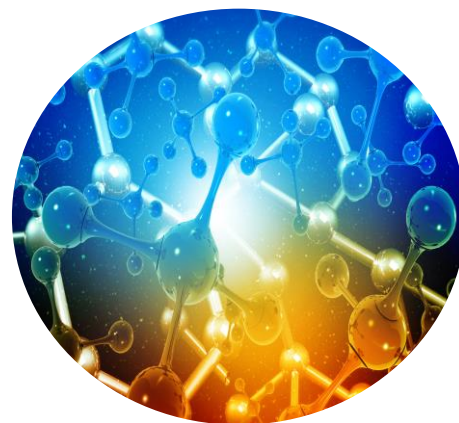


TN-0144

Comparing Two Different Simplified Liquid Extraction Sorbents for Low Level Steroids from Plasma

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Introduction

Simplified Liquid Extraction (SLE) is a fast and easy sample preparation technique for cleaning up matrices before LC/MS-MS analysis. To obtain good results at low steroid levels, baseline noises need to be monitored and ensured that products are selected in order to get the most accurate analysis. This can be hard to control using a natural diatomaceous earth SLE sorbent, which leads to irreproducible results and wasted time in the laboratory. Novum™ PRO SLE is a synthetic sorbent that undergoes additional manufacturing requirements to help get accurate MS detection at extremely low level extractions from complex matrices. In this application, a low level extraction using Novum PRO is compared to Biotage® ISOLUT+ SLE from urine. This also leads to high recoveries for steroid panels and this application provides an effective, easy SLE sample preparation method paired with a high efficiency Kinetex® 5 µm Biphenyl Core-Shell LC column for good separation and fast run times.

SLE Method		
Pre-treatment:	190 µL plasma with 10 µL spike standards solution	
96-Well Plate:	Novum PRO MAX 96-Well Plate	
Part No.:	8E-S539-5GA	
Load:	200 µL spiked plasma + 200 µL Sodium phosphate buffer.	
Elution:	2x 900 µL Ethyl acetate	
LC Conditions		
Column:	Kinetex 5 µm Biphenyl	
Dimensions:	50 x 4.6 mm	
Part No.:	00B-4627-E0	
Mobile Phase:	A: Water B: Methanol	
Gradient:	Time (min)	%B
	0	80
	1.5	95
	3	95
	3.1	80
	5	80
Flow Rate:	1 mL/min	
Temperature:	Ambient	
Injection:	1 µL	
Detection:	6500+ MS/MS (ESI+)	
System:	Agilent® Technologies 1200 Series	

Figure 1.
11-deoxycorticosterone Extracted from Plasma using DCM
Comparing SLE products

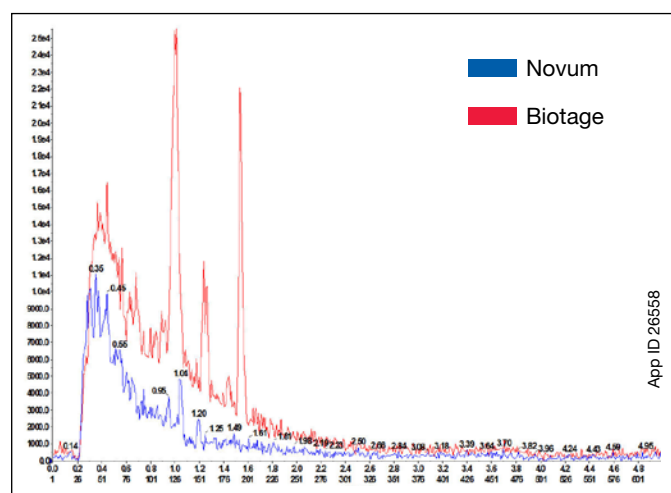
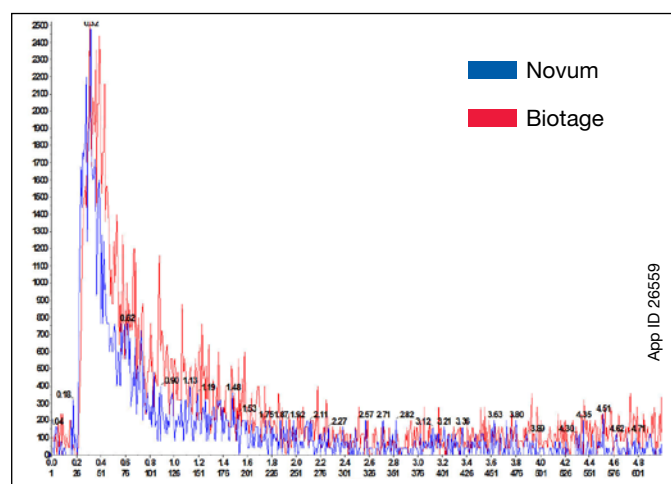


