La Frano Lab Metabolomics Analysis

Materials & Methods

Metabolomics analysis was performed on plasma using protein precipitation extraction with ultra-performance liquid chromatography tandem quadrupole mass spectrometry (UPLC-MS), as previously described (McMichael et al. 2021). For each sample, 25 µL of plasma was added to a 1.5 mL tube before the addition of 10 µL of 1 µM internal standard solution, followed by 750 µL or 500 µL chilled methanol. Samples were then vortexed 30 seconds prior to being centrifuged at 15,000 x G for 10 min at 4°C. The supernatant was transferred to 1.5 mL high performance liquid chromatography (HPLC) amber glass vials, dried by centrifugal vacuum evaporation, and reconstituted in 100 µL 3:1 acetonitrile:methanol solution with the internal standard 1-cyclohexyl ureido, 3-dodecanoic acid (CUDA) at 100 nM. The reconstituted solution was vortexed 30 seconds and placed on ice for 10 minutes. The solution was then centrifuged at 10,000 x G for 3 minutes at room temperature after being transferred to microfilter tubes. The supernatant was then transferred to a HPLC vial to be analyzed using the UPLC-MS.

UPLC-MS analysis was conducted on a Waters Acquity I-Class UPLC (Waters, Milford, MA, USA) coupled with a 4000 QTrap (Sciex, Framingham, MA) and quantified with AB Sciex MultiQuant version 3.0. Primary metabolomics used multiple reaction monitoring (MRM). Metabolites were separated using a 150 X 2.0 mm, 3µm Luna NH2 column (Phenomenex, Torrance, CA) with mobile phases consisting of solvent A as 20 mM ammonium acetate, 20 mM ammonium hydroxide and solvent B as 10 mM ammonium hydroxide in 75:25 % v/v (volume %) acetonitrile/methanol and detected by negative ion mode electrospray ionization. Gradient, mobile phase, and instrument parameters for the UPLC are listed in Table 1. MRMs and other parameters for the MS are listed in Table 2.

Primary metabolomics metabolite identities were confirmed using pure standards in order to establish retention time and MRM, as well as optimize instrument parameters. Standards included those within the Mass Spectrometry Metabolite Library of Standards (MSMLS; Sigma-Aldrich, St. Louis, MO, USA), as well as individually purchased standards from Sigma-Aldrich, Cambridge Isotope Laboratories, Inc (Tewksbury, MA, USA), and Cerilliant Corporation (Round Rock, TX). Some acylcarnitine species were identified based on MRM only. Surrogate standards used in the primary metabolomics assay included succinate-$^{13}C_4$, sorbitol-1,1,2,3,4,5,6,6-d8, and adenine-2-d1 utilized to monitor extraction efficiency and recovery percentage for each sample analyzed. Surrogates were purchased from Santa Cruz Biotechnology, Inc, (Dallas, TX, USA), CDN Isotopes Inc. (Pointe-Claire, Quebec, Canada), and Cambridge Isotope Laboratories, Inc. The internal standard CUDA (Sigma Aldrich), included in the reconstitution solvent that was added post-extraction, was used for controlling instrument and injection parameters. All raw data were normalized to the internal standard CUDA (Sigma Aldrich).

For quality control purposes, compounds whose background (as determined by method blank response) was greater than 50% of the average sample response or that had more than 1/3 of samples with signal to noise less than 3:1, were excluded from the dataset. To assess reproducibility, five replicates of the current study samples were separately extracted and analyzed. A pooled plasma sample collected from

a different study was used as a long-term reference QC sample for an inter-study assessment of data. All samples were run in a single batch.

Reference

McMichael LE, Heath H, Johnson CM, Fanter R, Alarcon N, Quintana-Diaz A, Pilolla K, Schaffner A, Jelalian E, Wing RR, Brito A, Phelan S, & La Frano MR (2021). Metabolites involved in purine degradation, insulin resistance, and fatty acid oxidation are associated with prediction of Gestational diabetes in plasma. Metabolomics, 17(12), 105. https://doi.org/10.1007/s11306-021-01857-5.

