

Table S1 Ingredient composition of the experimental diets fed to mice.

Low-fat diet (D12450J)		High-fat diet (D12492)	
Ingredient	g/kg	Ingredient	g/kg
Casein, 30 Mesh	200	Casein, 30 Mesh	200
L-Cystine	3	L-Cystine	3
Corn Starch	506.2	Corn Starch	0
Maltodextrin 10	125	Maltodextrin 10	125
Sucrose	68.8	Sucrose	68.8
Cellulose, BW200	50	Cellulose, BW200	50
Soybean Oil	25	Soybean Oil	25
Lard*	20	Lard*	245
Mineral Mix S10026	10	Mineral Mix S10026	10
DiCalcium Phosphate	13	DiCalcium Phosphate	13
Calcium Carbonate	5.5	Calcium Carbonate	5.5
Potassium Citrate, 1 H ₂ O	16.5	Potassium Citrate, 1 H ₂ O	16.5
Vitamin Mix V10001	10	Vitamin Mix V10001	10
Choline Bitartrate	2	Choline Bitartrate	2
FD&C Yellow Dye #5	0.04	FD&C Blue Dye #1	0.05
FD&C Blue Dye #1	0.01		
Energy	kcal%	Energy	kcal%
Protein	20	Protein	20
Carbohydrate	70	Carbohydrate	20
Fat	10	Fat	60
Saturated fatty acid / unsaturated fatty acid ratio	0.8:1	Saturated fatty acid / unsaturated fatty acid ratio	9.8:1
Total kcal/gm	3.85	Total kcal/gm	5.24

*Typical analysis of cholesterol in lard = 0.72 mg/gram.

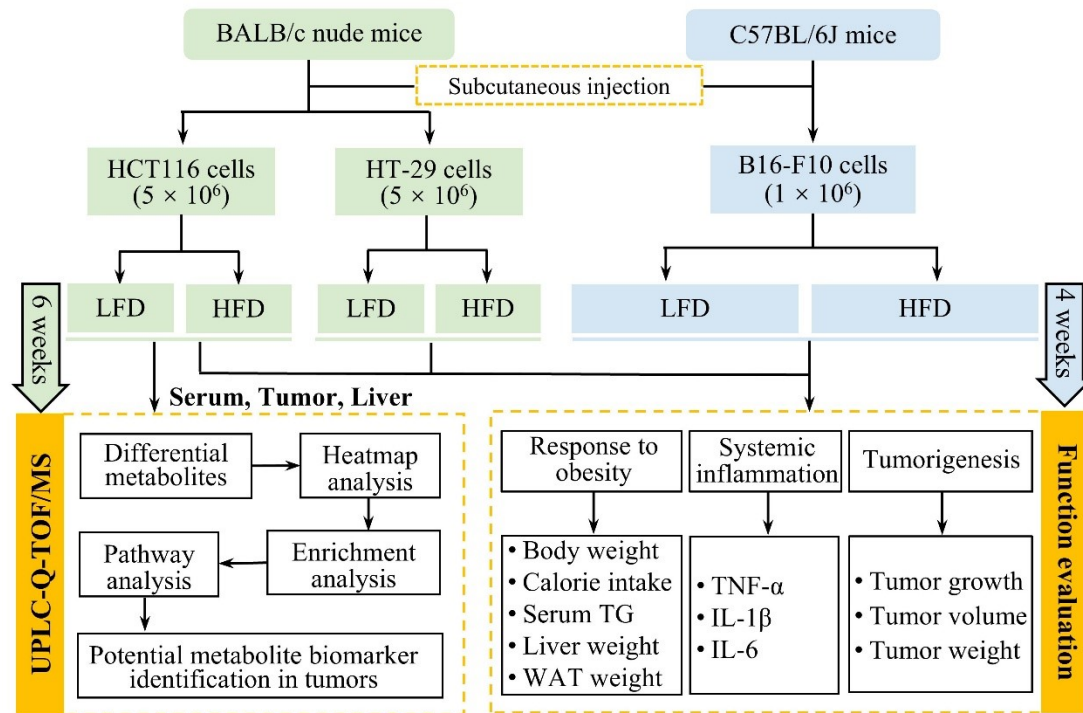


Fig. S1. The experimental design flowchart.

