Lomatogonium Rotatum for Treatment of Acute Liver Injury in Mice: A Metabolomics Study

Renhao Chen [1], Qi Wang [2], Lanjun Zhao [1], Shinlin Yang [1], Zhifeng Li [1,*], Yulin Feng [2], Jiaqing Chen [3], Choon Nam Ong [4] and Hui Zhang [5,*]

[1] National Pharmaceutical Engineering Center for Solid Preparation in Chinese Herb Medicine, Jiangxi University of Traditional Chinese Medicine, Nanchang 330002, China
[2] State Key Laboratory of Innovative Drug and Efficient Energy-Saving Pharmaceutical Equipment, Nanchang 330006, China
[3] NUS Graduate School for Integrative Sciences and Engineering, National University of Singapore, 119077, Singapore
[4] Saw Swee Hock School of Public Health, National University of Singapore, 117549, Singapore
[5] NUS Environmental Research Institute, National University of Singapore, 117411, Singapore
* Correspondence: Hui Zhang, zhanghui@u.nus.edu; Zhifeng Li, lizhifeng1976@hotmail.com

Figure Captions:

Figure S1. Total ion chromatograms of QC samples and selected top 12 peaks, A: GC-MS, B: LC-MS (ESI+), C: LC-MS (ESI-).

Figure S2. Plots of PCA and OPLS-DA. A: PCA plot of GC-MS data (R^2X: 0.774; Q^2: 0.603); B: OPLS-DA plot of GC-MS data (R^2X: 0.717; R^2Y: 0.948; Q^2: 0.631); C: PCA plot of LC-MS data (R^2X: 0.62; Q^2: 0.53); D: OPLS-DA plot of LC-MS data (R^2X: 0.737; R^2Y: 0.878; Q^2: 0.815).

Figure S3. MS/MS spectra (ESI+ & ESI-) of identified metabolites and the comparison with major fragments of metabolites in HMDB database.

Figure S4. Result of metabolic pathway analysis through MetPA software.

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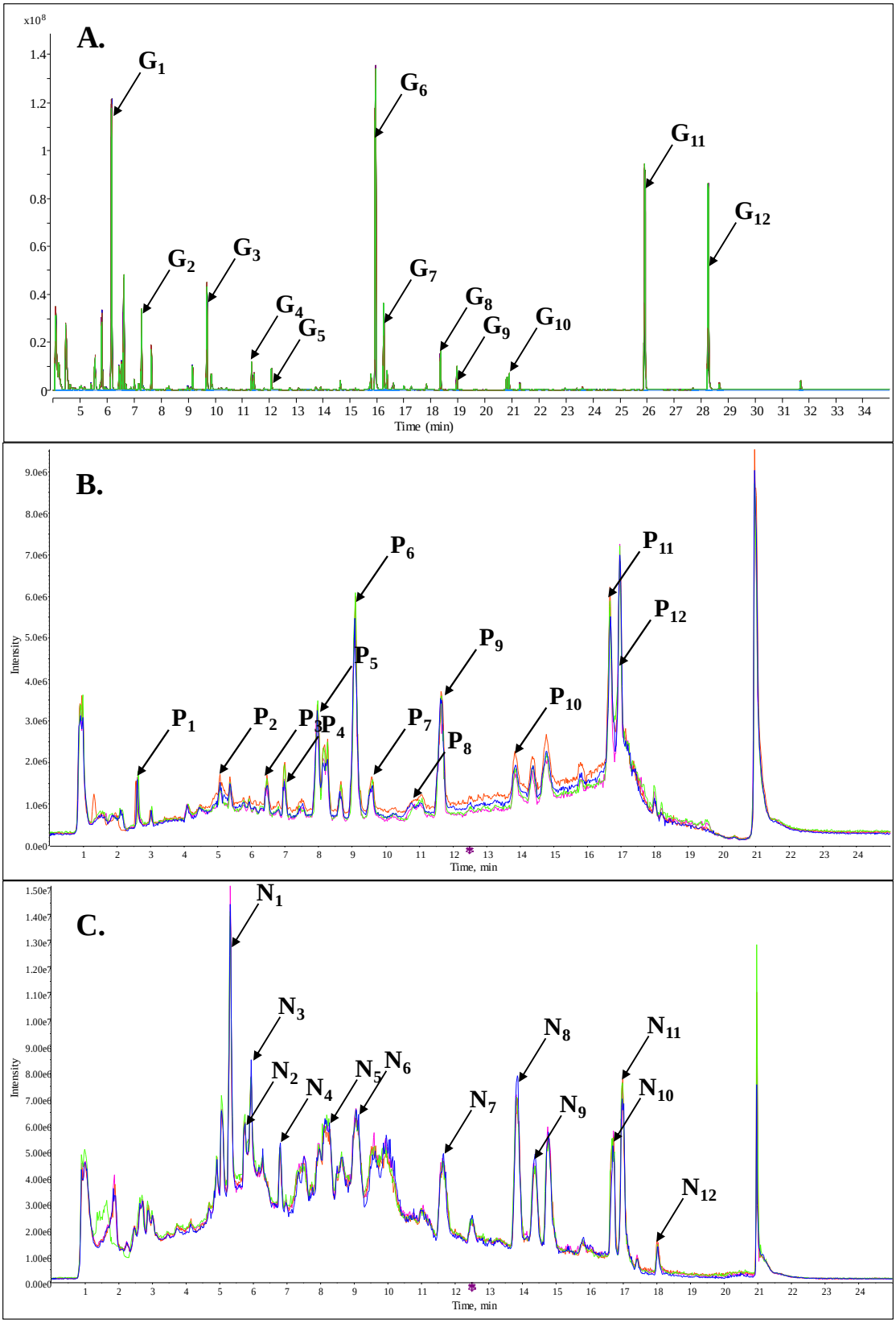

