

Supporting Information

Global metabolomic profiling of trastuzumab resistant gastric cancer cells reveals major metabolic pathways and metabolic signatures based on UHPLC-Q Exactive-MS/MS

Wenhu Liu, ‡^a Qiang Wang, ‡^b and Jinxia Chang ^{*a}

^a School of Pharmacy, School of Basic Medical Sciences, North Sichuan Medical College, Nanchong 637100, China. E-mail: jinxiachang@163.com; wenhuliu@cqu.edu.cn

^b Department of Laboratory Medicine, Affiliated Hospital of North Sichuan Medical College; Faculty of Laboratory Medicine, Center for Translational Medicine, North Sichuan Medical College, Nanchong 637000, China. E-mail: wqiang_1981@126.com

Figure legends

Fig. S1 The TICs from all QC samples based on UHPLC-Q Exactive-MS/MS in POS (A) and NEG ion modes (B).

Fig. S2 Spearman's correlation coefficients were calculated to assess the experiment reproducibility. The lower-left half shows pairwise scatter plots of QC samples. The upper-right half shows pairwise Spearman's correlation coefficients for the same comparison.

Fig. S3 The TICs of standard compound (2-chloro-L-phenylalanine) based on UHPLC-Q Exactive-MS/MS in POS (A) and NEG ion modes (B).

Fig. S4 (A, B) OPLS-DA scores plot between NCI N87 and NCI N87/R cells, (C, D) MKN45 and MKN45/R cells in POS and NEG ion modes respectively.

Fig. S5 Heatmap produced by hierarchical clustering analysis of NCI N87 and NCI N87/R cells (A, B), MKN45 and MKN45/R cells (C, D) in POS and NEG ion modes respectively. The increased and decreased variables are represented by range of red and blue intensities, respectively.

Table S1. Response stability of internal standard from QCs.

Table S2. Differential metabolites shared in NCI N87, NCI N87/R and MKN45, MKN45/R cell lines.

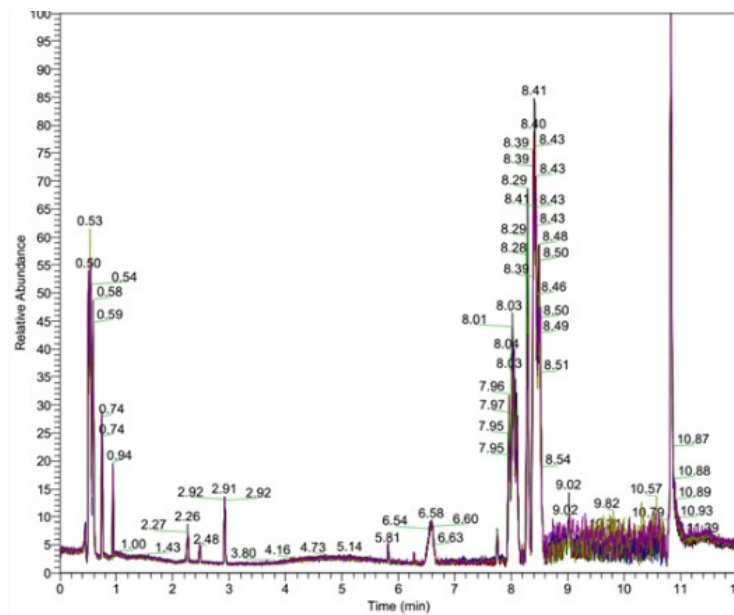
Table S3. Differential metabolites in NCI N87 and NCI N87/R cells.

Table S4. Differential metabolites in MKN45 and MKN45/R cells.

Fig. S1 The TICs from all QC samples based on UHPLC-Q Exactive-MS/MS in POS (A) and

NEG (B) ion modes.

