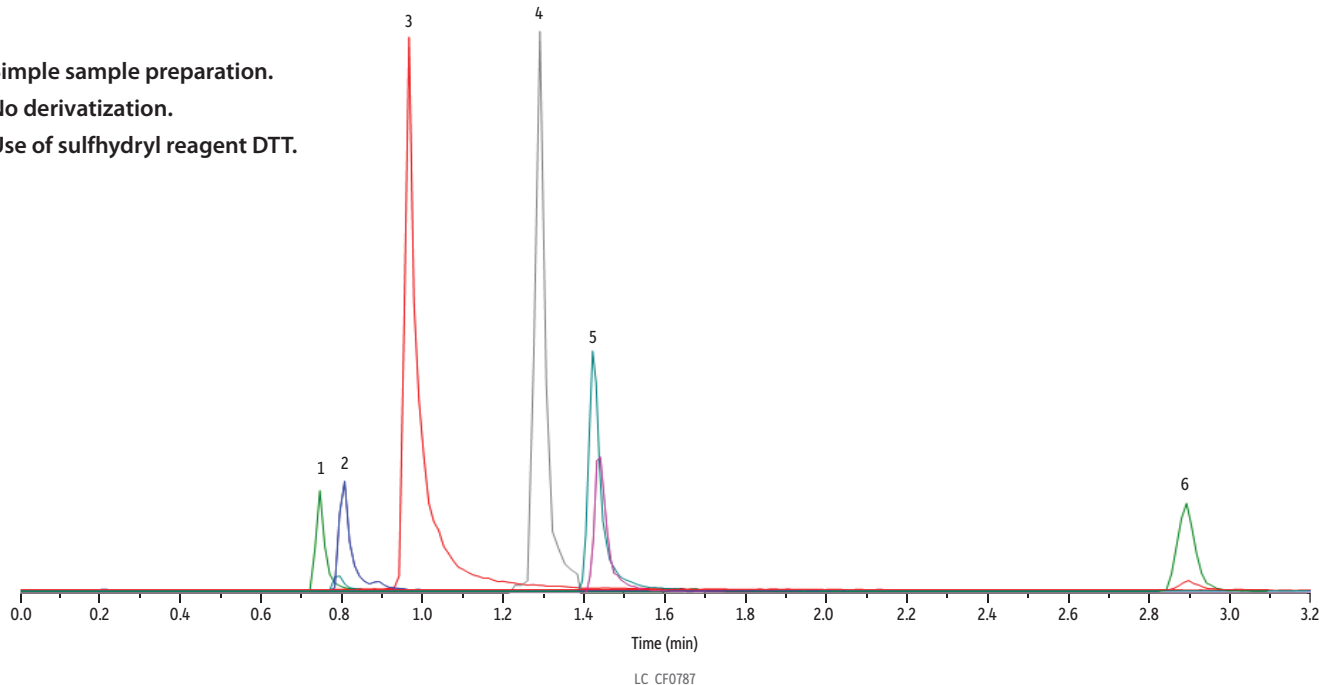


Methionine Pathway Metabolites and Methylmalonic Acid in Plasma on Raptor Polar X

- Simple sample preparation.
- No derivatization.
- Use of sulfhydryl reagent DTT.



Peaks	tr (min)	Precursor Ion	Product Ion 1	Product Ion 2	Polarity
1. L-Methionine	0.75	150.0	104.1	-	+
2. L-Homocysteine	0.81	136.2	90.1	118.1	+
3. L-Cysteine	1.00	122.1	76.0	-	+
4. Succinic acid	1.33	117.0	72.9	55.1	-
5. DL-Cystathionine	1.45	221.3	119.8	113.9	-
6. Methylmalonic acid	2.92	117.0	55.1	72.9	-

Column Raptor Polar X (cat.# 9311A52)
Dimensions: 50 mm x 2.1 mm ID
Particle Size: 2.7 µm
Pore Size: 90 Å
Guard Column: Raptor Polar X EXP guard column cartridge 5 mm, 2.1 mm ID, 2.7 µm (cat.# 9311A0252)
Temp.: 40 °C

Standard/Sample
Conc.: 100 ng/mL
Inj. Vol.: 5 µL

Mobile Phase
A: Water, 0.5% formic acid
B: Acetonitrile

Time (min)	Flow (mL/min)	%A	%B
0.00	0.6	15	85
1.00	0.6	45	55
3.00	0.6	90	10
3.01	0.6	15	85
4.00	0.6	15	85

Detector SCIEX 4500
Ion Source: Electrospray
Ion Mode: ESI+/ESI-
Instrument Shimadzu Nexera X2

Sample Preparation A 100 ng/mL standard mix of all of the analytes in this method was prepared in plasma. A 100 µL aliquot was taken from the standard and mixed with 20 µL of 0.5 M dithiothreitol (DTT). The sample was vortexed for 10 seconds and then left to incubate at room temperature in darkness for 30 minutes. After 30 minutes, 300 µL of methanol was added, and the sample was vortexed for 10 seconds and then centrifuged for 10 minutes at 4000 rpm. 100 µL of the supernatant was added to a 2 mL vial (cat.# 21142) containing a vial insert (cat.# 21776) and capped with a short screw cap (cat.# 24498).