RSC Advances

PAPER

View Article Online
View Journal | View Issue

Cite this: *RSC Adv.*, 2020, **10**, 26109

Rapid characterization of the chemical constituents of Sanhua decoction by UHPLC coupled with Fourier transform ion cyclotron resonance mass spectrometry†

Yanhui Zhao,[a] Miao Wang,[b] Lin Sun,[a] Xue Jiang,[a] Min Zhao *[a] and Chunjie Zhao *[a]

Sanhua decoction, a famous Chinese herbal formula has been widely used for the treatment of stroke. In our study, a rapid, swift and straightforward analytical method with the help of UHPLC-FT-ICR-MS/MS was successfully developed for the first time to separate and identify the chemical constituents of Sanhua decoction. Chromatography was performed on a Universal XB C_{18} column (150 mm × 2.1 mm, 1.8 μm) using a mobile phase containing 0.1% formic acid–water (A) and acetonitrile (B). A total of 137 compounds in Sanhua decoction were identified or tentatively characterized. The findings revealed the fact that Sanhua decoction mainly contains flavonoids (in *Aurantii fructus immaturus* and *Rheum palmatum* L.), anthraquinones (in *Rheum palmatum* L.), coumarins (in *Notopterygii Rhizoma Et Radix*), phenylpropanoid glycosides, alkaloids and lignans (in *Magnoliae Officmalis Cortex*), which made up the key ingredients existing in Sanhua decoction. This study is hoped to be meaningful for the characterization of components in other traditional Chinese medicines, and lay the foundation for research on the pharmacology of Sanhua decoction.

Received 21st March 2020
Accepted 23rd May 2020

DOI: 10.1039/d0ra02264k

rsc.li/rsc-advances

1. Introduction

Traditional Chinese medicines (TCMs) play an important role in Chinese medicine due to their wide application and efficacy, particularly in the management of chronic diseases.[1] TCMs are composed of complex chemical constituents for multi-target treatment, and produce synergistic effects and reduce side effects. Although there has been a lot of research on TCMs, the active ingredients of most traditional Chinese medicine prescriptions remain unknown. Therefore, developing a fast and efficient analysis method to separate and identify complex constituents in TCMs is necessary for illustrating the pharmacological material basis of TCMs.

Sanhua decoction (SHD), a famous Chinese herbal formula, is recorded in the classic traditional Chinese medicine (TCM) book Suwen Bingji Qiyi Baomingji (Plain Questions: Discourse on Mechanism for Preserving Life). It consists of four crude herbs: Zhishi (*Aurantii fructus immaturus*), Dahuang (*Rheum palmatum* L.), Houpu (*Magnoliae Officmalis Cortex*) and Qianghuo (*Notopterygii Rhizoma Et Radix*). SHD is used widely for the treatment of stroke. It can improve ischemic cerebral edema and brain blood barrier (BBB) permeability.[2] It has been documented that Zhishi has neuroprotective, antioxidant, anti inflammatory and anti-apoptotic effects.[3] Dahuang can protect neurons from hypoxic-ischemic brain damage.[4] Houpu can protect neural damage from cerebral ischemia and reperfusion by suppressing cerebral inflammation and improving BBB function.[5] Qianghuo has a function of anti-thrombosis, increasing cerebral blood flow and improving cerebral blood circulation.[6] However, the chemical material basis of SHD has not been studied in detail until now.

As the complexity of traditional Chinese medicine, it is very important to establish a sensitive and reliable detection method. Chromatography-mass spectrometry (LC-MS) is widely used for its high sensitivity and specificity. Ultra high performance liquid chromatography (UHPLC) coupled with Fourier transform ion cyclotron resonance mass spectrometry (FT-ICR-MS) as the most powerful instrument with high resolution, accuracy and sensitivity play a role in analyzing different kinds of complex samples.[7–10] It provides an unambiguous elemental composition and information about the isotopic abundance of ions to help determine the compositions and structures of chemical constituents. By usage of this technology, Li *et al.* characterized 179 constituents in *Rhodiola crenulata* as well as 37 prototypes and 142 metabolites in rats.[11] Guan *et al.* characterized 120 constituents in Sijunzi decoction[12] and Liu *et al.*

[a]*School of Pharmacy, Shenyang Pharmaceutical University, Wenhua Road 103, Shenhe District, Shenyang 110016, Liaoning Province, China. E-mail: lab433@163.com*
[b]*School of Life Science and Biopharmaceutics, Shenyang Pharmaceutical University, Wenhua Road 103, Shenyang, Liaoning Province, China*

† Electronic supplementary information (ESI) available. See DOI: 10.1039/d0ra02264k

characterized 174 constituents in Gegenqinlian decoction as well as 107 prototypes and 67 metabolites in rats.[13] The results show that this method is useful in detecting and indenting chemical constituents and metabolites by information about accurate molecular weight and MS^2.

In this study, a rapid and simple approach by UHPLC-FT-ICR-MS was established for the systematical characterization of the constituents of SHD. This constituent characterization and structural elucidation provided significant information for a pharmacological study of SHD.

2. Material and methods

2.1 Chemicals and materials

The purchase of key ingredients including, *Aurantii fructus immaturus* (batch number: 170 901; source: Sichuan China), *Rheum palmatum* L. (batch number: 20 181 101; source: Gansu China), *Magnoliae Officmalis Cortex* (batch number: 181 001; source: Sichuan China) and *Notopterygii Rhizoma Et Radix* (batch number: 13 112 501; source: Sichuan China) was made from Guoda pharmacy (Shenyang, China), which followed identification by Professor Jingming Jia (Department of TCM, Shenyang Pharmaceutical University, Shenyang, China). The key source of the reference compounds (purity > 98%), including sennoside B, nodakenin, narirutin, sennoside A, neohesperidin, naringenin, hesperetin, nobiletin was Shanghai Yuanye Bio-Technology Co., Ltd. (Shanghai, China); naringin, hesperidin and honokiol was attained from the National Institute for the Control of Pharmaceutical and Biological Products (Beijing, China). In addition, acetonitrile of HPLC grade, together with formic acid of LC-MS grade was attained from Fisher Scientific (Fair Lawn, NJ, USA), followed by attaining the purified water from Wahaha (Hangzhou, China).

2.2 Preparation of SHD for analysis

Following the documentary records of SHD, four constituting herbs that included *Aurantii fructus immaturus* (40 g), *Rheum palmatum* L. (40 g), *Magnoliae Officmalis Cortex* (40 g) and *Notopterygii Rhizoma Et Radix* (40 g) were soaked in water for 60 min, then decocted two times in 80 mL water for a period of 1 h each time in a glass flask and the ensuing solution was mixed and dried with the help of lyophilization. Prior to the analysis, dissolution the 0.5 g of dried powder was dissoluted by 5 mL methanol-aqueous (1 : 1) solution, soaking for 15 min with 80 °C water.

2.3 Instrument and analytical conditions

The sample was analyzed by the UHPLC-DAD-FT-ICR-MS system contains an Agilent 1260 UHPLC system combined with a Bruker Solarix7.0T FT-ICR-MS system (Germany) and Bruker Compass-Hystar workstation (Bruker, Germany). Chromatography was carried out on a Universal XB C_{18} column (150 mm × 2.1 mm, 1.8 μm; Kromat, USA) at the temperature of 35 °C. The mobile phase contained 0.1% formic acid–water (A) and acetonitrile (B). The gradient conditions were as follows: 8–12% (B) in 0 to 10 min, 12–13% (B) in 10 to 12 min, 13–18% (B) in 13 to 18 min, 18–24% (B) in 17 to 28 min, 24–30% (B) in 28 to 31 min, 30–36% (B) in 31 to 35 min, 36–55% (B) in 35 to 37 min, 55–90% (B) in 37 to 47 min, which was delivered at a flow rate of 0.2 mL min^{-1} and injection volume was 2 μL.

The mass spectra were performed with positive as well as negative electrospray ionization (ESI) modes, followed by setting the optimized conditions are as follows: capillary voltage, 4.5 kV; plate offset, 500 v; nebulizer gas pressure, 4.0 bar; dry gas flow rate, 8 L min^{-1}; dry gas temperature, 200 °C; ion accumulation time, 0.15 s, and flight time, 0.6 ms. Full scan mass spectrometry data were recorded between *m*/*z* 100 and 1200 amu and the collision energy was ranged from 10 eV to 30 eV for MS/MS experiments.