Table 1. Primary Metabolomics Assay UPLC parameters

Time (min)	A%	B%
0	10	90
10	95	5
11	95	5
13	10	90
15	10	90

Solvent A = 20 mM ammonium acetate, 20 mM ammonium hydroxide in water
Solvent B = 10 mM ammonium hydroxide in 75:25 % v/v (volume %) acetonitrile/methanol
Flow Rate = 0.3 mL/min
Column = 150 x 2.0 mm Luna NH2, 3 um, 100 angstrom (Phenomenex, Torrance, CA)
Column Temp = 30 °C

Table 2. Primary Metabolomics UPLC/ESI QTRAP metabolite-specific parameters

Metabolite	Q1	Q3	DP	CE
CITRATE	191.0	111.0	-25	-15
N-ACETYLNEURAMINATE	308.1	86.9	-25	-16
L-GLUTAMIC ACID	146.0	127.9	-60	-15
ASCORBATE	175.0	114.9	-25	-15
N-ACETYL-D-GLUCOSAMINE	220.1	59.0	-60	-40
GLUCONIC ACID	195.1	129.0	-60	-15
QUINATE	191.1	85.0	-25	-24
(S)-DIHYDROOROTATE	157.0	112.9	-25	-15
MALONATE	103.0	59.0	-25	-15
GLYCERATE	105.0	75.1	-25	-15
SHIKIMATE	173.0	93.0	-60	-16
SUCCINATE	117.0	73.1	-25	-15
URACIL	111.0	42.1	-25	-24
(R)-MALATE	133.0	115.0	-25	-10
DAMP	330.1	134.0	-25	-28
ISOCITRIC ACID	191.0	111.1	-25	-15
D-GLUCOSAMINE 6-PHOSPHATE	258.0	78.9	-60	-40
(S,S)-TARTARIC ACID	149.0	87.0	-25	-10
D-(+)-TREHALOSE	341.1	89.0	-60	-20
D-GLUCURONOLACTONE	175.0	113.0	-25	-15
(2-AMINOETHYL)PHOSPHONATE	124.0	79.0	-60	-28
ADENOSINE 5'-DIPHOSPHATE	426.0	158.9	-60	-20
GALACTITOL	181.1	101.0	-60	-20
PYRIDINE-2,3-DICARBOXYLATE	166.0	122.0	-25	-15
D-GLUCURONIC ACID	193.0	73.0	-25	-20
1-METHYLADENOSINE	280.1	148.1	-60	-20
DEOXYURIDINE	227.1	42.0	-60	-28
3-HYDROXY-3-METHYLGLUTARATE	161.0	99.0	-25	-15
3-METHYLCROTONYL-COA	848.1	426.0	-60	-40
NICOTINIC ACID ADENINE DINUCLEOTIDE PHOSPHATE	742.1	620.0	-60	-20
N-ACETYL-L-ASPARTIC ACID	174.0	88.0	-25	-10
INOSINE 5'-DIPHOSPHATE	427.0	159.0	-25	-32
ADENOSINE 2',3'-CYCLIC MONOPHOSPHATE	328.0	134.0	-25	-20
2'-DEOXYURIDINE 5'-TRIPHOSPHATE	467.0	159.0	-60	-20
2'-DEOXYGUANOSINE 5'-DIPHOSPHATE	426.0	79.0	-90	-40
DIHYDROXYACETONE PHOSPHATE	181.0	79.0	-25	-40
PHOSPHO(ENOL)PYRUVIC ACID	167.0	79.0	-25	-15
D-MANNOSE 6-PHOSPHATE	259.0	96.7	-60	-16