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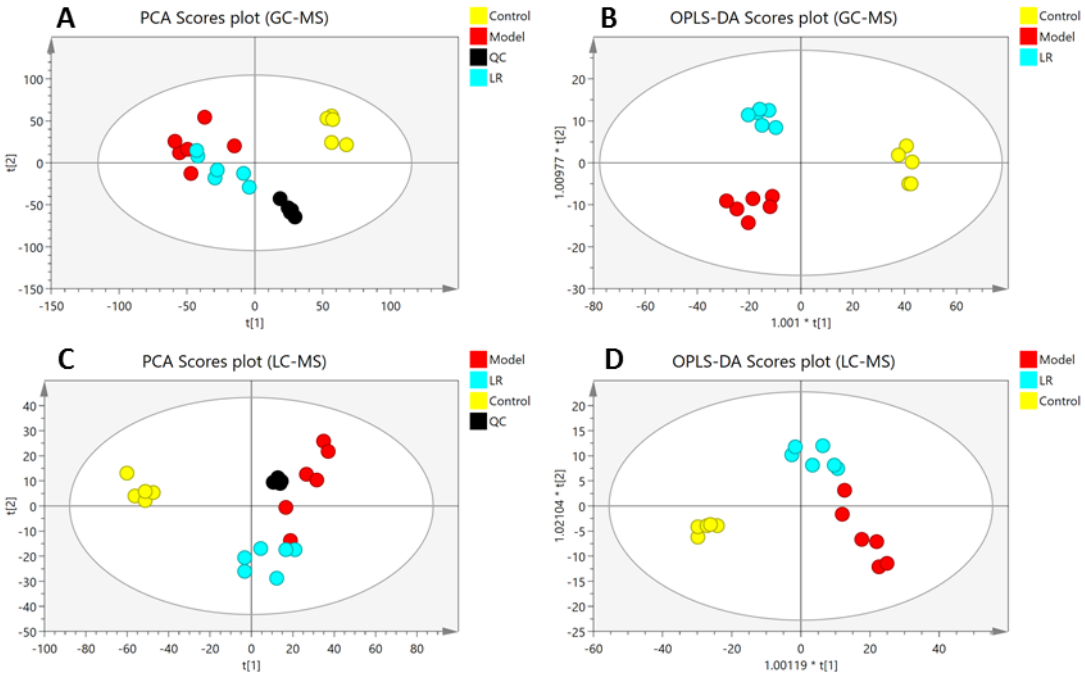