3. Results and discussion

The Fig. 1 reveals the base peak ion chromatograms (BPC) of SHD in positive as well as negative ion modes, together with the respective compounds. The extract ion chromatograms (EIC) of each molecule weight were correspondingly attained for detecting the associated compound followed by presenting in support information Fig. S1.† Comparing with reference compounds, eleven peaks from SHD were accurately identified by the retention times and MS fragments in positive ion mode. The peak of 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11 in Fig. 1C were sennoside B, nodakenin, narirutin, naringin, sennoside A, hesperidin, neohesperidin, naringenin, hesperetin, honokiol, nobiletin, respectively. The others were predicted by Bruker workstation, whether or not the mass error values between the identified molecular weights with the measured molecular weights were below 3.0 ppm. If yes, it can infer the chemical structure of each component by the retention time and MS/MS data with published literature.[14–28] In the end, a total of 137 compounds were identified and characterized, whereas their layouts were presented in support information Fig. S2.†

The MS/MS spectra of representative compounds are shown in Fig. 2, and displaying deduced fragmentation pathways are shown in Fig. 3. Except that, the formula, retention time, MS data, calculated *m*/*z*, error, ion mode and MS/MS data of other detected components were listed in Table 1.

3.1 Characterization of flavonoids in SHD

Flavonoids have a typical cyclohexene structure in the C ring, which is unstable and easy to break it for Diels–Alder reaction (RDA), and get the characteristic fragment ions 1,3A and 1,3B. Flavonoid glycosides have methoxy group, firstly losing the glycosyl groups, followed by losing the methyl groups, at last cleavage of the C ring. The mass spectral fragmentation of flavonoids by hesperidin as an example was described. In positive ion mode, adduct ion at *m*/*z* 611.19650 ($[M + H]^+$) indicating an elemental composition of $C_{28}H_{34}O_{15}$. The MS/MS spectrum (Fig. 3A) is presented that the main fragment ions are *m*/*z* 449.14312, 303.08581, 288.09732, 153.02175 as well as 136.04821. Among them, *m*/*z* 449.14312 and 303.08581 are inferred by cleaving of the glycosidic linkage and losing $C_6H_{10}O_5$ (162 Da) or $C_{12}H_{20}O_9$ (308 Da) from the ion of $[M - H]^+$,

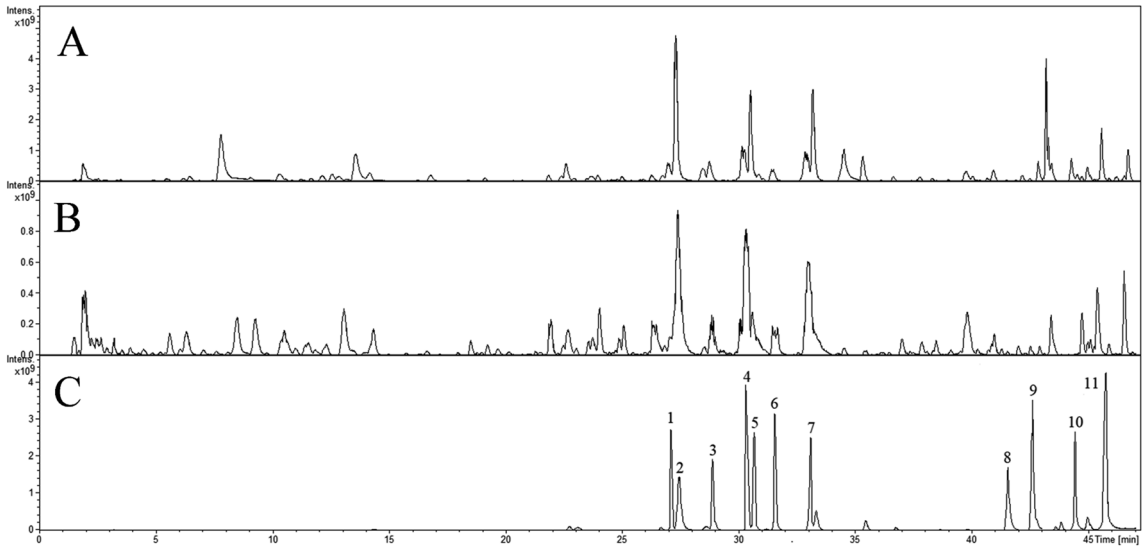

Fig. 1 The base peak ion chromatograms (BPC) of SHT in both positive (A) and negative (B) ion modes and the corresponding compounds (C). (1) Sennoside B, (2) nodakenin, (3) narirutin, (4) naringin, (5) sennoside A, (6) hesperidin, (7) neohesperidin, (8) naringenin, (9) hesperetin, (10) honokiol, (11) nobiletin.

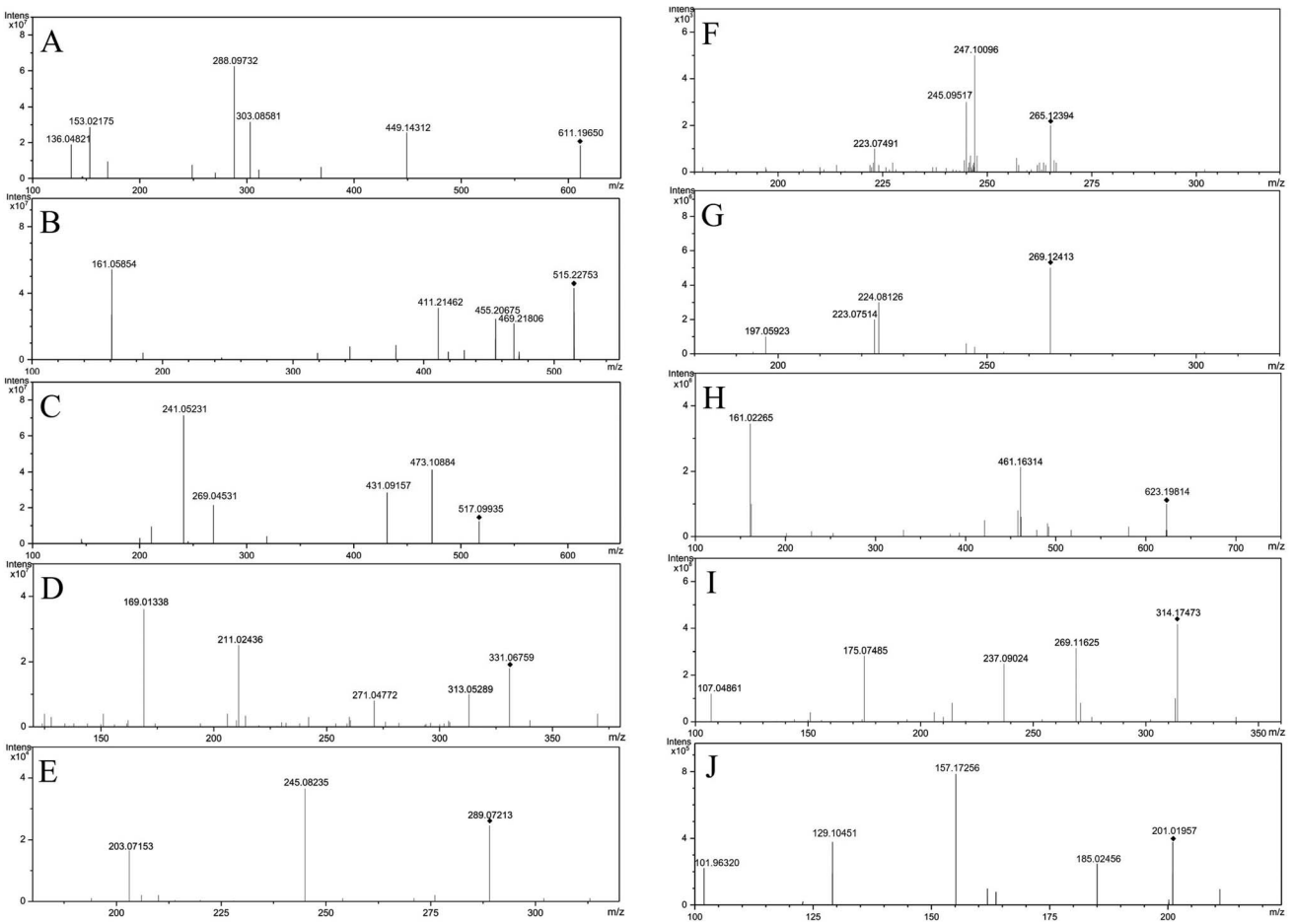

Fig. 2 The MS/MS spectra of the typical compounds. (A) Hesperidin, (B) nomilin, (C) emodin-*O*-malonyl-glucose, (D) galloyl-glucoside, (E) epicatechin, (F) honokiol, (G) magnolol, (H) acteoside, (I) magnocurarine, (J) bergaptol.