(A)



(B)

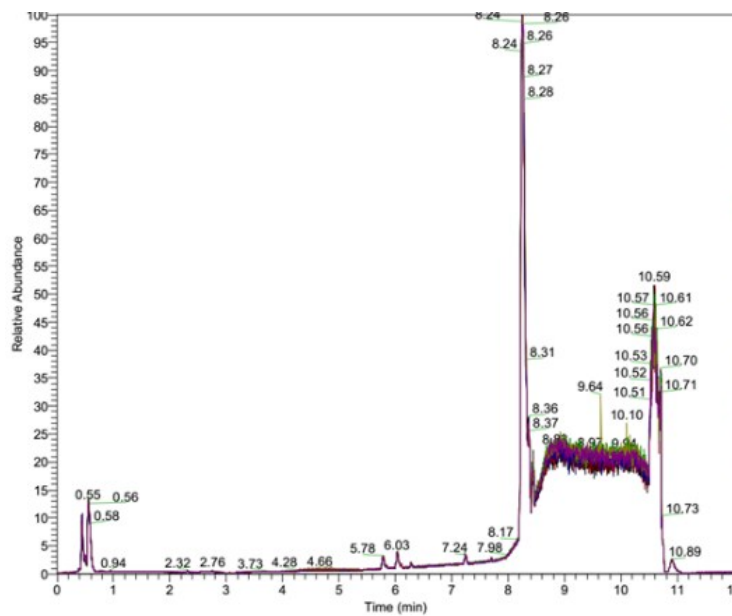


Fig. S2 Spearman's correlation coefficients from QC samples were calculated to assess the

experiment reproducibility in POS (A) and NEG (B) ion models. The lower-left half shows pairwise scatter plots of QC samples. The upper-right half shows pairwise Spearman's correlation coefficients for the same comparison.

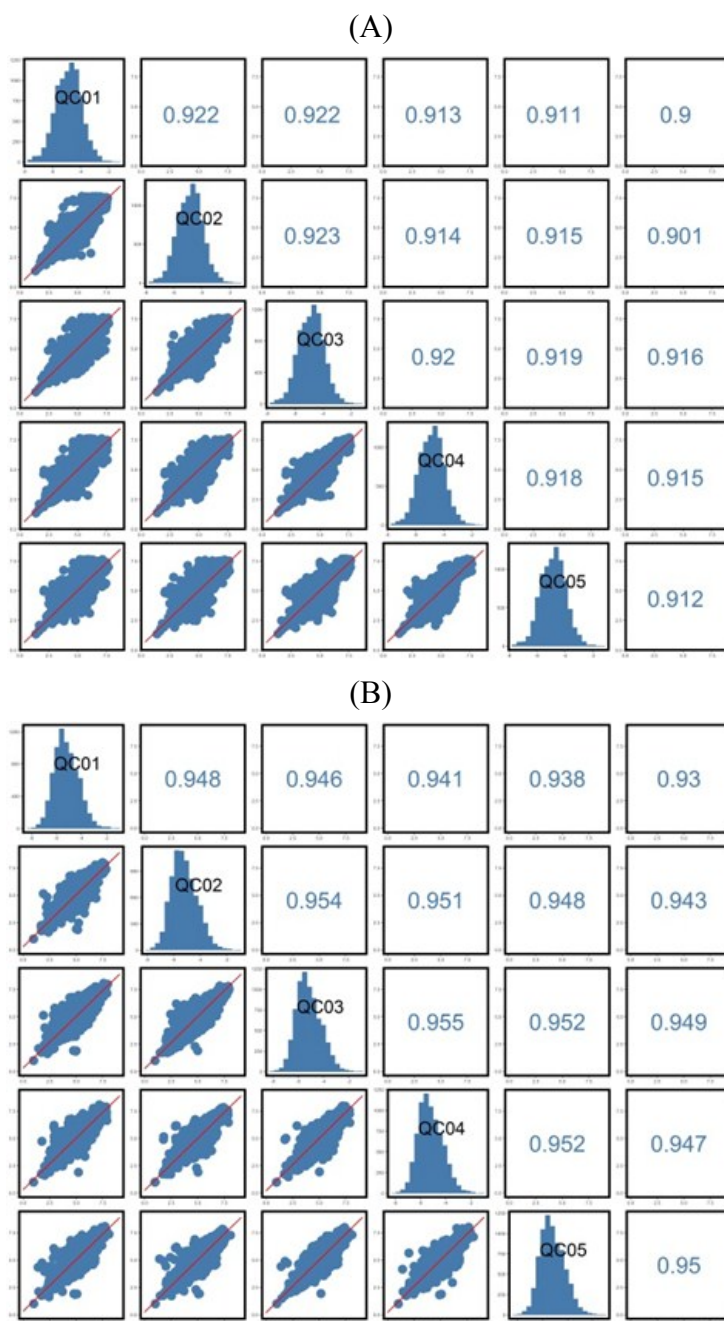
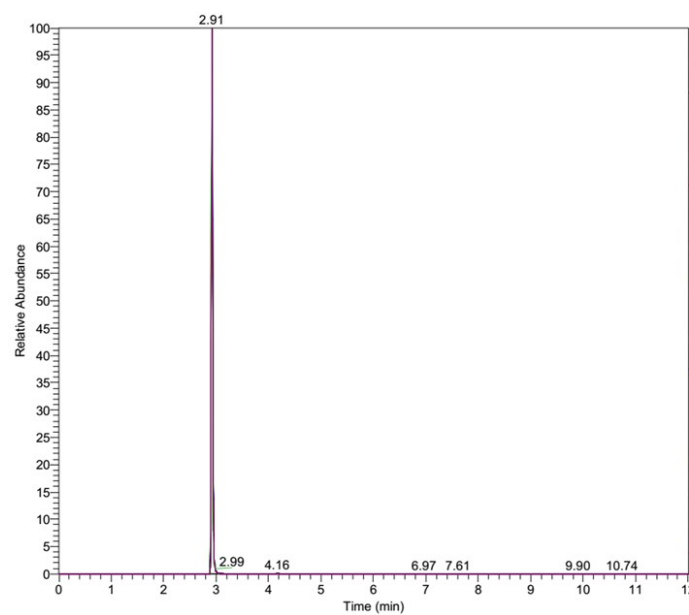


