volume: 20 µL
Flow Rate: 0.5 mL/min
Detection: MS/MS (SCIEX API 4000[™]), ESI+
Sample: 1. 25-OH-Vitamin D3
 2. 25-OH-Vitamin D2

	25-OH Vitamin D2	25-OH Vitamin D3
Recovery	96 %	84 %
RSD (n=8)	9 %	9 %



App ID 24552

APPLICATIONS

Ordering Information

Kinetex[®] EVO C18 Core-Shell LC Columns

2.6 µm Minibore Columns (mm)					SecurityGuard [™] ULTRA Cartridges [†]
Phases	30 x 2.1	50 x 2.1	100 x 2.1	150 x 2.1	3/pk
EVO C18	00A-4725-AN	00B-4725-AN	00D-4725-AN	00F-4725-AN	AJ0-9298 for 2.1 mm ID

2.6 µm MidBore [™] Columns (mm)				SecurityGuard [™] ULTRA Cartridges [†]
Phases	50 x 3.0	100 x 3.0	150 x 3.0	3/pk
EVO C18	00B-4725-YO	00D-4725-YO	00F-4725-YO	AJ0-9297 for 3.0 mm ID

2.6 µm Analytical Columns (mm)				SecurityGuard [™] ULTRA Cartridges [†]
Phases	50 x 4.6	100 x 4.6	150 x 4.6	3/pk
EVO C18	00B-4725-E0	00D-4725-E0	00F-4725-E0	AJ0-9296 for 4.6 mm ID

[†] SecurityGuard ULTRA Cartridges require holder, Part No.: AJ0-9000

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Strata[®] DE Supported Liquid Extraction

Part No.	Description	Unit
8E-S325-FGB	Strata DE SLE 200 µL 96-Well Plate	2/pk
8E-S325-5GB	Strata DE SLE 400 µL 96-Well Plate	2/pk
8B-S325-KDG	Strata DE SLE 12 cc Tube	20/pk
8B-S325-VFF	Strata DE SLE 60 cc Tube	16/pk

Presston[™] 100 Positive Pressure Manifold

Part No.	Description
AH0-9334	Presston 100 Positive Pressure Manifold, 96-Well Plate
AH0-9342	Presston 100 Positive Pressure Manifold, 1 mL Tube Complete Assembly
AH0-9347	Presston 100 Positive Pressure Manifold, 3 mL Tube Complete Assembly
AH0-9343	Presston 100 Positive Pressure Manifold, 6 mL Tube Complete Assembly

Presston 100 Tube Adapter Kits (for AH0-9334)

Part No.	Description
AH0-9344	1 mL Tube Adapter Kit
AH0-9345	3 mL Tube Adapter Kit
AH0-9346	6 mL Tube Adapter Kit



WARRANTY Phenomenex warrants that for a period of 12 months following delivery, the Presston 100 Positive Pressure Manifold you have purchased will perform in accordance with the published specifications and will be free from defects in materials or workmanship. In the event that the Presston 100 Positive Pressure Manifold does not meet this warranty, Phenomenex will repair or replace defective parts. Please visit www.phenomenex.com/Presston for complete warranty information.



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