

Testosterone from Female Human Plasma by LC/MS/MS using Strata-X-A and Kinetex 1.7 µm C18

Column: Kinetex® 1.7 µm C18 100 Å, LC Column 30 x 2.1 mm, Ea

Dimensions: 30 x 2.1 mm ID

Order No: 00A-4475-AN

Elution Type: Gradient

Eluent A: 0.1% Formic Acid +1 mM Amm Formate in Water

Eluent B: 0.1% Formic Acid +1 mM Amm Formate in ACN

Gradient Profile:	Step No.	Time (min)	Pct A	Pct B
	1	0	90	10
	2	2.5	10	90
	3	3.5	10	90
	4	3.6	90	10

Flow Rate: 0.4 mL/min

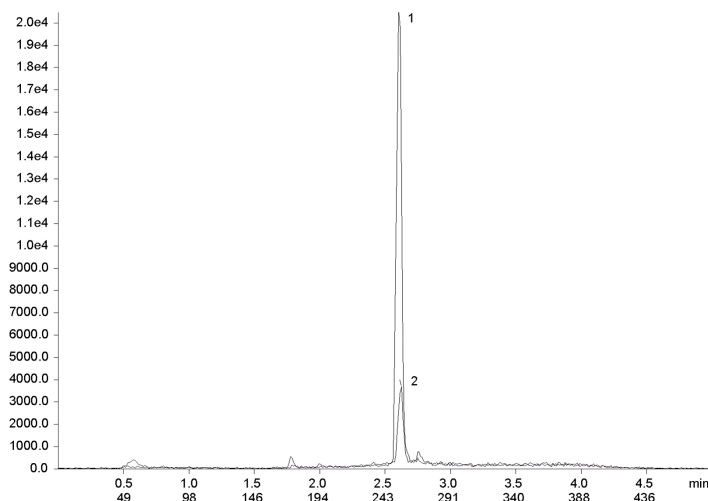
Col. Temp.: 55 °C

Detection: Mass Spectrometer (MS) @ amu (ambient)

Detector Info: SCIEX



Products used in this application:



ANALYTES:

- 1 Testosterone (304.3 > 124.0; 304.3 > 112.0)
Retention Time: 2.62 min
- 2 Testosterone-d3 (307.3 > 112.0; 307.3 > 124.0)
Retention Time: 2.61 min



Sample Preparation Details

for UHPLC Application ID No.: 19762

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PRODUCT DESCRIPTION:

Strata™-X-A 33 µm Polymeric Strong Anion, 30 mg / 3 mL, Tubes , 50/Pk

Order No.: 8B-S123-TBJ

SOLID PHASE EXTRACTION (SPE) PROCEDURE:

Note: The solvent volumes shown below are for a 30 mg bed mass.

The solvent volumes will need to be adjusted for a smaller or larger bed mass.

Condition:

Load:

Into individually labeled test tube combine 0.25 mL serum sample (or calibrator or QC sample), 1 mL DI water and 0.1 mL working internal standard solution (2 ng/mL).
Proceed to SPE procedure

Wash:

Dry:

Dry for 5 min under high vacuum

Elute:

Final Prep and Analysis:

Following evaporation of elution solvent @ 50-55 C under gentle nitrogen stream;
Add 50 µL 25% hydroxylamine solution and heat at 60-65 C for 5-10 min, then add 200 µL 5%

Inject: 20 µL on HPLC Mass Spectrometer (MS) @ amu (ambient)

ANALYTES:	Spiked Conc. (ng/mL)	Log P	pKa	% Rec	%RSC (n=0)
1 Testosterone	0				
2 Testosterone-d3	0.5				

Note: This method is designed as a convenient starting point for further investigation and can be tailored to meet your extraction goals.
Call your local Phenomenex Representative for assistance in method development and optimization techniques.

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For more information contact your Phenomenex Representative at info@phenomenex.com



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