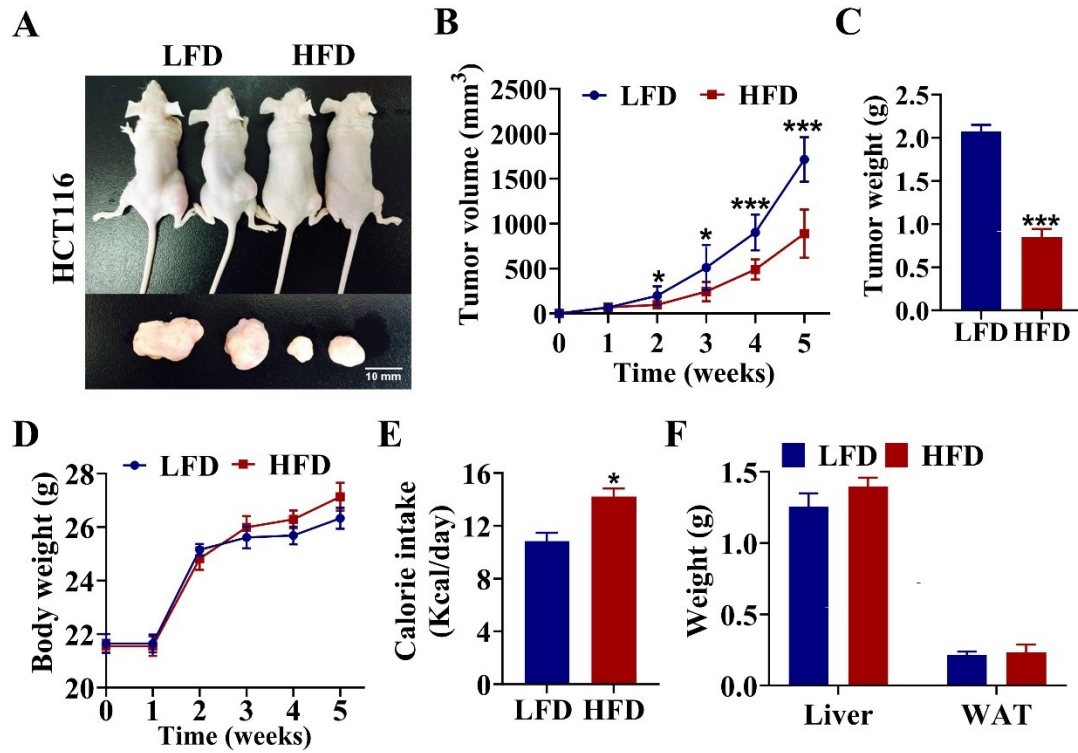


Fig. S2. Pilot study reveals that HFD inhibited HCT116 CRC xenograft tumor growth in BALB/c nude mice. (A) Morphology of xenograft tumors, scale bar: 10 mm; (B) Tumor volume; (C) Tumor weight; (D) Body weight; (E) Calorie intake; (F) Weight of liver and WAT. Data are expressed as mean $\pm$ SD ($n = 3$). * $p < 0.05$ and *** $p < 0.001$, LFD group vs. HFD group.

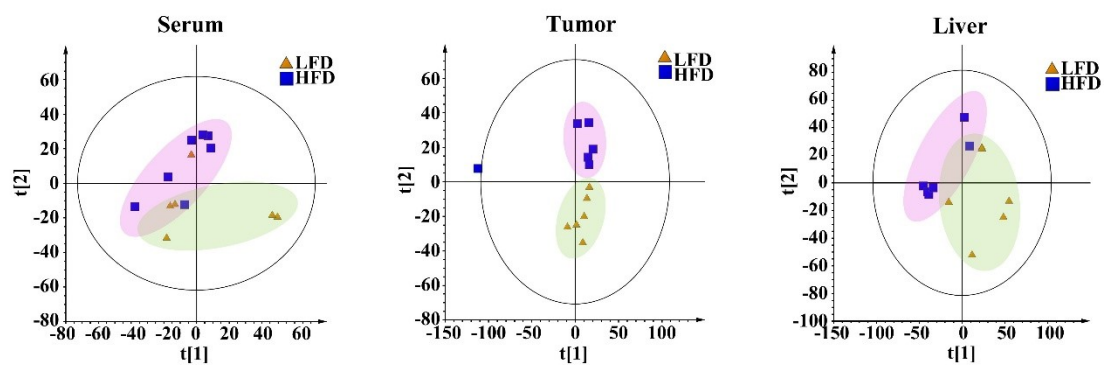


Fig. S3. The PCA score plots in serum, tumor, and liver samples from HCT116 xenograft nude mice between the LFD and HFD groups through untargeted metabolomics.

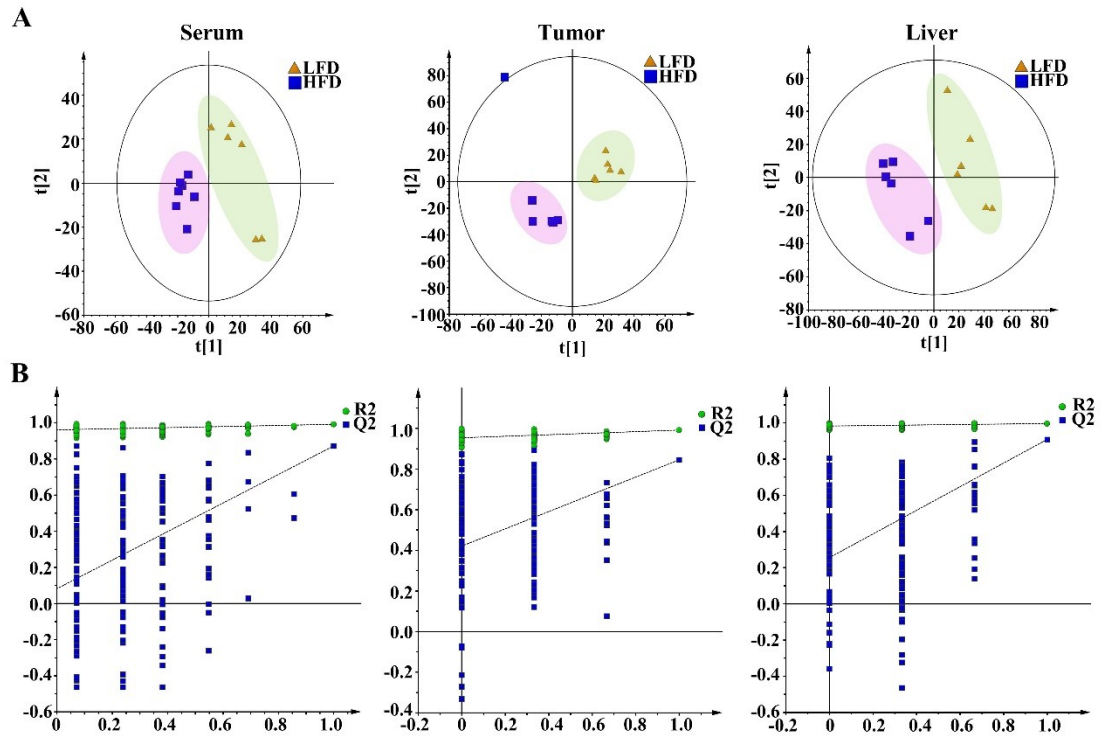


Fig. S4. PLS-DA score plots and permutation test evaluation in serum, tumor, and liver samples from HCT116 xenograft nude mice between the LFD and HFD groups. (A) PLS-DA score plots; (B) Permutation test.

