Supplementary material

Matrix-assisted Laser Desorption/Ionization Time-of-Flight Mass Spectrometry Combined with Chemometrics to Identify the Origin of Chinese Medicinal Materials

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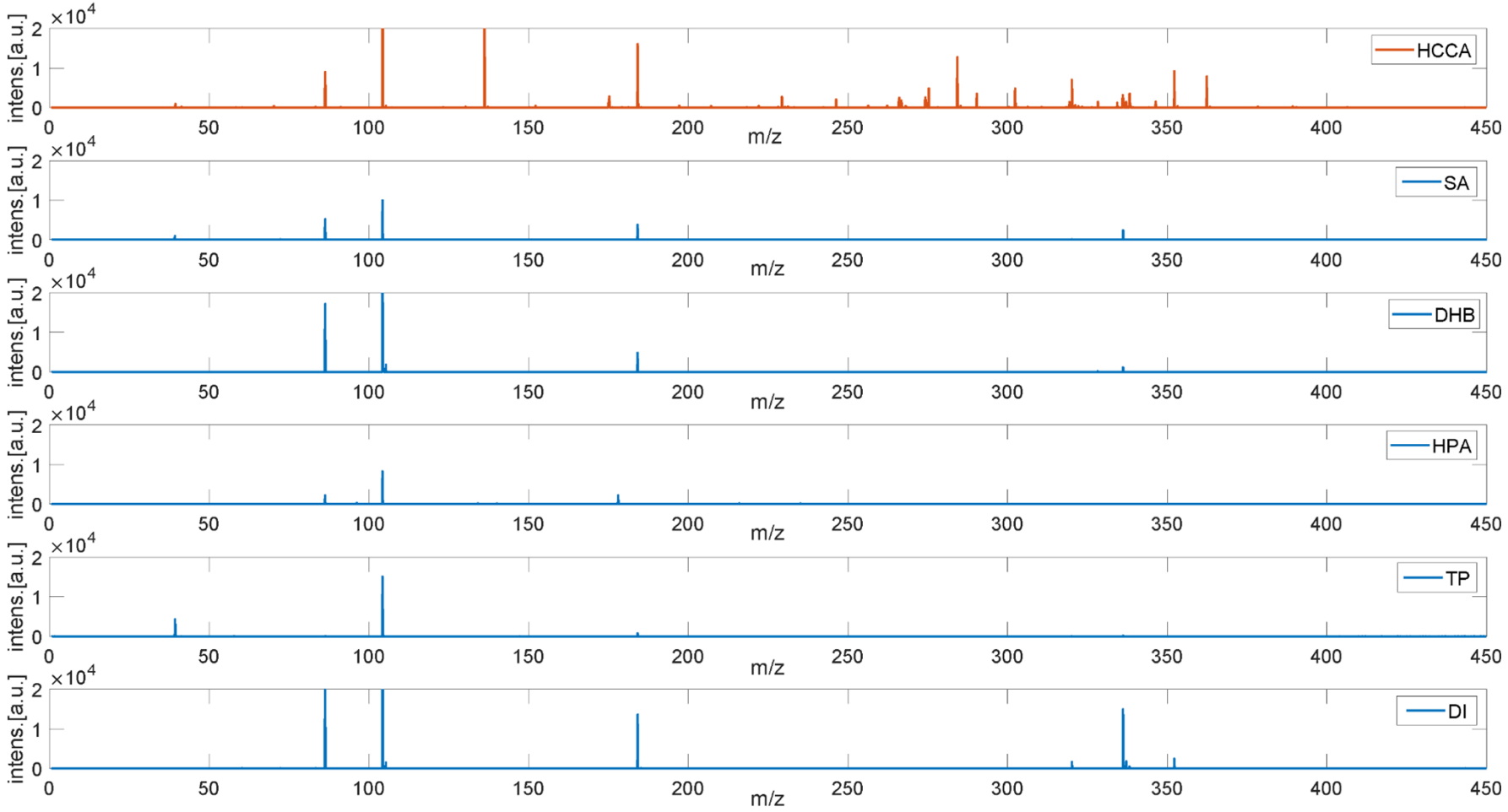


Figure S1. The MALDI-TOF MS profiles of AMK using different matrix (HCCA, cyano-4-hydroxycinnamic acid; SA, sinapinic acid, DHB, 2,5-dihydroxylbenzoic acid; HPA, 3-hydroxypicolinic acid; TP, 5,10,15,20-tetrakis (pentafluorophenyl) porphyrin; DI, dithranol)