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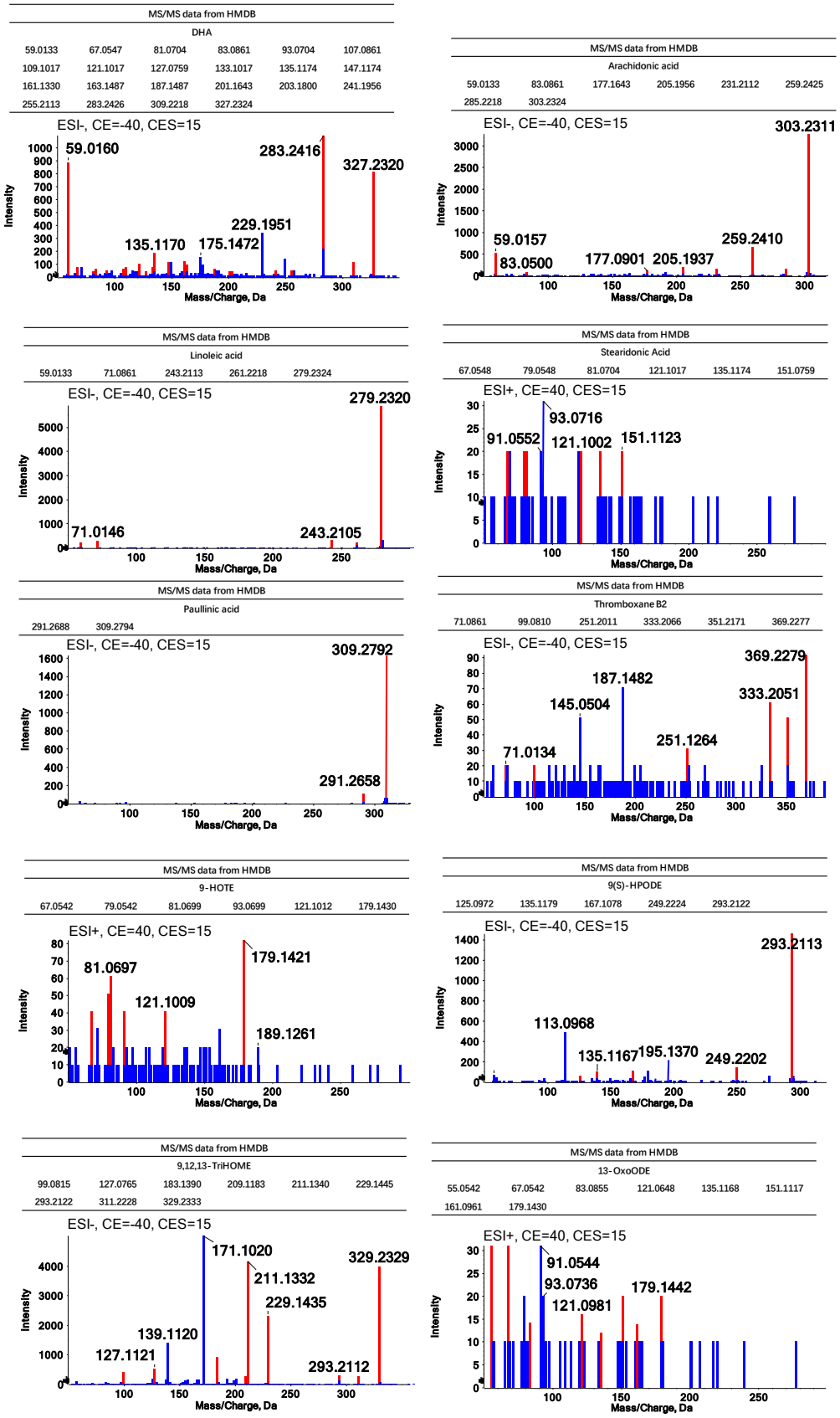

