Metabolic effects of agro-infiltration on *N. benthamiana* accessions

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Supplementary Figure 1. Heatmaps of untargeted (A) and identified (B) metabolite data of control plants of *N. benthamiana* NWA, WA, QLD and RA4.

Supplementary Figure 2. Heatmap of molecular features significantly different between leaf conditions.

Supplementary Figure 3. Heatmaps of primary (A) and secondary (B) metabolites in agroinfiltrated and control leaves.

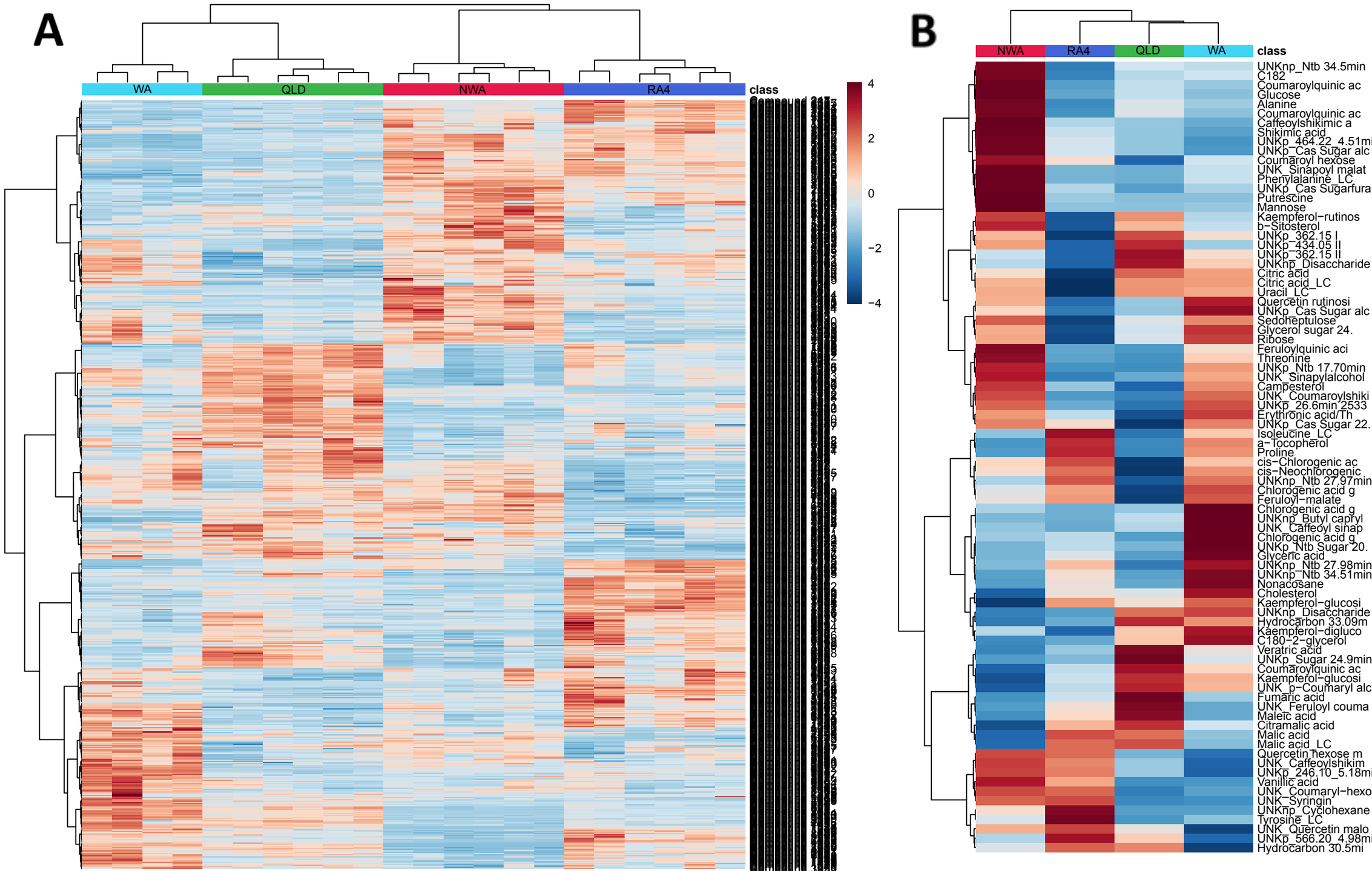
Supplementary Table 1. LC-MS metabolite profiling – untargeted data of control plants of *N. benthamiana* NWA, WA, QLD and RA4.

