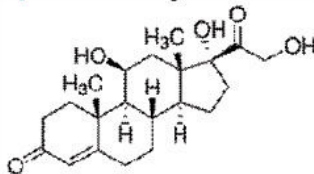


SUPPORTED LIQUID-LIQUID EXTRACTION Plate
Compound: Hydrocortisone

SUPPORTED LIQUID-LIQUID EXTRACTION METHOD:

(Supported liquid-liquid extraction plate)
 (Catalog #OC21DA260-HPGB)

Load:

100 μ L spiked human plasma (250 ng/mL) with 100 μ L water
 Let stand for 5 minutes, then apply a short pulse of positive pressure.


Elute:

1.5 mL of ethyl acetate twice. Let solvent stand for 5 minutes,
 and then apply a gentle stream of positive pressure till all solvent passes through.



Evaporate and Reconstitute:
 200 μ L of 35% acetonitrile

HPLC METHOD:

Column: Orochem Reliasil C8 HPLC column
 4.6x50mm, 5 μ m.
Part number: R3BI-102
Mobile phase: Water/acetonitrile 65:35
Flow rate: 0.5 mL/min
Injection volume: 20 μ L
Retention time: 2.53 min

MS/MS CONDITIONS:

Mass spectrometer: API3000
Ion source: Turbo Ionspray
Polarity: Positive
Scan mode: MRM: primary,
 Q1: 363.1, Q3: 121;
 secondary,
 Q1: 363.1, Q3: 327.1
IS: 4000
Temp. 500
DP: 20
CAD: 12
CE: 25
CXP: 10

RECOVERY (CV%)
 96% (9.0%); n=5

