

APPLICATIONS

Isolation of Low Level Testosterone from Plasma by LC-MS/MS Analysis in Conjunction with NovumTM PRO MAX Simplified Liquid Extraction (SLE)

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Overview

In this study testosterone was isolated from plasma using Novum PRO MAX 96-Well Plates SLE sample preparation. These samples were then injected onto LC-MS/MS for analysis. Testosterone was successfully isolated giving excellent recovery and lowest level of quantitation. Novum PRO utilized an optimized manufacturing process to provide less background noise which leads to cleaner samples post sample preparation.

Sample Preparation

Pretreatment:	200 μ L spiked plasma + 200 μ L 50 mM Sodium phosphate buffer, pH unadjusted
Load:	Novum PRO MAX 96-Well Plate, wait 5 minutes
Part No.:	8E-S539-5GA
Elute:	2 x 900 μ L 1:3 Hexane:Ethyl acetate
Dry:	Under Nitrogen
Add:	50 μ L Dansyl chloride/Sodium bicarbonate (50:50)
Reconstitute:	50 μ L 80:20 Water:Methanol
Inject:	Inject 20 μ L onto LC-MS/MS

LC-MS/MS Conditions

Column: Kinetex[®] 1.7 μ m Biphenyl 100 \AA
Dimension: 50 x 2.1 mm
Part No.: [00B-4628-AN](#)
Mobile Phase: A: 0.1 % Formic acid in Water
 B: 0.1 % Formic acid in Methanol

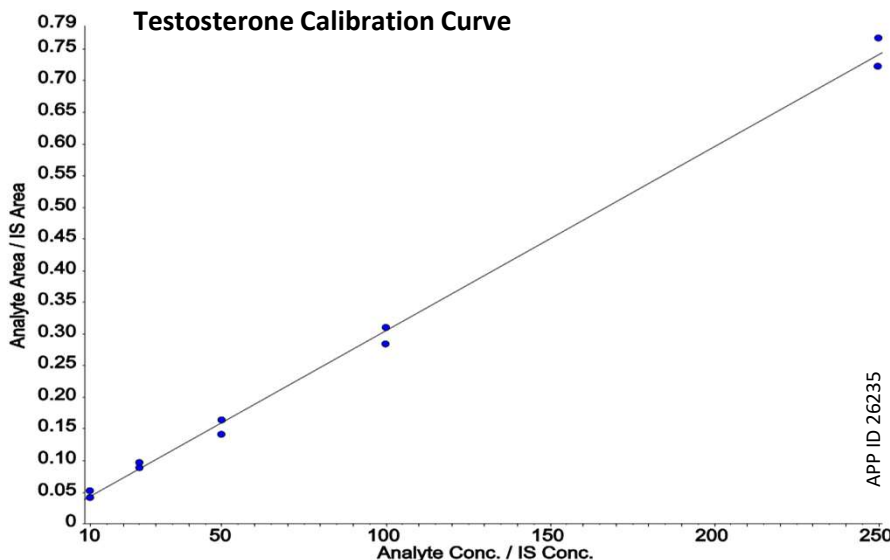
Gradient:	Time (min)	%B
	0	60
	2	95
	3	95
	3.1	40
	5	40

Flow Rate: 0.4 mL/min
Temperature: Ambient
Detection: SCIEX[®] 6500 MS/MS

MS/MS Conditions

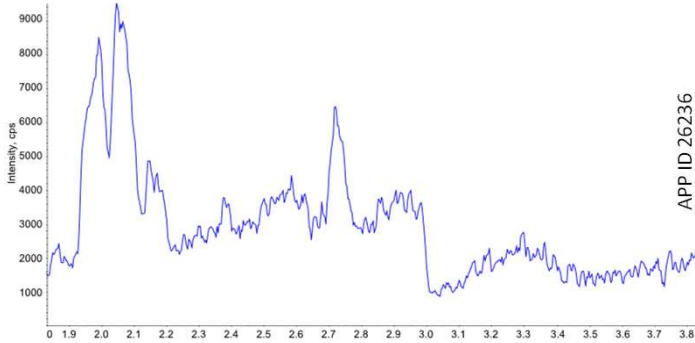
Ion Source: Positive
CUR: 23
CAD: High
GS1: 60
GS2: 50
IS: 2500
Source Temp: 450 $^{\circ}$ C

Q1	Q3	Time	ID	DP	CE	CXP
289.1	97	50	testo_1	27	27	15
289.1	109	50	testo_2	27	30	18
292.1	97	50	testo-D3	27	27	15
292.1	109	50	testo-D3	27	30	18

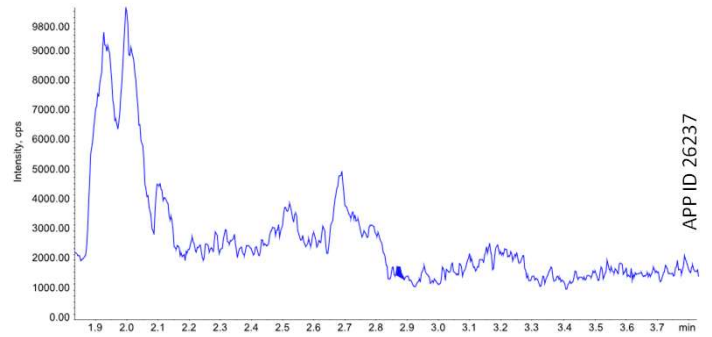


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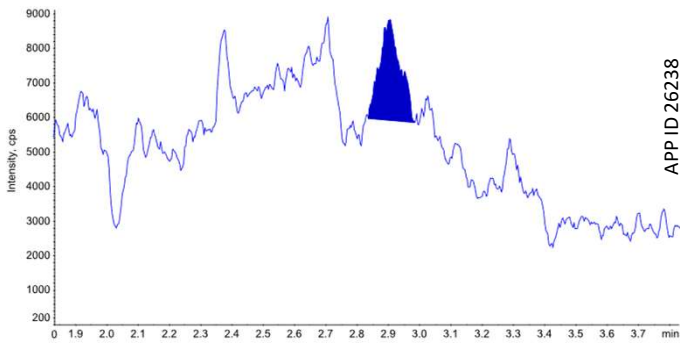
Plasma Blank



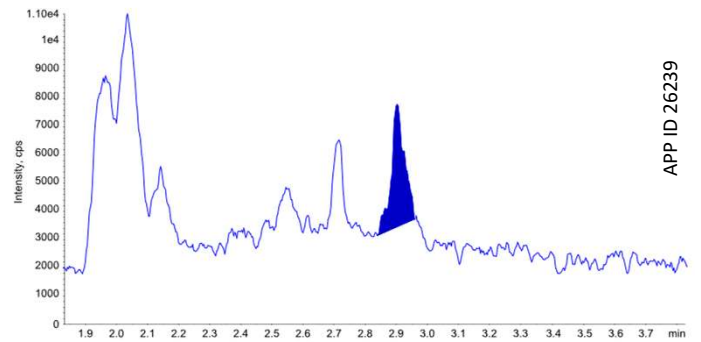
Solvent Blank



10 pg/mL Testosterone (LLOQ)



20 pg/mL Testosterone



Testosterone	% Recovery	% CV
20 pg/mL	91	10
40 pg/mL	108	9
80 pg/mL	101	12
150 pg/mL	105	12
300 pg/mL	103	12

APPLICATIONS

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Novum is patent pending.

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