2-AMINOETHYL DIHYDROGEN PHOSPHATE	140.0	78.8	-25	-16
ADENOSINE-5'-DIPHOSPHOGLUCOSE	588.1	346.1	-60	-20
D-FRUCTOSE 6-PHOSPHATE	259.0	96.8	-60	-15
URIDINE 5'-DIPHOSPHOGALACTOSE	565.0	323.0	-60	-24
2'-DEOXYURIDINE 5'-MONO-PHOS-PHATE	307.0	195.0	-60	-15
FLAVIN ADENINE DINUCLEOTIDE	784.1	437.1	-90	-40
O-PHOSPHO-DL-SERINE	184.0	96.8	-25	-15
URIDINE 5'-DIPHOSPHO-N-ACETYLGLUCOSAMINE	606.1	385.0	-90	-33
URIDINE 5'-DIPHOSPHOGLUCURONIC ACID	579.0	403.0	-60	-25
CYTIDINE 5'-DIPHOSPHATE	402.0	78.9	-60	-40
INDOXYL SULFATE	212.0	77.0	-25	-40
2,4-DIHYDROXYPYRIMIDINE-5-CARBOXYLIC ACID	155.0	68.1	-25	-20
HYDROXYPYRUVATE	103.0	59.0	-25	-15
N-ACETYL-DL-GLUTAMIC ACID	188.1	128.0	-25	-15
6-HYDROXYNICOTINATE	138.0	94.0	-25	-15
ANILINE-2-SULFONIC ACID	172.0	80.0	-60	-29
S-CARBOXYMETHYL-L-CYSTEINE	178.0	90.9	-60	-15
THYMIDINE 5'-MONOPHOSPHATE	321.0	97.0	-25	-40
D-(+)-RAFFINOSE	503.2	179.0	-90	-20
D-SACCHARIC ACID	209.0	85.1	-25	-15
3-HYDROXYBUTANOIC ACID	103.0	59.1	-60	-15
D-(+)-GALACTURONIC ACID	193.0	59.0	-25	-20
CYTIDINE 5'-TRIPHOSPHATE	482.0	384.0	-60	-20
ADENOSINE 5'-DIPHOSPHORIBOSE	558.1	346.0	-60	-28
COENZYME A	766.1	408.0	-90	-40
2'-DEOXYCYTIDINE 5'-DIPHOSPHATE	386.0	78.8	-60	-40
URIDINE 5'-DIPHOSPHATE	403.0	158.9	-90	-20
3-METHYGLUTARIC ACID	145.1	83.0	-25	-20
MALEIC ACID	115.0	70.9	-25	-15
4-HYDROXYPHENYLACETATE	151.0	107.0	-25	-20
MANDELIC ACID	151.0	106.9	-25	-15
GLUTARATE	131.0	87.0	-25	-15
OXALOACETATE	131.1	87.0	-25	-15
2,3-DIHYDROXYBENZOATE	153.0	109.0	-60	-10
2-HYDROXYPHENYLACETIC ACID	151.0	106.9	-25	-15
10-HYDROXYDECANOATE	187.1	141.1	-25	-20
ETHYLMALONIC ACID	131.0	87.1	-25	-15
2-QUINOLINECARBOXYLIC ACID	172.0	128.0	-25	-10
3-AMINO-4-HYDROXYBENZOIC ACID	152.0	108.0	-60	-16

2,5-DIHYDROXYBENZOATE	153.0	107.9	-60	-24
2-METHYLMALEATE	129.0	84.9	-25	-15
ITACONATE	129.0	85.0	-25	-10
AZELAIC ACID	187.1	125.0	-60	-16
SUBERIC ACID	173.1	111.1	-25	-20
2',4'-DIHYDROXYACETOPHENONE	151.0	108.9	-25	-16
(RS)-MEVALONIC ACID LITHIUM SALT	147.1	59.0	-25	-20
3-METHOXY-4-HYDROXYMANDELATE	197.0	137.0	-25	-24
HOMOGENTISATE	167.0	123.0	-25	-10
3-HYDROXYPHENYLACETATE	151.0	107.1	-25	-10
4-METHYLCATECHOL	123.0	108.0	-60	-20
3-(4-HYDROXYPHENYL)LACTATE	181.1	162.9	-60	-15
XYLITOL	151.1	71.0	-25	-20
RIBITOL	151.1	89.0	-25	-15
MANNOSE	179.1	89.0	-25	-10
ALPHA-D-GLUCOSE	179.1	89.0	-60	-10
MELIBIOSE	341.1	179.0	-25	-15
MALTOSE	341.1	179.1	-25	-10
URATE_	167.0	123.9	-60	-15
ALLANTOIN_	157.0	96.9	-60	-16
N(PAI)-METHYL-L-HISTIDINE_	168.1	151.0	-60	-20
PYRIDOXAL 5_PHOSPHATE_	246.0	79.0	-60	-40
MANNOSE 6_PHOSPHATE_	259.0	96.7	-60	-16
FRUCTOSE 6_PHOSPHATE_	259.0	96.8	-60	-15
XANTHOSINE 5_MONOPHOSPHATE_	363.0	79.0	-60	-40
THIAMINE MONOPHOSPHATE_	343.1	97.0	-60	-20
LACTOSE_	341.1	101.0	-60	-15
DEOXYCYTIDINE 5_DIPHOSPHATE_	386.0	78.8	-60	-40
BENZOATE_	121.0	77.0	-60	-15
HIPPURATE_	178.1	134.0	-25	-15
HOMOVANILLATE_	181.1	137.0	-25	-15
BIOTIN_	243.1	200.1	-60	-20
MYO_INOSITOL_	179.1	161.0	-25	-10
L-GLUTAMINE	145.1	127.0	-25	-15
INOSINE 5'-PHOSPHATE	347.0	78.8	-25	-40
L-SERINE	104.0	74.0	-90	-15
CITRULLINE	174.1	131.0	-90	-15
ADENINE	134.0	106.9	-25	-20
THYMIDINE	241.1	42.0	-60	-24
XANTHINE	151.0	108.0	-25	-20
URIDINE-5-MONOPHOSPHATE	323.0	79.0	-60	-40
URIDINE	243.1	199.9	-60	-15
CARNOSINE	225.1	154.0	-60	-16