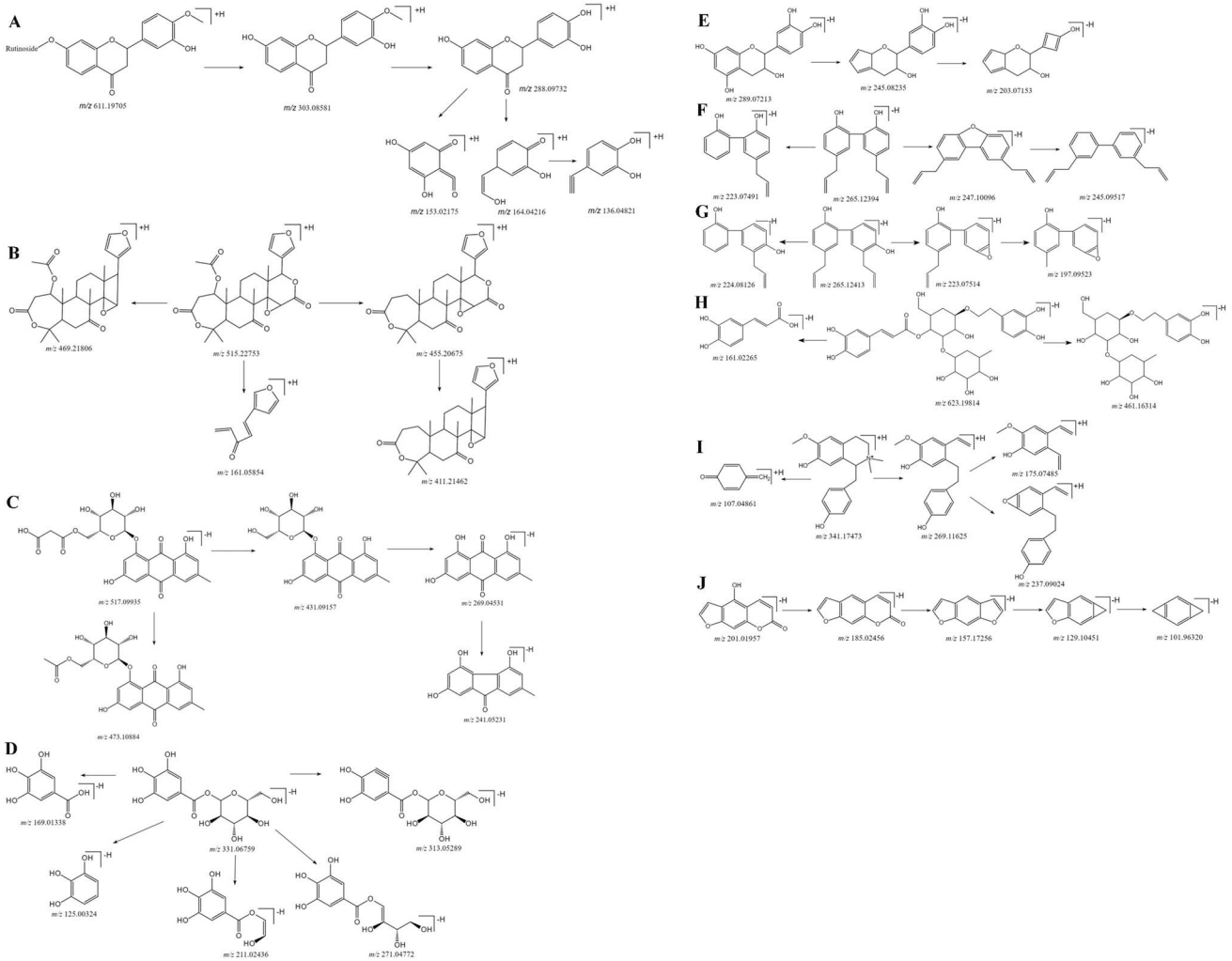

Fig. 3 The possible fragmentation pathways of the typical compounds. (A) Hesperidin, (B) nomilin, (C) emodin-*O*-malonyl-glucose, (D) galloyl-glucoside, (E) epicatechin, (F) honokiol, (G) magnolol, (H) acteoside, (I) magnocurarine, (J) bergaptol.

m/*z* 288.09732 is the result of losing CH_3 (15 Da) from *m*/*z* 303.08581, and *m*/*z* 153.02175 and *m*/*z* 136.04821 was due to the Diels–Alder reaction from *m*/*z* 288.09732. The characterization of other flavonoids in SHD was performed based on the fragmentation patterns and related literatures.[14–18]

3.2 Characterization of triterpenoids in SHD

Triterpenoids are prone to lose H_2O (18 Da), CO (28 Da) and CO_2 (44 Da), due to the presence of lactone rings. Nomilin was selected to illustrate the fragmentation pathways. In positive ion mode, adduct ion at *m*/*z* 515.22753 ($[M + H]^+$) indicating an elemental composition of $C_{28}H_{34}O_9$. The MS/MS spectrum (Fig. 3B) is presented that the main fragment ions are *m*/*z* 469.21806, 455.20675, 411.21462, 161.05854. Among them, *m*/*z* 469.21806 is inferred by losing CO and H_2O (46 Da) from $[M + H]^+$, *m*/*z* 455.20675 is presumed by losing $C_2H_4O_2$ (60 Da) from $[M + H]^+$, *m*/*z* 411.21462 is the result of losing CO_2 (44 Da) from *m*/*z* 455.20675, and *m*/*z* 161.05854 is estimated by losing $C_{18}H_{26}O_7$ (354 Da) from $[M + H]^+$. The characterization of other triterpenoids in SHD was performed based on the fragmentation patterns and related literatures.[15,19]

3.3 Characterization of anthraquinones in SHD

The common characteristic of MS^2 in anthraquinones is the continuous loss of CO (28 Da). Anthraquinone glycosides can lose glycosyl and CO, continually. Aglycones can be judged by $[aglycone - H]^-$. Anthraquinone glycosides can lose methyl-glucuronide (190 Da), cinnamoyl (148 Da), malonyl (86 Da), acetyl-glucose (204 Da), and glucose (162 Da). Emodin-*O*-malonylglucose was selected to illustrate the fragmentation pathways. In negative ion mode, adduct ion at *m*/*z* 517.09935 ($[M - H]^-$) indicating an elemental composition of $C_{24}H_{22}O_{13}$. The mass spectrum (Fig. 3C) shows that the main fragment ions are *m*/*z* 473.10884, 431.09157, 269.04531 and 241.05131. Among them, *m*/*z* 473.10884 is presumed by losing of CO_2 (44 Da) from $[M - H]^-$, *m*/*z* 431.09157 is inferred by losing of the $C_3H_2O_3$ (86 Da) from $[M - H]^-$, and *m*/*z* 269.04531 is speculated by

Table 1 UHPLC-FT-ICR-MS analysis of SHD[a]