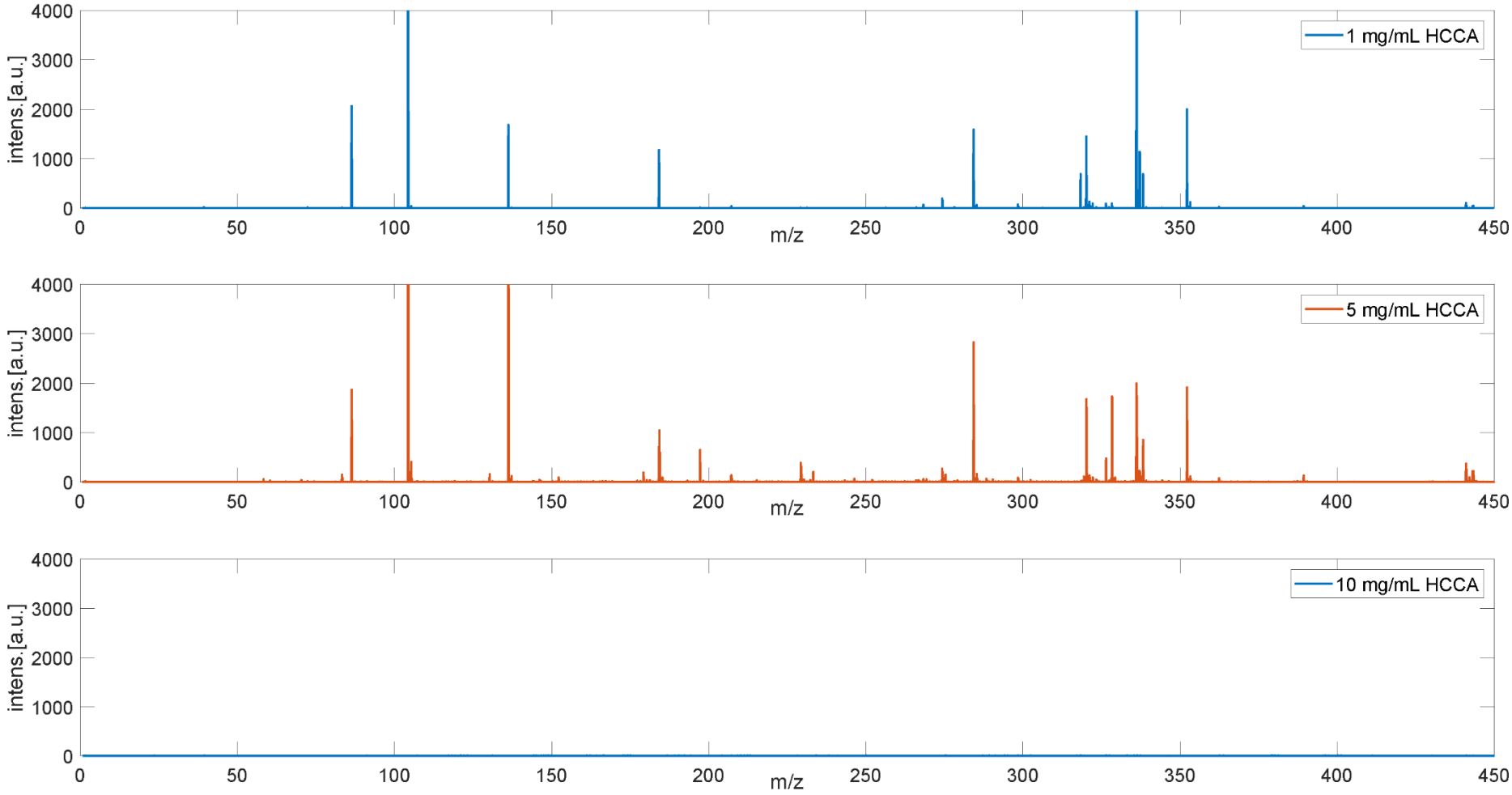


Figure S2. The MALDI-TOF MS profiles of AMK using different HCCA concentrations

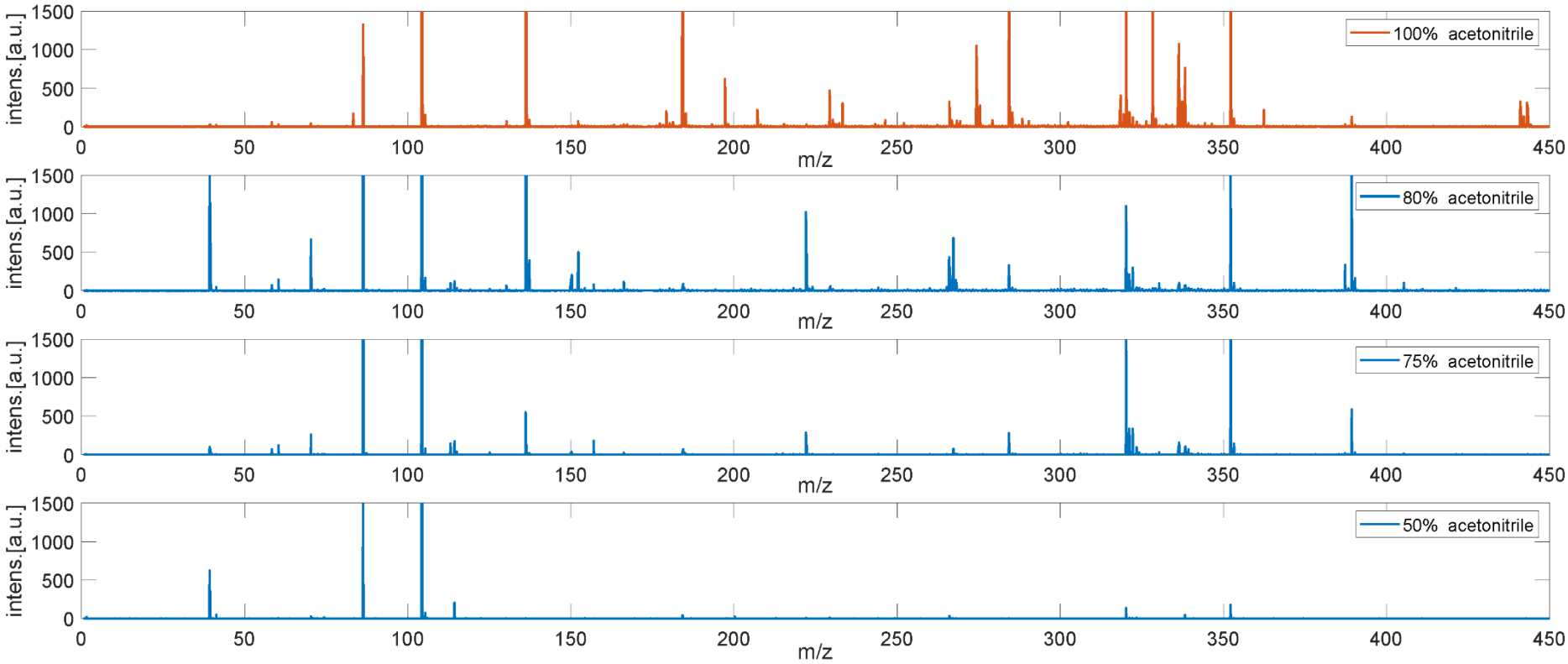


Figure S3. The MALDI-TOF MS profiles of AMK using solvent of acetonitrile and water with different volume ratio

