

Supplementary Table 1 Detailed ingredients of the MCD diet and MCD control diet

Diet	MCD diet		MCD control diet	
	gm	kcal	gm	kcal
Protein	17	16	17	16
Carbohydrate	66	63	65	62
Fat	10	21	10	21
		100		100
	4.2		4.2	
Ingredient (gm)				
L-Alanine	3.5	14	3.5	14
L-Arginine	12.1	48.4	12.1	48.4
L-Asparagine-H ₂ O	6	24	6	24
L-Aspartate	3.5	14	3.5	14
L-Cystine	3.5	14	3.5	14
L-Glutamine	40	160	40	160
Glycine	23.3	93.2	23.3	93.2
L-Histidine-HCl-H ₂ O	4.5	18	4.5	18
L-Isoleucine	8.2	32.8	8.2	32.8
L-Leucine	11.1	44.4	11.1	44.4
L-Lysine-HCl	18	72	18	72
L-Phenylalanine	7.5	30	7.5	30
L-Proline	3.5	14	3.5	14
L-Serine	3.5	14	3.5	14
L-Threonine	8.2	32.8	8.2	32.8
L-Tryptophan	1.8	7.2	1.8	7.2
L-Tyrosine	5	20	5	20
L-Valine	8.2	32.8	8.2	32.8
Total L-Amino Acids	171.4	685.6	171.4	685.6
Sucrose	455.3	1821.2	452.3	1809.2
Corn Starch	150	600	150	600
Maltodextrin 10	50	200	50	200
Cellulose	30	0	30	0
Corn Oil	100	900	100	900
Mineral Mix S10001	35	0	35	0
Sodium Bicarbonate	7.5	0	7.5	0
Vitamin Mix V10001	10	40	10	40
L-Methionine	0	0	3	12
Choline Bitartrate	0	0	2	0
Total	1009.2	4246.8	1011.2	4246.8

MCD: methionine-choline-deficient.

Supplementary Table 2 Changes in of differential metabolites as determined by GC-MS in the MCD group compared to those in the control group at 2 weeks

Metabolites	MZ	RT(min)	VIP	P-value	Fold change	Average (MCD)	Average (Control)
Arachidic acid	117	17.32451	2.11907	3.256E-18	0.3757095	121.319259	322.907082
Hexadecane	71	5.921848	2.12664	2.4362E-17	1.81709779	105.358619	57.9818103
Elaidic acid	129	28.52395	1.25608	2.9306E-16	0.19375394	4.54217003	23.4429811
3-Hydroxynorvaline	131	9.715868	2.12478	7.4918E-16	2.15331213	16.198138	7.52242918
2-Aminoethanethiol	130	7.505784	2.01895	7.2635E-15	1.45504748	20.1818506	13.8702351
Pentadecanoic acid	117	26.75852	1.27319	2.3871E-14	0.20259598	0.33821819	1.66942202
p-benzoquinone	121	5.579929	2.11137	5.3984E-14	1.52639007	14.100864	9.23804752
Spermine	144	22.73695	1.7685	1.3975E-13	0.0928121	0.26467608	2.85174114
Glucose-1-phosphate	217	8.865014	1.84602	6.129E-13	3.43495946	33.6025093	9.78250536
3,4-Dihydroxyphenylglycol	147	27.26112	1.0833	1.0581E-12	0.19971126	5.62992477	28.1903225
N-Acetylisatin	263	13.02163	2.15218	1.2136E-12	1.55435007	28.5024957	18.3372435
1-Methylhydantoin	116	7.39099	1.98661	2.8E-12	1.50470188	144.885397	96.2884402
N-Acetyl-D-galactosamine	71	20.48059	1.9809	2.8714E-12	1.60181675	68.1811902	42.5649128
Oleic acid	129	25.1285	1.92711	1.593E-11	0.32101751	24.2929453	75.6748301
Putrescine	174	9.636674	1.32041	2.0046E-11	0.0673124	1.35193089	20.0844258