L-ASPARTATE	132.0	88.0	-25	-10
2'-DEOXYCYTIDINE 5'-MONOPHOSPHATE	306.0	79.0	-25	-40
L-ASPARAGINE	131.0	113.3	-25	-15
PYRIDOXINE	168.1	150.0	-25	-15
THEOPHYLLINE	179.1	164.0	-60	-20
3-SULFINO-L-ALANINE	152.0	88.0	-25	-15
O-SUCCINYL-L-HOMOSERINE	218.1	117.4	-60	-15
4-PYRIDOXATE	182.0	138.0	-25	-15
O-PHOSPHO-L-SERINE	184.0	96.8	-25	-15
PHOSPHONOACETATE	139.0	78.8	-25	-24
N-METHYL-L-GLUTAMATE	160.1	142.1	-25	-10
NALPHA-ACETYL-L-LYSINE	187.1	145.1	-25	-20
N-ACETYL-DL-METHIONINE	190.1	147.9	-25	-15
GUANOSINE 3',5'-CYCLIC MONOPHOSPHATE	344.0	133.0	-60	-40
S-(5'-ADENOSYL)-L-HOMOCYSTEINE	383.1	133.9	-60	-32
GUANOSINE 5'-MONOPHOSPHATE	362.1	78.9	-60	-28
N-ACETYL-L-ALANINE	130.1	88.0	-25	-15
D-PANTOTHENIC ACID	218.1	87.9	-60	-15
4-IMIDAZOLEACETIC ACID	125.0	80.9	-25	-15
3',5'-CYCLIC AMP	328.0	134.0	-60	-28
L-METHIONINE SULFOXIMINE	179.0	78.0	-25	-15
FORMYL-L-METHIONYL PEPTIDE	176.0	128.0	-25	-10
STACHYOSE	665.2	383.1	-60	-40
N-ACETYL-DL-SERINE	146.0	116.0	-25	-10
XANTHURENIC ACID	204.0	160.0	-25	-16
MELATONIN	231.1	231.0	-60	-15
4-HYDROXY-2-QUINOLINECARBOXYLIC ACID	188.0	144.0	-25	-20
FERULATE	193.1	178.1	-25	-15
GLYCOCHOLATE	464.3	74.0	-90	-40
METHYLMALONATE	117.0	73.0	-25	-15
4-HYDROXYBENZALDEHYDE	121.0	92.0	-60	-28
ALPHA-KETOGLUTARIC ACID	145.0	100.9	-60	-15
4-QUINOLINECARBOXYLIC ACID	172.0	128.0	-25	-15
MANNITOL	181.1	59.1	-60	-24
D-SORBITOL	181.1	71.0	-25	-20
NAD	662.1	540.1	-25	-16
HYPOTAURINE	108.0	63.9	-25	-16
L-THREONINE	118.1	74.1	-25	-15
PURINE	119.0	92.0	-90	-24
CYTIDINE	242.1	108.9	-60	-15

