

ANALYSIS OF NEUROTRANSMITTERS AND RELATED METABOLITES

Description: LC-MS/MS method for the sensitive detection and quantification of several neurotransmitters and its related metabolites. The detection limits depend on matrix type and input quantity. Samples are extracted and measured by a Waters I-Class Plus LC System paired with a Sciex 6500+ QTRAP.

Analytes are reported as μM or pmol/mio cells or pmol/mg tissue or pmol/mL plasma or urine.

Container: Eppendorf Tube or equivalent.

Normal Volume: Plasma / cell culture medium (100 μl); Tissue (25 mg)¹; Cells (1 mio).

Minimal Volume: Plasma / cell culture medium (10 μL); Tissue (10 mg)¹; Cells (0.5 mio).

Sample Collection: Please see our detailed sample collection protocols.

Quantification: Absolute, using a >6 point calibration curve and $r^2 > 98\%$ for most of the components. This analysis is a multi-component method for which the conditions are necessarily not ideal for all compounds. Results should be critical considered and reported as fold-changes towards control groups instead of absolute quantification for: 2-oxoadipic acid, GHB, Glutaric acid and homovanillic acid. If wished, peak areas can be provided

Please note: For human material, note any known presence of infectious agents

List of analytes reported

(D- and L- enantiomers are not distinguished)

Compound name	Identifier	Formula	Monoisotopic mass
2-aminoadipic acid	HMDB0302754	C ₆ H ₁₁ NO ₄	161.07
D-2-hydroxyglutaric acid	HMDB0000606	C ₅ H ₈ O ₅	148.04
2-oxoadipic acid	HMDB0000225	C ₆ H ₈ O ₅	160.04
3-hydroxyglutaric acid	HMDB0000428	C ₅ H ₈ O ₅	148.04
3-O-Methyl-a-methyl-dopa	HMDB0060747	C ₁₀ H ₁₃ NO ₄	211.08
5-Hydroxyindoleacetic acid	HMDB0000763	C ₁₀ H ₉ NO ₃	191.06
Dopamin	HMDB0000073	C ₈ H ₁₁ NO ₂	153.08
gamma-Aminobutyric acid	HMDB0000112	C ₄ H ₉ NO ₂	103.06
gamma-Hydroxybutyrate	HMDB0000710	C ₄ H ₈ O ₃	104.05
Glutaric acid	HMDB0000661	C ₅ H ₈ O ₄	132.04

¹ Pulverized/crushed (deep-frozen) and exact weight noted

Compound name	Identifier	Formula	Monoisotopic mass
Homovanillic acid	HMDB0000118	C9H10O4	182.06
Kynurenic acid	HMDB0000715	C10H7NO3	189.04
L-DOPA	HMDB0000181	C9H11NO4	197.07
Quinolinic acid	HMDB0000232	C7H5NO4	167.02

LC conditions

Column	Waters HSS T3 150 x 2.1mm
Temperature	45° C
Mobile phase A	Water + 0.02% formic acid
Mobile phase B	ACN:MeOH 1:1 + 0.02% formic acid
Flow	0.3-0.4 ml/min

Notes

Samples need to be snap-frozen and stored at -80°C.

Variations in sampling procedures will affect metabolite measurements.

¹ Pulverized/crushed (deep-frozen) and exact weight noted