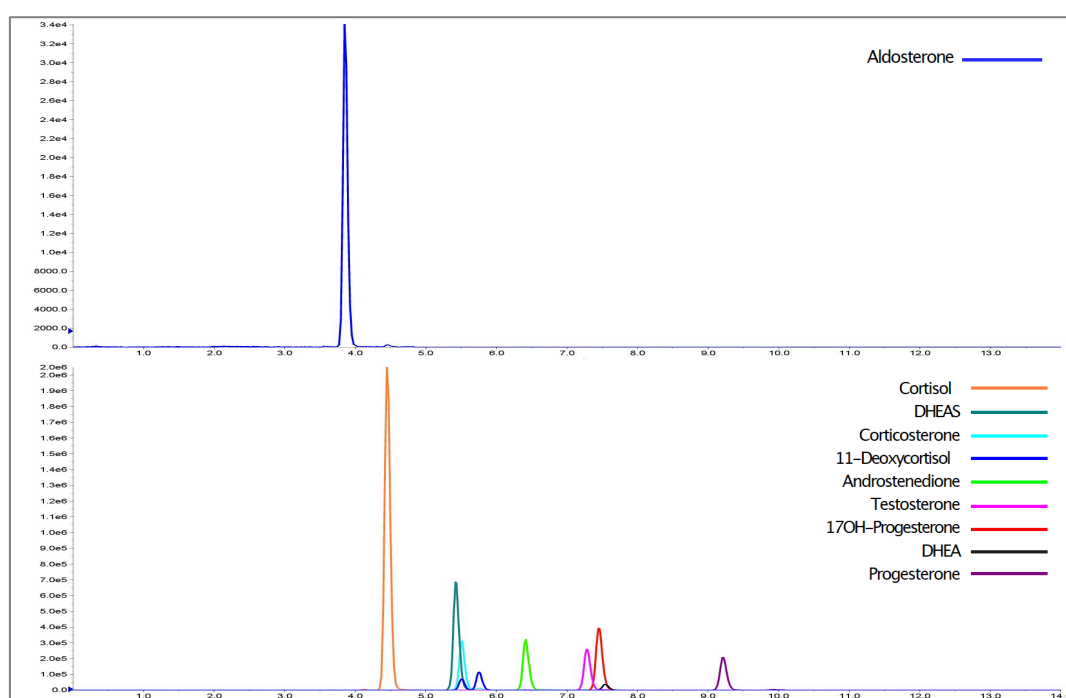


FLOMASS[®] STEROIDS IN SERUM (AUTOMATABLE ON 96-WELLS PLATE)

Steroid hormones are lipids and originate from a common precursor: cholesterol. Transformation of cholesterol to steroid hormones has an extreme physiological importance. Steroid hormones are involved in metabolism, growth and reproduction. They are in the circulatory flow, transported by specific proteins named carriers, allowing the hormone to reach its target. Spontaneous or inherited genetic mutations that may affect the synthesis of these enzymes are responsible of the alteration of normal levels of steroid hormones. Disorders of organs involved in steroids production and regulation can also bring to a steroid pathological level. It is useful to determinate steroid hormones profile rather than individual steroid analyte



HPLC-MS/MS system conditions

Ionization: ESI/APCI positive mode, except Aldosterone analyzed in negative mode

MS/MS: specific MRM

Injection volume: 20 μ L (variable according to instrumental sensitivity)

Running time: 17 min

Column heater: 30 °C

Sample preparation

- Aliquot 200 µL of serum in a precipitation microplate well
- Add to every sample 400 µL of Sol Precipitant + 8 µL of Internal Standard mix sufficient for the number of samples required. Vortex
- Cover the microplate with a foil and vortex for 15 min
- Centrifuge the microplate for 60 min at maximum speed
- Remove the foil and transfer 400 µL of the supernatant into a collection microplate
- Evaporate the solution until dry with nitrogen flow
- Resuspend the solution with 30 µL of Mobile Phase B
- Cover the microplate with a foil and vortex for 10 min
- Add 30 µL of Mobile Phase A
- Cover the microplate with a foil and vortex for 5 min
- Insert the microplate in the autosampler rack and analyze with HPLC-MS/MS technique

Performance

ANALYTE	LINEARITY (µg/l)	LLOD (µg/l)	LLOQ (µg/l)	CV% INTRA	CV% INTER
Aldosterone	0.014 - 37	0.004	0.014	3.08 - 14.21	3.08 - 13.36
Androstenedione	0.006 - 53	0.002	0.006	1.23 - 11.90	7.07 - 9.72
Corticosterone	0.025 - 225	0.008	0.025	1.29 - 7.56	3.19 - 4.64
Cortisol	0.109 - 6500	0.033	0.109	1.41 - 5.48	5.09 - 6.49
11-Deoxycortisol	0.014 - 56	0.004	0.014	3.67 - 14.24	3.13 - 9.69
DHEA (derivatized)	0.103 - 216	0.031	0.103	3.42 - 9.67	6.65 - 11.40
Progesterone	0.032 - 64	0.010	0.032	1.97 - 8.45	2.92 - 5.97
17-OH-Progesterone	0.014 - 56	0.004	0.014	2.37 - 4.10	5.94 - 8.84
DHEAS	0.710 - 12600	0.213	0.710	1.12 - 11.00	2.89 - 4.84
Testosterone	0.007 - 53	0.002	0.007	1.19 - 5.81	5.17 - 6.58

Expected values and results interpretation

EUM130960	FloMass® Steroids in Serum (Automatable on 96-wells plate)	96 assays
EUM01041	7-Levels Calibrators, lyophil.	2 x 7 x 1.0 mL
EUM01051	3-Levels Controls, lyophil.	2 x 3 x 2.5 mL
EUM00C01	Analytical Column	1 pc
EUM00A12	Precolumn	4 pcs