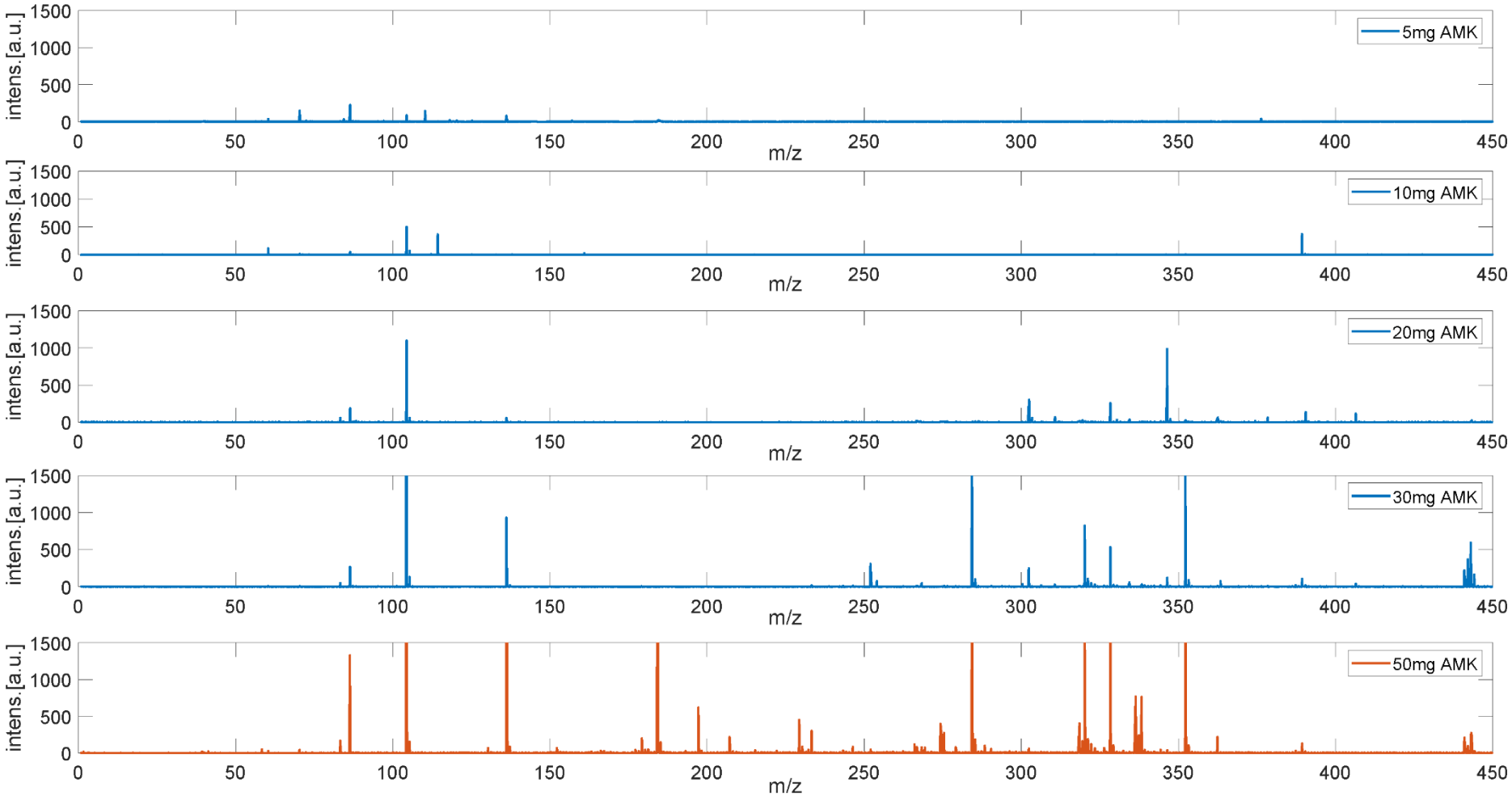


Figure S4. The MALDI-TOF MS profiles of AMK with different weight

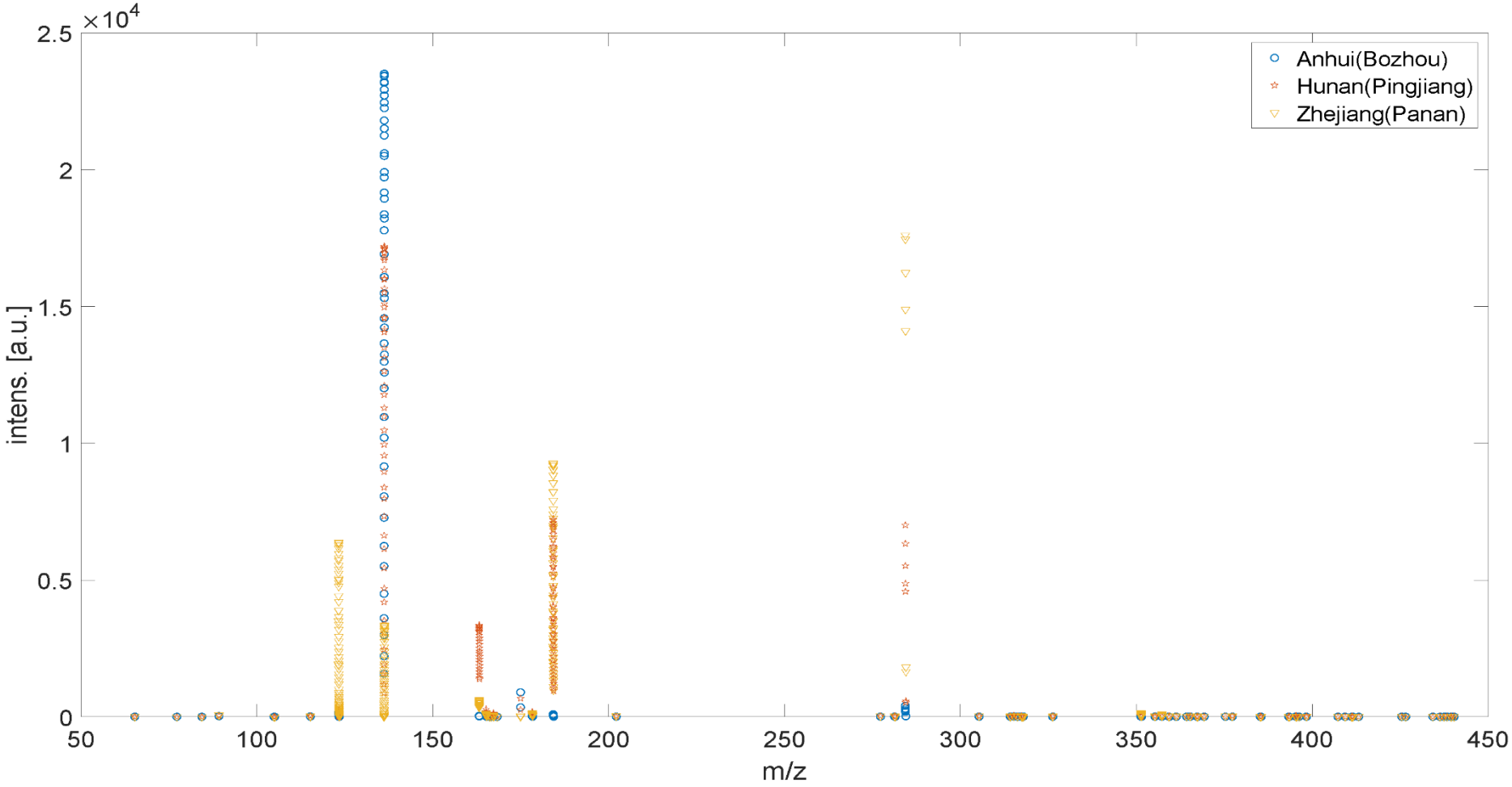


Figure S5. The selected 1000 variables of AMK samples from 3 different origins

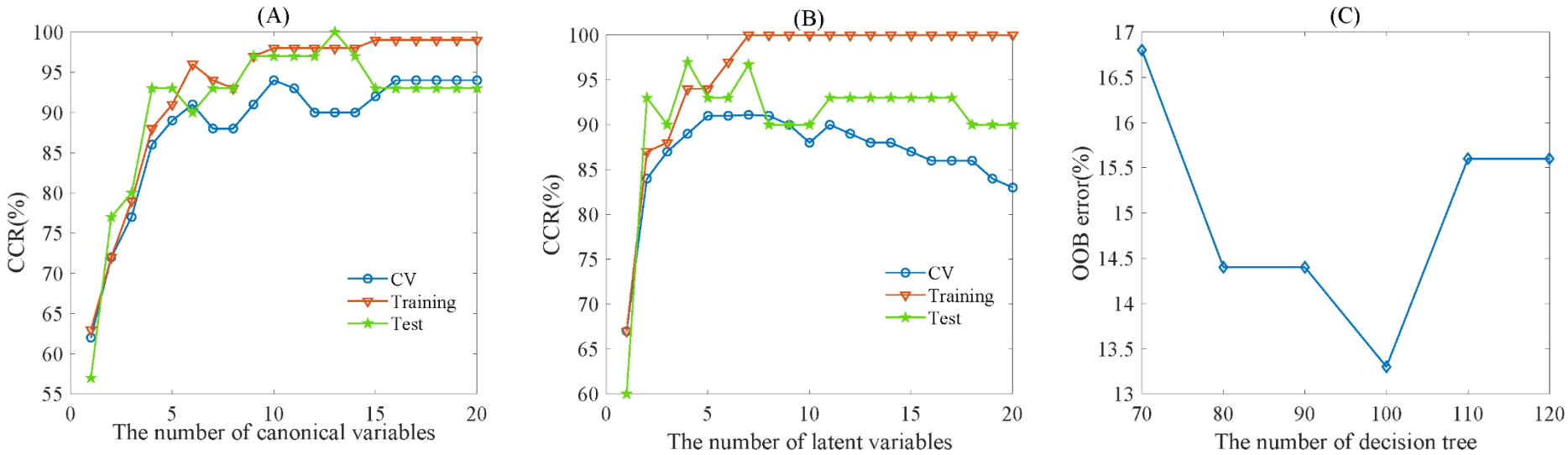
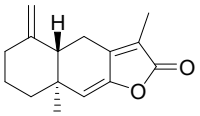
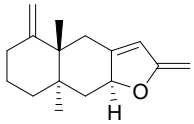
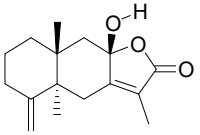
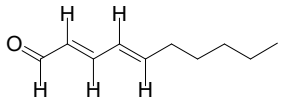
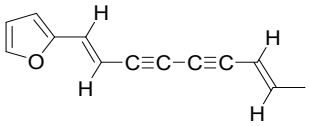
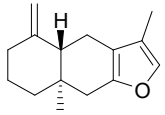
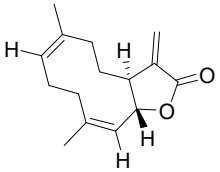
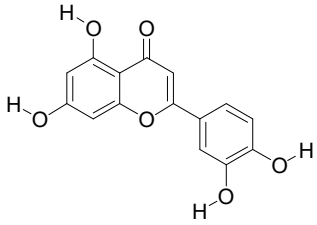
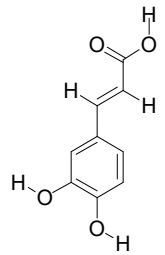
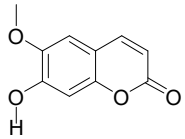