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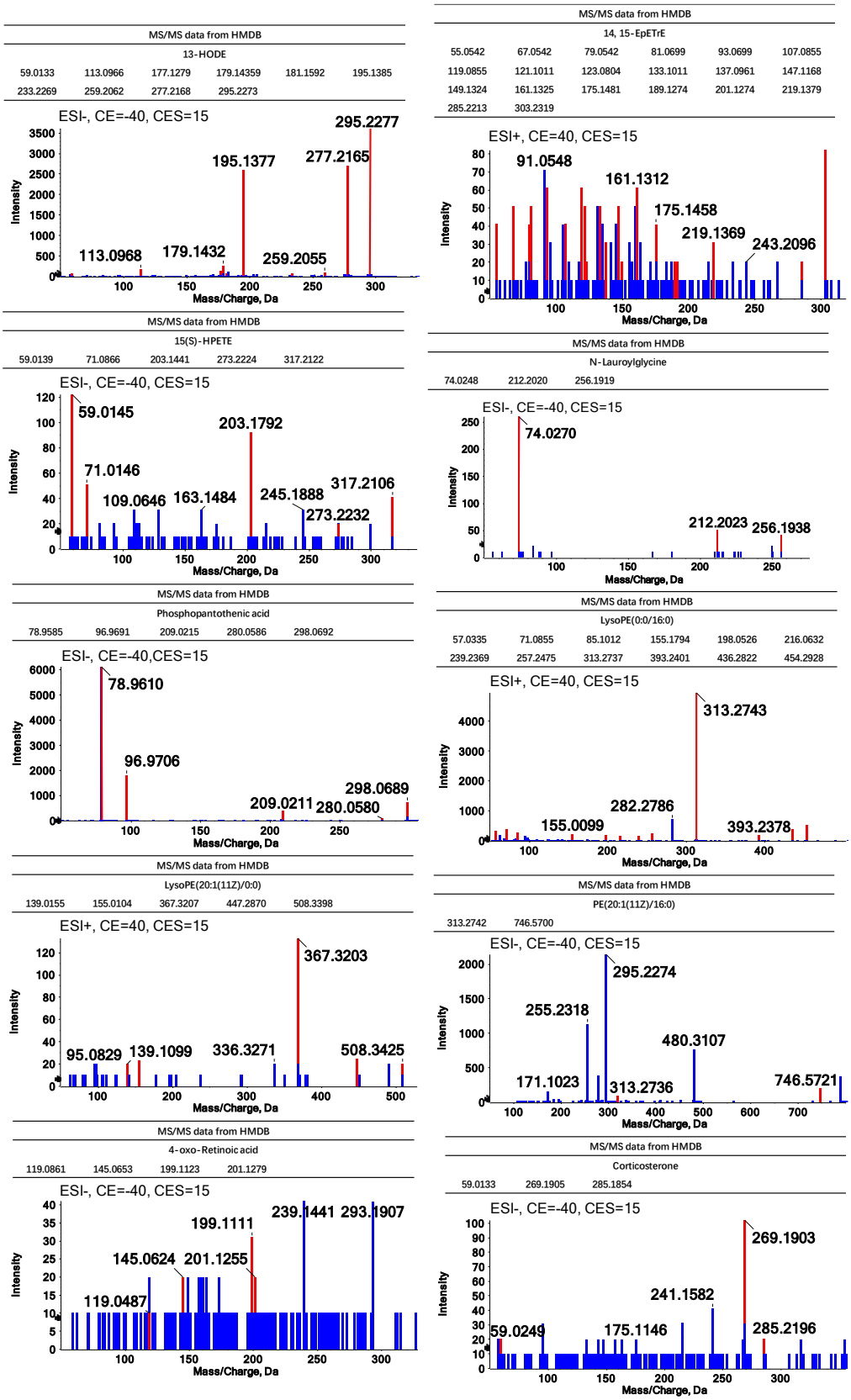
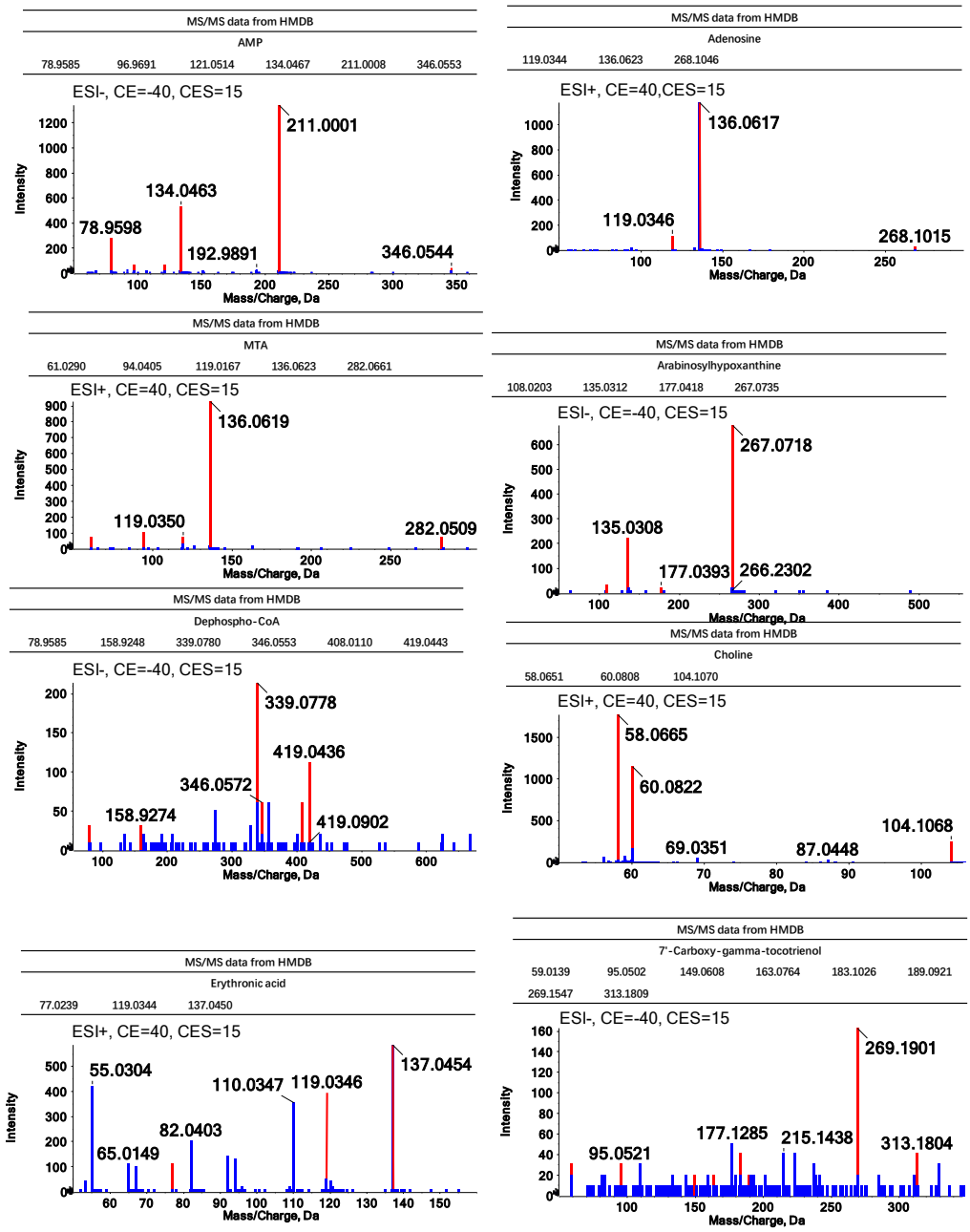