Ceholic acid	253	35.81975	1.91406	3.4919E-11	0.25000421	56.4083707	225.629683
Ttetraicosane	71	5.877989	1.91053	6.5147E-11	1.50292401	312.889109	208.186912
3-Aaminopropionitrile	71	13.61563	1.88943	1.2868E-10	1.58676554	6.108543	3.84968216
5-Aalpha-Dihydroprogesterone	253	41.39163	1.8642	1.5763E-10	2.01704834	1.02975332	0.51052486
2-Furoic Acid	55	22.70568	1.87327	3.3037E-10	8.60110081	5.66360966	0.65847498
N-cyclohexylformamide	57	8.404311	1.95956	3.6422E-10	25.5293936	3.94090877	0.1543675
Diglycerol	207	15.2439	1.82547	8.5963E-10	1.6492677	7.11182466	4.31211057
8-Aminocaprylic acid	174	12.48007	1.81345	1.1279E-09	1.45680989	14.9988815	10.2957026
Maleamate	151	7.047265	1.83121	1.3414E-09	1.40662733	10.9979543	7.8186696
Ffumaric acid	80	10.46577	1.67546	2.2802E-09	1.51655908	1.36902443	0.9027175
1-Monopalmitin	205	20.92656	1.59224	3.5612E-09	5.5768926	1.45531373	0.26095423
2-Hhydroxybutanoic acid	131	6.097009	1.8591	4.2503E-09	2.40401996	11.1132933	4.62279576
3-Hydroxypyridine	152	7.55446	1.82088	4.3785E-09	1.52569941	3.34205434	2.19050641
Gglycerol	205	9.584259	1.74951	1.6465E-08	0.39121744	102.609716	262.28308
Cholestane-3,5,6-triol, (3?5?6?-	195	37.0944	2.04249	2.2196E-08	9.00470336	8.84810608	0.98260939
Sstearic acid	117	22.30491	1.61898	2.5072E-08	0.77306574	621.112234	803.440384
Behenic acid	132	28.94067	1.6562	3.1915E-08	0.57851618	33.7991797	58.4239139
Hheptadecanoic acid	117	14.96926	1.78021	5.5526E-08	1.37445541	788.793065	573.894983

Dithioerythritol	221	6.61901	1.7363	1.4371E-07	1.55301442	15.1098913	9.72939534
Perillyl Alcohol	187	15.89798	1.66685	2.1363E-07	456742.024	0.45674202	0.000001
Palmitic acid	117	17.23046	1.4729	2.2335E-07	0.71020847	927.751067	1306.30808
Alanine	116	7.103686	1.52297	2.329E-07	0.49437613	74.9933906	151.692984
Zymosterol	129	38.98858	1.17742	3.1422E-07	2.04765484	170.787429	83.4063562
Pyruvic acid	174	6.339347	1.95742	3.732E-07	9.105951	2.50728761	0.27534605
24,25-Dihydrolanosterol	83	37.99151	1.96283	4.4568E-07	5.76846612	21.2158078	3.67789415
4-Aminophenol	142	24.10197	2.052	4.5185E-07	1.44504897	2.71648659	1.87985782
Noradrenaline	174	13.28223	1.54699	7.653E-07	1.34763584	20.2075347	14.9948036
Phosphomycin	72	12.12994	1.38273	1.0396E-06	5.72174495	3.14282343	0.54927709
Uridine	139	38.03379	1.78296	1.1493E-06	1.54400048	5.5437708	3.59052402
4-Nitrophenol	196	10.21402	1.82869	1.257E-06	3.35757719	9.30374753	2.77097056
Cis-1,2-Dihydronaphthalene-1,2-diol	191	6.91554	1.61728	1.4415E-06	0.20180266	0.06259558	0.31018213
2-Deoxyerythritol	183	6.597413	1.07888	2.0277E-06	2.64815032	0.3407603	0.12867861
Gallic acid	281	8.165004	1.51375	2.6155E-06	1.8107924	30.7518952	16.9825626
4-Vinylphenol dimer	192	9.362955	1.71317	2.7161E-06	1.50279808	17.8128446	11.8531191
Cholestan-3beta-ol	398	38.87631	1.80218	4.1447E-06	5.36086118	312.765334	58.3423677
2-Deoxy-D-galactose	175	15.91132	1.31117	6.0859E-06	5.66609837	1.75374966	0.30951628