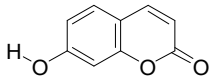
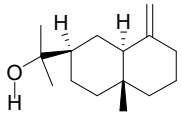


Figure S6. Correct classification rate (CCR) obtained by cross-validation in the cross-validation (CV), training set and test set, respectively, as a function of (A) the *CVs* of PCA-LDA, (B) the *LVs* of PLS-DA and (C) the number of decision tree for RF.

Table S1. The information about bioactive compounds in AMK

Compounds (abbreviations)	Structural formula	Molecular weight	Manufacturers	Solvent	The concentration of solution (μg/mL)	CAS No.	Class
Atractylenolide I (AT I)		230.1301	Yuanye (China, Shanghai)	Methanol/ Acetonitrile	252	73069-13-3	Sesquiterpenoids
Atractylenolide II (AT II)		232.1458	Yuanye (China, Shanghai)	Methanol/ Acetonitrile	52.4	73069-14-4	Sesquiterpenoids
Atractylenolide III (AT III)		248.1407	Yuanye (China, Shanghai)	Methanol/ Acetonitrile	57.6	73030-71-4	Sesquiterpenoids

Trans-2,4-Decadienal (DEC)		152.1196	MACKLIN (China, Shanghai)	n-Hexane/ Acetonitrile	1063	25152-84-5	aldehydes
Atractylodin (ATLODIN)		182.0726	Yuanye (China, Shanghai)	n-Hexane/ Acetonitrile	551	55290-63-6	Polyacetylenes
Atractylon (ATLON)		216.1509	Yuanye (China, Shanghai)	Methanol/ Acetonitrile	439	6989-21-5	Sesquiterpenoids
Costunlide (COS)		232.1458	Bomei (China, Hefei)	Methanol/ Acetonitrile	146	553-21-9	sesquiterpenes
Luteolin (LUT)		286.0472	Yuanye (China, Shanghai)	Methanol/ Acetonitrile	420	491-70-3	Flavonoids