Figure S3. Continued.



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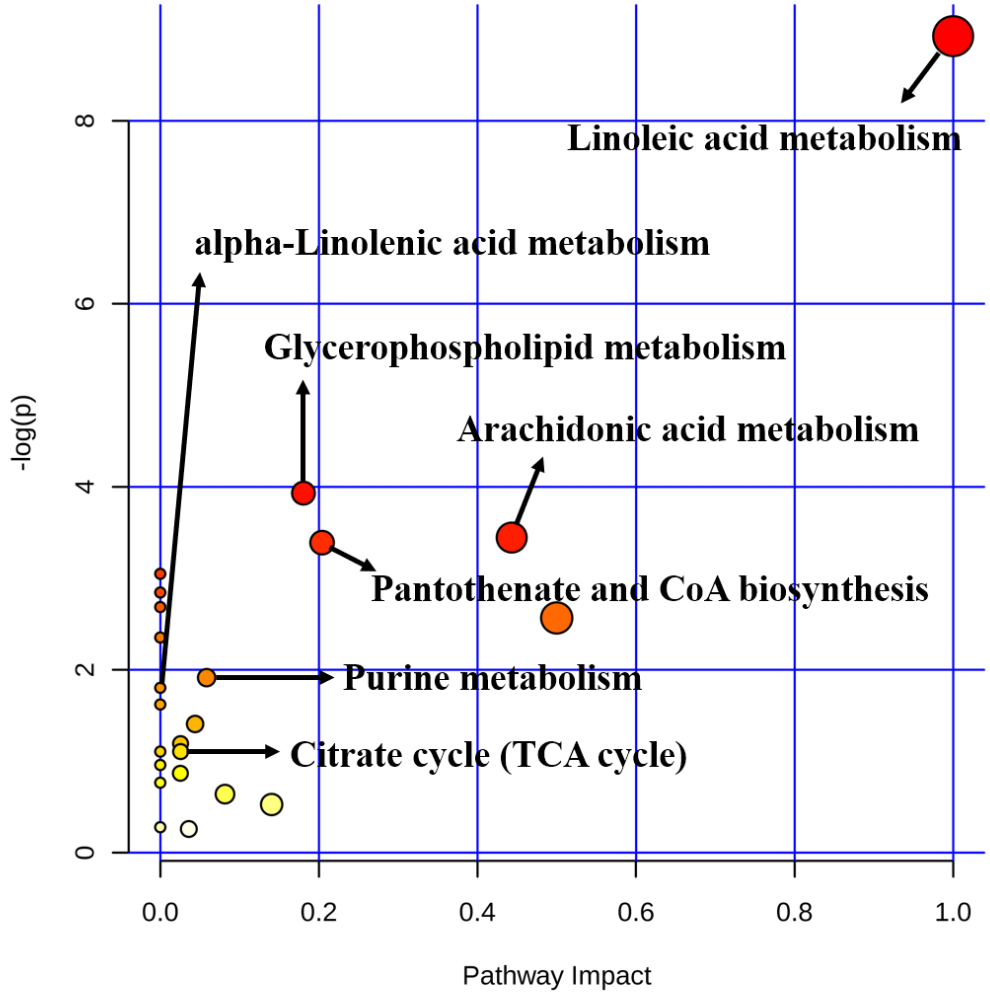

Figure S4. Result of metabolic pathway analysis through MetPA software.

Table S1. The peak areas and retention times of top 12 peaks in QC samples (GC-MS).

NO.	Peak area		Retention time	
	Mean ± SD (× 10^7)	**RSD%**	**Mean ± SD**	**RSD%**
G1	26.75 ± 0.69	2.60	6.17 ± 0.00	0.06
G2	5.04 ± 0.17	3.29	7.29 ± 0.00	0.04
G3	10.08 ± 0.27	2.68	9.70 ± 0.00	0.03
G4	1.86 ± 0.11	5.94	11.35 ± 0.00	0.02
G5	1.49 ± 0.10	6.55	12.10 ± 0.00	0.02
G6	37.69 ± 0.31	0.83	15.95 ± 0.00	0.03
G7	7.59 ± 0.15	1.97	16.25 ± 0.00	0.02
G8	3.01 ± 0.07	2.47	18.35 ± 0.00	0.02
G9	2.08 ± 0.07	3.29	18.97 ± 0.00	0.02
G10	1.34 ± 0.07	4.97	20.89 ± 0.00	0.01
G11	23.73 ± 0.49	2.08	25.93 ± 0.00	0.01
G12	22.88 ± 0.46	2.00	28.28 ± 0.00	0.01

Table S2. The peak areas and retention times of top 12 peaks in QC samples (LC-MS).