Supplementary Table 2. LC-MS metabolite profiling – untargeted data of agroinfiltrated plants of *N. benthamiana* NWA, WA, QLD and RA4.

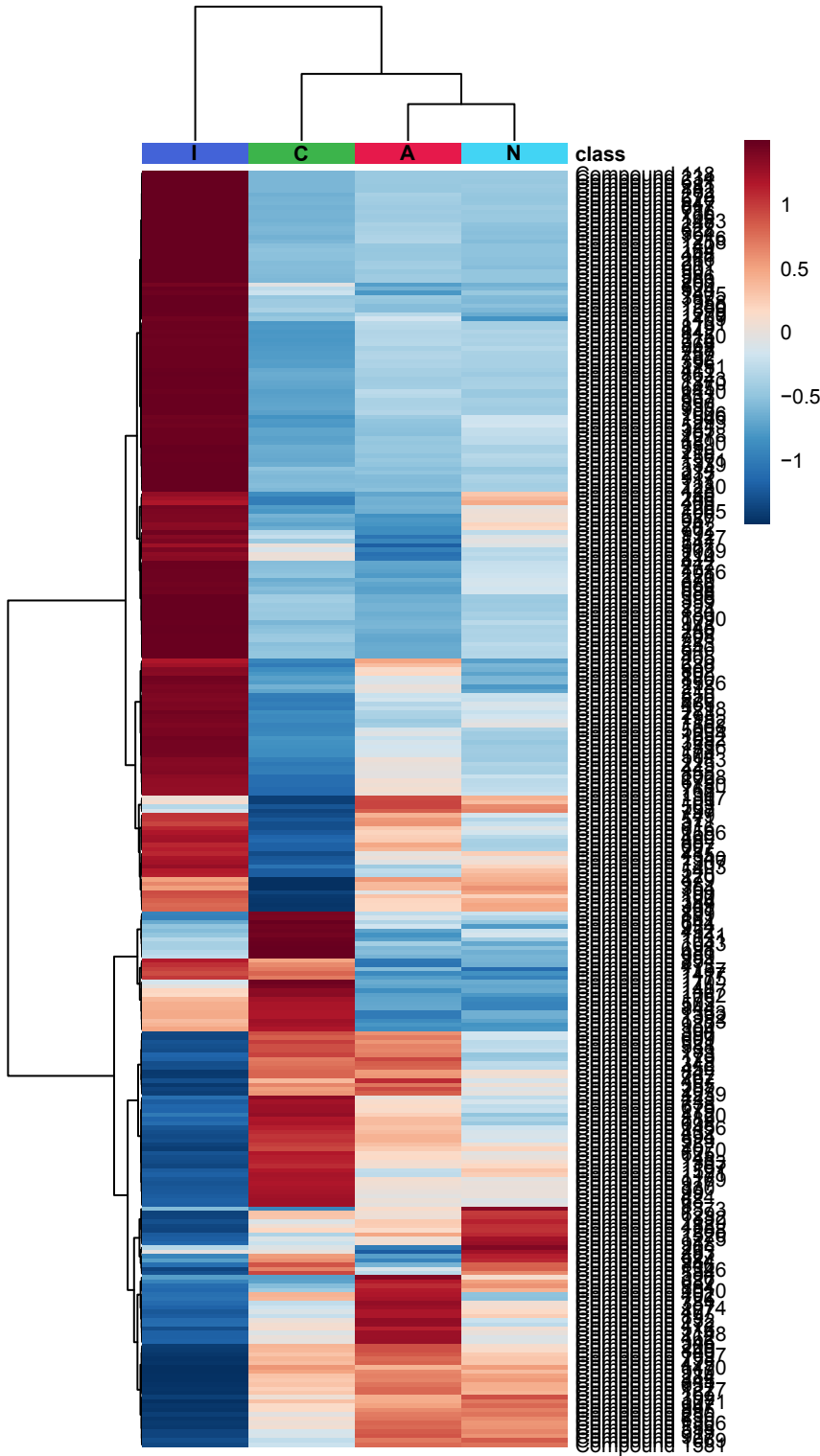
Supplementary Table 3. Levels of identified metabolites in agroinfiltrated and control leaves.

