

METHOD AT A GLANCE

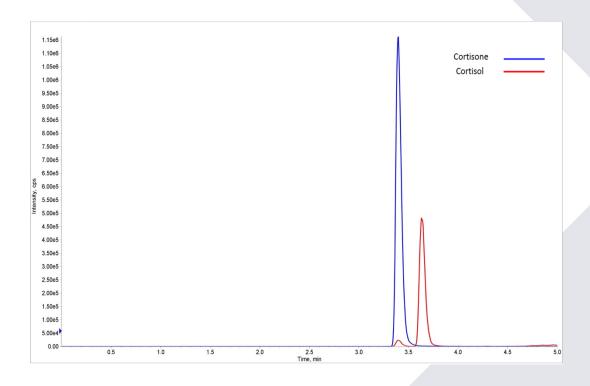
FLOMASS® CORTISOL AND CORTISONE IN SALIVA

The interest in the use of the salivary matrix is increasing because, in addition to providing metabolic information, it can be collected through a simple, non-invasive method and without medical supervision.

Cortisol and Cortisone are steroid hormones synthesized in the adrenal glands starting from cholesterol. Cortisol and Cortisone are a useful target in the evaluation of mineralocorticoid or glucocorticoid excess, adrenal congenital hyperplasia and adrenal insufficiency. Furthermore, salivary cortisol increases in Cushing's syndrome, decrease in Addison's disease and can be associated with cardiovascular disease, anti-inflammatory and immunosuppressive effects.

The evaluation of salivary Cortisol and Cortisone is a good indicator of concentrations related to the proportion of free Cortisol and Cortisone, therefore not linked to transport proteins, in serum and plasma. For this reason, the analytes are at very low concentrations in the saliva samples (about 5 times lower than plasma and serum).

Therefore, it is necessary to have a sensitive and specific analytical technology to measure these low concentrations of Cortisol and Cortisone present in saliva.





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HPLC-MS/MS system conditions

Ionization: ESI positive mode

MS/MS: specific MRM

Injection volume: 10 µL (variable according to instrumental sensitivity)

Running time: 6 min Column heater: 30°C

Sample preparation

- Prepare a mix with 180 μ L of Sol 1 Dilution + 20 μ L of Internal Standard sufficient for the number of samples to be analyzed
- Add 200 µL saliva in a vial
- Add 200 μL of Mix Solution obtained in previous step of the procedure
- Add 1 mL of Sol 2 Extraction and vortex 1 min
- Centrifuge for 5 min at 10000 rpm to achieve separation of the two phases
- Transfer 800 μ L of supernatant and dispense into a 1.5 mL vial
- Evaporate at 50°C under nitrogen flow
- Resuspend with 30 μL of Sol 3 Reconstitution and vortex 30 sec
- Add 30 µL of Sol 1 Dilution and vortex 15 sec
- Centrifuge for 5 min at 10000 rpm
- Transfer 60 μL of supernatant in a vial
- Inject 10-20 μL and analyze with HPLC-MS/MS technique

Performance

ANALYTE	LINEARITY (ng/mL)	LLOD (ng/mL)	LLOQ (ng/mL)	CV% INTRA	CV% INTER
Cortisol	0.019 - 200	0.0057	0.019	2.3 – 5.4	4.8 – 6.2
Cortisone	0.009 - 500	0.0027	0.009	2.5 – 4.5	5.3 – 6.2

Ordering guide

EUM14200	FloMass® Cortisol and Cortisone in Saliva	200 assays
EUM14041	6-Levels Calibrators, lyophil.	2 x 6 x 1.0 mL
EUM14051	3-Levels Controls, lyophil.	2 x 3 x 2.0 mL
EUM00C14	Chromatographic Column	1 pc
EUM00A06	Precolumn	4 pcs

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