

Metabolite profiling.

Sample processing

Frozen brain tissue samples were homogenized in 4 volumes of HPLC water (J. T. Baker, Center Valley Pa.) using a TissueLyser II (Qiagen, Hilden, Germany) with 3mm tungsten beads at 20 Hz in two 2-minute cycles. Homogenates were then aliquoted for profiling.

Data acquisition

Polar metabolites were profiled using liquid chromatography tandem mass spectrometry (LC-MS). Briefly, positive ionization mode data were acquired using a 6495 triple quadrupole mass spectrometer coupled to a 1290 Infinity II U-HPLC system (Agilent, Santa Clara, CA). Plasma or brain homogenates (10 μ L) were extracted using 90 μ L of 74.9:24.9:0.2 (v/v/v) acetonitrile/methanol/formic acid containing stable isotope-labeled internal standards (0.2 ng/ μ L valine-d8, Isotec; and 0.2 ng/ μ L phenylalanine-d8 (Cambridge Isotope Laboratories, Inc., Tewksbury MA)). The samples were centrifuged (10 min, 9,000 \times g, 4°C) and the supernatants (10 μ L) were injected onto a 150 \times 2.1 mm Atlantis HILIC column (Waters). The column was eluted isocratically at a flow rate of 250 μ L/min with 5% mobile phase A (10 mM ammonium formate and 0.1% formic acid in water) for 1 minute followed by a linear gradient to 40% mobile phase B (acetonitrile with 0.1% formic acid) over 10 minutes. MS data were acquired using multiple reaction monitoring and retention times, mass transitions, and collision energies were determined using authentic reference standards. Other MS parameters were: ion spray voltage, 3.0 kV; source temperature, 200°C; nozzle voltage, 500 V; gas flow, 14 L/min; nebulizer, 40 psi; sheath gas, 250°C; sheath gas flow, 1 L/min; iFunnel high pressure RF, 90; and low pressure RF, 90. Raw data were processed using MassHunter software (Agilent, Santa Clara, CA) for automated peak integration. Metabolite peaks were manually reviewed for quality of integration and compared against standard reference standards to confirm identities.

Supplement

Table SX. Multiple reaction monitoring MS parameters

Compound	Parent Ion (m/z)	Product Ion (m/z)	RT (min)	Collision Energy
valine-d8	126.1	80.2	7.42	9
phenylalanine-d8	174.1	128.1	6.70	17
1-methylhistamine	126.1	109.0	9.95	13
1-methylnicotinamide	139.1	122.0	8.25	13
2-deoxycytidine	228.1	112.0	5.95	9
3-chloro-L-tyrosine	216.0	169.9	6.35	13
3-hydroxyanthranilic acid	154.1	136.0	1.85	9
3-hydroxykynurenine	225.1	208.1	6.50	9
3-methyloxytriamine	168.1	151.0	6.10	9
3-nitro-L-tyrosine	227.1	180.9	1.95	13
5-adenosylhomocysteine	385.1	136.1	8.20	21
5-aminolevulinic acid	132.1	86.1	7.50	13
5-HIAA	192.1	146.0	1.90	17
5-hydroxytryptophan	221.1	204.0	6.12	9
5-methylthioadenosine	298.1	136.0	4.10	21
acetylcholine	147.1	88.0	8.50	13
adenine	136.1	119.0	5.45	24
adenosine	268.1	136.0	4.95	17
ADMA	203.2	70.2	9.95	33
alanine	90.1	44.3	7.80	13
alpha-glycerophosphocholine	258.1	104.1	10.70	17
aminoisobutyric acid	104.1	30.3	7.75	13
anserine	241.1	109.0	10.75	25
anthranilic acid	138.1	120.0	1.95	9
arginine	175.1	70.2	9.30	33
argininosuccinate	291.1	70.2	9.30	35
asparagine	133.1	74.1	7.97	17
aspartate	134.1	74.1	8.05	17
atenolol	267.2	145.0	6.80	29
atorvastatin	559.3	440.1	1.75	25
beta-alanine	90.1	72.1	7.90	5
betaine	118.1	58.2	9.00	33
butyrobetaine	147.1	88.1	8.85	17
carnitine	162.1	85.1	8.90	21
C2 carnitine	204.1	85.1	8.72	19
C3 carnitine	218.4	85.1	8.35	20
C4 carnitine	232.4	85.1	8.00	20
C5:1 carnitine	244.5	85.1	7.85	20
C5 carnitine	246.5	85.1	7.77	20
C3-DC carnitine	248.4	85.1	8.90	20
C4-OH carnitine	248.4	85.1	8.90	20
C6 carnitine	260.2	85.0	7.60	19
C3-DC-CH3 carnitine	262.4	85.1	8.70	20