Supplementary Table 4. Metabolite library of *N. benthamiana* accessions.

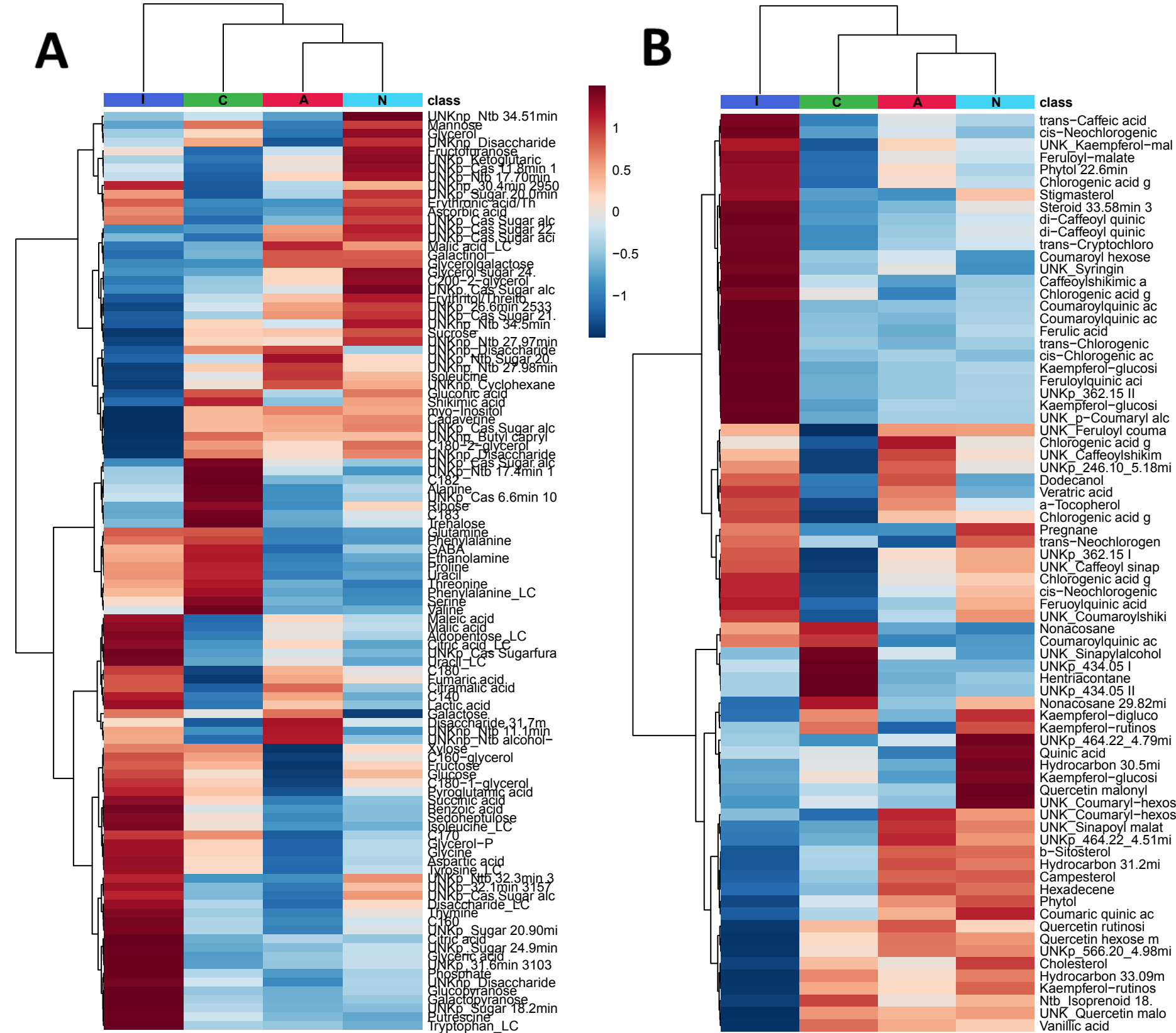
Supplementary Figure 1.



Supplementary Figure 2.



Supplementary Figure 3.



