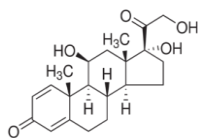
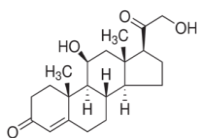


Supported Liquid-liquid Extraction Plate

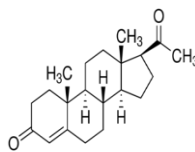
Compounds :



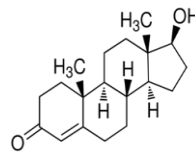
Prednisolone



Corticosterone



Progesterone



Testosterone

SPE EXTRACTION METHOD:

(Supported liquid-liquid extraction plate: catalog number: OC21DA400-HPGB, 400 mg/well)

Pre-treat 100 μ L of human plasma with 100 μ L of water



Load pretreated plasma; let it flow by gravity, or apply a pulse of pressure or vacuum



Dry the bed for 5 minutes



Elute with 1 mL of hexane/ethyl acetate(1:1) twice. Let it flow by gravity



Evaporate eluate under nitrogen at 40C.
Reconstitute with 40% ACN

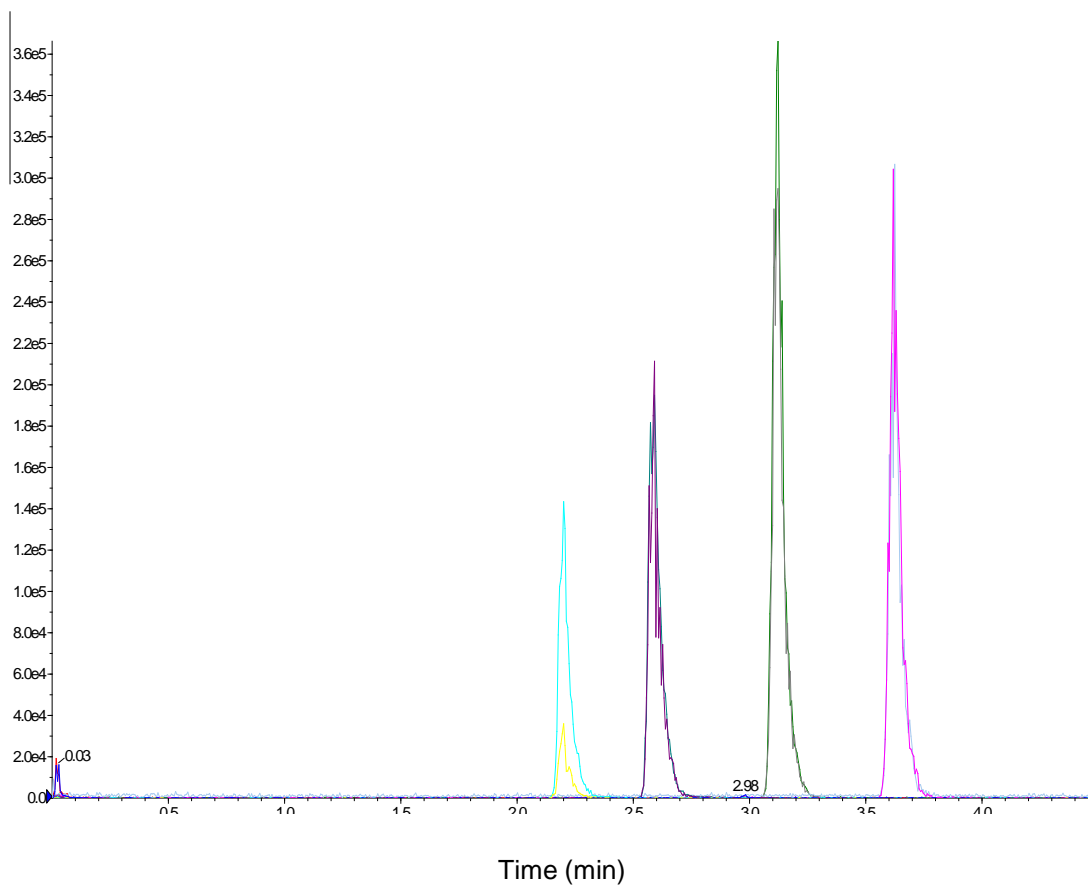
MS/MS CONDITIONS:

Mass spectrometer: API3000
Ion source: Turbo IonSpray
Polarity: Positive
IS: 5000
Temp. 500
DP: 30
CAD: 12
EP: 10
CE: various by analytes
CXP: 10
Scan mode: MRM

HPLC METHOD:

Column: Orochem Reliasil C8 HPLC column 4.6x50 mm, 3 μ m.
Part number: R3BI-102
Mobile phase: A: 0.1% formic acid in water
 B: 0.1% formic acid in ACN
Gradient: Initial 40% B for 0.5 min, then gradient to 80% B at 2.5 min, and hold 2 min
Flow rate: 0.5 mL/min
Injection volume: 10 μ L
Retention Time:
 Prednisolone– 2.1 min
 Corticosterone– 2.5 min
 Progesterone – 3.6 min
 Testosterone – 3.1 min

Steroids	Quantifier ions	Qualifier ions
	Q1/Q3	Q1/Q3
Estradiol	273.3/107	273.3/135.2
Testosterone	289.2/109.1	289.2/97
Progesterone	315.3/109.1	315.3/97
Corticosterone	347.2/121	347.2/329.5
Prednisolone	361/147.2	361/307.4
Aldosterone	361/315.2	361/299.7



RESULTS:

Chemicals	Testosterone	Progesterone	Corticosterone	Prednisolone
Recovery(%)	85.8	85.0	86.4	85.8
CV(%)	9.9	7.7	5.4	13.9