No.	t_R (min)	Identification	Formula	Monoisotopic mass (*m*/*z*)	Detected mass (*m*/*z*)	Ion mode	ppm	MS/MS (*m*/*z*)	Main source
1	2.07	Quinic acid	$C_7H_{12}O_6$	193.07066	193.07024	$[M + H]^+$	2.21	147.04051, 129.05446	C
2	2.64	Galloyl-glucoside	$C_{13}H_{16}O_{10}$	331.06707	331.06759	$[M - H]^-$	−1.57	313.05289, 271.04772, 211.02436, 169.01338, 125.00325	R
3	2.91	1-Galloyl-di-glucoside	$C_{19}H_{26}O_{15}$	493.11989	493.12035	$[M - H]^-$	−0.93	373.07705, 331.06771, 313.05667, 271.04642	R
4	3.21	Gallic acid	$C_7H_6O_5$	169.01425	169.01447	$[M - H]^-$	−1.30	151.00284, 125.02324	R
5	4.85	Catechin-3-*O*-glucoside	$C_{21}H_{24}O_{11}$	451.12459	451.12518	$[M - H]^-$	−1.31	289.07014, 245.08287	R
6	5.59	Rebouoside C	$C_{20}H_{30}O_{12}$	461.16645	461.16674	$[M - H]^-$	−0.64	299.11268, 137.05927	M
7	6.04	Chlorogenic acid	$C_{16}H_{18}O_9$	353.08781	353.08824	$[M - H]^-$	−1.23	191.09726, 179.76826, 173.10064	N
8	7.02	Catechin-5-*O*-glucoside	$C_{21}H_{24}O_{11}$	451.12459	451.12504	$[M - H]^-$	−1.01	289.07156, 245.08176, 203.07109	R
9	7.79	Magnocurarine	$C_{19}H_{24}NO_3^+$	314.17507	314.17473	$[M + H]^+$	1.07	269.11618, 237.08974, 175.07514, 107.04887	M
10	10.40	Catechin	$C_{15}H_{14}O_6$	289.07176	289.07211	$[M - H]^-$	−1.22	245.08146, 205.04962, 203.06974	R
11	10.63	Cryptochlorogenic acid	$C_{16}H_{18}O_9$	353.08781	353.08825	$[M - H]^-$	−1.25	191.10029, 179.14054, 173.10065	N
12	10.64	Syringin	$C_{17}H_{24}O_9$	395.13125	395.13094	$[M + Na]^+$	0.80	372.08647, 343.94804, 208.09042, 137.03937	M
13	13.55	Magnoflorine	$C_{20}H_{24}NO_4^+$	342.16998	342.16967	$[M + H]^+$	0.91	297.10956, 282.08696, 265.08507, 237.08972	M
14	16.29	(*R*)-Oblongine	$C_{19}H_{24}NO_3^+$	314.17507	314.17475	$[M + H]^+$	1.02	269.11625, 237.09024, 175.07485, 107.04861	M
15	16.62	Epicatechin	$C_{15}H_{14}O_6$	289.07176	289.07213	$[M - H]^-$	−1.26	245.08235, 203.07153	R
16	18.49	Teupolioside	$C_{35}H_{46}O_{20}$	785.25097	785.24931	$[M - H]^-$	2.11	649.28262, 623.38565, 477.34832	M
17	19.22	Aloe-emodin-*O*-di-glucoside	$C_{27}H_{30}O_{15}$	593.15119	593.15142	$[M - H]^-$	−0.38	431.09851, 269.04559, 239.03448	R
18	19.31	Apigenin-6,8-di-*C*-glucoside	$C_{27}H_{30}O_{15}$	595.16575	595.16502	$[M + H]^+$	1.23	577.15636, 505.13492, 475.12405	C
19	20.32	Xanthoplanine	$C_{21}H_{25}O_4N^+$	356.18563	356.18528	$[M + H]^+$	1.01	311.12628, 296.10825, 280.10864	M
20	20.74	Asimilobine	$C_{17}H_{17}O_2N$	268.13321	268.13294	$[M + H]^+$	0.99	251.10647, 236.08474, 219.08076, 191.08617	M
21	21.26	Echinacoside	$C_{35}H_{46}O_{20}$	785.25097	785.24959	$[M - H]^-$	1.76	649.28357, 623.38647, 477.34187	M
22	21.76	Chysoeriol-6,8-di-*C*-glucoside	$C_{28}H_{32}O_{16}$	625.17631	625.17579	$[M + H]^+$	0.84	607.16748, 535.14662, 505.13284	C
23	21.95	1-Cinnamoyl-6-*O*-(glucosyl)-galloyl-glucoside	$C_{28}H_{32}O_{16}$	623.16176	623.16247	$[M - H]^-$	−1.15	461.10814, 331.06702, 313.05827, 211.02305	R
24	22.31	Scopoletin	$C_{10}H_8O_4$	193.04954	193.04935	$[M + H]^+$	0.95	178.02482, 165.05373, 122.03784	C
25	22.45	2-*O*-Cinnamyl-β-D-glucoside	$C_{15}H_{18}O_7$	309.09798	309.09841	$[M - H]^-$	−1.39	237.07692, 201.01967, 146.96590	R
26	22.55	Emodin-*O*-glucuronic acid methyl ester	$C_{22}H_{20}O_{11}$	459.09329	459.09358	$[M - H]^-$	−0.65	269.04543, 241.05106, 225.05490, 213.05525	R
27	23.60	Decuroside V	$C_{20}H_{20}O_{10}$	447.12617	447.12576	$[M + Na]^+$	0.91	263.08937, 245.08472	N
28	23.73	Xanthotoxol	$C_{11}H_6O_4$	203.03389	203.03379	$[M + H]^+$	0.45	173.07564, 145.04536, 117.05273	C
29	23.73	Bergaptol	$C_{11}H_6O_4$	201.01933	201.01957	$[M - H]^-$	−1.17	185.02456, 157.17256, 129.10451, 101.96320	N
30	23.76	1-*O*-Cinnamyl-β-D-glucoside	$C_{15}H_{18}O_7$	309.09798	309.09843	$[M - H]^-$	−1.47	248.95978, 161.04574, 146.96419	R
31	23.77	Bergaptol-*O*-β-D-glucopyranoside	$C_{17}H_{16}O_9$	365.08671	365.08641	$[M + H]^+$	0.81	201.03287, 159.17264, 131.10514	N
32	24.94	Natsudaidain-3-*O*-glucoside	$C_{27}H_{32}O_{14}$	581.18648	581.18599	$[M + H]^+$	0.85	419.13174, 273.07487, 153.01846	C
33	24.97	Narirutin-4′-*O*-glucoside	$C_{33}H_{42}O_{19}$	743.23931	743.23875	$[M + H]^+$	0.75	581.18682, 435.12794, 273.07526	C
34	25.16	Eriocitrin	$C_{27}H_{32}O_{15}$	597.18140	597.18096	$[M + H]^+$	0.73	435.12791, 289.06947, 153.01849	C
35	25.35	Resveratrol-4′-*O*-glucoside	$C_{20}H_{22}O_8$	389.12419	389.12479	$[M - H]^-$	−0.83		R

Table 1 (*Contd.*)