Supplementary Table 4.

	Metabolites[a]	Analytical platform	Identification features (retention time (RT) in min.) RT (GC/MS)	RI (GC/MS)	RT (LC/MS)	m/z [M-H]- (LC/MS)	Identification Extract	Level[c]	Standard[d]	Compound class
1	**Fructofuranose**	GCMS	18.13	1794.6			Polar	3	No	Sugar
2	**Fructose**	GCMS	18.53	1824.3			Polar	1	Yes	Sugar
3	**Galactinol**	GCMS	30.55	2972.4			Polar	1	Yes	Sugar alcohol
4	**Galactopyranose**	GCMS	19.32	1884.5			Polar	1	Yes	Sugar
5	**Galactose**	GCMS	19.22	1877.0			Polar	1	Yes	Sugar
6	**Gluconic acid**	GCMS	20.65	1990.9			Polar	1	Yes	Sugar acid
7	**Glucopyranose**	GCMS	20.41	1972.0			Polar	3	No	Sugar
8	**Glucose**	GCMS	19.66	1911.1			Polar	1	Yes	Sugar
9	**Mannose**	GCMS	18.73	1839.5			Polar	1	Yes	Sugar
10	**myo-Inositol**	GCMS	21.74	2082.3			Polar	1	Yes	Sugar
11	**Ribose**	GCMS	16.38	1667.9			Polar	1	Yes	Sugar
12	**Sedoheptulose**	GCMS	22.11	2118.7			Polar	1	Yes	Sugar
13	**Xylose**	GCMS	16.06	1646.3			Polar	1	Yes	Sugar
14	**LC_Aldopentose**	LCMS			0.74	194.06	Polar	3	No	Sugar
15	**UNKp_Sugar 21.9min**	GCMS	21.93	2098.1			Polar	3	No	Sugar
16	**UNKp_Sugar 22.7min**	GCMS	22.71	2166.5			Polar	3	No	Sugar
17	**UNKp_Sugar acid 17.4min**	GCMS	17.44	1744.3			Polar	3	No	Sugar acid
18	**UNKp_Sugar alcohol 15.4min**	GCMS	15.44	1603.0			Polar	3	No	Sugar alcohol
19	**UNKp_Sugar alcohol 16.7min**	GCMS	16.68	1688.8			Polar	3	No	Sugar alcohol
20	**UNKp_Sugar alcohol 16.98min**	GCMS	16.99	1711.1			Polar	3	No	Sugar alcohol
21	**UNKp_Sugar alcohol 17.1min**	GCMS	17.06	1716.5			Polar	3	No	Sugar
22	**UNKp_Sugar alcohol 19.8min**	GCMS	19.79	1921.9			Polar	3	No	Sugar alcohol
23	**UNKp_Sugarfuranose 17.6min**	GCMS	17.63	1758.0			Polar	3	No	Sugar
24	**UNKp_Sugar 20.3min**	GCMS	20.26	1959.5			Polar	3	No	Sugar
25	**UNKp_Sugar 18.2min**	GCMS	18.05	1785.4			Polar	3	No	Sugar
26	**UNKp_Sugar 20.0min**	GCMS	19.97	1935.3			Polar	3	No	Sugar
27	**UNKp_Sugar 20.9min**	GCMS	20.84	2005.8			Polar	3	No	Sugar
28	**UNKp_Sugar 24.9min**	GCMS	24.87	2365.1			Polar	3	No	Sugar
29	**Sucrose**	GCMS	26.87	2563.7			Polar	1	Yes	Sugar
30	**Trehalose**	GCMS	28.47	2734.9			Polar	1	Yes	Sugar
31	**Disaccharide 31.7min**	GCMS	31.70	3107.0			Polar	3	No	Sugar
32	**LC_Disaccharide**	LCMS			0.69	341.12	Polar	3	No	Sugar
33	**Citric acid**	LCMS/GCMS	18.32	1808.7	0.86	191.03	Polar	1	Yes	TCA cycle
35	**Ketoglutaric acid**	GCMS	18.54	1827.2			Polar	1	Yes	TCA cycle
36	**Succinic acid**	GCMS	10.69	1301.4			Polar	1	Yes	TCA cycle
37	**Fumaric acid**	GCMS	11.39	1345.3			Polar	1	Yes	TCA cycle
38	**Maleic acid**	GCMS	10.62	1299.1			Polar	1	Yes	TCA cycle
39	**Malic acid**	LCMS/GCMS	13.62	1482.8	0.72	133.02	Polar	1	Yes	TCA cycle
