Supplementary data

Identification of potential diagnostic biomarkers of cerebral infarction by using gas chromatography-mass spectrometry and chemometrics

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Table S1. Baseline characteristics of CI patients and healthy control. Values are described as mean ± SD, or %.

characteristics	Age (year)	Male gender (%)	TG (mmol/L)	TC (mmol/L)	LDL (mmol/L)	HDL (mmol/L)
Patients (n=33)	50.38 ± 8.72	50%	2.72 ± 0.50	4.28 ± 1.56	1.86 ± 1.26	1.87 ± 0.03
healthy control (n=40)	45.43 ± 6.57	50%	1.22 ± 0.41	3.89 ± 1.35	2.82 ± 0.13	1.28 ± 0.12

TG: Triglyceride; TC: Total Cholesterol; LDL: Low density lipoprotein; HDL: High density lipoprotein.

Table S2. Precision test of 20 reference standards (RT, retention time, n=6).

Peak No.	Metabolites	Precision test		
		RT (min)	Content (%)	RSD(%)
1	Methyl hexanoate (C6:0)	2.11±0.0043	0.392±0.0222	5.66
2	Methyl octanoate (C8:0)	2.66±0.0032	0.601±0.0164	2.72
3	Methyl decanoate (C10:0)	3.29±0.0018	0.811±0.0458	5.65
4	Methyl undecanoate (C11:0)	3.61±0.0021	0.525±0.0269	5.13
5	Methyl dodecanoate (C12:0)	3.95±0.0037	1.081±0.0486	4.49
6	Methyl tridecanoate (C13:0)	4.30±0.0011	0.697±0.0202	2.90
7	Methyl myristate (c14:0)	4.68±0.0054	1.404±0.0449	3.20
8	Methyl myristoleate (C14:1)	4.90±0.0019	0.776±0.0191	2.46
9	Methyl pentadecanoate (C15:0)	5.10±0.0036	0.874±0.0162	1.85
10	Methyl cis-10-pentadecenoate (C15:1)	5.37±0.0027	0.860±0.0148	1.73
11	Methyl palmitate (C16:0)	5.61±0.0023	2.346±0.0548	2.33
12	Methyl palmitoleate (C16:1)	5.86±0.0032	0.933±0.0175	1.87
13	Methyl heptadecanoate (C17:0)	6.22±0.0034	0.680±0.0039	0.58
14	Methyl cis-10-heptadecenoate (C17:1)	6.55±0.0033	0.968±0.0127	1.31
15	Methyl stearate (C18:0)	7.00±0.0051	1.794±0.0316	1.76
16	Methyl linolelaidate (C18:2n6t)	7.58±0.0049	0.940±0.0168	1.79
17	Methyl linoleate (C18:2n6c)	7.89±0.0059	0.962±0.0185	1.92
18	Methyl γ-linolenate (C18:3n3)	8.31±0.0045	0.889±0.0150	1.68
19	Methyl linolenate (C18:3n6)	8.66±0.0052	0.879±0.0172	1.96
20	cis-4,7,10,13,16,19-Docosahexaenoic acid methyl ester (C22:6ns)	13.98±0.0042	0.770±0.0293	3.81

Table S3. Stability and reproducibility tests of 14 investigated fatty acid (RT, retention time, n=6)