No.	t_R (min)	Identification	Formula	Monoisotopic mass (*m*/*z*)	Detected mass (*m*/*z*)	Ion mode	ppm	MS/MS (*m*/*z*)	Main source
								245.08196, 227.07138, 199.07567, 135.04384	
36	25.47	Isolindleyin	$C_{23}H_{26}O_{11}$	477.14024	477.14067	$[M-H]^-$	−0.90	313.05691, 211.02328, 169.01307, 125.02387	R
37	25.79	Lindleyin	$C_{23}H_{26}O_{11}$	477.14024	477.14071	$[M-H]^-$	−1.12	313.05659, 211.02454, 169.01340, 125.02321	R
38	26.19	Limonin	$C_{26}H_{30}O_8$	471.20134	471.20060	$[M+H]^+$	0.83	425.19343, 339.19426, 161.05897	C
39	26.19	Rhein-8-*O*-β-D-glucoside	$C_{21}H_{18}O_{11}$	445.07763	445.07816	$[M-H]^-$	−1.21	285.07559, 267.03326, 257.07879, 241.07041	R
40	26.28	Nicotiflorin	$C_{27}H_{30}O_{15}$	595.16575	595.16515	$[M+H]^+$	1.00	419.13403, 390.09214, 287.07785	C
41	26.28	Naringenin-7-*O*-glucoside	$C_{21}H_{22}O_{10}$	435.12857	435.12833	$[M+H]^+$	0.56	272.07462, 227.18968, 177.22102, 165.13608	C
42	26.29	Eriodictyol	$C_{15}H_{12}O_6$	289.07066	289.07051	$[M+H]^+$	0.53	153.01692, 135.06538, 107.04796	C
43	26.42	Limonin-17-β-D-glucoside	$C_{32}H_{42}O_{14}$	649.25018	649.25108	$[M-H]^-$	−1.39	487.19725, 425.18954, 339.18671	C
44	26.49	Neoeriocitrin	$C_{27}H_{32}O_{15}$	597.18140	597.18134	$[M+H]^+$	0.52	435.12786, 289.06814, 153.01794	C
45	26.55	Isosakuranetin	$C_{16}H_{14}O_5$	287.09140	287.09138	$[M+H]^+$	0.38	153.01718, 133.065095	C
46	27.07	Alhagidin	$C_{34}H_{44}O_{20}$	771.23532	771.23656	$[M-H]^-$	−1.61	635.18786, 609.19823, 161.02394	M
47	27.17	Cinnamoyl-6-*O*-galloyl-diglucoside	$C_{28}H_{32}O_{16}$	623.16176	623.16185	$[M-H]^-$	−1.50	475.10891, 331.06765, 313.05664	R
48	27.25	Nodakenin	$C_{20}H_{24}O_9$	407.13476	407.13536	$[M-H]^-$	−1.48	247.04625, 229.14536, 175.41575	N
49	27.29	Marmesin	$C_{14}H_{14}O_4$	247.09649	247.09655	$[M+H]^+$	−0.28	153.01714	C
50	27.38	Veronicastroside	$C_{27}H_{30}O_{15}$	595.16575	595.16502	$[M+H]^+$	0.84	577.15147, 457.10763, 295.05903	C
51	27.40	Sennoside B	$C_{42}H_{38}O_{20}$	861.18837	861.18831	$[M-H]^-$	−1.71	699.13514, 655.14385, 389.08791	R
52	27.44	Emodin-*O*-di-glucoside	$C_{27}H_{30}O_{15}$	593.15119	593.15112	$[M-H]^-$	−1.24	269.04549, 240.04318, 225.05493	R
53	28.01	Cassiachromone	$C_{13}H_{12}O_4$	231.06628	231.06653	$[M-H]^-$	−1.09	269.04549, 240.04321	R
54	28.96	Narirutin	$C_{27}H_{32}O_{14}$	581.18648	581.18594	$[M+H]^+$	0.94	435.12779, 419.13317, 273.07528	C
55	29.56	Isorhoifolin	$C_{27}H_{30}O_{14}$	579.17083	579.17056	$[M+H]^+$	0.47	449.14346, 303.08368, 153.01728	C
56	29.64	Chrysophanol-1,8-*O*-di-glucoside	$C_{27}H_{30}O_{14}$	577.15628	577.15658	$[M-H]^-$	−0.52	415.10387, 253.04962, 225.05395	R
57	30.04	Acteoside	$C_{29}H_{36}O_{15}$	623.19814	623.19902	$[M-H]^-$	−0.36	461.16314, 161.02265	M
58	30.15	1-Dihydro-*p*-coumaroyl-6-*O*-galloyl-glucoside	$C_{22}H_{24}O_{12}$	479.11950	479.12006	$[M-H]^-$	−1.16	461.10874, 313.05591, 169.01294, 125.02296	R
59	30.21	Prunin	$C_{21}H_{22}O_{10}$	435.12857	435.12887	$[M+H]^+$	−0.68	273.07329, 153.01674	C
60	30.22	Natsudaidai	$C_{21}H_{22}O_9$	419.13366	419.13349	$[M+H]^+$	0.39	390.09212, 389.09134, 371.08154	C
61	30.36	Naringin	$C_{27}H_{32}O_{14}$	581.18648	581.18605	$[M+H]^+$	0.75	435.12862, 419.13274, 273.07564	C
62	30.80	Sennoside A	$C_{42}H_{38}O_{20}$	885.18486	885.18538	$[M+Na]^+$	−0.59	701.13594, 657.14538, 391.08807, 270.03807	R
63	30.94	Isoacteoside	$C_{29}H_{36}O_{15}$	623.19814	623.19925	$[M-H]^-$	−1.77	461.16741, 161.02568	M
64	30.98	Laccaic acid-D-*O*-glucoside	$C_{22}H_{20}O_{12}$	475.08740	475.08820	$[M-H]^-$	−0.91	431.09851, 269.03452, 253.05063, 240.04252	R
65	31.45	Rhoifolin	$C_{27}H_{30}O_{14}$	579.17083	579.17057	$[M+H]^+$	0.57	433.10764, 271.05967, 153.02389	C
66	31.45	Neodiosmin	$C_{28}H_{32}O_{15}$	609.18140	609.18107	$[M+H]^+$	0.77	301.07091, 286.04659	C
67	31.48	Isosakuranin	$C_{22}H_{24}O_{10}$	449.14422	449.14408	$[M+H]^+$	0.96	301.06877, 287.09012, 286.04485	C
68	31.59	Hesperidin	$C_{28}H_{34}O_{15}$	611.19705	611.19650	$[M+H]^+$	0.10	449.14312, 303.08581, 288.09732, 153.02175,	C
69	32.03	Nodakenetin	$C_{14}H_{14}O_4$	245.08193	245.08221	$[M-H]^-$	−1.14	229.10047, 211.64807, 175.42487	N

Table 1 (*Contd.*)

No.	t_R (min)	Identification	Formula	Monoisotopic mass (*m*/*z*)	Detected mass (*m*/*z*)	Ion mode	ppm	MS/MS (*m*/*z*)	Main source
70	32.36	Physcion-1-*O*-glucuronic acid	$C_{22}H_{20}O_{11}$	459.09329	459.09368	$[M - H]^-$	−0.86	283.06124, 268.03815, 212.04737, 184.05123	R
71	32.78	Diosmin	$C_{28}H_{32}O_{15}$	607.16684	607.16710	$[M - H]^-$	−0.42	299.10025, 284.29568	C
				609.18140	609.18081	$[M + H]^+$	0.96	301.06863 286.04525	
72	32.89	Neohesperidin	$C_{28}H_{34}O_{15}$	611.19705	611.19637	$[M + H]^+$	0.53	449.14228, 303.08417, 153.01734	C
73	32.89	Hesperetin-7-*O*-glucoside	$C_{22}H_{24}O_{11}$	465.13914	465.13958	$[M + H]^+$	−0.92	303.08652, 288.03284, 153.01532	C
74	32.90	Sakuranetin	$C_{22}H_{24}O_{10}$	449.14422	449.14477	$[M + H]^+$	−1.22	301.06945, 287.08765, 286.045473	C
75	32.97	Magnoloside E	$C_{28}H_{34}O_{15}$	609.18249	609.18261	$[M - H]^-$	0.19	447.15213, 315.11019, 161.02613	M
76	33.05	Homoeriodictyol	$C_{16}H_{14}O_6$	303.08631	303.08630	$[M + H]^+$	0.06	288.04856, 153.01712, 117.03375	C
77	33.18	Limocitrin-3-*O*-(3-hydroxy-3-methylglutarate)-glucoside	$C_{29}H_{32}O_{17}$	653.17123	653.17394	$[M + H]^+$	−0.73	539.13891, 347.07494	C
78	33.40	1-Dihydrocinnamoyl-6-*O*-galloyl-glucoside	$C_{22}H_{24}O_{11}$	463.12459	463.12503	$[M - H]^-$	−0.97	331.06693, 313.05656, 211.02564, 169.01332	R
79	34.30	Magnolignan B	$C_{18}H_{20}O_5$	315.12380	315.12417	$[M - H]^-$	−0.95	267.10167, 249.09178, 221.09658, 133.06619	M
80	34.51	Meranzin	$C_{15}H_{16}O_4$	261.11214	261.11203	$[M + H]^+$	0.42	243.10142, 159.04375, 103.05456	C
81	34.59	Hesperidone hydrate	$C_{15}H_{18}O_5$	296.14925	296.14925	$[M + HH_4]^+$	0.00	403.12656, 418.12351, 165.05352	C
82	35.89	Diosmetin-7-*O*-(6″-*O*-acetyl) neohesperidoside	$C_{30}H_{34}O_{17}$	667.18688	667.18653	$[M + H]^+$	−0.26	301.07182, 286.05412	C
83	36.14	Natsudaidain-3-*O*-(3-hydroxy-3-methylglutarate)-glucoside	$C_{33}H_{40}O_{18}$	725.22874	725.22824	$[M + H]^+$	0.47	581.18621, 419.13224	C
84	36.29	Magnolignan A	$C_{18}H_{20}O_4$	299.12888	299.12924	$[M - H]^-$	−1.18	239.10865, 221.10746	M
85	37.01	Physcion-8-*O*-glucuronic acid	$C_{22}H_{20}O_{11}$	459.09329	459.09369	$[M - H]^-$	−0.89	283.06138, 268.03804, 239.03501, 224.04837	R
86	38.08	Cinnamoyl-6-*O*-galloyl-glucoside	$C_{22}H_{22}O_{11}$	461.10894	461.10948	$[M - H]^-$	−1.18	401.08794, 313.05874, 285.03987, 271.04612	R
87	39.00	Didymin	$C_{28}H_{34}O_{14}$	595.20213	595.20196	$[M + H]^+$	0.70	433.14783, 287.09001, 153.01794	C
88	39.08	Beta-hydroxyacteoside	$C_{29}H_{36}O_{16}$	639.19306	639.19315	$[M - H]^-$	−0.14	503.14287, 477.16173, 161.02408, 133.03174	M
89	39.39	6-Methoxy-8-*O*-β-D-glucoside	$C_{20}H_{24}O_9$	407.13476	407.13521	$[M - H]^-$	−1.12	253.04989, 201.09011, 146.96598	R
90	39.47	Magnoloside A	$C_{29}H_{36}O_{15}$	623.19705	623.19645	$[M + H]^+$	0.95	461.16607, 161.02574	M
91	39.51	Chrysophanol isomer	$C_{15}H_{10}O_4$	253.05063	253.05093	$[M - H]^-$	−1.23	225.05517, 210.03072, 197.06054	R
92	39.52	Chrysophanol-1-*O*-glucoside	$C_{21}H_{20}O_9$	415.10346	415.10402	$[M - H]^-$	−1.35	253.04884, 225.05627, 209.05942	R
93	39.70	Emodin-*O*-glucoside	$C_{21}H_{20}O_{10}$	431.09837	431.09867	$[M - H]^-$	−0.7	269.04562, 241.04997, 225.05524	R
94	39.74	3,5,6,7,8,3′,4′-Heptamethoxy flavone	$C_{22}H_{24}O_9$	433.14931	433.14910	$[M + H]^+$	0.49	418.12356, 403.12656, 165.05354	C
95	39.79	1-*P*-Coumaroyl-6-*O*-galloyl-di-glucoside	$C_{29}H_{36}O_{16}$	639.19306	639.19367	$[M - H]^-$	−0.96	477.13972, 313.05688, 211.02412, 141.03624	R
96	39.87	Sakuranin	$C_{16}H_{14}O_5$	287.09142	287.09127	$[M + H]^+$	0.46	153.01721, 133.06492	C
97	39.88	Poncirin	$C_{28}H_{34}O_9$	595.20213	595.20168	$[M + H]^+$	0.73	433.14812, 287.09110, 153.16712	C
98	39.93	Magnoloside M	$C_{29}H_{36}O_{15}$	623.19705	623.19643	$[M + H]^+$	0.99	461.16597, 161.02576	M
99	39.98	6′-*O*-*trans*-Feruloylnodakenin	$C_{30}H_{32}O_{12}$	583.18213	583.18148	$[M - H]^-$	1.07	247.10278, 229.44072, 175.28676	N
				607.17865	607.17861	$[M + Na]^+$	−0.02	249.11947, 177.29759	
100	40.24	Chrysophanol	$C_{15}H_{10}O_4$	253.05063	253.05094	$[M - H]^-$	−1.21	225.05537, 210.03104, 202.88598	R
101	40.93	Marmin	$C_{19}H_{24}O_5$	333.16965	333.16942	$[M + H]^+$	0.70	163.03837, 107.08582	C