Table S2 The differential metabolites in serum samples from HCT116 xenograft nude mice between the LFD and HFD groups.

Name	Fold change	VIP	CompMW	Time	t-test	ESI
Glutathione, oxidized	2.795293944	1.739	612.1514	1.218	0.011	+
LysoPC (20:0)	2.563295848	1.953	551.3932	17.122	0.002	+
9(S)-HODE	2.386671486	1.614	296.2348	7.213	0.01	-
α -Linolenic Acid	2.057653416	1.867	278.2241	7.214	0.005	+
Phosphohydroxypyruvic acid	1.698192493	1.439	183.9722	0.993	0.045	+
Uric acid	1.564823563	1.432	168.0283	1.064	0.047	+
LysoPE (0:0/20:0)	1.556170353	1.527	509.3471	9.946	0.017	-
LysoPC (18:0)	1.464085696	1.82	523.3608	9.624	0.006	+
4-Hydroxybenzaldehyde	1.440929749	1.52	122.0325	0.759	0.032	+
L-Methionine	1.384149716	1.483	149.0509	1.218	0.038	+
LysoPE (0:0/22:2(13Z,16Z))	1.380317353	1.506	533.3466	8.832	0.019	-
α -D-Glucose/D-Galactose/D-Fructose	1.375541818	1.472	180.0633	0.864	0.023	-
Glucoheptonic acid	1.373636233	1.469	226.0688	0.862	0.023	-
LysoPE (0:0/18:2(9Z,12Z))	1.330529041	1.424	477.284	7.436	0.048	+
LysoPE (0:0/18:0)	1.320422841	1.498	481.3149	9.02	0.035	+
Choline/Neurine	1.301341855	2.294	103.0992	10.111	0	+
5-Methylcytidine	1.204972315	1.477	257.1022	0.991	0.039	+
Oleic Acid	0.829894586	1.541	282.2557	8.264	0.015	-
Phosphocholine	0.826450318	1.806	183.0652	10.812	0.007	+
LysoPC (18:1(11Z))	0.799960128	1.81	521.3466	8.888	0.007	+
LysoPE (0:0/20:1(11Z))	0.784040454	1.51	507.3311	7.881	0.034	+
Metyrosine	0.770037174	1.365	195.0895	6.549	0.038	-
LysoPC (20:4(5Z,8Z,11Z,14Z))	0.755236293	1.817	543.3294	8.61	0.007	+
Nervonyl carnitine	0.733058379	1.534	101.1202	0.914	0.03	+
Acetylcarnitine	0.719965659	1.578	203.1153	1.218	0.025	+
Hypoxanthine	0.664803554	1.714	136.0374	0.892	0.005	-

Pyroglutamic acid	0.663882579	1.77	129.0428	1.218	0.003	-
Retinoic acid	0.644834125	1.51	300.208	7.432	0.034	+
LysoPC (14:0)	0.640379931	1.809	467.3007	7	0.007	+
Oxoglutaric acid	0.625898229	1.576	146.0215	0.919	0.013	-
Palmitoyl-L-carnitine	0.62546454	2.184	399.3328	7.728	0	+
Erythrose	0.613867842	1.545	120.0424	0.911	0.015	-
LysoPE (0:0/20:5(5Z,8Z,11Z,14Z,17Z))	0.572758949	1.569	499.2687	6.879	0.013	-
Allantoin	0.555554364	1.444	158.0441	0.897	0.026	-
Urea	0.545253866	1.47	60.03205	0.967	0.04	+
L-Phenylalanine	0.539614118	1.536	165.0787	4.91	0.03	+
L-Threonine	0.499653546	1.326	119.0584	0.871	0.046	-
LysoPE (0:0/22:1(13Z))	0.476649273	2.296	535.3623	9.162	0	+
D-Leucic acid	0.471719125	1.401	132.0788	3.729	0.032	-
MG (0:0/20:4(5Z,8Z,11Z,14Z)/0:0)	0.471392268	1.38	378.2738	12.19	0.036	-
Stearic acid	0.468136124	2.037	284.2707	12.047	0	-
Ornithine	0.458502022	1.464	132.0896	0.779	0.041	+
Taurocholic acid	0.452816992	2.239	515.2973	7.335	0	+
LysoPC (16:1(9Z))	0.450625231	2.27	493.3156	7.314	0	+
LysoPE (18:1(11Z)/0:0)	0.408951029	1.945	479.3006	7.116	0.001	-
LysoPE (0:0/16:1(9Z))	0.362235539	2.098	451.269	7.087	0	-
Chenodeoxycholic acid	0.358488812	1.31	392.2917	5.732	0.049	-
Octadecanedioic acid	0.355519353	1.392	314.2458	8.013	0.034	-
7-ketodeoxycholic acid	0.336341802	1.341	406.271	5.488	0.043	-
Arachidonic Acid	0.330563651	2.104	304.2388	8.476	0.001	+
Cortisone	0.217185546	2.161	360.1963	5.42	0	-
MG (0:0/16:1(9Z)/0:0)	0.210661707	1.617	328.2609	8.519	0.01	-
Cholic acid	0.197510328	1.503	408.2864	5.145	0.019	-
Cortisol	0.098960733	2.004	362.2089	4.624	0.002	+
Glycocholic acid	0.050695447	1.351	465.3081	4.536	0.041	-

MG (0:0/20:5(5Z,8Z,11Z,14Z,17Z)/0:0)	0.046262916	1.583	376.2606	4.68	0.024	+
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Name, differential metabolite name; VIP, the contribution rate of different substances to the OPLS-DA model; CompMW, the molecular weight of the compound; Time, retention time; *t*-test, significant values in *t*-test; Fold change, the value of the ratio of the mean value in high-fat diet group to low-fat diet group. ESI, electrospray ionization. “+” indicates positive ion mode; “-” indicates negative ion mode.