Figure 2.
Progesterone Extraction from Plasma using 25:75 Ethyl Acetate/
Hexane Comparing SLE products



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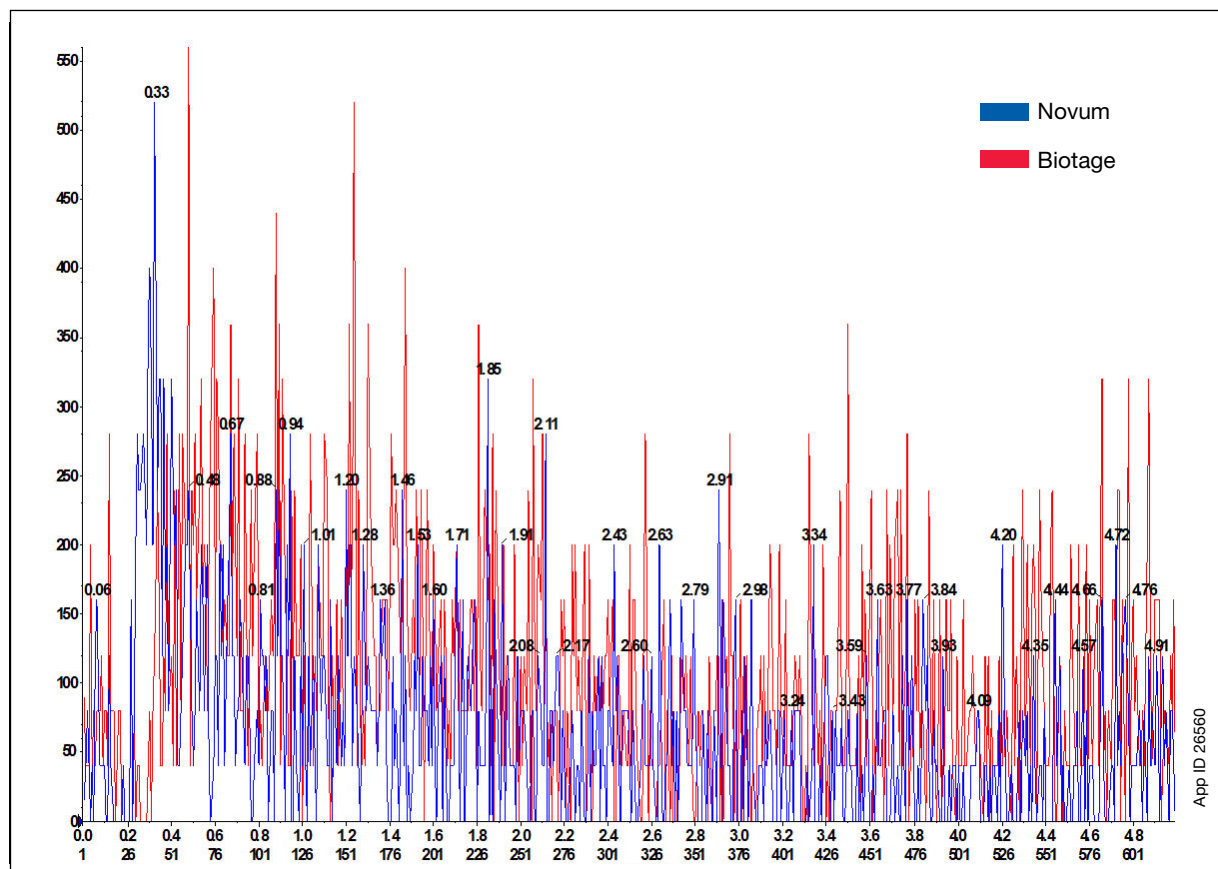
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Figure 3.
Progesterone Extraction from Plasma using 90:10 DCM/Ethyl acetate Comparing SLE Products



Results and Discussion

Diverse extraction solvents were tested for the SLE products to ensure that the best solution is selected. DCM, 90:10 DCM/ Ethyl acetate, and 25:75 Ethyl Acetate/Hexane were all tested. Using a comparison between Novum PRO and Biotage SLE is observed in **Figures 1, 2, and 3** all with different extraction solvents.