3-Hexenedioic acid	165	23.51869	1.58231	6.3112E-06	2.13069691	5.3723031	2.52138306
Uric acid	57	41.45434	1.59267	6.3172E-06	1.90515637	30.7385783	16.1344122
Linoleic acid	67	22.51888	1.5245	7.0032E-06	0.33459	37.0892902	110.849966
Proline	142	8.239865	1.20212	1.1186E-05	0.60133917	35.2580797	58.6326008
Bis(2-hydroxypropyl)amine	160	23.06775	1.07547	1.436E-05	11.1999492	0.56400283	0.05035762
Cis-gondoic acid	117	16.111	1.45755	1.5508E-05	0.29171955	6.35406222	21.7814068
Tricetin	233	27.13133	1.34834	2.556E-05	1.28640497	45.1338019	35.0852205
Oxamide	102	10.2002	1.4549	2.5896E-05	4.7064E-06	0.000001	0.21247828
Prunin degr. Prod.	219	45.30366	1.19984	2.6507E-05	4.93075284	0.35332928	0.07165828
Erythrose	205	12.0167	1.47551	2.691E-05	1.85143038	19.3977868	10.4771894
L-cysteine	218	13.20717	1.45458	3.3365E-05	3.9645091	1.4126024	0.35631206
Mandelonitrile	190	22.60168	1.43881	5.5978E-05	484857.716	0.48485772	0.000001
Diethyl phthalate	149	13.5979	1.30906	5.7885E-05	1.31668975	4.2517843	3.22914665
Ethyl cinnamate	131	12.03603	1.30051	6.7682E-05	3.3763312	1.0526112	0.31176183
O-acetylserine	116	12.51153	1.32175	9.6809E-05	1.25933788	1.73436191	1.37720142
Kyotorphin	57	8.068523	1.03909	9.822E-05	1.89616224	4.43825833	2.34065326
2-Hydroxypyridine	147	6.137094	1.22312	0.00011119	1.25379328	39.7391286	31.69512
Palmitoleic acid	117	18.70499	1.67869	0.00014561	4.0251082	4.43427462	1.10165352

Ceyclocleucine	156	10.77189	1.05768	0.000202	0.22491031	1.0783498	4.79457695
Levoglucozan	173	14.45395	1.57508	0.00020861	0.4118904	0.11576613	0.28106053
Sedoheptulose	204	31.98197	1.12301	0.00022487	0.11750551	0.33602197	2.85962735
Tryptophan	203	22.1649	1.20211	0.00023886	0.52134865	3.33095601	6.38911405
Octadecanol	103	20.78398	1.25613	0.00025401	1.98452838	7.05724391	3.55613152
5-Aminovaleric acid	174	14.02358	1.91352	0.00027834	0.04677151	14.9905398	320.505784
Methyl Palmitoleate	87	19.90244	1.26012	0.00032878	0.45583116	4.32394752	9.48585335
Quinic acid	255	40.77018	1.23178	0.00033396	30.273485	3.26638605	0.10789594
Nicotinoylglycine	207	5.956709	1.48397	0.00034747	1.28221399	21.6731136	16.9028834
Tyrosine	218	17.81478	1.28754	0.00042876	0.60128767	36.3409324	60.4385126
Cehrysin	147	29.9369	1.24676	0.00049521	0.66822511	14.747849	22.0701808
Analyte 420	72	10.82596	1.15262	0.00059079	19.5523256	0.12637609	0.00646348
2-Butyne-1,4-diol	107	8.558498	1.08417	0.00059962	1.40267861	2.57174446	1.83345239
Pphytosphingosine	204	18.82935	1.22111	0.00062465	0.55416287	122.679403	221.377883
2-Monopalmitin	103	21.81071	1.68888	0.00071371	4.14137999	3.23593878	0.78136727
Cortexolone	177	43.96971	1.47892	0.00073789	597811.755	0.59781175	0.000001
Ethanolamine	174	7.84101	1.28264	0.00076387	0.6395158	9.16348981	14.3287935
5-Methoxytryptamine	174	6.969069	1.08335	0.00077004	0.60811545	3.12210739	5.13407013