Peak No.	Metabolites	Stability test			Reproducibility test		
		RT(min)	Content (%)	RSD(%)	RT(min)	Content (%)	RSD(%)
1	Myristic acid	4.73±0.0046	0.013±0.0012	8.96	4.73±0.0016	0.013±0.0006	4.33
2	Pentadecanoic acid	5.16±0.0031	0.001±0.0008	5.73	5.16±0.0017	0.001±0.0001	5.73
3	Palmitic acid	5.72±0.0094	0.582±0.0229	3.93	5.72±0.0048	0.536±0.0119	2.22
4	Palmitoleic acid	5.94±0.0068	0.045±0.0038	8.52	5.94±0.0024	0.043±0.0027	6.14
5	Stearic acid	6.40±0.0136	0.218±0.0076	3.50	6.40±0.0081	0.224±0.0020	0.89
6	9-Octadecenoic acid	7.20±0.0074	0.789±0.0372	4.71	7.21±0.0084	0.719±0.0112	1.56
7	Linoleic acid	7.49±0.0096	0.657±0.0329	5.01	7.49±0.0053	0.599±0.0075	1.26
8	6,9,12-Octadecatrienoic acid	8.05±0.0054	0.005±0.0004	8.12	8.06±0.0075	0.0046±0.0003	5.95
9	9,12,15-Octadecatrienoic acid	8.44±0.0055	0.032±0.0019	5.97	8.42±0.0093	0.028±0.0008	2.79
10	8,11,14-Eicosatrienoic acid	8.79±0.0039	0.019±0.0012	6.34	8.79±0.0066	0.020±0.0006	3.10
11	Arachidonic acid	10.63±0.0089	0.153±0.0066	4.33	10.63±0.0078	0.139±0.0010	0.70
12	Eicosapentaenoic acid	10.96±0.0098	0.009±0.0007	8.55	10.95±0.0029	0.008±0.0004	4.81
13	Docosapentaenoic acid	11.86±0.0074	0.005±0.0004	7.78	11.86±0.0088	0.008±0.0004	5.26
14	Docosahexaenoic acid	13.87±0.0038	0.060±0.0038	6.33	13.86±0.0120	0.051±0.0014	2.80

Table S4. Result from key Metabolic Pathway Analysis with MetaboAnalyst 3.0

No.	Pathway name	total cmpd	hits	raw p	-log (p)	impact
1	Fatty acid biosynthesis	49	4	5.0802E-6	12.19	0.0
2	Linoleic acid metabolism	15	1	0.042868	3.1496	0.0
3	Fatty acid elongation in mitochondria	27	1	0.076019	2.5768	0.0
4	Fatty acid metabolism	50	1	0.13681	1.9891	0.02959
5	Arachidonic acid metabolism	62	1	0.16715	1.7889	0.21669