Caffeic acid (CAF)		180.0417	Bomei (China, Hefei)	Methanol/ Acetonitrile	120	331-89-5	phenylpropanoids
Scopoletin (SCO)		192.0417	Bomei (China, Hefei)	Methanol/ Acetonitrile	484	92-61-5	coumarins
7-Hydroxycoumarin (HYD)		162.0311	Bomei (China, Hefei)	Methanol/ Acetonitrile	440	93-35-6	coumarins
β-Eudesmol (EUD)		222.1978	Yuanye (China, Shanghai)	Methanol/ Acetonitrile	547	473-15-4	Sesquiterpenoids

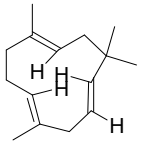
α-Caryophyllene (CAR)		204.1873	Zhenzhun (China, Shanghai)	Methanol/ Acetonitrile	50	6753-98-6	Sesquiterpenoids

Table S2. The detailed sampling information

No.	Collecting province (city)	Collecting time (Year/Month/Day)	Geographical coordinates			Temperature (°C)	Humidity (%)	Weight (g)
			East longitude	Northern latitude	Altitude (metre)			
1	Anhui (Bozhou)	2020/12/2	115°37' 40"	33°44' 8"	19.39	13.8	41.0	22.7
2			115°37' 40"	33°44' 8"	15.59	13.7	39.0	20.7
3			115°37' 40"	33°44' 8"	21.19	13.1	42.0	19.9
4			115°37' 40"	33°44' 8"	21.52	12.5	40.0	19.4
5			115°37' 40"	33°44' 8"	17.94	12.1	40.0	16.7
6			115°37' 40"	33°44' 8"	19.86	14.1	40.0	19.5
7			115°37' 40"	33°44' 8"	23.87	14.4	38.0	25.0
8			115°37' 39"	33°44'10"	18.09	14.5	38.0	21.7
9			115°37' 39"	33°44' 9"	21.26	14.8	38.0	23.7
10			115°37' 39"	33°44' 9"	26.61	15.4	42.0	20.3
11			115°37' 40"	33°44' 8"	20.99	16.4	37.0	18.5
12			115°37' 41"	33°44' 8"	24.01	13.2	42.0	20.9
13			115°37' 40"	33°44' 8"	13.35	12.3	41.0	21.7

14	115°37' 41"	33°44' 8"	18.82	11.8	43.0	19.4
15	115°37' 41"	33°44' 7"	21.16	11.5	42.0	20.9
16	115°40' 54"	33°45' 43"	18.63	14.7	40.0	21.1
17	115°40' 54"	33°45' 43"	18.71	14.8	41.0	23.0
18	115°40' 54"	33°45' 43"	17.49	15.4	44.0	14.8
19	115°40' 54"	33°45' 43"	17.79	14.5	39.0	19.2
20	115°40' 54"	33°45' 43"	16.09	15.0	40.0	22.4
21	115°40' 54"	33°45' 41"	20.4	14.5	30.0	23.6
22	115°40' 54"	33°45' 41"	24.14	16.3	40.0	23.3
23	115°40' 54"	33°45' 41"	21.47	15.7	39.0	20.4
24	115°40' 54"	33°45' 41"	23.95	15.9	38.0	16.3
25	115°40' 54"	33°45' 41"	21.82	16.1	39.0	20.2
26	115°40' 54"	33°45' 42"	20.42	13.1	44.0	21.3
27	115°40' 54"	33°45' 42"	23.55	13.9	42.0	22.0
28	115°40' 54"	33°45' 42"	20.51	14.2	43.0	21.6
29	115°40' 54"	33°45' 42"	20.44	14.2	43.0	23.6
30	115°40' 54"	33°45' 42"	24.29	14.2	47.0	19.0

31			115°41’ 3"	33°45’ 35"	18.15	14.2	46.0	23.7
32			115°41’ 3"	33°45’ 35"	21.34	13.7	40.0	19.9
33			115°41’ 3"	33°45’ 35"	19.26	13.5	43.0	34.0
34			115°41’ 3"	33°45’ 35"	18.45	13.1	42.0	19.2
35			115°41’ 1"	33°45’ 35"	7.84	13.5	45.0	29.5
36			115°41’ 0"	33°45’ 34"	19.36	14.3	40.0	15.6
37			115°41’ 1"	33°45’ 35"	21.73	13.9	45.0	20.3
38			115°41’ 1"	33°45’ 35"	23.77	13.5	41.0	16.6
39			115°41’ 0"	33°45’ 34"	20.98	14.1	42.0	16.3
40			115°41’ 1"	33°45’ 35"	20.39	14.7	41.0	31.0
41			115°41’ 1"	33°45’ 34"	21.06	15.5	38.0	23.9
42			115°41’ 1"	33°45’ 34"	20.19	16.0	38.0	17.4
43			115°41’ 1"	33°45’ 34"	22.11	16.3	38.0	17.4
44			115°41’ 2"	33°45’ 34"	16.7	16.5	38.0	28.7
45			115°41’ 2"	33°45’ 34"	36.02	16.4	40.0	21.5
46	Hunan (Yueyang)	2020/11/30	113°47' 30"	28°57' 19"	463.3	10.2	76.0	10.7
47			113°47' 30"	28°57' 19"	463.3	10.2	76.0	10.6