Fig. S3 The TICs of standard compound (2-chloro-L-phenylalanine) based on UHPLC-Q

Exacte-MS/MS in POS (A) and NEG(B) ion modes respectively.

(A)



(B)

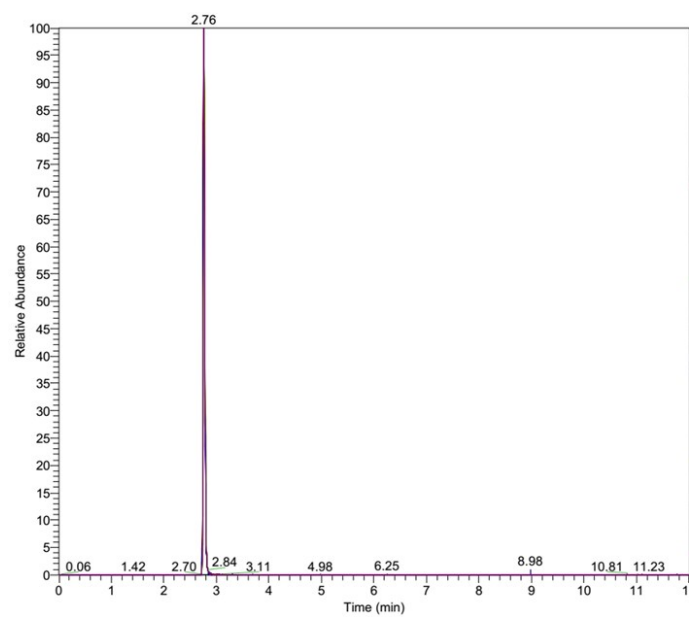


Fig. S4 OPLS-DA score plots between NCI N87 and NCI N87/R cells, MKN45 and MKN45/R

cells in POS and NEG ion modes respectively.