In **Figure 1**, there is a noticeable peak present at low levels for the Biotage® SLE product that is not present in the Novum™ PRO with a DCM extraction solvent. This is because the Novum PRO product is lab manufactured to reduce natural deviation that can occur when using a diatomaceous earth sorbent that could offer

variety from well to well. For the most part, Novum PRO offers a cleaner baseline using different extraction solvents at low levels for steroid extraction, such as 5-10 pg/mL that would be necessary for a clinical laboratory. In conclusion, Novum PRO makes it possible to get lower extractions for routine steroid extraction to make getting accurate results easier, and with a simple method that is fast yet leads to accurate quantifiable results.



Kinetex® Ordering Information

3.5 µm Minibore and Analytical Columns (mm)						SecurityGuard™ ULTRA Cartridges [†]	
Phases	50 x 2.1	150 x 2.1	100 x 4.6	150 x 4.6	250 x 4.6	3/pk	3/pk
XB-C18	—	—	00D-4744-E0	00F-4744-E0	—	—	AJO-8768
PAH	00B-4764-AN	00F-4764-AN	00D-4764-E0	00F-4764-E0	00G-4764-E0	AJO-9535	AJO-9533
						for 2.1 mm ID	for 4.6 mm ID

5 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
Biphenyl	00A-4627-AN	00B-4627-AN	00D-4627-AN	—	AJO-9209
					for 2.1 mm ID

5 µm MidBore™ Columns (mm)					SecurityGuard™ ULTRA Cartridges [†]
Phases	30 x 3.0	50 x 3.0	100 x 3.0	150 x 3.0	3/pk
Biphenyl	—	00B-4627-Y0	00D-4627-Y0	00F-4627-Y0	AJO-9208
					for 3.0 mm ID

5 µm Analytical Columns (mm)					SecurityGuard™ ULTRA Cartridges [†]
Phases	50 x 4.6	100 x 4.6	150 x 4.6	250 x 4.6	3/pk
Biphenyl	00B-4627-E0	00D-4627-E0	00F-4627-E0	00G-4627-E0	AJO-9207
					for 4.6 mm ID

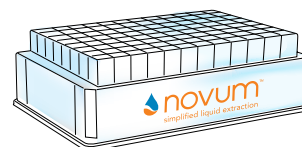
[†]SecurityGuard ULTRA Cartridges require holder, Part No.: [AJO-9000](#)



Novum™ Ordering Information

Novum SLE 96-Well Plates		
Part No.	Description	Unit
8E-S138-FGA	Novum SLE MINI 96-Well Plate	1/pk
8E-S138-5GA	Novum SLE MAX 96-Well Plate	1/pk
8E-S539-FGA	Novum PRO SLE MINI, 96-Well Plate	1/pk
8E-S539-5GA	Novum PRO SLE MAX, 96-Well Plate	1/pk

Accessories		
Collection Plates (deep well, polypropylene)		Unit
AH0-7192	96-Well Collection Plate, 350 µL/well Conical	50/pk
AH0-7193	96-Well Collection Plate, 1 mL/well Conical	50/pk
AH1-7022	96-Well Collection Plate, 2 mL/well Conical	50/pk
AH0-8636	96-Well Collection Plate, 2 mL Round/Round, 8 mm	50/pk
AH1-7025	96-Well Collection Plate, 1 mL/well Round, 7 mm	50/pk
Sealing Mats		Unit
AH0-8597	Sealing Mats, Pierceable, 96-Square Well, Silicone	50/pk
AH0-8598	Sealing Mats, Pre-Slit, 96-Square Well, Silicone	50/pk
AH0-7362	Sealing Tape Pad	10/pk
Vacuum and Positive Pressure Manifolds		Unit
AH0-8950	96-Well Plate Manifold, Universal with Vacuum Gauge	ea
VM12	12-Position Vacuum Manifold Set	ea
VM24	24-Position Vacuum Manifold Set	ea
AH1-7033	Presston 1000 Positive Pressure Manifold, 96-Well Plate	ea



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