41	**Citramalic acid**	GCMS	13.19	1456.1			Polar	1	Yes	TCA cycle
42	**Erythritol/Threitol**	GCMS	13.89	1500.1			Polar	2	No	Cell wall
43	**Erythronic acid/Threonic acid**	GCMS	14.21	1521.4			Polar	1	Yes	Sugar acid
44	**Ethanolamine**	GCMS	10.03	1264.5			Polar	1	Yes	Other
45	**Glyceric acid**	GCMS	11.05	1324.9			Polar	1	Yes	Sugar acid
46	**Glycerol**	GCMS	10.10	1269.0			Polar	1	Yes	Cell wall
47	**Glycerol sugar 24.6min**	GCMS	24.60	2343.2			Polar	2	No	Glyceride
48	**Galactosylglycerol**	GCMS	24.18	2299.1			Polar	1	Yes	Glyceride
49	**Glycerol-3-phosphate**	GCMS	16.97	1709.5			Polar	1	Yes	Glyceride
50	**C14:0**	GCMS	18.62	1832.0			Non-polar	1	Yes	Fatty acid
51	**C16:0**	GCMS	21.13	2034.1			Non-polar	1	Yes	Fatty acid
52	**C16:0-glycerol**	GCMS	26.90	2571.3			Non-polar	1	Yes	Fatty acid
53	**C17:0**	GCMS	21.93	2099.2			Non-polar	1	Yes	Fatty acid
54	**C18:0**	GCMS	23.38	2225.9			Non-polar	1	Yes	Fatty acid
55	**C18:0-1-glycerol**	GCMS	28.67	2769.8			Non-polar	1	Yes	Fatty acid
56	**C18:0-2-glycerol**	GCMS	28.66	2755.9			Non-polar	1	Yes	Fatty acid
57	**C18:2**	GCMS	23.02	2193.0			Non-polar	1	Yes	Fatty acid
58	**C18:3**	GCMS	23.15	2203.2			Non-polar	1	Yes	Fatty acid
59	**C20:0-2-glycerol**	GCMS	30.38	2952.5			Non-polar	1	Yes	Fatty acid
60	**Dodecanol**	GCMS	14.70	1553.9			Non-polar	1	Yes	Hydrocarbon
61	**Hentriacontane**	GCMS	31.50	3086.1			Non-polar	1	Yes	Hydrocarbon
62	**Hexadecene**	GCMS	18.59	1829.2			Non-polar	1	Yes	Hydrocarbon
63	**Hydrocarbon 30.5min**	GCMS	30.50	2960.4			Non-polar	3	No	Hydrocarbon
64	**Hydrocarbon 31.2min**	GCMS	31.20	3049.5			Non-polar	3	No	Hydrocarbon
65	**Hydrocarbon 33.09min**	GCMS	33.09	3272.8			Non-polar	3	No	Hydrocarbon
66	**Nonacosane**	GCMS	29.48	2849.0			Non-polar	1	Yes	Hydrocarbon
67	**Nonacosane 29.82min**	GCMS	29.82	2885.5			Non-polar	2	No	Hydrocarbon
68	**Cholesterol**	GCMS	31.92	3135.6			Non-polar	1	Yes	Sterol
69	**Campesterol**	GCMS	32.77	3236.1			Non-polar	1	Yes	Sterol
70	**b-Sitosterol**	GCMS	33.45	3317.7			Non-polar	1	Yes	Sterol
71	**Stigmasterol**	GCMS	32.97	3260.2			Non-polar	1	Yes	Sterol
72	**UNKnp_Pregnane**	GCMS	18.73	1839.7			Non-polar	3	No	Sterol
73	**Steroid 33.58min 3333**	GCMS	33.58	3333.0			Non-polar	3	No	Sterol
74	**a-Tocopherol**	GCMS	31.79	3120.5			Non-polar	1	Yes	Isoprenoid
75	**Ntb_Isoprenoid 18.6min**	GCMS	18.86	1865.6			Non-polar	3	No	Isoprenoid
76	**Phytol**	GCMS	18.53	1824.1			Non-polar	1	Yes	Isoprenoid
77	**Phytol 22.6min**	GCMS	22.58	2155.7			Non-polar	2	No	Isoprenoid
78	**Alanine**	GCMS	7.40	1109.5			Polar	1	Yes	Amino acid
79	**Aspartic acid**	GCMS	14.09	1513.1			Polar	1	Yes	Amino acid