C7 carnitine	274.5	85.1	7.40	20
C5-DC carnitine	276.5	85.1	8.35	20
C8 carnitine	288.2	85.1	7.30	23
C9 carnitine	302.5	85.1	7.13	20
C10:2 carnitine	312.6	85.4	7.20	20
C10 carnitine	316.3	85.1	7.05	31
C12:1 carnitine	342.6	85.1	6.95	20
C12 carnitine	344.6	85.1	6.85	20
C14:2 carnitine	368.5	85.2	6.80	20
C14:1 carnitine	370.5	85.2	6.70	20
C14 carnitine	372.3	85.1	6.75	27
C16 carnitine	400.3	85.1	6.60	35
C16-OH carnitine	416.7	85.1	7.05	20
C18:2 carnitine	424.7	85.1	6.60	20
C18:1 carnitine	426.4	85.2	6.50	31
C18 carnitine	428.7	85.1	6.48	20
C18:1-OH carnitine	442.7	85.1	7.00	20
C20:4 carnitine	448.7	85.1	6.45	20
C20 carnitine	456.7	85.1	6.38	20
C22:6 carnitine	472.6	85.1	6.40	20
C24:4 carnitine	504.8	85.1	5.12	20
C26 carnitine	540.9	85.1	6.20	20
cAMP	330.1	136.2	6.92	25
carnosine	227.1	110.1	9.48	25
carvedilol	407.2	100.0	5.65	35
cGMP	346.1	134.9	6.35	35
choline	105.1	61.3	8.35	17
citrulline	176.1	70.1	9.30	25
cotinine	177.1	80.1	4.98	25
creatine	132.1	90.1	8.30	9
creatinine	114.1	44.3	6.42	17
cystamine	153.1	108.0	9.25	9
cysteine	122.0	59.2	7.45	29
cytosine	112.1	94.9	5.65	21
dimethylglycine	104.1	58.2	8.35	13
dopamine	154.1	91.1	6.20	25
epinephrine	184.1	166.0	6.60	5
GABA	104.1	87.1	7.55	9
glucosamine	180.1	162.1	8.60	5
glutamate	148.1	84.0	7.60	17
glutamine	147.1	84.1	8.05	17
glycine	76.0	30.4	7.92	9
guanidinoacetic acid	118.1	30.4	7.85	21
guanine	152.1	135.0	5.65	20
histamine	112.1	95.0	9.50	17
histidine	156.1	110.0	9.55	13
homocysteine	136.1	90.0	7.05	9
hydroxyproline	132.1	86.1	7.90	13
hypoxanthine	137.1	119.0	4.72	24

isoleucine	132.1	86.1	7.10	9
kynurenic acid	190.1	144.1	5.10	21
kynurenine	209.1	94.0	6.60	13
leucine	132.1	86.1	6.95	9
lisinopril	406.2	84.1	9.65	25
lysine	147.1	84.2	9.55	21
melatonin	233.1	174.0	2.50	13
metformin	130.1	60.2	7.20	9
methionine	150.1	61.1	7.05	21
methionine sulfoxide	166.1	74.1	8.65	13
metoprolol	268.2	56.2	5.75	33
mevalonic acid	149.1	65.1	1.65	29
N-carbamoyl-beta-alanine	133.1	115.0	4.05	5
niacinamide	123.1	80.1	3.70	21
NMMA	189.1	70.2	9.50	29
ornithine	133.1	70.2	9.42	21
phenylalanine	166.1	120.1	6.70	13
phosphocholine	185.1	87.1	11.00	17
phosphoethanolamine	142.0	44.3	9.50	9
pipecolic acid	130.1	84.1	7.98	17
proline	116.1	70.1	8.10	21
putrescine	89.1	72.2	9.62	9
pyridoxine	170.1	152.0	6.05	13
pyroglutamic acid	130.1	84.1	8.02	13
quinolinic acid	168.0	149.9	4.40	9
sarcosine	90.1	44.3	8.30	11
SDMA	203.1	70.3	9.80	33
serine	106.1	60.2	7.65	11
serotonin	177.1	160.0	6.30	11
sildenafil	475.2	58.2	6.10	35
spermidine	146.2	72.1	12.00	15
spermine	202.2	112.1	9.80	19
taurine	126.0	44.3	6.05	23
thiamine	266.1	122.1	9.92	15
threonine	120.1	74.0	7.65	25
thymidine	243.1	127.0	4.90	11
thymine	127.1	110.0	2.95	16
thyroxine	777.7	731.5	5.60	23
triiodothyronine	651.8	605.6	5.65	27
trimethylamine-N-oxide	76.1	58.2	8.10	23
tryptophan	205.1	187.9	6.45	7
tyramine	138.1	121.0	6.27	11
tyrosine	182.1	136.1	6.80	11
uracil	113.0	70.2	10.60	20
uric acid	169.0	107.2	1.80	12
valine	118.1	72.2	7.40	11
verapamil	455.3	165.0	1.75	31
xanthosine	285.1	153.1	4.25	11
xanthurenic acid	206.1	160.0	6.00	17