Table S3 The differential metabolites in tumor samples from HCT116 xenograft nude mice between the LFD and HFD groups.

Name	Fold change	VIP	CompMW	Time	t-test	ESI
Glutaconic acid	4.78659585	1.454	130.0267	2.055	0.039	-
Lactosylceramide (d18:1/12:0)	4.62354683	1.723	805.5567	9.828	0.016	+
Ketoleucine	2.00138677	1.942	130.0631	3.622	0.002	-
Indolelactic acid	1.85060886	1.623	205.0734	4.043	0.026	+
Succinic acid	1.84292837	1.828	118.0267	1.395	0.004	-
SM (d18:1/18:0)	1.62000695	1.712	730.5941	10.115	0.017	+
Hypoxanthine	1.61552156	1.641	136.0373	0.884	0.015	-
Levoglucozan	1.45195828	2.088	162.0528	1.343	0	-
Uracil	1.54314157	1.612	112.0274	1.111	0.018	-
Metyrosine	1.42503761	1.747	195.0893	6.553	0.008	-
4-Hydroxybenzaldehyde	1.51225803	1.459	122.0368	3.856	0.038	-
Palmitic acid	1.35660433	1.621	256.2404	8.909	0.017	-
Pyroglutamic acid	1.21503679	1.396	129.0428	1.014	0.049	-
LysoPE (0:0/16:0)	0.8362464	1.488	453.2826	7.689	0.046	+
LysoPC (16:0)	0.79004131	1.48	495.3289	7.979	0.047	+
Choline/Neurine	0.78840017	2.146	103.099	8.2	0.001	+
Phosphocholine	0.76737305	2.029	183.0651	7.977	0.002	+
LysoPE (0:0/20:1(11Z))	0.74638919	1.918	507.3314	8.504	0.002	-
MG (0:0/15:0/0:0)	0.73611343	1.699	316.2608	6.181	0.011	-
MG (0:0/22:5(4Z,7Z,10Z,13Z,16Z)/0:0)	0.73255044	1.597	404.2918	6.453	0.029	+
Taurine	0.69399636	1.469	125.0143	0.954	0.049	+
Stearic acid	0.65603840	2.024	284.2715	8.811	0.001	-
SM (d18:1/16:0)	0.65883998	1.773	702.5634	9.94	0.012	+
Ubiquinone-1	0.63463544	2.072	250.1201	5.641	0	-
MGDG (18:2(9Z,12Z)/18:3(9Z,12Z,15Z))	0.62503115	1.955	776.55	12.32	0.004	+
Glycocholic Acid	0.61770932	1.557	465.3089	8.463	0.024	-

LysoPE (0:0/20:5(5Z,8Z,11Z,14Z,17Z))	0.61386784	1.47	499.2687	6.747	0.036	-
LysoPE (0:0/20:3(8Z,11Z,14Z))	0.60583633	1.573	503.3029	7.835	0.022	-
LysoPE (0:0/22:6(4Z,7Z,10Z,13Z,16Z,19Z))	0.6037403	1.83	525.283	6.602	0.008	+
Creatine	0.58845337	1.799	131.0688	0.887	0.01	+
D-Leucic acid	0.57236208	1.804	132.072	0.886	0.01	+
LysoPE (0:0/18:0)	0.54980807	2.12	481.3158	8.825	0	-
LysoPC (18:3(9Z,12Z,15Z))	0.5403627	1.928	517.3147	7.041	0.004	+
LysoPE (0:0/22:1(13Z))	0.53292337	1.587	535.3628	10.698	0.021	-
LysoPE (0:0/20:4(5Z,8Z,11Z,14Z))	0.48632747	2.037	501.2846	17.149	0.001	-
LysoPE (0:0/20:0)	0.4691106	1.719	509.3474	10.021	0.01	-
CPA (16:0/0:0)	0.39749263	1.436	392.2322	11.036	0.042	-
LysoPE (0:0/24:6(6Z,9Z,12Z,15Z,18Z,21Z))	0.38715951	1.462	553.3159	7.556	0.037	-
Heme	0.35873738	1.703	614.1663	1.217	0.017	+
Chenodeoxycholic Acid	0.34151006	2.27	392.2919	5.737	0	-
3-Methyl-L-histidine	0.31841937	1.68	169.0847	0.786	0.02	+
Glutathione	0.26480458	1.49	307.0835	1.218	0.033	-
Carnosine	0.09271843	1.839	226.1062	0.786	0.008	+
Homocarnosine	0.07114963	1.704	240.1219	0.781	0.011	-

Name, differential metabolite name; VIP, the contribution rate of different substances to the OPLS-DA Model; CompMW, the molecular weight of the compound; Time, retention time; *t*-test, significant values in *t*-test; Fold change, the value of the ratio of the mean value of high-fat diet group to low-fat diet group. ESI, electrospray ionization. “+” indicates positive ion mode; “-” indicates negative ion mode.

Table S4 The differential metabolites in liver samples from HCT116 xenograft nude mice between the LFD and HFD groups.