80	**Cadaverine**	GCMS	8.10	1148.5			Polar	1	Yes	Amino acid
81	**GABA**	GCMS	14.24	1523.4			Polar	1	Yes	Amino acid
82	**Glutamine**	GCMS	17.73	1764.9			Polar	1	Yes	Amino acid
83	**Glycine**	GCMS	10.69	1302.8			Polar	1	Yes	Amino acid
84	**Isoleucine**	LCMS/GCMS	10.46	1289.4	0.96	130.09	Polar	1	Yes	Amino acid
86	**Phenylalanine**	LCMS/GCMS	15.52	1608.7	2.38	164.08	Polar	1	Yes	Amino acid
88	**Proline**	GCMS	10.55	1294.4			Polar	1	Yes	Amino acid
89	**Putrescine**	GCMS	17.25	1730.0			Polar	1	Yes	Amino acid
90	**Pyroglutamic acid**[b]	GCMS	14.11	1514.7			Polar	1	Yes	Amino acid
91	**Serine**	GCMS	11.56	1355.2			Polar	1	Yes	Amino acid
92	**Threonine**	GCMS	11.96	1379.5			Polar	1	Yes	Amino acid
94	**Tryptophan**	LCMS			3.66	203.09	Polar	1	Yes	Amino acid
95	**Tyrosine**	LCMS			1.51	180.08	Polar	1	Yes	Amino acid
96	**Valine**	GCMS	9.19	1215.0			Polar	1	Yes	Amino acid
97	**Shikimic acid**	GCMS	18.20	1799.2			Polar	1	Yes	Phenylpropanoid
98	**Quinic acid**	LCMS			0.92	191.06	Polar	1	Yes	Phenylpropanoid
99	**trans-Caffeic acid**	GCMS	22.22	2123.4			Polar	1	Yes	Phenylpropanoid
100	**Caffeoylshikimic acid**	LCMS			4.07	335.08	Polar	2	No	Phenylpropanoid
101	**di-Caffeoyl quinic acid**	LCMS			3.07	515.15	Polar	2	No	Phenylpropanoid
102	**di-Caffeoyl quinic acid (3,5-di-CQA)**	LCMS			3.38	515.15	Polar	2	No	Phenylpropanoid
103	**UNK_Caffeoyl sinapoyl quinic acid**	LCMS			5.62	559.09	Polar	2	No	Phenylpropanoid
104	**UNK_Caffeoylshikimic acid**	LCMS			5.18	335.07	Polar	2	No	Phenylpropanoid
105	**Chlorogenic acid glycoside I**	LCMS			5.44	515.13	Polar	2	No	Phenylpropanoid
106	**Chlorogenic acid glycoside II**	LCMS			5.62	515.13	Polar	2	No	Phenylpropanoid
107	**Chlorogenic acid glycoside III**	LCMS			5.73	515.13	Polar	2	No	Phenylpropanoid
108	**Chlorogenic acid glycoside IV**	LCMS			5.85	515.13	Polar	2	No	Phenylpropanoid
109	**Chlorogenic acid glycoside V**	LCMS			6.01	515.18	Polar	2	No	Phenylpropanoid
110	**cis-Chlorogenic acid**	LCMS			3.96	353.09	Polar	1	Yes	Phenylpropanoid
111	**cis-Neochlorogenic acid**	LCMS			2.66	353.09	Polar	1	Yes	Phenylpropanoid
112	**cis-Neochlorogenic acid II**	LCMS			3.25	353.09	Polar	1	Yes	Phenylpropanoid
113	**trans-Chlorogenic acid**	LCMS			3.41	353.09	Polar	1	Yes	Phenylpropanoid
114	**trans-Cryptochlorogenic acid**	LCMS			3.59	353.09	Polar	1	Yes	Phenylpropanoid
115	**trans-Neochlorogenic acid**	LCMS			2.51	353.09	Polar	1	Yes	Phenylpropanoid
116	**Coumaric quinic acid (CoQ)**	LCMS			3.24	337.10	Polar	2	No	Phenylpropanoid
117	**Coumaroyl hexose**	LCMS			3.51	325.10	Polar	2	No	Phenylpropanoid
118	**Coumaroylquinic acid**	LCMS			4.88	337.10	Polar	2	No	Phenylpropanoid