Table 1 (*Contd.*)

No.	t_R (min)	Identification	Formula	Monoisotopic mass (*m*/*z*)	Detected mass (*m*/*z*)	Ion mode	ppm	MS/MS (*m*/*z*)	Main source
102	41.15	2-(2′-hydroxypropyl)-5-methyl-7-hydroxychromone	$C_{13}H_{14}O_4$	233.08193	233.08217	$[M-H]^-$	−1.01	189.05568, 146.96597, 129.97617	R
103	41.29	Magnolignan C	$C_{18}H_{20}O_4$	299.12888	299.12928	$[M-H]^-$	−1.33	258.09247, 239.10852, 197.06244	M
104	41.55	Naringenin	$C_{15}H_{12}O_5$	273.07575	273.07562	$[M+H]^+$	0.48	153.17823, 133.06712	C
105	42.12	Procyanidin B	$C_{30}H_{26}O_{12}$	577.13515	577.13535	$[M-H]^-$	−0.49	425.08747, 407.07637, 289.07107	R
106	42.26	Physcion-8-*O*-glucuronic acid methyl ester	$C_{23}H_{22}O_{11}$	473.10894	473.10946	$[M-H]^-$	−1.11	283.06094, 268.03811, 240.04325	R
107	42.27	Emodin-*O*-malonyl-glucose	$C_{24}H_{22}O_{13}$	517.09876	517.09935	$[M-H]^-$	−1.15	473.10884, 431.09157, 269.04531, 241.05131	R
108	42.51	Chrysophanol-8-*O*-glucoside	$C_{21}H_{20}O_9$	415.10346	415.10400	$[M-H]^-$	−1.30	253.04984, 225.05618, 209.05959	R
109	42.51	Emodin	$C_{15}H_{10}O_5$	269.04555	269.04588	$[M-H]^-$	−1.22	241.05016, 225.05524, 197.06027	R
110	42.53	Aloe-emodin-*O*-glucoside	$C_{21}H_{20}O_{10}$	431.09885	431.09886	$[M-H]^-$	−1.14	269.04462, 239.03647, 211.02524	R
111	42.61	Physcion-1-*O*-glucoside	$C_{22}H_{22}O_{10}$	445.11402	445.11460	$[M-H]^-$	−1.30	427.03257, 283.06205, 269.04607	R
112	42.61	Physcion	$C_{16}H_{12}O_5$	283.06120	283.06162	$[M-H]^-$	−1.49	283.07696, 268.04557, 240.04266	R
113	42.62	Hesperetin	$C_{16}H_{14}O_6$	303.08631	303.08608	$[M+H]^+$	0.77	153.01741, 110.04783	C
114	42.85	Magnatriol	$C_{15}H_{14}O_3$	241.08702	241.08730	$[M-H]^-$	−1.17	223.07597, 197.09716, 133.06719	M
115	42.97	Magnolignan E	$C_{18}H_{18}O_4$	297.11323	297.11365	$[M-H]^-$	−1.39	225.09132, 184.05786, 183.04759	M
116	43.29	5-Hydroxy-6,7,3′,4′,5′-pentamethoxyflavone	$C_{20}H_{20}O_8$	389.12309	389.12283	$[M+H]^+$	0.67	374.09631, 359.08221, 356.08174, 328.07931	C
117	43.54	5-*o*-Demethylnobiletin	$C_{20}H_{20}O_7$	373.12818	373.12781	$[M+H]^+$	0.73	358.10001, 343.07224, 325.06554, 297.06973	C
118	43.78	Demethylnobiletin	$C_{20}H_{20}O_8$	389.12309	389.12282	$[M+H]^+$	0.79	359.08112, 360.07831, 341.06984	C
119	44.32	Obovatol	$C_{18}H_{18}O_3$	281.11832	281.11866	$[M-H]^-$	−1.20	267.10174, 249.08513	M
120	44.54	Sinensetin	$C_{20}H_{20}O_7$	373.12818	373.12790	$[M+H]^+$	0.74	175.01549, 147.03194, 119.01593	C
121	44.59	Honokiol	$C_{18}H_{18}O_2$	265.12340	265.12413	$[M-H]^-$	−0.42	224.08126, 223.07514, 197.05923	M
122	44.59	*O*-Prenyl-umbelliferone	$C_{14}H_{14}O_3$	229.08072	229.08726	$[M-H]^-$	−1.04	160.01707, 142.17814	N
123	44.74	Aloe-emodin	$C_{15}H_{10}O_5$	269.04555	269.04598	$[M-H]^-$	−1.60	253.05121, 240.04247, 211.03968	R
124	44.94	Magnaldehyde D	$C_{16}H_{14}O_3$	253.08702	253.08735	$[M-H]^-$	−1.32	235.07963, 207.08634, 194.04012	M
125	45.13	Phelloptein	$C_{17}H_{16}O_5$	323.08899	323.08875	$[M+Na]^+$	0.76	269.07627, 231.01796	N
126	45.25	Notopterol	$C_{21}H_{22}O_5$	353.13945	353.13988	$[M-H]^-$	−1.21	203.17578, 159.09786, 147.63478	N
				377.13594	377.13559	$[M+Na]^+$	0.95	205.20652, 161.11072	
127	45.38	Rhein	$C_{15}H_8O_6$	283.02481	283.02504	$[M-H]^-$	−0.82	257.04507, 239.03461, 211.03958	R
128	45.38	Danthron	$C_{14}H_8O_4$	239.03498	239.03530	$[M-H]^-$	−1.33	186.73096, 141.51394, 130.71196	R
129	45.58	Nobiletin	$C_{21}H_{22}O_8$	403.13874	403.13819	$[M+H]^+$	1.41	373.09359, 355.08584, 327.08476	C
130	45.69	Nomilin	$C_{28}H_{34}O_9$	515.22756	515.22753	$[M+H]^+$	0.05	469.21806, 455.20675, 411.21462, 161.05854	C
131	45.88	Notoptol	$C_{21}H_{22}O_5$	353.13945	353.13987	$[M-H]^-$	−1.19	225.17472, 207.13978, 189.14075	N
				377.13594	377.13563	$[M+Na]^+$	0.97	227.19076, 209.14289	
132	46.17	Magnaldehyde E	$C_{16}H_{14}O_3$	253.08702	253.08735	$[M+H]^+$	−1.32	225.09424, 184.05624, 183.05074	M
133	46.17	Magnolol	$C_{18}H_{18}O_2$	265.12340	265.12394	$[M-H]^-$	−0.39	247.10096, 245.09517, 223.07491	M
134	46.19	*p*-Hydroxyphenethyl anisate	$C_{16}H_{16}O_4$	295.09408	295.09386	$[M+Na]^+$	0.73	153.07368, 138.08654	N
135	46.54	Obovaaldehyde	$C_{16}H_{14}O_4$	269.08193	269.08215	$[M-H]^-$	−0.80	152.01478, 124.02018	M
136	46.70	Isosinensetin	$C_{20}H_{20}O_7$	373.12818	373.12783	$[M+H]^+$	0.94	358.10398, 343.08158, 315.08276	C
137	46.70	Tangeretin	$C_{20}H_{20}O_7$	395.11012	395.10998	$[M+Na]^+$	0.35	278.54947, 276.55982, 243.12708	C