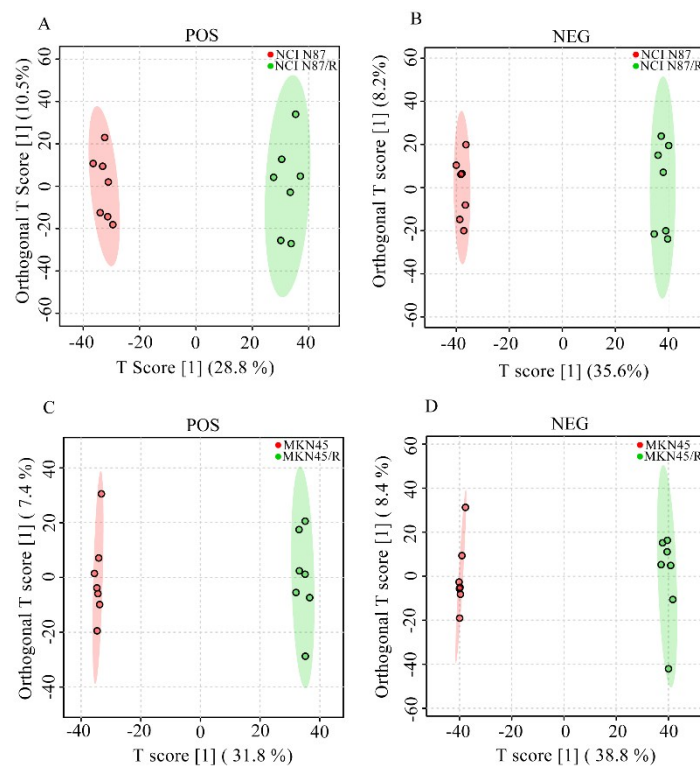


Fig. S5 Heatmap produced by hierarchical clustering analysis of NCI N87 and NCI N87/R cells

(A, B), MKN45 and MKN45/R cells (C, D) in POS and NEG ion modes respectively. The increased and decreased variables are represented by range of red and blue intensities, respectively.

