MassChrom® Amino Acids and Acylcarnitines from Dried Blood



LC-MS/MS Assay with Succinylacetone Upgrade Set

- > Each analyte safeguarded by its own internal standard
- Screening of many metabolic disorders in a single run such as PKU, MSUD and tyrosinemia type I

Amino Acids and Succinylacetone	Internal Standard
Alanine	Alanine-D4
Arginine	Arginine-D7
Aspartic acid	Aspartic acid-D3
Citrulline	Citrulline-D2
Glutamic acid	Glutamic acid-D5
Glycine	Glycine- ¹³ C ₂ - ¹⁵ N
Leucine	Leucine-D3
Methionine	Methionine-D3
Ornithine	Ornithine-D6
Phenylalanine	Phenylalanine-D5
Proline	Proline-D7
Tyrosine	Tyrosine-D4
Valine	Valine-D8
Succinylacetone	Succinylacetone-13C ₅

Internal Standard	
Carnitine-D9	
C2-Carnitine-D3	
C3-Carnitine-D3	
C4-Carnitine-D3	
C5-Carnitine-D9	
C5DC-Carnitine-D6	
C6-Carnitine-D3	
C8-Carnitine-D3	
C10-Carnitine-D3	
C12-Carnitine-D3	
C14-Carnitine-D3	
C16-Carnitine-D3	
C18-Carnitine-D3	

Specifications

Limit of quantification

Amino acids with SUAC: 0.5-5 µmol/l Acylcarnitines: 0.05-0.5 µmol/l

Linearity

Amino acids with SUAC: 250-2500 µmol/l Acylcarnitines: 11-240 µmol/l

Recovery

Amino acids with SUAC: 51-94 % Acylcarnitines: 62-89 %

Intraassay

Amino acids with SUAC: CV = 4-10 %Acylcarnitines: CV = 5-20 %

Interassay

Amino acids with SUAC: CV = 6-9 %Acylcarnitines: CV = 9-28 %

Analysis time: < 2 min

Cutoff thresholds

Amino acids: 22-834 µmol/l
Free carnitine: 50-60 µmol/l
Acylcarnitines: 0.1-57 µmol/l
SUAC: 1.16 µmol/l

Pre-analytic Treatment

Specimen collection: whole blood from the heel of the newborn taken onto a filter card. Collection between 24th and 72nd hour of life depending on individual national guideline. Stability: storage at low humidity (< 30 %) and ambient temperature is sufficient if analysis is performed within 24 to 48 hours. For longer storage deep-freeze samples with humidity excluded below -18 °C.

Sample Preparation

- > Punch out 3.2 mm blood spots from the filter cards into a 96 Well Plate.
- > Add 200 µl Internal Standard and agitate for 20 min.
- > Transfer supernatant into a new 96 Well Plate (or with 55000/F centrifuge for 2 min).
- > Evaporate supernatant.
- > Add 60 µl Derivatisation Reagent and incubate for 15 min at 60 °C.
- > Evaporate to dryness.
- > Add 100 µl Reconstitution Buffer and agitate for 1 min.
- > Inject 10 µl into the LC-MS/MS system.

The sample preparation for the analysis of succinylacetone differs from this. Please see the instruction manual.

MS Parameters

For newborn screening any LC-MS/MS system with sufficient sensitivity is suitable. No analytical HPLC column is required.

Injection volume: 10 µl

Flow gradient: 20-600 µl/min

MS/MS Mode:

Amino acids: Neutral Loss Scan/MRM

Acylcarnitines: Parent Ion Scan

Ordering Information

These products are not available in all countries

These Chromsystems assays allow the fast and reliable determination of amino acids, acylcarnitines and succinylacetone from dried blood spot samples as part of the newborn screening for amino acid and fatty acid metabolic disorders using tandem mass spectrometry. Sample preparation is based on effective extraction of the analytes from the filter card with subsequent derivatisation. The use of stable isotope labelled internal standards for calibration and measurement ensures reliable and precise quantification of the analytes. Compared with the underivatised method (57000 or 57000/F), this method is suited for highest sensitivity. One pipetting step per sample can be saved with the assay that uses well filter plates (55000/F).

Order no.	Product
55000	MassChrom® Amino Acids and Acylcarnitines from Dried Blood
	For 960 determinations using 96 Well Plates
55111	Succinylacetone Upgrade Set for 960 determinations,
	consisting of Internal Standard and Reconstitution Buffer
55000/F	MassChrom® Amino Acids and Acylcarnitines from Dried Blood
	For 960 determinations using 96 Well Filter Plates
	Components available separately
55001	Mobile Phase, 1000 ml
55002	Mobile Phase, 10 x 1000 ml
55004	Internal Standard (lyoph.), 4 x 50 ml
55044	Internal Standard, Succinylacetone, 3 x 50 ml
55005	Derivatisation Reagent, 30 ml
55006	Reconstitution Buffer, 100 ml
55007	Rinsing Solution, 1000 ml
55008	Extraction Buffer, 200 ml
55010	96 Well Plates, 10 pcs.
55010/F	96 Well Plates, 5 pcs.
55057	96 Well Filter Plates, 5 pcs.
55011	Protective Sheets for 96 Well Plates, 10 pcs.
	Startup Accessories
57014	Pierceable Heat Seals for 96 Well Plates, 6 pcs.
55013	Pierceable Adhesive Seals for 96 Well Plates, 10 pcs.
55015	Restrictor Capillary, 1 pc.
55016	Adapter Collar for centrifugation, 2 pcs.
55099	Tuning Mix, Analytes and Internal Standards, 2 ml
55098	Tuning Mix Succinylacetone, Analyte and Internal Standard, 1 ml
55033	PEEK Prefilter, 2 μm, 5 pcs.
15010	PEEK Prefilter Housing, 1 pc.



Chromsystems MassCheck® Controls

Dried Blood Spot Controls

Bi-Level (I + II), 2×3 spots

Level I, 1 x 3 spots Level II, 1 x 3 spots

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MassCheck® Amino Acids, Acylcarnitines and Succinylacetone