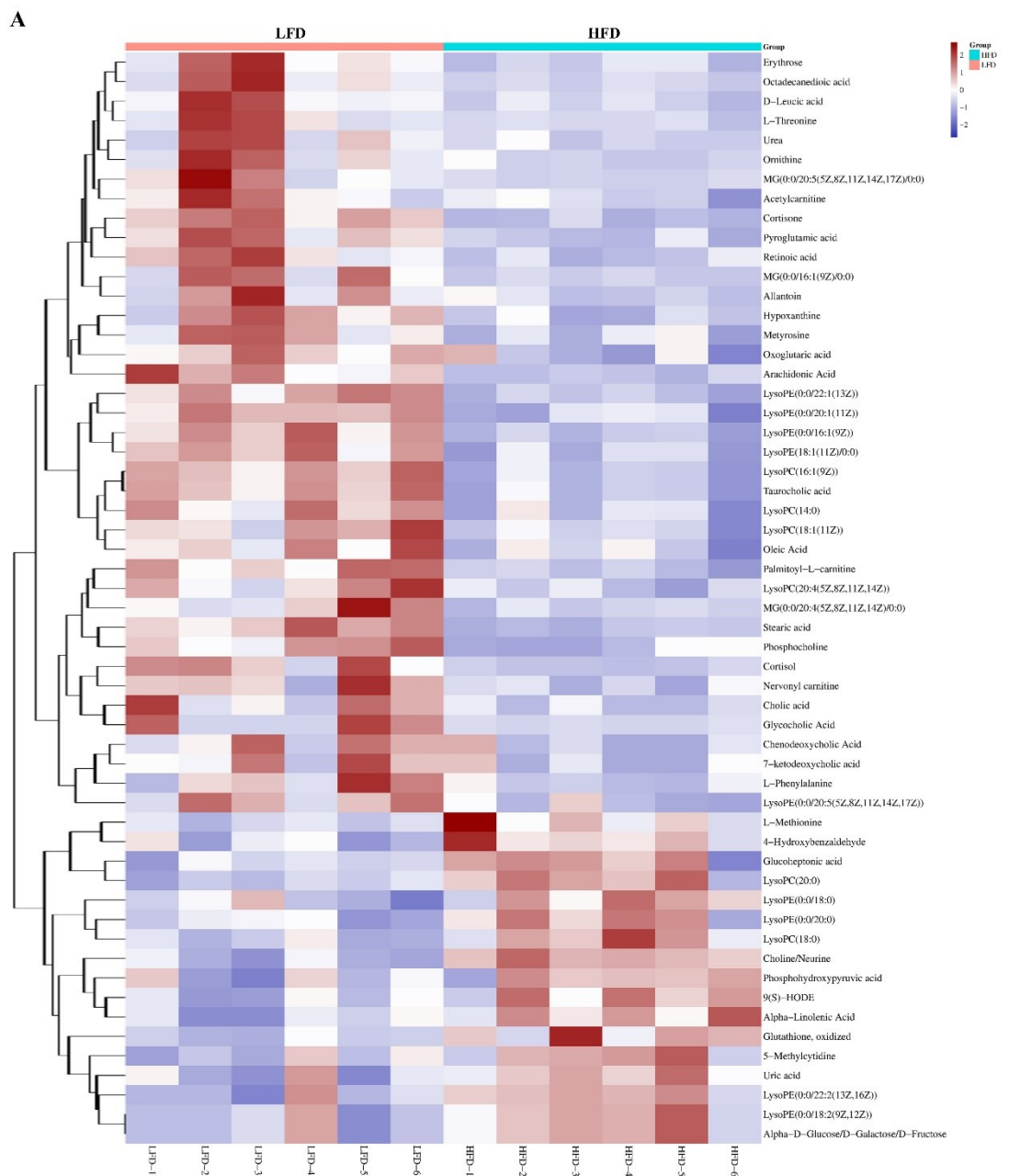
Name	Fold change	VIP	CompMW	Time	t-test	ESI
DG (18:3(9Z,12Z,15Z)/20:4(8Z,11Z,14Z,17Z)/0:0)	28.98028746	-4.857	638.4864	12.132	0.019	+
LysoPE (18:1(11Z)/0:0)	10.28877987	-3.363	479.3001	6.157	0.011	-
Nonadecanoic acid	8.236320667	-3.042	298.2871	10.078	0.003	-
Urocanic acid	6.079699426	-2.604	138.0431	1.366	0.03	-
2-Methylhippuric acid	5.759725274	-2.526	193.0737	3.931	0	-
MG (0:0/22:6(4Z,7Z,10Z,13Z,16Z,19Z)/0:0)	5.668629586	-2.503	402.2755	6.908	0.014	+
Tetracosahexaenoic acid	5.278031643	-2.4	356.2701	6.676	0.001	+
Carnosine	4.927992294	-2.301	226.1065	0.777	0.009	-
Inosine	4.860147168	-2.281	268.0806	1.244	0.039	-
MG (0:0/22:5(4Z,7Z,10Z,13Z,16Z)/0:0)	4.429058338	-2.147	404.2911	6.455	0.038	+
LysoPE (0:0/22:4(7Z,10Z,13Z,16Z))	4.39541687	-2.136	529.3155	6.405	0.023	-
Chenodeoxycholic acid	4.284123293	-2.099	392.2919	6.796	0.026	-
L-Histidine	3.837056477	-1.94	155.0696	0.795	0.005	-
L-Glutamine	3.76069681	-1.911	146.0688	0.995	0.01	+
L-Serine	3.758090997	-1.91	105.0421	1.019	0.01	+
Acetylcarnitine	3.716642697	-1.894	203.1155	3.938	0.003	-
5-Methylcytidine	3.680750602	-1.88	257.1017	0.86	0.003	+
L-arginine/D-arginine	3.217633484	-1.686	174.1113	0.827	0.013	+
3-Hydroxy-L-proline	3.179941004	-1.669	131.0584	1.331	0.003	-
Tauroursodeoxycholic acid	3.012580933	-1.591	499.2952	5.101	0.005	-
Linoleic acid	2.920064021	-1.546	280.24	8.41	0.017	-
α -D-glucose/D-galactose/D-fructose	2.913998228	-1.543	180.0635	0.857	0.006	-
β -Alanine	2.913998228	-1.543	89.04661	0.876	0.01	-
Retinol	2.856007959	-1.514	286.2276	11.574	0.015	+
LysoPE (0:0/20:0)	2.735870608	-1.452	509.3447	10.47	0.014	+
L-aspartic acid	2.681425183	-1.423	133.0377	0.952	0.008	-