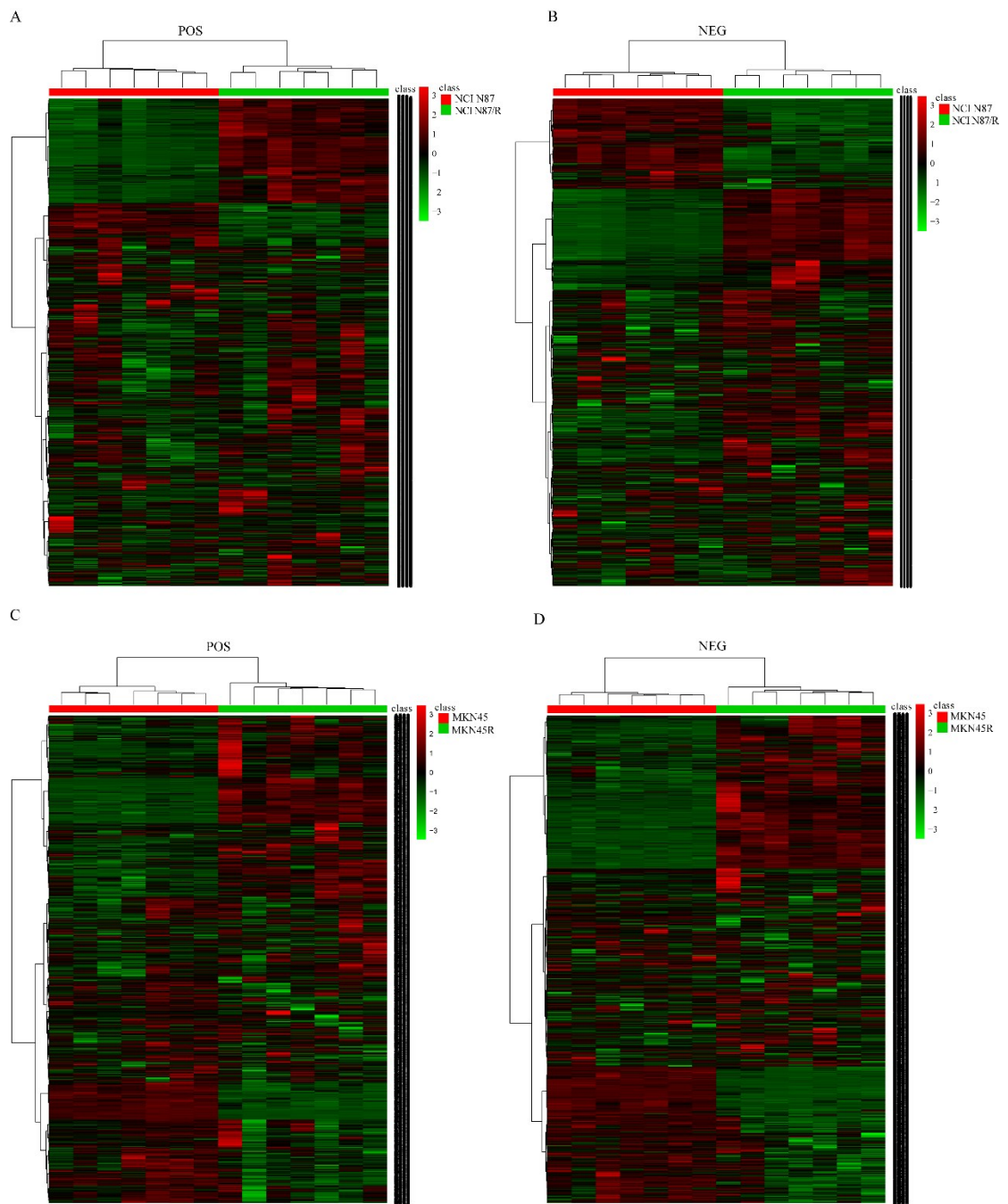


Table S1. Response stability of internal standard from QC samples.

Internal standard	Positive	Negative
-------------------	----------	----------

2-chloro-L-phenylalanine	m/z 200.0471	RT(s) 175.5295	RSD 4.53%	m/z 198.033	RT(s) 165.894	RSD 1.88%
--------------------------	-----------------	-------------------	--------------	----------------	------------------	--------------

Table S2. Differential metabolites shared in NCI N87, NCI N87/R and MKN45, MKN45/R cell lines

No	Metabolites	HMDB	KEGG	Tread (NCI N87R/NCI N87)	Tread (MKN45R/MKN45)
1	Gluconic acid	HMDB0000625	C00257	Up	Up
2	NAD	HMDB0000902	C00003	Up	Up
3	creatine	HMDB0000064	C00300	Up	Down
4	L-valine	HMDB0000883	C00183	Up	Up
5	L-threonine	HMDB0000167	C00188	Up	Up
6	pantothenic Acid	HMDB0000210	C00864	Up	Up
7	L-carnitine	HMDB0000062	C00318	Up	Down
8	fumaric acid	HMDB0000134	C00122	Up	Up
9	guanosine	HMDB0000133	C00387	Up	Up
10	pyroglutamic acid	HMDB0000267	C01879	Up	Up
11	2'-deoxyguanosine 5'-monophosphate	HMDB0001044	C00362	Up	Down
12	thiamine	HMDB0000235	C00378	Up	Up
13	S-adenosylmethionine	HMDB0001185	C00019	Up	Down
14	glyceric acid	HMDB0000139	C00258	Up	Up
15	palmitoylethanolamide	HMDB0002100	C16512	Up	Up
16	L-histidine	HMDB0000177	C00135	Up	Up
17	citrulline	HMDB0000904	C00327	Up	Up
18	L-proline	HMDB0000162	C00148	Up	Up
19	phosphoric acid	HMDB0002142	C00009	Up	Up
20	S-acetyldihydrolipoamide-E	HMDB0006878	C16255	Up	Down
21	L-tyrosine	HMDB0000158	C00082	Up	Up
22	adenosine	HMDB0000050	C00212	Up	Up
23	adenine	HMDB0000034	C00147	Up	Up
24	taurine	HMDB0000251	C00245	Up	Up
25	inosine	HMDB0000195	C00294	Up	Up
26	phosphoenolpyruvic acid	HMDB0000263	C00074	Up	Up
27	citric acid	HMDB0000094	C00158	Up	Up
28	NADP	HMDB0000217	C00006	Up	Up
29	phosphocholine	HMDB0001565	C00588	Up	Up
30	L-norleucine	HMDB0001645	C01933	Up	Down
31	malic acid	HMDB0000744	C00711	Up	Up
32	3-phosphoglyceric acid	HMDB0000807	C00597	Up	Up