48			113°47'30"	28°57'19"	463.3	10.2	76.0	10.6
49			113°47'30"	28°57'19"	463.3	10.2	76.0	12.6
50			113°47'30"	28°57'19"	463.3	10.2	76.0	9.6
51			113°48'4"	28°59'28"	462.28	10.2	76.0	21.4
52			113°48'4"	28°59'28"	462.28	10.2	76.0	18.5
53			113°48'4"	28°59'28"	462.28	10.2	76.0	10.7
54			113°48'4"	28°59'28"	462.28	10.2	76.0	10.0
55			113°48'4"	28°59'28"	462.28	10.2	76.0	11.1
56			113°35'38"	28°39'13"	75.5	11.2	68.0	11.7
57			113°35'38"	28°39'13"	75.5	11.2	68.0	21.9
58			113°35'38"	28°39'13"	75.5	11.2	68.0	11.5
59			113°35'38"	28°39'13"	75.5	11.2	68.0	19.2
60			113°35'38"	28°39'13"	75.5	11.2	68.0	12.2
61	Hunan (Yueyang)	2020/12/01	114°2'17"	28°47'26"	158.59	15.3	56.0	6.3
62			114°2'17"	28°47'26"	158.59	15.3	56.0	12.4
63			114°2'17"	28°47'26"	158.59	15.3	56.0	7.5
64			114°2'17"	28°47'26"	158.59	15.3	56.0	20.2

65	114°2'17"	28°47'26"	158.59	15.3	56.0	3.5
66	114°2'17"	28°47'26"	158.59	13.2	56.0	11.8
67	114°2'17"	28°47'26"	158.59	13.2	56.0	9.7
68	114°2'17"	28°47'26"	158.59	13.2	56.0	20.2
69	114°2'17"	28°47'26"	158.59	13.2	56.0	12.7
70	114°2'17"	28°47'26"	158.59	13.2	56.0	6.5
71	114°2'21"	28°47'32"	149.32	15.3	59.0	9.3
72	114°2'21"	28°47'32"	149.32	15.3	59.0	10.4
73	114°2'21"	28°47'32"	149.32	15.3	59.0	11.4
74	114°2'21"	28°47'32"	149.32	15.3	59.0	7.4
75	114°2'21"	28°47'32"	149.32	15.3	59.0	10.9
76	114°2'17"	28°47'27"	168.32	11.6	65.0	10.7
77	114°2'17"	28°47'27"	168.32	11.6	65.0	10.3
78	114°2'17"	28°47'27"	168.32	11.6	65.0	9.0
79	114°2'17"	28°47'27"	168.32	11.6	65.0	11.1
80	114°2'17"	28°47'27"	168.32	11.6	65.0	20.0
81	114°2'18"	28°47'28"	151.97	11.7	67.0	19.5

82			114°2'18"	28°47'28"	151.97	11.7	67.0	19.1
83			114°2'18"	28°47'28"	151.97	11.7	67.0	9.3
84			114°2'18"	28°47'28"	151.97	11.7	67.0	21.7
85			114°2'18"	28°47'28"	151.97	11.7	67.0	20.6
86			114°2'16"	28°47'25"	150.00	12.7	72.0	11.7
87			114°2'16"	28°47'25"	150.00	12.7	72.0	20.8
88			114°2'16"	28°47'25"	150.00	12.7	72.0	10.1
89			114°2'16"	28°47'25"	150.00	12.7	72.0	10.3
90			114°2'16"	28°47'25"	150.00	12.7	72.0	10.4
91	Zhejiang (Jinhua)	2020/12/03	120°23'26"	28°56'47"	411.71	9.9	74.0	5.1
92			120°23'26"	28°56'47"	60.08	9.4	76.0	7.8
93			120°23'26"	28°56'46"	367.68	10.2	70.0	7.5
94			120°23'26"	28°56'47"	397.65	9.0	82.0	10.5
95			120°23'26"	28°56'47"	402.81	8.7	86.0	7.1
96			120°23'26"	28°56'47"	395.62	8.5	90.0	6.2
97			120°23'27"	28°56'46"	378.68	8.3	91.0	6.1
98			120°23'26"	28°56'46"	346.42	8.3	91.0	7.5