Lactic acid	2.642676811	-1.402	90.03184	0.988	0	-
2-Acetolactic acid	2.620786808	-1.39	132.0425	0.972	0.02	-
DG (18:1(11Z)/22:6(4Z,7Z,10Z,13Z,16Z,19Z)/0:0)	2.581124981	-1.368	666.5169	16.516	0.026	+
SM(d18:1/16:0)	2.540301965	-1.345	702.563	12.083	0.012	+
MGDG (18:2(9Z,12Z)/18:3(9Z,12Z,15Z))	2.406606052	-1.267	776.55	11.917	0.021	+
D-Glucuronic acid/D-Fructuronic acid	2.380063393	-1.251	194.0429	0.884	0.016	-
Stearic acid	2.371828999	-1.246	284.2712	10.076	0.002	-
Acetylcholine	2.366902044	-1.243	145.1099	3.474	0.008	+
SM (d18:1/22:0)	2.348923942	-1.232	786.6568	12.07	0	+
Gluconic acid/Galactonic acid	2.347296357	-1.231	196.0585	0.889	0.002	-
Phosphorylcholine	2.177994031	-1.123	169.0524	3.399	0.002	+
Levogluconan	2.165950091	-1.115	162.0529	1.35	0.022	-
L-Tryptophan	2.15995312	-1.111	204.0892	3.394	0.002	+
4-Hydroxybenzaldehyde	2.039195366	-1.028	122.0367	1.227	0.016	+
Stearidonic acid	1.892115293	-0.92	276.2074	8.119	0.036	+
Spermidine	1.780151467	-0.832	145.1575	0.769	0.003	+
α -Linolenic acid	1.742308384	-0.801	278.2237	6.488	0.009	+
9(S)-HODE	1.620006947	-0.696	296.2341	6.485	0.025	+
Phenylpyruvic acid	1.614402149	-0.691	164.0466	1.226	0.022	+
Ribonic acid	1.53581027	-0.619	166.0478	0.925	0.027	-
L-Threonine	1.527317498	-0.611	119.0578	1.025	0.009	+
Taurocholic acid	1.512567997	-0.597	515.2904	4.487	0.048	-
L-Proline	1.446934886	-0.533	115.0627	0.914	0.008	+
Ornithine	1.418140036	-0.504	132.0892	0.789	0.003	+
Homocarnosine	1.329607108	-0.411	240.1218	0.785	0.021	+
L-Methionine	1.319507911	-0.4	149.0503	0.98	0.004	+
Riboflavin (Vitamin B2)	1.310393404	-0.39	376.1374	3.975	0.003	+
LysoPC (14:0)	1.298638603	-0.377	467.2985	8.459	0.018	+
L-Leucine/L-Isoleucine	1.295042999	-0.373	131.0939	0.953	0.023	+

Homoserine lactone	1.257013375	-0.33	101.0473	1.032	0.005	+
LysoPC (18:0)	1.237990291	-0.308	523.3603	9.958	0	+
L-Phenylalanine	1.139183377	-0.188	165.0784	1.891	0.031	+
Choline/Neurine	1.126619228	-0.172	103.0989	9.89	0.001	+
Phosphohydroxypyruvic acid	0.922103118	0.117	183.9774	17.165	0.001	+
SM(d18:2/24:0)	0.865736566	0.208	812.6725	17.134	0.007	+
Phosphoric acid	0.853817714	0.228	97.9766	1.767	0.001	+
CPA (16:0/0:0)	0.839149637	0.253	392.2306	7.904	0.021	+
D-Glucosamine/D-Galactosamine	0.777546036	0.363	179.0804	2.287	0.018	+
Urea	0.740719899	0.433	60.03199	0.989	0.048	+
Niacinamide	0.708087719	0.498	122.0472	1.135	0.011	+
Pyroglutamic acid	0.689680461	0.536	129.0422	1.218	0.007	+
Uracil	0.678302164	0.56	112.027	1.132	0.003	+
LysoPE (0:0/16:0)	0.639492791	0.645	453.2843	7.69	0.013	-
L-Carnitine	0.625031151	0.678	161.1046	0.897	0.008	+
C-2 Ceramide	0.598739352	0.74	341.2909	10.495	0.011	+
Lactosylceramide (d18:1/12:0)	0.598324482	0.741	805.5572	9.96	0.041	+
D-2-Aminobutyric acid	0.574349177	0.8	103.0629	0.954	0.004	+
Traumatic acid	0.57236208	0.805	228.1355	4.858	0.038	+
Sphinganine	0.540737382	0.887	301.2965	7.061	0.002	+
LysoPE (0:0/14:0)	0.536258308	0.899	425.2534	6.823	0.017	-
Urocortisol	0.50768305	0.978	366.2399	4.786	0.016	-
Niacin (Nicotinic acid)	0.474342158	1.076	123.0315	1.194	0.047	+
Palmitoyl-L-carnitine	0.470739232	1.087	399.3332	7.765	0	+
LysoPC (16:1(9Z))	0.470087101	1.089	493.315	7.321	0.038	+
Retinoic acid	0.44596426	1.165	300.2083	5.475	0.005	+
Sphingosine	0.442270218	1.177	299.2813	9.333	0	+
5(S)-HEPE	0.432268616	1.21	318.2189	5.479	0.004	+
C-2 Ceramide	0.38209413	1.388	341.2926	9.353	0.001	-