Pelargonic acid	117	10.61902	1.07994	0.00115022	1.52833972	3.784613	2.47629041
5-Methylresorcinol	253	35.82096	1.08818	0.00116523	0.47701511	0.52026656	1.09067104
Analyte 476	191	11.63044	1.15519	0.00196357	107863.198	0.1078632	0.000001
Adenosine	192	29.35886	1.30671	0.00228271	9.2323131	0.18175435	0.01968676
Cerotinic acid	132	35.43625	1.22235	0.00250555	1.38730405	7.80474845	5.62583843
Lactic acid	117	5.681062	1.09918	0.00267415	0.15799421	55.5745514	351.750566
D-Talose	205	17.50724	1.01	0.00306633	1.5432266	393.276026	254.840103
Capric Acid	117	11.8661	1.08906	0.00355983	2.14915046	0.57294905	0.26659327
Octanal	86	18.77049	1.02977	0.00369759	185252.89	0.18525289	0.000001
lanosterol	75	42.94944	1.1187	0.00384307	1.35541279	4.7232994	3.48476821
2-Indanone	205	12.62989	1.26621	0.00408952	1.68894172	3.52277937	2.08579096
4-Cholesten-3-one	56	8.288051	1.02485	0.00496478	1.22536642	0.39497614	0.32233308
Nonanoic acid methyl ester	87	22.41519	1.19613	0.00784424	1.27265877	6.90665765	5.42695167
Mannitol	205	21.18538	1.57273	0.01052543	0.52992404	1.24142319	2.34264366
Hydantoin, 5-(4-hydroxybutyl)-	217	25.47083	1.04265	0.01075508	0.59558425	29.860048	50.1357246
Adipic acid	185	22.53616	1.20323	0.01888652	531734.807	0.53173481	0.000001

MCD: methionine-choline-deficient.

Supplementary Table 3 Changes in of differential metabolites as determined by GC-MS in the MCD group compared to those in the control group at 4 weeks

Metabolites	MZ	RT(min)	VIP	P-value	Fold change	Average (MCD)	Average (Control)
11-Bbeta-prostaglandin-F-2-alpha	191	25.74259	1.52828	1.0377E-19	0.1415182	0.14328	1.01244929
Ceholic acid	253	35.81975	2.2211	7.4207E-16	0.04791785	16.0047444	334.00384
Mono(2-ethylhexyl)phthalate	221	10.25353	2.38415	7.5598E-16	66.7573951	2.98157619	0.04466286
3-Hydroxynorvaline	131	9.715868	2.28436	1.6499E-15	7.607427	49.7446363	6.53895677
Arachidic acid	117	17.32451	1.94169	4.7239E-15	0.48226451	229.964492	476.843073
Behenic acid	132	28.94067	2.06308	7.5427E-15	0.46170786	40.3609577	87.4166577
Ceholesterol	129	37.42861	2.21532	1.2523E-14	2.18802449	563.805863	257.67804
Ppentadecanoic acid	117	26.75852	2.22716	1.3175E-14	0.05113198	0.12436458	2.43222675
Cetadiol	267	36.13898	2.12367	1.6412E-14	7.8403E-07	0.000001	1.27546463
21-Hhydroxypregnenolone	157	18.47285	2.18265	5.3004E-13	6.5058E-07	0.000001	1.53708966
Bis(2-hydroxypropyl)amine	160	23.06775	1.30517	1.1902E-12	4.12334817	0.9532411	0.23118133
Ttetracosane	71	5.877989	2.03344	1.1855E-11	1.90943482	461.520497	241.70529
Nicotinoylglycine	207	5.956709	2.00513	1.67E-11	2.01263136	20.1330003	10.0033223
Sstearic acid	117	22.30491	1.93498	2.1618E-11	0.73479394	575.29705	782.936573
N-cyclohexylformamide	57	8.404311	1.78808	4.4146E-11	7.14007199	4.96487782	0.69535403