99			120°23'27"	28°56'47"	335.44	8.3	91.0	10.2
100			120°23'28"	28°56'46"	371.96	8.4	91.0	10.0
101	Zhejiang (Taizhou)	2020/12/04	120°49'2"	29°12'5"	512.7	-[a]	-	19.2
102			120°49'2"	29°12'5"	512.7	-	-	12.3
103			120°49'2"	29°12'5"	512.7	-	-	10.6
104			120°49'2"	29°12'5"	512.7	-	-	19.9
105			120°49'2"	29°12'5"	512.7	-	-	19.4
106			120°49'2"	29°12'5"	512.7	-	-	19.8
107			120°49'2"	29°12'5"	512.7	-	-	10.6
108			120°49'2"	29°12'5"	512.7	-	-	21.8
109			120°49'2"	29°12'5"	512.7	-	-	10.5
110			120°49'2"	29°12'5"	512.7	-	-	19.1
111			120°49'2"	29°12'5"	512.7	-	-	21.3
112			120°49'2"	29°12'5"	512.7	-	-	20.5
113			120°49'2"	29°12'5"	512.7	-	-	20.1
114			120°49'2"	29°12'5"	512.7	-	-	20.0
115			120°49'2"	29°12'5"	512.7	-	-	22.6

116	Zhejiang (Shaoxing)	2020/12/04	120°49'11"	29°15'26"	433.3	12.5	49.0	19.6
117			120°49'13"	29°15'9"	446.92	15.3	78.0	22.0
118			120°49'13"	120°15'9"	438.83	16.8	60.0	23.2
119			120°49'13"	29°15'9"	440.19	17.7	66.0	19.8
120			120°49'12"	29°15'8"	438.81	19	58.0	21.4

[a] Samples No. 101 to 115 were collected from local farmers' warehouse, no more detailed temperature and humidity information were recorded.

Table S3. Identification of chemical compounds by MALDI-TOF MS

No	Compound name	Molecular formula	Molecular weight	Ion type	Detected mass (m/z)	Mass error (ppm)
1	HYD	$C_9H_6O_3$	162.0311	$[M+H]^+$	163.021	-110
				$[M+K]^+$	201.012	85
2	DEC	$C_{10}H_{16}O$	152.1196	$[M+Na]^+$	175.117	44
				$[M+K]^+$	191.074	-49
3	CAF	$C_9H_8O_4$	180.0417	$[M+H]^+$	181.039	-58
4	ATLODIN	$C_{13}H_{10}O$	182.0726	$[M+H]^+$	183.062	-101
5	SCO	$C_{10}H_8O_4$	192.0417	$[M+H]^+$	193.031	-96
				$[M+Na]^+$	215.003	-133
6	ATLON	$C_{15}H_{20}O$	216.1509	$[M+H]^+$	217.152	-31
7	AT I	$C_{15}H_{18}O_2$	230.1301	$[M+H]^+$	231.118	-87
8	AT II	$C_{15}H_{20}O_2$	232.1458	$[M+H]^+$	233.139	-63
9	CAR	$C_{15}H_{24}$	204.1873	$[M+K]^+$	243.156	21
10	EUD	$C_{15}H_{26}O$	222.1978	$[M+Na]^+$	245.203	63
11	AT III	$C_{15}H_{20}O_3$	248.1407	$[M+H]^+$	249.182	134
12	COS	$C_{15}H_{20}O_2$	232.1458	$[M+Na]^+$	255.111	-96
13	LUT	$C_{15}H_{10}O_6$	286.0472	$[M+H]^+$	287.032	-80
				$[M+K]^+$	324.969	-129

Table S4. The sensitivity and specificity of cross-validation (CV), training set and test set obtained by PCA-LDA, PLS-DA, RF classification methods.

Models		Sensitivity[a]			Specificity[b]		
		Anhui	Hunan	Zhejiang	Anhui	Hunan	Zhejiang
PCA-LDA	CV	100.0	94.3	85.0	100.0	94.6	97.1
	Training set	100.0	100.0	95.0	100.0	98.2	100.0
	Test set	100.0	80.0	100.0	100.0	100.0	90.0
PLS-DA	CV	100.0	85.7	85.0	100.0	100.0	100.0
	Training set	100.0	100.0	100.0	95.0	100.0	100.0
	Test set	100.0	90.0	100.0	95.0	100.0	100.0
RF	Training set	100.0	100.0	95.0	100.0	98.2	100.0
	Test set	100.0	100.0	100.0	100.0	100.0	100.0

[a] The sensitivity is defined by TP/(TP + FN). TP is true positives and FN is false negatives.

[b] The specificity is defined by TN/(TN + FP). TN is true negatives and FP is false positives.

Table S5. The confusion matrix of cross-validation (CV), training set and test set obtained by PCA-LDA, PLS-DA, RF classification methods.

			PCA-LDA			PLS-DA			RF		
			Predicted class								
			Anhui	Hunan	Zhejiang	Anhui	Hunan	Zhejiang	Anhui	Hunan	Zhejiang
CV	Actual class	Anhui	35	0	0	35	0	0	-	-	-
		Hunan	0	33	2	1	30	4	-	-	-
		Zhejiang	0	3	17	0	3	17	-	-	-
Training		Anhui	35	0	0	35	0	0	35	0	0
		Hunan	0	35	0	0	35	0	0	35	0
		Zhejiang	0	1	19	0	0	20	0	1	19
Test		Anhui	10	0	0	10	0	0	10	0	0
		Hunan	0	8	2	1	9	0	0	10	0
		Zhejiang	0	0	10	0	0	10	0	0	10