[a] Ps: C represented *Aurantii fructus immaturus*; R represented *Rheum palmatum* L.; M represented *Magnoliae Officmalis Cortex*; N represented *Notopterygii Rhizoma Et Radix*.

cleaving of the glycosidic linkage and losing $C_6H_{10}O_5$ (162 Da) from *m/z* 431.09157, *m/z* 241.05131 is the result of losing CO (28 Da) at *m/z* 269.04531. The characterization of anthraquinone compounds in SHD is inferred from this cleavage rule and related literatures.[20,21]

3.4 Characterization of glucosides in SHD

Glucosides are formed by the esterification of acid with glucose. According to the type of acid, it can be divided into cinnamoyl-glucose, dihydrocinnamoyl-glucose, *p*-cinnamoyl-glucose, dihydro-*p*-cinnamoyl glucose, galloyl-glucose, and resveratrol-galloyl-glucose in rhubarb. They are vulnerable to loss of neutral fragments 148 Da, 150 Da, 164 Da, 166 Da, 170 Da, and 228 Da, respectively. Galloyl-glucose was selected to illustrate the fragmentation pathways. In negative ion mode, adduct ion at *m/z* 331.06759 ($[M - H]^-$) indicating an elemental composition of $C_{13}H_{16}O_{10}$. The mass spectrum (Fig. 3D) shows that the main fragment ions are *m/z* 313.05289, 271.04772, 211.02436, 169.01338, and 125.00325. Among them, *m/z* 313.05289 is presumed by losing H_2O (18 Da) from $[M - H]^-$, *m/z* 271.04772 is inferred by losing $C_2H_4O_2$ (60 Da) from $[M - H]^-$, *m/z* 211.02436 is the result of losing $C_4H_8O_4$ (120 Da) from $[M - H]^-$, *m/z* 169.01338 is speculated by cleaving of the glycosidic linkage and losing glucose $C_6H_{10}O_5$ (162 Da) from $[M - H]^-$, and *m/z* 125.00325 is deduced by losing CO_2 (44 Da) from *m/z* 169.01338. The characterization of glucosides in SHD is concluded by referring to the cleavage rule and related literature.[21]

3.5 Characterization of epicatechin in SHD

Epicatechin was selected to illustrate the fragmentation pathways for catechins. The adduct ion at 289.07213 ($[M - H]^-$) indicating an elemental composition of $C_{15}H_{14}O_6$, and the MS/MS spectrum (Fig. 3E) is presented that the main fragment ions are *m/z* 245.08235, 203.07153. Among them, *m/z* 245.08235 is presumed by losing CO_2 (44 Da) from $[M - H]^-$, and *m/z* 203.07153 is inferred by losing of C_2H_2O (42 Da) from *m/z* 245.08235. The characterization of catechins in SHD is concluded by referring to the cleavage rule and related literatures.[21–23]

3.6 Characterization of lignans in SHD

Most of the lignans have the presence of allyl and hydroxyl groups, which relative positions are different, their cleavage methods will be different. Honokiol and magnolol were selected to illustrate the fragmentation pathways. Magnolol showed *m/z* 265.12394 ($[M - H]^-$) in the negative ion mode, which indicated the elemental composition of $C_{18}H_{18}O_2$. The mass spectrum (Fig. 3F) shows that the main fragment ions are *m/z* 247.10096, 245.09517, 223.07491. Among them, *m/z* 247.10096 is presumed by losing H_2O (18 Da) from $[M - H]^-$, *m/z* 245.09517 is inferred by losing H_2 (2 Da) from *m/z* 247.10096, and *m/z* 223.07491 is obtained by losing C_3H_6 (42 Da) from $[M - H]^-$. Honokiol showed *m/z* 265.12413 ($[M - H]^-$) in the negative ion mode, which indicated the elemental composition of $C_{18}H_{18}O_2$. The mass spectrum (Fig. 3G) shows that the main fragment ions are *m/z* 224.08126, 223.07514, 197.05923. Among them, *m/z* 224.08126 is presumed by losing C_3H_5 (41 Da) from $[M - H]^-$, *m/z* 223.07514 is inferred by losing C_3H_6 (42 Da) from $[M - H]^-$, and *m/z* 197.05923 is obtained by losing of C_2H_2 (26 Da) by *m/z* 223.07514. The characterization of lignans in SHD is concluded by referring to the cleavage rule and related literatures.[24,25]

3.7 Characterization of phenylpropanoid glycosides in SHD

The phenylpropanoid glycosides are composed of glucose and phenethyl alcohol, which mainly based on the loss of coffee groups (162 Da). Verbascoside is chosen as an example to explain the cleavage pathway. It showed *m/z* 623.19814 ($[M - H]^-$) in the negative ion, which indicated an elemental composition of $C_{29}H_{36}O_{15}$. The mass spectrum (Fig. 3H) shows that its main fragment ion is *m/z* 461.16314, 161.02265. Among them, *m/z* 461.16314 is presumed by losing $C_9H_6O_3$ (162 Da) from $[M - H]^-$, and *m/z* 161.02265 is inferred by losing $C_{20}H_{30}O_{12}$ (462 Da) from $[M - H]^-$. The characterization of phenylpropanoid glycosides in SHD is concluded by referring to the cleavage rule and related literatures.[24,25]

3.8 Characterization of alkaloids in SHD

The main rupture method of alkaloids is the loss of N. (*R*)-Oblongine is chosen as an example to explain the cleavage pathway. It showed an *m/z* 314.17473 ($[M + H]^+$) in the positive ion mode, which indicated the elemental composition of $C_{19}H_{24}NO_3^+$. The mass spectrum (Fig. 3I) shows that the main fragment ions are *m/z* 269.11625, 237.09024, 175.07485, 107.04861. Among them, *m/z* 269.11625 is presumed by losing C_2H_6NH (45 Da) from $[M + H]^+$, *m/z* 237.09024 is inferred by losing CH_4O (32 Da) at *m/z* 269.11625, *m/z* 175.07485 is obtained by losing C_6H_6O (94 Da) from *m/z* 269.11625, and *m/z* 107.04861 is speculated by losing $C_{12}H_{18}NO_2$ (207 Da) from $[M + H]^+$. The characterization of alkaloids in SHD is concluded by referring to the cleavage rule and related literature.[25]

3.9 Characterization of coumarins in SHD

The main cleavage mode of coumarins is loss of CO and CO_2, while coumarin glycosides lose glucose firstly, and then break by the above-mentioned cleavage law. Bergaptol is chosen as an example to explain the cleavage pathway. It showed an *m/z* 201.01957 ($[M - H]^-$) in the negative ion mode, which indicated an elemental composition of $C_{11}H_6O_4$. The mass spectrum (Fig. 3J) shows that the main fragment ions are *m/z* 185.02456, 157.17256, 129.10451, 101.96320. Among them, *m/z* 185.02456 is presumed by losing OH (17 Da) from $[M - H]^-$, *m/z* 157.17256 is presumed by losing CO (28 Da) from *m/z* 185.02456, *m/z* 129.10451 is inferred by losing CO (28 Da) from *m/z* 157.17256, and *m/z* 101.96320 is obtained by losing CO (28 Da) from *m/z* 129.10451. The characterization of coumarins in SHD is concluded by referring to the cleavage rule and related literatures.[26–28]

4. Conclusions

In our study, a rapid, swift and straightforward analytical method with the help of UHPLC-FT-ICR-MS/MS was successfully developed for the first time to separate and identify the chemical constituents of SHD. A total of 137 compounds in SHD were identified or tentatively characterized, and 11 compounds were accurately identified by reference. The research indicated that SHD mainly contains 42 flavonoids (in *Aurantii fructus immaturus* and *Rheum palmatum* L.), 3 triterpenoids (in *Aurantii fructus immaturus*), 23 anthraquinones (in *Rheum palmatum* L.), 14 glucosides, 5 catechins and 2 anthranone (in *Rheum palmatum* L.), 11 phenylpropanoid glycosides, 5 alkaloids and 11 lignans (in *Magnoliae Officmalis Cortex*), 16 coumarins (in *Notopterygii Rhizoma Et Radix* and in *Aurantii fructus immaturus*),which has laid the foundation for prospective research associated with SHD. And it is hoped to be useful to identify and characterize the components in other TCMs. And it provided a basis for finding the prototype components and metabolites in the serum of containing SHD.