Cehrysin	147	29.9369	2.00782	8.1238E-11	0.46395794	10.9389826	23.5775308
Diglycerol	207	15.2439	2.33516	8.9862E-11	0.02145163	0.25972509	12.1074775
N-Acetyl-L-leucine	71	9.461075	1.97851	1.547E-10	2.13352186	13.6538987	6.39969945
3,4-Dihydroxyphenylglycol	147	27.26112	1.89357	4.1713E-10	0.33199674	10.71865	32.2854074
Ppalmitoleic acid	117	18.70499	1.80662	4.2036E-10	0.39382469	4.0240221	10.2178004
2-Butyne-1,4-diol	107	8.558498	1.71261	9.2448E-10	17.2442969	2.4067367	0.13956711
Noradrenaline	174	13.28223	1.92679	1.0837E-09	1.5480021	22.045146	14.241031
Gglucuronic acid	160	18.0372	2.17189	1.1275E-09	25.9496869	0.55972396	0.02156958
4-Vinylphenol dimer	192	9.362955	1.92763	1.2531E-09	1.91603944	17.1063768	8.92798786
Gglutaraldehyde	221	8.603746	1.85317	3.014E-09	1.71687173	1.71570846	0.99932245
Ffarnesol	107	5.825323	1.29579	3.5086E-09	13.5643984	55.6459116	4.10235013
Tricetin	233	27.13133	1.73542	9.7525E-09	0.4931175	19.1968258	38.9295162
Cortexolone	177	43.96971	2.31895	1.1073E-08	925698.6	0.9256986	0.000001
Oectanal	86	18.77049	1.78586	1.4883E-08	1.42560186	20.9998536	14.7305178
Zymosterol	129	38.98858	1.74499	2.8204E-08	1.64677695	237.469183	144.202397
S-carboxymethylcysteine	75	13.40518	1.61743	5.9896E-08	8.51652604	1.38377507	0.16248116
Tetradecanoic acid, methyl ester	87	15.78893	1.97157	6.8803E-08	0.02881273	0.05961999	2.06922382
Quinaldic Acid	143	19.29844	1.77269	7.8825E-08	10.655041	1.09545911	0.10281135

2-Aminoethanethiol	130	7.505784	1.72003	9.0317E-08	1.84172256	18.5102853	10.0505286
Adipic acid	185	22.53616	1.37536	2.1431E-07	6.19089893	1.02852268	0.16613463
Heptadecanoic acid	117	14.96926	1.24911	2.2001E-07	0.61402597	441.010749	718.228169
Maleamate	151	7.047265	1.77808	2.4096E-07	2.0174556	11.6905252	5.79468772
2-Hydroxypyridine	147	6.137094	1.67996	3.6416E-07	1.63396611	43.8942705	26.8636359
1-Methylhydantoin	116	7.39099	1.66508	4.5527E-07	1.94926616	132.873016	68.165661
5-Methylresorcinol	253	35.82096	1.43092	5.4457E-07	0.12193645	0.16734943	1.37243157
Uric acid	57	41.45434	1.66591	5.4776E-07	1.77389774	51.3705785	28.9591544
Dehydroascorbic Acid	173	12.94381	1.1908	5.8706E-07	2.15385149	0.23893021	0.11093161
Methyl Palmitoleate	87	19.90244	1.87459	7.8975E-07	0.16107391	3.96560354	24.6197757
Fumaric acid	80	10.46577	1.63004	8.1337E-07	1.5352897	1.94580606	1.26738691
Analyte 476	191	11.63044	1.80892	1.1152E-06	18.4272055	0.28018868	0.01520516
Lignoceric acid	117	30.58297	1.52469	1.2706E-06	0.60786271	91.1015258	149.871877
3-Aminopropionitrile	71	13.61563	1.60959	1.9262E-06	1.53743717	5.37235403	3.4943568
3-Hydroxypalmitic acid	195	41.94402	1.42255	5.4126E-06	9.71682019	1.45109682	0.14933865
DL-dihydrosphingosine	204	10.91404	1.4927	6.6013E-06	0.51418777	23.5509442	45.8022254
Guanosine	144	31.91643	1.28273	2.0757E-05	50.5777431	0.39675078	0.00784437
Cholestane-3,5,6-triol, (3S,5S,6S)-	195	37.0944	1.61209	2.125E-05	2.40531724	8.75981317	3.64185357