Figure S1. The typical total ion chromatograms (TICs) of serum samples in nontargeted global metabolites' study from healthy control (the black picks), cerebral infarction (the red picks) and quality control (the blue picks).
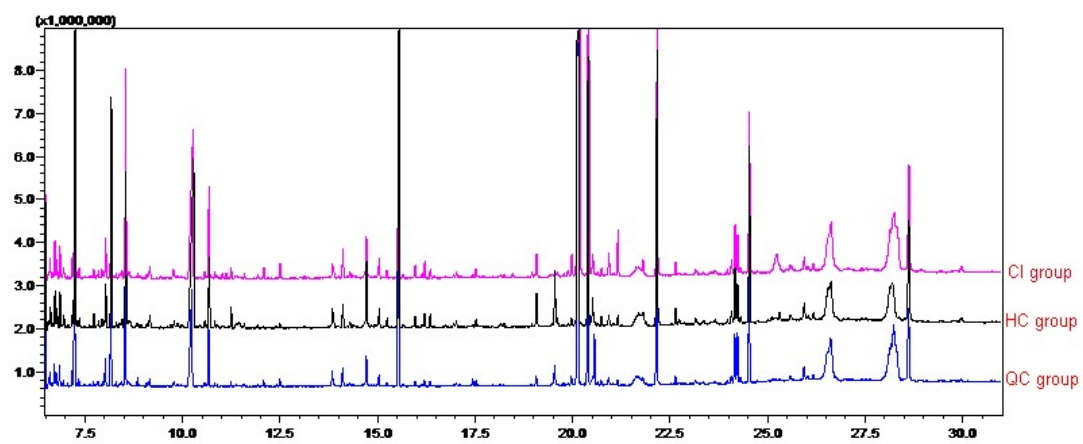
Figure S2. The variable importance measure of serum samples in nontargeted global metabolites' study from healthy participants, CI patients and QC samples obtained by random forest (top 15 metabolites).
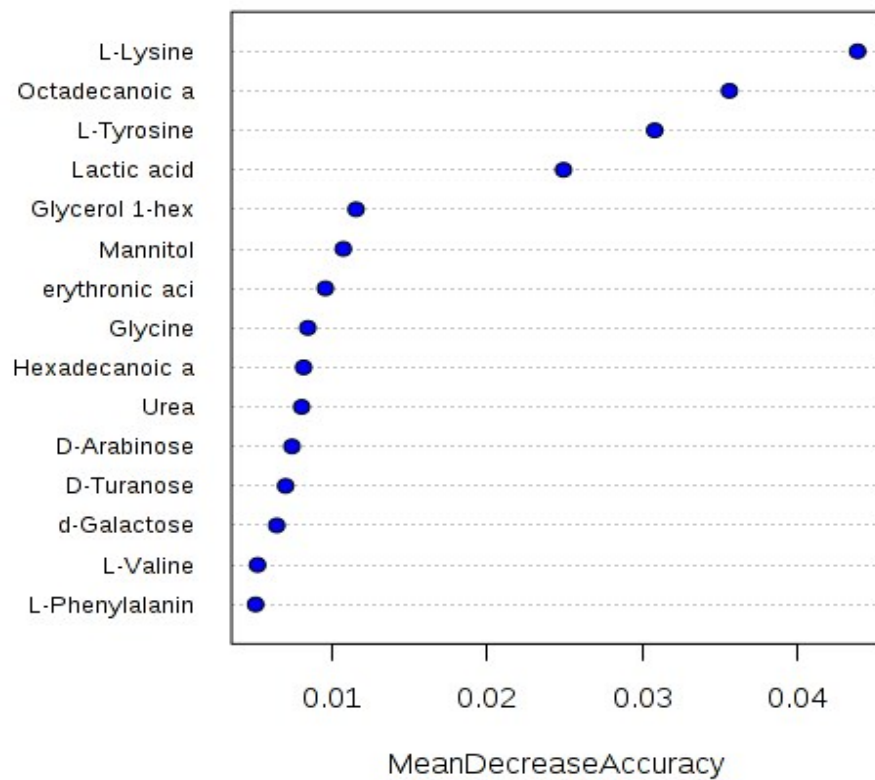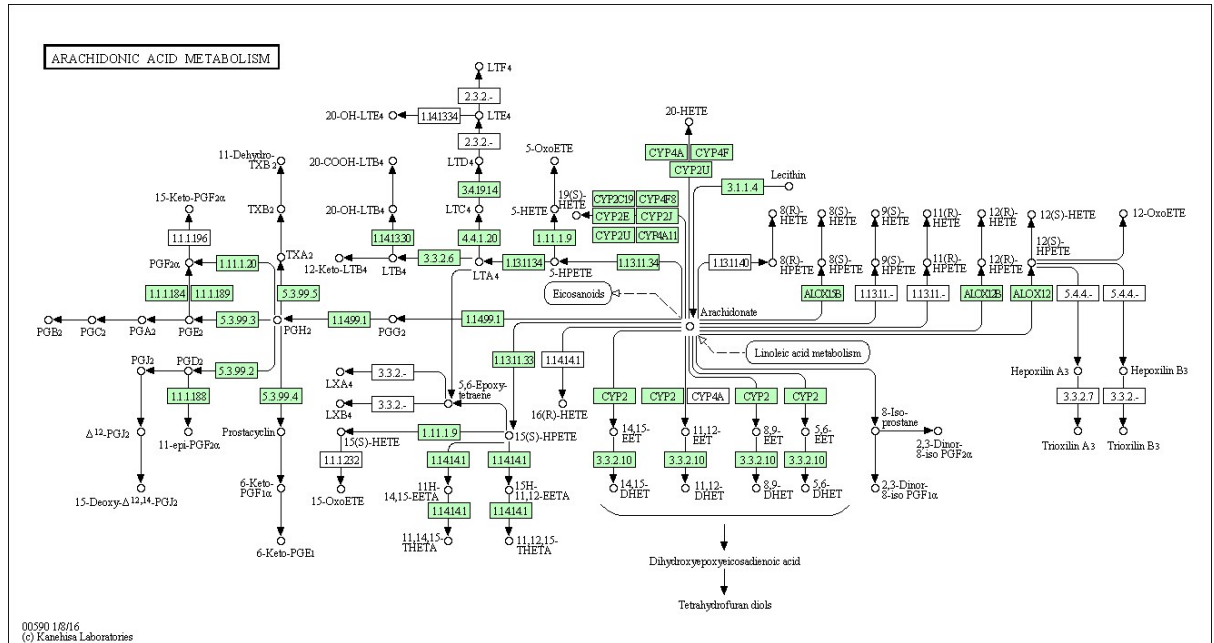
Figure S3. Arachidonic acid metabolism (impact:0.22)

Figure S4.Fatty acid metabolism (impact:0.03)