In this study, the structure of the compound, except for the comparison of the reference, was obtained through the combination of retention time, accurate primary and secondary mass spectrometry information, cracking rules of the same compound and literature comparison. The identification of compound structure by this method is limited to known compounds, because this method only mass spectrometry for identification.

Abbreviations

TCMs	Traditional Chinese medicines
SHD	Sanhua decoction
Zhishi	*Aurantii fructus immaturus*
Dahuang	*Rheum palmatum* L
Houpu	*Magnoliae Officmalis Cortex*
Qianghuo	*Notopterygii Rhizoma Et Radix*
BBB	Brain blood barrier
UHPLC-FT-ICR-MS/MS	Ultra high performance liquid chromatography coupled with Fourier transform ion cyclotron resonance mass spectrometry
BPC	Base peak ion chromatograms
EIC	Extract ion chromatograms
RDA	Diels–Alder reaction

Conflicts of interest

The authors declare that there are no conflicts of interest.

Acknowledgements

The authors gratefully acknowledge the facilities and assistance provided by Dr Fei Han of the Bruker Corporation.

References

1 L. Zhang, J. B. Yan, X. M. Liu, Z. G. Ye, X. H. Yang, R. Meyboom, K. Chan, D. Shaw and P. Duez, *J. Ethnopharmacol.*, 2012, **140**, 519–525, DOI: 10.1016/j.jep.2012.01.058.

2 C. Zhang, G. Q. Zheng and H. J. Huang, *Chin. J. Integr. Tradit. West. Med. Intensive Crit. Care*, 2007, **6**, 352–356, DOI: 10.3321/j.issn:1008-9691.2007.06.009.

3 J. Bi, H. Zhang, J. Lu and W. Lei, *Mol. Med. Rep.*, 2016, **14**, 5408–5414, DOI: 10.3892/mmr.2016.5919.

4 Y. Ma, X. Xia, J. M. Cheng and Y. Q. Kuang, *Neurochem. Res.*, 2014, **39**, 1809–1816, DOI: 10.1007/s11064-014-1395-y.

5 X. Liu, X. Chen, Y. Zhu, K. Wang and Y. Wang, *Metab. Brain Dis.*, 2017, **32**, 1109–1118, DOI: 10.1007/s11011-017-0004-6.

6 X. Wu, Y. B. Zhang, L. Zhang and X. W. Yang, *J. Chromatogr. B: Anal. Technol. Biomed. Life Sci.*, 2018, **1092**, 244–251, DOI: 10.1016/j.jchromb.2018.06.006.

7 S. Forcisi, F. Moritz, B. Kanawati, D. Tziotis, R. Lehmann and P. Schmitt-Kopplin, *J. Chromatogr. A*, 2013, **1292**, 51–56, DOI: 10.1016/j.chroma.2013.04.017.

8 D. F. Smith, A. Kiss, F. E. Leach, E. W. Robinson, L. PasaTolic and R. M. Heeren, *Anal. Bioanal. Chem.*, 2013, **405**, 6069–6076, DOI: 10.1007/s00216-013-7048-1.

9 Y. N. Wang, M. Zhao, Y. B. Yu, M. Wang and C. J. Zhao, *RSC Adv.*, 2016, **6**, 39642–39651, DOI: 10.1039/c6ra01428c.

10 Y. N. Wang, M. Zhao, Y. F. Ou, B. W. Zeng, X. Y. Lou, M. Wang and C. J. Zhao, *J. Chromatogr. B: Anal. Technol. Biomed. Life Sci.*, 2016, **1020**, 120–128, DOI: 10.1039/c6ra01428c.

11 Y. Li, Y. Y. Zhao, X. Li, T. F. Liu, X. W. Jiang and F. Han, *J. Pharm. Biomed. Anal.*, 2017, **149**, 318–328, DOI: 10.1016/j.jpba.2017.10.032.

12 Z. Guan, M. Wang, Y. Cai, H. M. Yang, M. Zhao and C. J. Zhao, *J. Chromatogr. B: Anal. Technol. Biomed. Life Sci.*, 2018, **1086**, 11–22, DOI: 10.1016/j.jchromb.2018.04.009.

13 T. Liu, X. M. Tian, Z. Q. Li, F. Han, B. Ji, Y. L. Zhao and Z. G. Yu, *J. Chromatogr. B: Anal. Technol. Biomed. Life Sci.*, 2018, **1079**, 69–84, DOI: 10.1016/j.jchromb.2018.02.001.

14 H. F. Chen, W. G. Zhang, J. B. Yuan, Y. G. Li, S. L. Yang and W. L. Yang, *J. Pharm. Biomed. Anal.*, 2012, **59**, 90–95, DOI: 10.1016/j.jpba.2011.10.013.

15 Y. He, Z. Li, W. Wang, S. Sooranna, Y. Shi, Y. Chen, Y. Chen, C. Q. Wu, J. G. Zeng, Q. Tang and H. Q. Xie, *Molecules*, 2018, **23**, 2189, DOI: 10.3390/molecules23092189.

16 R. Tong, M. Peng, C. Tong, K. K. Guo and S. Y. Shi, *J. Chromatogr. B: Anal. Technol. Biomed. Life Sci.*, 2018, **1077–1078**, 1–6, DOI: 10.1016/j.jchromb.2018.01.031.

17 W. Y. Liu, C. Zhou and C. M. Yan, *Chin. J. Nat. Med.*, 2012, **10**, 456–463, DOI: 10.1016/S1875-5364(12)60087-9.

18 X. J. Zhao, Q. Q. Liu and T. Xing, *J. Southwest Jiaot. Univ.*, 2018, **40**, 20–29, DOI: 10.13718/j.cnki.xdzk.2018.11.003.

19 Y. Jiang, R. Liu, J. Chen, M. Liu, B. Liu, L. Z. Yi and S. Liu, *J. Chromatogr. A*, 2019, **1600**, 197–208, DOI: 10.1016/j.chroma.2019.04.051.

20 Y. Liu, L. Li, Y. Q. Xiao, J. Q. Yao, P. Y. Li, D. R. Yu and Y. L. Ma, *Food Chem.*, 2016, **192**, 531–540, DOI: 10.1016/j.foodchem.2015.07.013.

21 L. Zhang, H. Y. Liu, L. L. Qin, Z. X. Zhang, Q. Wang, Q. Q. Zhang, Z. W. Lu, S. L. Wei, X. Y. Gao and P. F. Tu, *J. Sep. Sci.*, 2015, **38**, 511–522, DOI: 10.1002/jssc.201400971.

22 J. Han, M. Ye, M. Xu, X. Qiao, H. B. Chen, B. R. Wang, J. H. Zheng and D. A. Guo, *Planta Med.*, 2008, **74**, 873–879, DOI: 10.1097/BCR.0b013e31818480b8.

23 Y. Li, M. Wang, Q. Zhang, Y. Tian, F. G. Xu and Z. J. Zhang, *Anal. Methods*, 2014, **6**, 1720–1727, DOI: 10.1039/c3ay41986j.

24 K. Guo, C. Tong, Q. Fu, J. Xu, S. Shi and Y. Xiao, *J. Pharm. Biomed. Anal.*, 2019, **170**, 153–160, DOI: 10.1016/j.jpba.2019.03.044.

25 K. P. Sun, K. M. Qin, W. D. Li, S. Y. Peng, J. Jin, B. Yang and B. C. Cai, *World Chin. Med.*, 2019, **14**, 287–291, DOI: 10.3969/j.issn.1673-7202.2019.02.007.

26 Y. H. Li, S. Y. Jiang, Y. L. Guan, X. Liu, L. M. Zhou, S. X. Huang, H. D. Sun, S. L. Peng, Y. Zhou, 2006, **64**, 405–411. DOI: 10.1365/s10337-006-0052-2.

27 K. Xu, S. Jiang, Y. Zhou, B. Xia, X. Xu, Y. Zhou, Y. Li, M. Wang and L. Ding, *J. Pharm. Biomed. Anal.*, 2011, **56**, 1089–1093, DOI: 10.1016/j.jpba.2011.07.034.

28 X. Liu, S. Jiang, K. Xu, H. Sun, Y. Zhou, X. Xu, J. Yi, Y. Gu and L. S. Ding, *J. Ethnopharmacol.*, 2009, **126**, 474–479, DOI: 10.1016/j.jep.2009.09.011.