

Steroid Panel Extraction from Plasma Using Novum PRO Simplified Liquid Extraction (SLE)

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Overview

Steroids can often be a difficult to extract from a complex matrix due to their sensitive nature and need for a clean background. Novum PRO SLE 96-well plates offer a synthetic SLE media that result in a fast sample preparation method, and with the addition of advanced manufacturing processes, the results lead to high recoveries and low variation between samples. 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 as demonstrated in **Figure 1** and resulted in recovery values that were greater than 80 % (**Table 1**).

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

Table 1. % Absolute recovery of Steroid Panel Extracted from Plasma using a Novum PRO MAX 96-Well Plate

Analyte	% Absolute Recovery	% CV
17-OH Progesterone	115	14
Corticosterone	106	6
Cortisol	80	15
Cortisone	82	12
Progesterone	105	14
Testosterone	114	10
11-deoxycorticosterone	106	6

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	%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+)

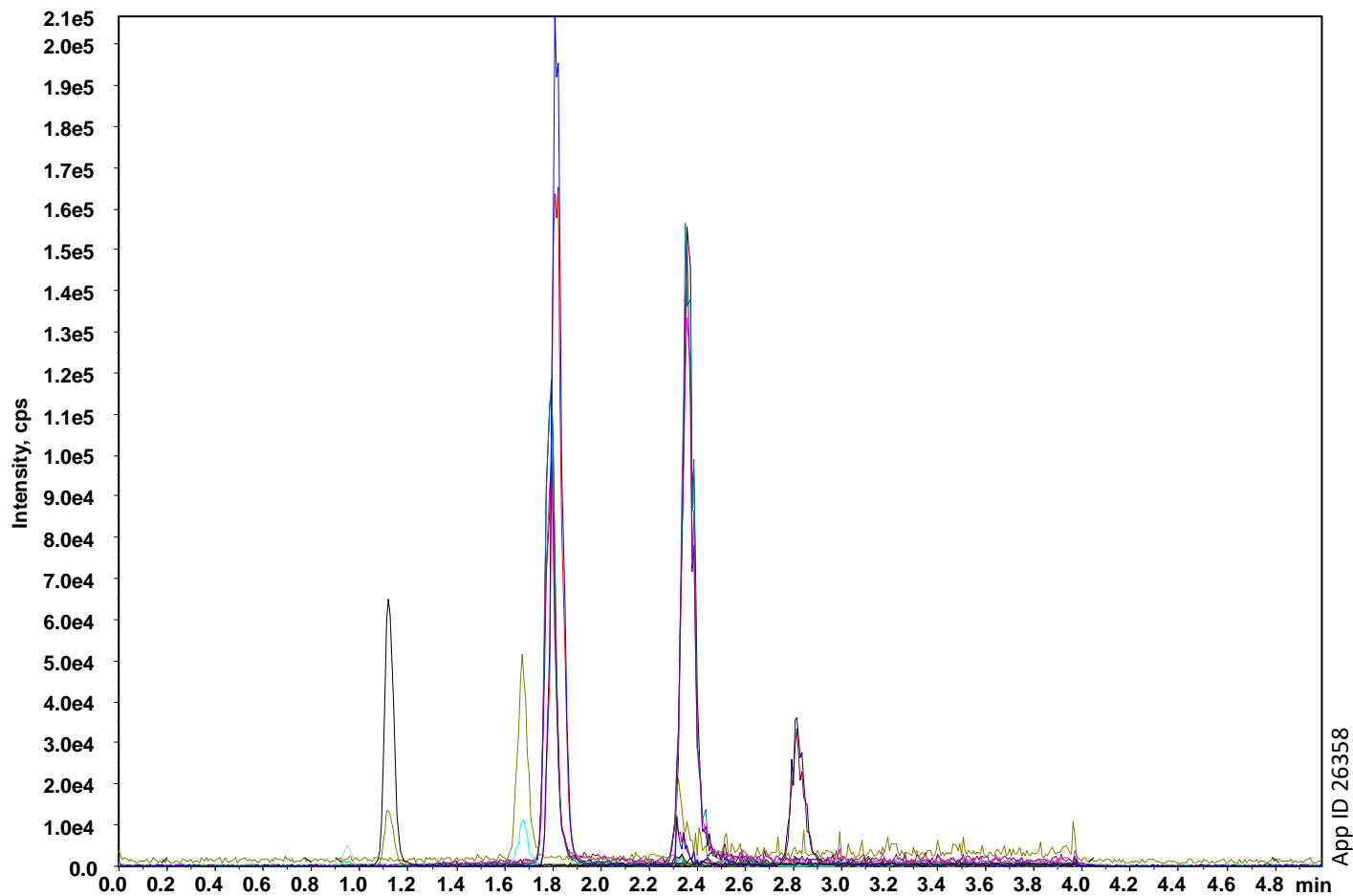
System: Agilent® Technologies 1200 Series

MS Conditions

Parameter	Value
CAD	3
CUR	26
GS1	60
GS2	60
IS	5500
TEM	640



Figure 1. Representative Chromatogram of Steroid Panel



Analytes:

Peak Number	Analyte	Retention Time
1	Cortisol	0.94
2	Cortisone	1.12
3	Corticosterone	1.68
4	11-deoxycorticosterone	1.79
5	Testosterone	1.81
6	17-OH progesterone	2.36
7	Progesterone	2.81



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Novum is patent pending.

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