TAURINE	124.0	80.0	-60	-20
NICOTINATE	122.0	77.9	-60	-15
INOSINE	267.1	134.9	-60	-20
CYTOSINE	110.0	67.0	-60	-15
L-METHIONINE	148.0	47.0	-25	-16
CREATINE	130.1	88.0	-25	-10
GUANOSINE	282.1	150.0	-60	-20
L-TYROSINE	180.1	119.0	-25	-20
HOMOSERINE	118.1	100.0	-25	-10
FOLIC ACID	440.1	175.0	-25	-40
DEOXYCYTIDINE	226.1	93.0	-60	-10
D-TRYPTOPHAN	203.1	116.0	-60	-20
5-OXO-D-PROLINE	128.0	82.0	-25	-20
UROCANATE	137.0	93.0	-25	-15
KYNURENINE	207.1	190.1	-25	-10
5-OXO-L-PROLINE	128.0	128.0	-25	-25
4-ACETAMIDOBUTANOATE	144.1	102.0	-25	-15
1,3-DIAMINOPROPANE	73.1	79.0	-25	-40
L-CYSTATHIONINE	221.1	134.0	-25	-15
L-ARGININE	173.1	131.1	-25	-10
4-HYDROXY-L-PROLINE	130.1	128.0	-60	-20
XANTHOSINE	283.1	151.0	-25	-20
THIAMINE	264.1	147.6	-25	-20
CYTIDINE 2',3'-CYCLIC MONO-PHOS-PHATE	304.0	110.0	-60	-10
3-NITRO-L-TYROSINE	225.1	162.9	-25	-15
2'-DEOXYGUANOSINE	266.1	150.0	-60	-40
LAUROYLCARNITINE	342.3	199.2	-25	-10
4-GUANIDINOBUTANOATE	144.1	102.1	-25	-10
ADENOSINE	266.1	134.0	-25	-10
CIS-4-HYDROXY-D-PROLINE	130.1	84.0	-60	-15
OPHTHALMIC ACID	288.1	270.0	-60	-15
3,5-DIIODO-L-TYROSINE	431.9	126.9	-60	-20
LUMICHROME	241.1	198.1	-60	-20
N-ACETYL-L-PHENYLALANINE	206.1	164.1	-60	-10
L-TRYPTOPHANAMIDE	202.1	201.7	-60	-15
THIOPURINE S-METHYLETHER	165.0	149.9	-60	-24
INDOLE-3-ACETAMIDE	173.1	130.1	-60	-15
3,5-DIIODO-L-THYRONINE	523.9	506.9	-60	-20
DETHIOBIOTIN	213.1	170.0	-60	-20
ADIPIC ACID	145.1	83.0	-25	-15
N-ACETYL-L-LEUCINE	172.1	130.0	-25	-15
5-HYDROXYINDOLEACETATE	190.1	145.8	-25	-15

SUCROSE	341.1	179.1	-25	-10
L_HISTIDINE_	154.1	136.9	-60	-15
NAD_QI	662.1	328.0	-25	-28
L-GLUTAMINE_QI	145.1	42.0	-25	-36
INOSINE 5'-PHOSPHATE_QI	347.0	135.0	-60	-32
CITRATE_QI	191.0	87.0	-25	-25
PURINE_QI	119.0	64.9	-90	-32
N-ACETYLNEURAMINATE_QI	308.1	169.9	-25	-15
CYTIDINE_QI	242.1	110.0	-60	-10
INOSINE_QI	267.1	108.0	-60	-40
N-ACETYL-D-GLUCOSAMINE_QI	220.1	100.0	-60	-10
(S)-DIHYDROOROTATE_QI	157.0	42.1	-25	-28
MALONATE_QI	103.0	41.0	-25	-40
L-METHIONINE_QI	148.0	100.0	-25	-10
ADENINE_QI	134.0	92.0	-25	-20
THYMIDINE_QI	241.1	125.0	-60	-10
GLYCERATE_QI	105.0	75.0	-25	-15
XANTHINE_QI	151.0	42.0	-25	-28
URIDINE-5-MONOPHOSPHATE_QI	323.0	96.9	-60	-20
URIDINE_QI	243.1	82.0	-60	-40
CARNOSINE_QI	225.1	109.9	-60	-20
SHIKIMATE_QI	173.0	111.0	-60	-10
SUCCINATE_QI	117.0	98.9	-25	-15
L-ASPARTATE_QI	132.0	71.0	-25	-20
2'-DEOXYCYTIDINE 5'-MONOPHOSPHATE_QI	306.0	195.0	-25	-20
GUANOSINE_QI	282.1	132.9	-25	-36
(R)-MALATE_QI	133.0	71.0	-60	-10
L-TYROSINE_QI	180.1	163.0	-25	-10
L-ASPARAGINE_QI	131.0	114.0	-25	-10
PYRIDOXINE_QI	168.1	122.0	-25	-16
DAMP_QI	330.1	195.0	-25	-10
FOLIC ACID_QI	440.1	310.9	-25	-24
D-GLUCOSAMINE 6-PHOSPHATE_QI	258.0	96.9	-60	-16
THEOPHYLLINE_QI	179.1	122.1	-60	-24
D-(+)-TREHALOSE_QI	341.1	119.0	-60	-20
O-SUCCINYL-L-HOMOSERINE_QI	218.1	73.0	-60	-28
D-GLUCURONOLACTONE_QI	175.0	85.0	-25	-10
ADENOSINE 5'-DIPHOSPHATE_QI	426.0	134.0	-90	-20
GALACTITOL_QI	181.1	71.0	-60	-20
4-PYRIDOXATE_QI	182.0	107.9	-25	-24
PYRIDINE-2,3-DICARBOXYLATE_QI	166.0	78.0	-25	-15
D-GLUCURONIC ACID_QI	193.0	85.0	-25	-20