Sitosterol	129	40.36715	1.45451	2.209E-05	1.54411991	609.746351	394.882772
Prostaglandin E2	225	6.327636	1.2014	2.7964E-05	3.19919001	1.41472098	0.44221224
Nonanoic acid methyl ester	87	22.41519	1.42976	2.9496E-05	0.64907679	5.11734441	7.88403548
3-Hexenedioic acid	165	23.51869	1.41493	3.0265E-05	2.8698E-07	0.000001	3.48460163
Hexadecane	71	5.921848	1.40121	3.4358E-05	1.6882908	102.708503	60.8357892
3-Hydroxypyridine	152	7.55446	1.38884	3.6799E-05	1.63103139	2.57692158	1.57993377
Ribose	103	14.76194	1.56118	4.184E-05	0.47591119	60.6960318	127.536466
Diethyl phthalate	149	13.5979	1.37716	8.2908E-05	1.40528425	5.6650627	4.03125753
5-Aminovaleric acid	174	14.02358	1.03963	0.00015843	0.15159158	33.4533618	220.680871
Glutathione	107	6.528893	1.60455	0.00016863	667817.677	0.66781768	0.000001
Maltotriitol	204	28.48848	1.15279	0.00024206	0.31879321	2.45926999	7.71431116
Uridine	139	38.03379	1.35048	0.00025091	1.24087322	4.19008554	3.37672333
p-benzoquinone	121	5.579929	1.26843	0.00031015	2.8103E-07	0.000001	3.55830768
Cycloleucine	156	10.77189	1.09435	0.00032838	0.29838509	1.90811501	6.39480686
2-Deoxytetronic acid	189	11.67288	1.55823	0.00036136	0.21192023	0.0313056	0.14772349
Maltose	204	28.90088	1.21984	0.0004707	0.54637215	8.03747428	14.7106222
Squalene	81	32.05698	1.09512	0.00059235	0.72211068	35.0104499	48.4834954
Urea	189	10.35712	1.06077	0.00066517	1.47442618	0.7772469	0.52715213

Cois-gondoic acid	117	16.111	1.2201	0.00068859	0.37575472	1.57064616	4.17997718
Sspermine	144	22.73695	1.32394	0.00070085	0.11432108	0.69036257	6.03880391
Methyl-beta-D-galactopyranoside	204	10.17833	1.45644	0.00079515	3.2988852	4.24707553	1.2874275
D-galacturonic acid	142	18.2446	1.19655	0.00085016	0.26801066	0.05704519	0.21284672
Dodecanol	243	13.22003	1.27383	0.0011684	3.64583089	0.96247449	0.26399318
Stigmasterol	83	39.39231	1.24989	0.00119931	1.38112926	82.969505	60.0736709
Pphloroglucinol	73	10.09054	2.01965	0.00140417	36.8680299	68.8877642	1.86849594
Pelargonic acid	117	10.61902	1.05767	0.00225471	1.50838395	5.21199867	3.45535278
Mmethyloctanoate	87	5.4365	1.02931	0.00229667	0.69452821	4.15332374	5.98006488
N-Ethylglycine	71	8.702305	1.86248	0.00235993	0.33759155	0.77151733	2.28535736
Llactitol	75	18.34837	1.05206	0.00263665	7.4240539	1.28815935	0.17351158
Aalpha-Santonin	204	42.47148	1.22081	0.00264977	1.3773423	1.11211717	0.80743703
Gglycolic acid	147	6.703727	1.00118	0.00297951	1.24806687	4.42453358	3.5451094
5-Aalpha-Dihydroprogesterone	253	41.39163	1.22758	0.00312758	1.32901364	1.3046229	0.98164749
2'-Ddeoxyguanosine	281	31.85468	1.12183	0.00375795	0.05286852	0.05197793	0.98315455
N-Carbamylglutamate	174	20.23807	1.15801	0.00429668	1.58943558	3.60323081	2.26698764
Ethyl cinnamate	131	12.03603	1.10123	0.00648426	0.28363798	0.10556333	0.37217629
Llysine	174	17.57405	1.08433	0.00691688	0.6856915	28.2995961	41.2716156

Elaidic acid	129	28.52395	1.03922	0.00828241	7.22988902	2.75666234	0.38128695
Oxalic acid	147	5.485517	1.1027	0.00836823	1.41894179	146.913044	103.537047
Analyte 2014	253	43.87129	1.0023	0.00864407	0.15300359	0.03676009	0.24025641
6-Phosphogluconic acid	147	25.93081	1.25645	0.01531349	0.61116304	8.24628468	13.4927739
D-erythro-sphingosine	204	13.30478	1.00755	0.01920033	0.76547696	129.291588	168.903305
6-Deoxy-D-glucose	117	15.31829	1.08878	0.03262034	0.53866881	1.68492003	3.12793316

MCD: methionine-choline-deficient.