119	**Coumaroylquinic acid (4-CoQA)**	LCMS			4.05	337.10	Polar	2	No	Phenylpropanoid
120	**Coumaroylquinic acid (5-CoQA)**	LCMS			4.56	337.10	Polar	2	No	Phenylpropanoid
121	**UNK_Coumaroylshikimic acid**	LCMS			3.88	319.15	Polar	2	No	Phenylpropanoid
122	**UNK_Coumaryl-hexose malic acid I**	LCMS			5.44	441.18	Polar	2	No	Phenylpropanoid
123	**UNK_Coumaryl-hexose malic acid II**	LCMS			5.66	441.18	Polar	2	No	Phenylpropanoid
124	**UNK_p-Coumaryl alcohol 4-O-glucoside**	LCMS			4.88	311.12	Polar	2	No	Phenylpropanoid
125	**Ferulic Acid**	LCMS			6.15	193.06	Polar	1	Yes	Phenylpropanoid
126	**Feruloyl-malate**	LCMS			6.01	309.14	Polar	1	Yes	Phenylpropanoid
127	**Feruloylquinic acid (trans-5-FQA)**	LCMS			4.36	367.11	Polar	2	No	Phenylpropanoid
128	**Feruoylquinic acid (trans-4-FQA)**	LCMS			4.75	367.11	Polar	2	No	Phenylpropanoid
129	**UNK_Feruloyl coumaroyl caffeoyl shikimic acid**	LCMS			6.03	675.13	Polar	2	No	Phenylpropanoid
130	**UNK_Sinapoyl malate**	LCMS			5.54	339.13	Polar	2	No	Phenylpropanoid
131	**UNK_Sinapylalcohol**	LCMS			4.72	207.02	Polar	2	No	Phenylpropanoid
132	**UNK_Syringin**	LCMS			5.63	371.14	Polar	2	No	Phenylpropanoid
133	**Kaempferol-diglucoside**	LCMS			3.90	609.15	Polar	2	No	Phenylpropanoid
134	**Kaempferol-glucoside I**	LCMS			4.55	447.09	Polar	1	Yes	Phenylpropanoid
135	**Kaempferol-glucoside II**	LCMS			4.88	447.10	Polar	1	Yes	Phenylpropanoid
136	**Kaempferol-glucoside III**	LCMS			5.59	447.10	Polar	1	Yes	Phenylpropanoid
137	**Kaempferol-rutinoside I**	LCMS			5.07	563.18	Polar	1	Yes	Phenylpropanoid
138	**Kaempferol-rutinoside II**	LCMS			5.38	593.16	Polar	1	Yes	Phenylpropanoid
139	**UNK_Kaempferol-malic acid glucoside**	LCMS			6.02	563.16	Polar	2	No	Phenylpropanoid
140	**Quercetin hexose malic acid**	LCMS			5.49	579.21	Polar	2	No	Phenylpropanoid
141	**Quercetin malonyl glucoside**	LCMS			5.38	549.21	Polar	1	Yes	Phenylpropanoid
142	**Rutin**	LCMS			4.96	609.16	Polar	1	Yes	Phenylpropanoid
143	**UNK_Quercetin malonyl glucoside**	LCMS			5.56	549.20	Polar	2	No	Phenylpropanoid
144	**Benzoic acid**	GCMS	9.74	1248.5			Polar	1	Yes	Phenylpropanoid
145	**Vanillic acid**	LCMS			4.45	167.08	Polar	1	Yes	Phenylpropanoid
146	**Veratric acid**	LCMS			6.72	181.13	Polar	1	Yes	Phenylpropanoid
147	**Ascorbic acid**	GCMS	19.99	1937.7			Polar	1	Yes	Vitamin
148	**Lactic acid**	GCMS	6.75	1071.0			Polar	1	Yes	Cell wall
149	**Phosphate**	GCMS	8.56	1178.0			Polar	1	Yes	Other
150	**UNKnp_30.4min 2950**	GCMS	30.40	2950.0			Non-polar	4	No	Unknown
151	**UNKnp_Butyl caprylate**	GCMS	11.36	1342.9			Non-polar	4	No	Unknown
152	**UNKnp_Cyclohexane acetic acid**	GCMS	10.65	1300.0			Non-polar	4	No	Unknown
