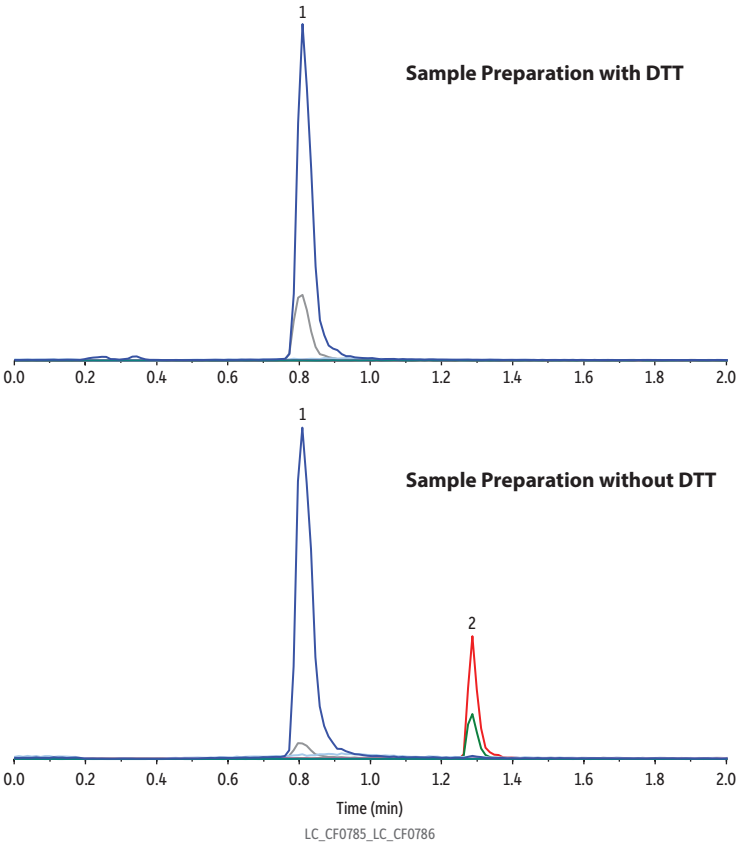
Comparison of the Homocysteine Monomer and Dimer in Plasma with and without the Use of DTT

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



LC_CF0785_LC_CF0786

Peaks	t_R (min)	Precursor Ion	Product Ion 1	Product Ion 2
1. L-Homocysteine	0.81	136.2	90.1	118.1
2. L-Homocystine	1.29	268.9	136.0	118.0

Column
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+
Instrument Shimadzu Nexera X2
Sample Preparation

With DTT
A 100 ng/mL standard mix of homocysteine (monomer) and homocystine (dimer) 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).

Without DTT
A 100 ng/mL standard mix of homocysteine (monomer) and homocystine (dimer) was prepared in plasma. A 100 µL aliquot was taken from the standard and was vortexed for 10 seconds. After vortexing, 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).