NO.	LC-MS [a]	M/Z	Intensity		Retention time	
			Mean ± SD (× 10^4)	RSD%	Mean ± SD	RSD%
P1	ESI+	166.09	43.10 ± 3.50	8.12	2.60 ± 0.03	0.96
P2	ESI+	287.06	26.11 ± 2.03	7.76	5.07 ± 0.00	0.10
P3	ESI+	274.27	46.72 ± 4.18	8.94	6.44 ± 0.00	0.08
P4	ESI+	432.24	28.85 ± 3.35	11.63	6.98 ± 0.01	0.14
P5	ESI+	520.34	62.24 ± 9.44	15.17	7.98 ± 0.01	0.10
P6	ESI+	496.34	229.53 ± 18.02	7.85	9.09 ± 0.01	0.06
P7	ESI+	522.35	29.46 ± 3.94	13.36	9.60 ± 0.01	0.12
P8	ESI+	601.33	7.14 ± 0.62	8.72	10.83 ± 0.05	0.47
P9	ESI+	524.37	117.52 ± 6.16	5.24	11.67 ± 0.01	0.08
P10	ESI+	329.25	17.12 ± 2.80	16.35	13.85 ± 0.01	0.04
P11	ESI+	760.58	56.53 ± 5.12	9.06	16.68 ± 0.01	0.03
P12	ESI+	758.57	111.25 ± 11.75	10.56	16.96 ± 0.01	0.03
N1	ESI-	285.04	113.50 ± 6.33	5.58	5.07 ± 0.00	0
N2	ESI-	514.28	561.74 ± 39.28	6.99	5.32 ± 0.01	0.09
N3	ESI-	453.29	102.40 ± 3.97	3.87	5.93 ± 0.01	0.08
N4	ESI-	471.06	61.13 ± 3.88	6.36	6.79 ± 0.00	0
N5	ESI-	564.33	93.70 ± 5.80	6.19	8.28 ± 0.01	0.06
N6	ESI-	540.33	155.90 ± 11.07	7.10	9.11 ± 0.01	0.06
N7	ESI-	480.31	143.84 ± 4.98	3.46	11.59 ± 0.01	0.05
N8	ESI-	327.23	282.50 ± 20.99	7.43	13.84 ± 0.01	0.04
N9	ESI-	303.23	159.25 ± 11.68	7.33	14.37 ± 0.01	0.04
N10	ESI-	255.23	69.30 ± 8.03	11.58	16.68 ± 0.01	0.03
N11	ESI-	281.25	133.64 ± 4.79	3.58	16.99 ± 0.00	0.03
N12	ESI-	283.26	62.78 ± 4.22	6.72	18.01 ± 0.00	0.03

[a] ESI+: ESI positive mode; ESI-: ESI negative mode.

Table S3. Identified metabolites and their relative levels in the control, model and LR groups.