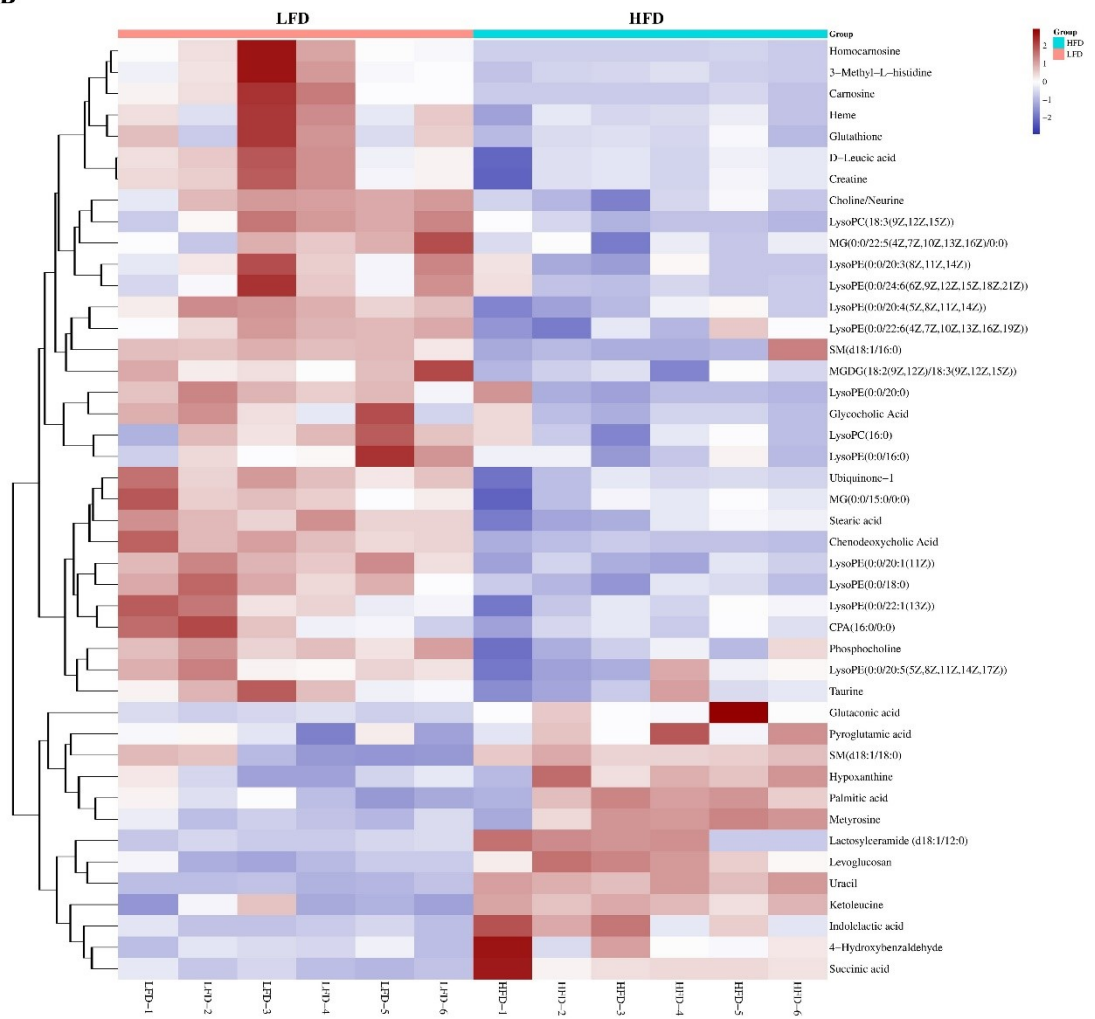
Guanine	0.349411743	1.517	152.0834	4.709	0.037	+
Cortisol	0.321078952	1.639	362.2085	4.627	0.001	+
Salicylamide	0.319968103	1.644	137.0473	3.985	0	+
Erythrose	0.306508715	1.706	120.04	3.985	0	+
Glycocholic acid	0.30333839	1.721	465.308	4.901	0.049	-
9(S)-HpODE	0.297095776	1.751	312.2295	7.486	0.002	-
LysoPE (0:0/16:1(9Z))	0.287772373	1.797	451.2691	7.111	0.016	-
Hexadecanedioic acid	0.257563488	1.957	286.2141	7.004	0.01	-
Cortisone	0.224533093	2.155	360.1932	4.632	0.012	-
Kynurenic acid	0.119244944	3.068	189.0424	3.502	0.048	+

Name, differential metabolite name; VIP, the contribution rate of different substances to the OPLS-DA Model; CompMW, the molecular weight of the compound; Time, retention time; *t*-test, significant values in *t*-test; Fold change, the value of the ratio of the mean value of high-fat diet group to low-fat diet group. ESI, electrospray ionization. “+” indicates positive ion mode; “-” indicates negative ion mode.



(to be continued)

B



(to be continued)