153	**UNKnp_33.57min**	GCMS	33.57	3330.0			Non-polar	4	No	Unknown
154	**UNKnp_Disaccharide/Steroid bound compound 33.62min**	GCMS	33.62	3328.8			Non-polar	4	No	Unknown
155	**UNKnp_Disaccharide/Steroid bound compound 34.51min**	GCMS	34.51	3432.3			Non-polar	4	No	Unknown
156	**UNKnp_Disaccharide/Steroid bound compound 34.59min**	GCMS	34.59	3431.4			Non-polar	4	No	Unknown
157	**UNKnp_11.1min 1326**	GCMS	11.10	1326.0			Non-polar	4	No	Unknown
158	**UNKnp_27.97min 2680**	GCMS	27.97	2680.0			Non-polar	4	No	Unknown
159	**UNKnp_27.98min 2681**	GCMS	27.98	2681.0			Non-polar	4	No	Unknown
160	**UNKnp_34.51min 3444**	GCMS	34.51	3444.0			Non-polar	4	No	Unknown
161	**UNKnp_34.5min 3439**	GCMS	34.50	3439.0			Non-polar	4	No	Unknown
162	**UNKnp_alcohol-Si-alcohol**	GCMS	33.86	3366.1			Non-polar	4	No	Unknown
163	**UNKp_26.6min 2533**	GCMS	26.60	2533.0			Polar	4	No	Unknown
164	**UNKp_31.6min 3103**	GCMS	31.60	3103.0			Polar	4	No	Unknown
165	**UNKp_32.1min 3157**	GCMS	32.10	3157.0			Polar	4	No	Unknown
166	**UNKp_11.8min 1369**	GCMS	11.76	1369.0			Polar	4	No	Unknown
167	**UNKp_6.6min 1060**	GCMS	6.60	1060.0			Polar	4	No	Unknown
168	**UNKp_17.4min 1745**	GCMS	17.40	1745.0			Polar	4	No	Unknown
169	**UNKp_17.70min 1763**	GCMS	17.70	1763.0			Polar	4	No	Unknown
170	**UNKp_32.3min 3178**	GCMS	32.30	3178.0			Polar	4	No	Unknown
93	**Thymine**	GCMS	11.49	1350.2			Polar	1	Yes	Nucleotide
171	**Uracil**	LCMS/GCMS	11.17	1332.0	0.86	111.0	Polar	1	Yes	Nucleotide
173	**UNKp_362.15 I**	LCMS			2.83	361.2	Polar	4	No	Unknown
174	**UNKp_362.15 II**	LCMS			2.92	361.2	Polar	4	No	Unknown
175	**UNKp_434.05 I**	LCMS			2.84	433.1	Polar	4	No	Unknown
176	**UNKp_434.05 II**	LCMS			2.96	433.1	Polar	4	No	Unknown
177	**UNKp_246.10_5.18min**	LCMS			5.18	245.1	Polar	4	No	Unknown
178	**UNKp_464.22_4.51min**	LCMS			4.51	463.2	Polar	4	No	Unknown
179	**UNKp_464.22_4.79min**	LCMS			4.79	463.2	Polar	4	No	Unknown
180	**UNKp_566.20_4.98min**	LCMS			4.98	565.2	Polar	4	No	Unknown

[a] Compound number in Figure 1C

[b] Derivatisation of glutamic acid produces the corresponding TMS derivatives in addition to TMS-pyroglutamic acid derivatives (Kanani H, Klapa MI (2007). Metabolic Engineering, 9 39-51).

[c] Identification confirmed by spectral and chromatographic properties comparison to authentic commercial standards

[d] Level of metabolite identification as in (Bino et al. (2004) Trends in Plant Science, 9:418–425; Sumner et al. (2007) Metabolomics, 3:211-221*)(1) Identified compounds; (2) Putatively annotated compounds; (3) Putatively characterized compound classes; (4) Unknown compounds*