DEOXYURIDINE_QI	227.1	183.9	-60	-15
KYNURENINE_QI	207.1	143.9	-25	-28
5-OXO-L-PROLINE_QI	128.0	82.0	-25	-20
4-ACETAMIDOBUTANOATE_QI	144.1	100.0	-25	-15
O-PHOSPHO-L-SERINE_QI	184.0	79.0	-25	-40
L-CYSTATHIONINE_QI	221.1	119.9	-25	-15
3-HYDROXY-3-METHYLGLUTARATE_QI	161.0	57.1	-25	-20
4-HYDROXY-L-PROLINE_QI	130.1	84.0	-25	-20
N-METHYL-L-GLUTAMATE_QI	160.1	116.1	-25	-10
XANTHOSINE_QI	283.1	107.8	-25	-40
3-METHYLCROTONYL-COA_QI	848.1	408.0	-60	-40
NICOTINIC ACID ADENINE DINUCLEOTIDE PHOSPHATE_QI	742.1	408.0	-25	-40
INOSINE 5'-DIPHOSPHATE_QI	427.0	79.0	-25	-40
2'-DEOXYURIDINE 5'-TRIPHOSPHATE_QI	467.0	369.0	-25	-20
2'-DEOXYGUANOSINE 5'-DIPHOSPHATE_QI	426.0	158.9	-60	-40
D-FRUCTOSE 6-PHOSPHATE_QI	259.0	79.0	-60	-40
URIDINE 5'-DIPHOSPHOGALACTOSE_QI	565.0	79.0	-60	-40
2'-DEOXYURIDINE 5'-MONO-PHOS-PHATE_QI	307.0	79.0	-60	-20
FLAVIN ADENINE DINUCLEOTIDE_QI	784.1	346.1	-60	-40
N-ACETYL-DL-METHIONINE_QI	190.1	141.9	-25	-15
GUANOSINE 3',5'-CYCLIC MONOPHOSPHATE_QI	344.0	150.0	-60	-20
O-PHOSPHO-DL-SERINE_QI	184.0	79.0	-25	-40
S-(5'-ADENOSYL)-L-HOMOCYSTEINE_QI	383.1	188.0	-60	-20
URIDINE 5'-DIPHOSPHO-N-ACETYLGLUCOSAMINE_QI	606.1	273.0	-60	-40
URIDINE 5'-DIPHOSPHOGLUCURONIC ACID_QI	579.0	323.0	-60	-23
CYTIDINE 5'-DIPHOSPHATE_QI	402.0	159.0	-60	-20
2,4-DIHYDROXYPYRIMIDINE-5-CARBOXYLIC ACID_QI	155.0	42.0	-25	-32
GUANOSINE 5'-MONOPHOSPHATE_QI	362.1	97.0	-60	-40
N-ACETYL-DL-GLUTAMIC ACID_QI	188.1	102.0	-25	-20
D-PANTOTHENIC ACID_QI	218.1	146.1	-60	-16
ANILINE-2-SULFONIC ACID_QI	172.0	108.0	-60	-21
THYMIDINE 5'-MONOPHOSPHATE_QI	321.0	195.0	-25	-10
D-SACCHARIC ACID_QI	209.0	55.0	-25	-40
4-IMIDAZOLEACETIC ACID_QI	125.0	40.0	-25	-32
D-(+)-GALACTURONIC ACID_QI	193.0	73.0	-25	-10
CYTIDINE 5'-TRIPHOSPHATE_QI	482.0	159.0	-120	-20
3',5'-CYCLIC AMP_QI	328.0	79.1	-120	-40
SORBITOL-1,1,2,3,4,5,6,6-d8	189.2	123.0	-60	-24

ADENINE-2-d1	116.1	99.0	-25	-25
SUCCINATE-13C4	134.5	92.0	-20	-17
CUDA	339.2	214.3	-30	-33

Abbreviations: QI, qualifier ion; Q1, precursor ion; Q3, product ion; DP, declustering potential; CE, collision energy