Class	Metabolites	HMDB	Model/Control [a]	LR/Control [b]	LR/Model [c]	Analytical Instrument [d]
Fatty acids	Docosahexaenoic acid (DHA)	HMDB0002183	↓***	↓	↑***	LC-MS (ESI-)
Fatty acids	Arachidonic acid	HMDB0001043	↓***	↓***	↑**	LC-MS (ESI-)
Fatty acids	Linoleic acid	HMDB0000673	↑***	↑***	↓***	LC-MS (ESI-)
Fatty acids	Stearidonic Acid	HMDB0006547	↑***	↑**	↓	LC-MS (ESI+)
Fatty acids	Paullinic acid	HMDB0035159	↑***	↑***	↓	LC-MS (ESI-)
Fatty acids	Thromboxane B2	HMDB0003252	↑***	↑***	↓	LC-MS (ESI-)
Fatty acids	9-HOTE	HMDB0010224	↑***	↑**	↓	LC-MS (ESI+)
Fatty acids	9(S)-HPODE	HMDB0006940	↑***	↑**	↓	LC-MS (ESI-)
Fatty acids	9,12,13-TriHOME	HMDB0004708	↑**	↑*	↓	LC-MS (ESI-)
Fatty acids	13-OxoODE	HMDB0004668	↑**	↑**	↓	LC-MS (ESI+)
Fatty acids	13-HODE	HMDB0004667	↑***	↑***	↓	LC-MS (ESI-)
Fatty acids	14,15-EpETrE	HMDB0004264	↑**	↑***	↓	LC-MS (ESI+)
Fatty acids	15(S)-HPETE	HMDB0004244	↑***	↑**	↓	LC-MS (ESI-)
Amino acids	L-Tyrosine	HMDB0000158	↓*	↓	↑	GC-MS
Amino acids	N-Lauroylglycine	HMDB0013272	↓***	↓**	↑	LC-MS (ESI-)

Amino acids	Phosphopantothenic acid	HMDB0001016	↑***	↑***	↓***	LC-MS (ESI-)
Lipids	Glycerol 3-phosphate	HMDB0000126	↓**	↓	↑*	GC-MS
Lipids	LysoPE(0:0/16:0)	HMDB0011473	↑***	↑***	↓	LC-MS (ESI+)
Lipids	LysoPE(20:1(11Z)/0:0)	HMDB0011512	↑**	↑***	↓	LC-MS (ESI+)
Lipids	PE(20:1(11Z)/16:0)	HMDB0009253	↑***	↑***	↓	LC-MS (ESI-)
Lipids	4-oxo-Retinoic acid	HMDB0006285	↑***	↑***	↓	LC-MS (ESI-)
Lipids	Corticosterone	HMDB0001547	↑***	↑**	↓	LC-MS (ESI-)
Nucleosides	Adenosine monophosphate (AMP)	HMDB0000045	↓**	↓**	↑*	LC-MS (ESI-)
Nucleosides	Adenosine	HMDB0000050	↓*	↓*	↑	LC-MS (ESI+)
Nucleosides	Inosine	HMDB0000195	↓**	↓**	↑	LC-MS (ESI+)
Nucleosides	5'-Methylthioadenosine (MTA)	HMDB0001173	↓***	↓***	↑	LC-MS (ESI+)
Nucleosides	Arabinosylhypoxanthine	HMDB0003040	↓***	↓***	↑	LC-MS (ESI-)
Nucleosides	Dephospho-CoA	HMDB0001373	↓***	↓***	↑	LC-MS (ESI-)
Others	Succinic acid	HMDB0000254	↑	↑	↓*	GC-MS
Others	Choline	HMDB0000097	↑***	↑*	↓**	LC-MS (ESI+)
Others	Erythronic acid	HMDB0000613	↓*	↓***	↑	LC-MS (ESI+)
Others	3-Hydroxybutyric acid	HMDB0000357	↓***	↓**	↓	GC-MS
Others	N-Acetylmannosamine	HMDB0001129	↑***	↑**	↓	GC-MS
Others	7'-Carboxy-gamma-tocotrienol	HMDB0012851	↑***	↑**	↓	LC-MS (ESI-)

[a] Comparison of relative levels of metabolites in the model group and the control group.

[b] Comparison of relative levels of metabolites in the LR group and the control group.

[c] Comparison of relative levels of metabolites in the LR group and the model group.

[d] ESI+: ESI positive mode; ESI-: ESI negative mode.

The hypothesis test for the difference between two means was conducted through t-test. *: $p < 0.05$; **: $p < 0.01$; ***: $p < 0.001$.