33	cellobiose	HMDB0000055	C06422	Down	Down
34	L-acetylcarnitine	HMDB0000201	C02571	Up	Down
35	niacinamide	HMDB0001406	C00153	Up	Down
36	L-leucine	HMDB0000687	C00123	Up	Up
37	uridine diphosphate glucuronic acid	HMDB0000935	C00167	Up	Down
38	guanine	HMDB0000132	C00242	Up	Up
39	choline	HMDB0000097	C00114	Up	Up
40	cytidine	HMDB0000089	C00475	Up	Down
41	L-glutamine	HMDB0000641	C00064	Up	Up
42	N-acetylneuraminic acid	HMDB0000230	C19910	Up	Down
43	L-glutamic acid	HMDB0000148	C00025	Up	Up
44	uridine 5'-diphosphate	HMDB0000295	C00015	Up	Down
45	phytosphingosine	HMDB0004610	C12144	Up	Down
46	uridine 5'-monophosphate	HMDB0000288	C00105	Up	Down
47	glyceraldehyde	HMDB0001051	C02154	Up	Down
48	L-aspartic acid	HMDB0000191	C00049	Up	Up
49	eicosenoic acid	HMDB0002231	C16526	Up	Up
50	L-methionine	HMDB0000696	C00073	Up	Up
51	phosphocreatine	HMDB0001511	C02305	Up	Up
52	L-phenylalanine	HMDB0000159	C00079	Up	Up
53	uridine diphosphate-N-acetylglucosamine	HMDB0000290	C00043	Up	Down
54	octadecanamide	HMDB0034146	C13846	Down	Up

Table S3. Differential metabolites in NCI N87 and NCI N87/R cells.

No	Metabolites	HMDB	KEGG	tread (NCI N87R/NCI
----	-------------	------	------	---------------------

87)				
1	butyrylcarnitine	HMDB0002013	C02862	Up
2	xanthine	HMDB0000292	C00385	Up
3	palmitoleic acid	HMDB0003229	C08362	Down
4	5'-methylthioadenosine	HMDB0001173	C00170	Up
5	nicotinamide	HMDB0001406	C00153	Up
6	cysteinylglycine	HMDB0000078	C01419	Up
7	sphinganine 1-phosphate	HMDB0001383	C01120	Up
8	lysoPE(18:0)	HMDB0011129	-	Up
9	acetylcholine	HMDB0000895	C01996	Up
10	N-acetyl-L-aspartic acid	HMDB0000812	C01042	Up
11	uridine diphosphate galactose	HMDB0000302	C00052	Up
12	pantothenic acid	HMDB0000210	C00864	Up
13	uracil	HMDB0000300	C00106	Up
14	guanosine monophosphate	HMDB0001397	C00144	Up
15	glutathione	HMDB0000125	C00051	Up
16	cytidine monophosphate N-acetylneuraminic acid	HMDB0001176	C00128	Up
17	adenosine 5'-monophosphate	HMDB0000045	C00020	Up
18	L-alanine	HMDB0000161	C00041	Down
19	stearic acid	HMDB0000827	C01530	Up
20	epicatechin	HMDB0001871	C09727	Up
21	propionylcarnitine	HMDB0000824	C03017	Up
22	mannitol	HMDB0000765	C00392	Up
23	cytidine monophosphate	HMDB0000095	C00055	Up
24	ADP	HMDB0001341	C00008	Up
25	citicoline	HMDB0001413	C00307	Up

Table S4. Differential metabolites in MKN45 and MKN45/R cells.

No	Metabolites	HMDB	KEGG	tread
----	-------------	------	------	-------

(MKN45R/MKN45)				
1	eicosapentaenoic Acid	HMDB0001999	C06428	Up
2	arachidonic acid	HMDB0001043	C00219	Up
3	betaine aldehyde	HMDB0001252	C00576	Up
4	D-erythrose 4-phosphate	HMDB0001321	C00279	Up
5	sphinganine	HMDB0000269	C00836	Down
6	cholesterol sulfate	HMDB0000653	C18043	Down
7	glycerophosphocholine	HMDB0000086	C00670	Down
8	adenosine monophosphate	HMDB0000045	C00020	Down
9	N-acetyl-glucosamine 1-phosphate	HMDB0001367	C04256	Down
10	mannose 6-phosphate	HMDB0001078	C00275	Up
11	tetradecanoylcarnitine	HMDB0005066	-	Up
12	lysoPC(16:0)	HMDB0010382	C04230	Up
13	L-lysine	HMDB0000182	C00047	Up
14	2-methylbutyrylcarnitine	HMDB0000378	-	Down
15	pyruvic acid	HMDB0000243	C00022	Down
16	sanguinarine	HMDB0029367	C06162	Down
17	allopurinol	HMDB0014581	-	Up
18	L-tryptophan	HMDB0000929	C00078	Up
19	dihydroxyacetone phosphate	HMDB0001473	C00111	Down
20	lysoPE(16:0/0:0)	HMDB0011503	-	Up
21	nicotinic acid	HMDB0001488	C00253	Up