C

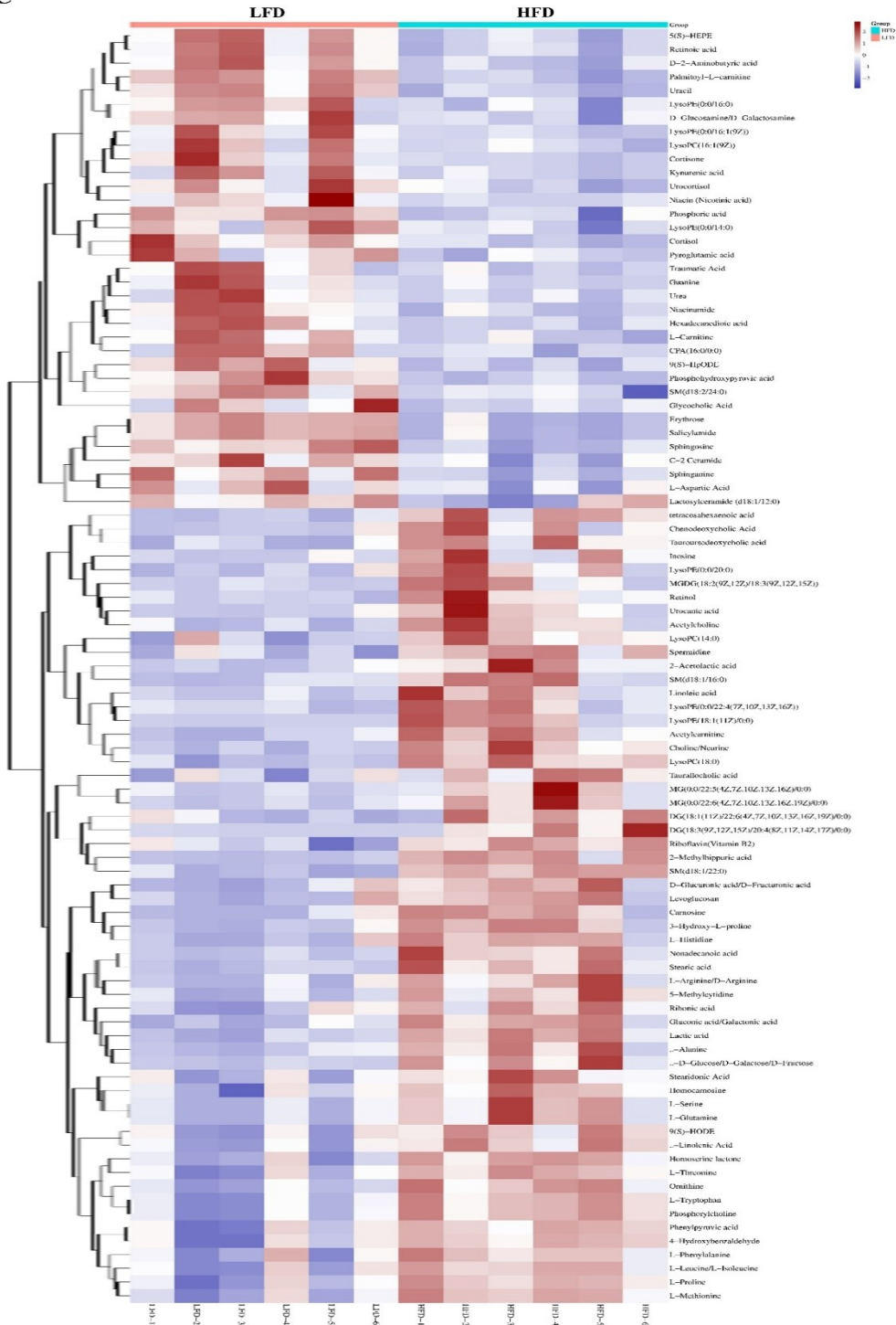


Fig. S5. The heatmap and relative abundances of the differential metabolites in serum, tumor, and liver. (A) Serum; (B) Tumor; (C) Liver. The more red colored, the higher the abundance of these metabolites, and the more blue colored, the lower the abundance of these metabolites.

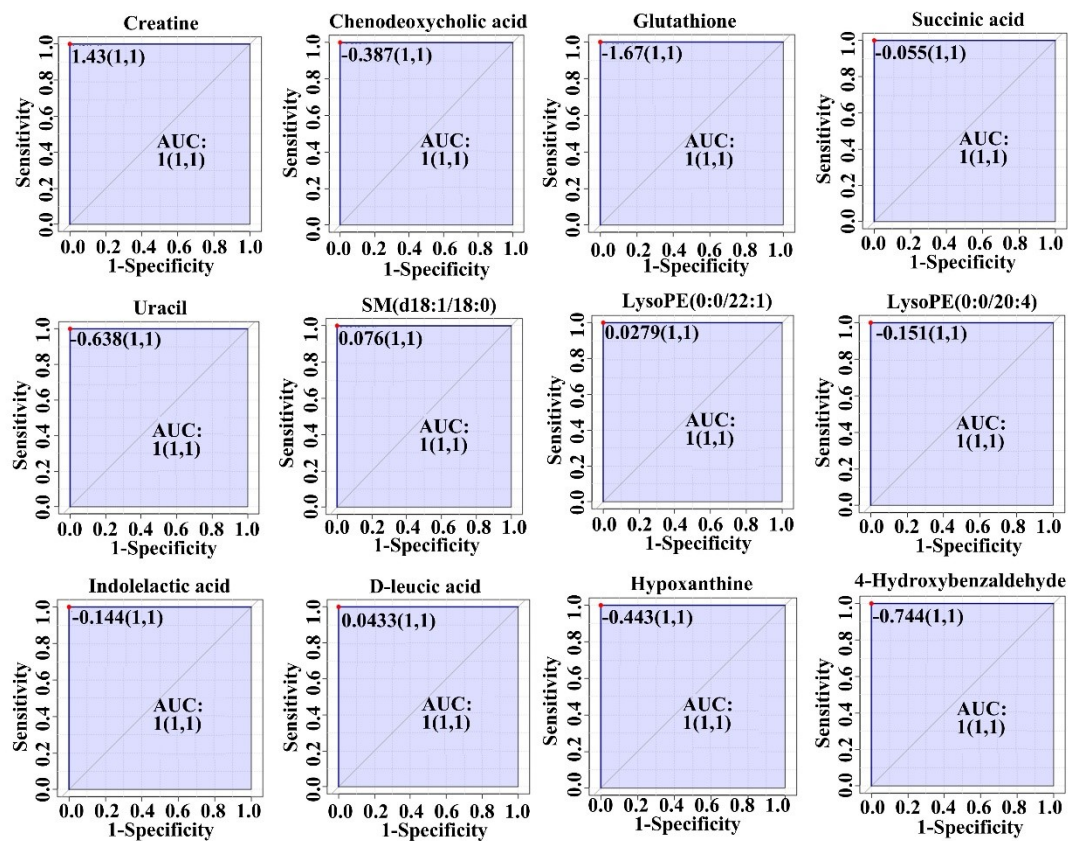


Fig. S6. Twelve potential metabolite biomarkers in tumors from HCT116 xenograft nude mice between LFD and HFD groups through univariate ROC curve analysis.