Supplementary material

Quality control of the herb pair

The main components of the compound of herb pair were identified by ultra-performance liquid chromatography-quadrupole time-of-flight tandem mass spectrometry (UHPLC-QTOF-MS/MS), and 8 compounds were identified by positive and negative dual-mode scanning based on the cleavage mode combined with references[1-5] (Supplementary Table 1 and Supplementary Figures 1-6).

The main components identified in the herb pair were classified into three categories: Polyphenols (catechin), paeoniflorin and its derivatives, and phenolic acid (ferulic acid). Among them, compounds 3 and 4 were the main components. An ion peak of m/z 525.16 was detected in the negative ion mode, which may have been the formate ion [M+HCOO]- of albiflorin and paeoniflorin in the negative ion mode. It was simultaneously observed that m/z 479.15 was the dehydrogenation ion of albiflorin and paeoniflorin.

These two compounds are isomers, and their secondary fragment ions include m/z 449.14, 357.11, 327.10, 195.06, and 165.05. In the negative ion mode, the molecular ion at m/z 479.15 $[M-H]^-$ loses one molecule of hydroxymethyl to generate the fragment ion at m/z 449.14 $[M-H-CH2O]^-$. This fragment further loses one molecule of benzoyl group to form the fragment ion at m/z 327.10 $[M-H-CH2O-C7H6O2]^-$. The fragment ion at m/z 357.11 was formed by the loss of one molecule of the the benzoyl group from the molecular ion at m/z 479.15 $[M-H]^-$, corresponding to $[M-H-C7H6O2]^-$. Subsequent reactions involving the loss of glucose and hydroxymethyl groups generate the fragment ions at m/z 195.06 $[M-H-C7H6O2-C6H10O5]^-$ and 165.05 $[M-H-C7H6O2-C6H10O5-CH2O]^-$, respectively.

Biosafety analysis of the herb pair

We conducted a biosafety assessment of the herb pair. All mice survived the 14-day observation period without exhibiting any apparent signs of toxicity or mortality. This observation indicates that the herb pair did not manifest any evident acute toxicity

(Supplementary Figure 7).
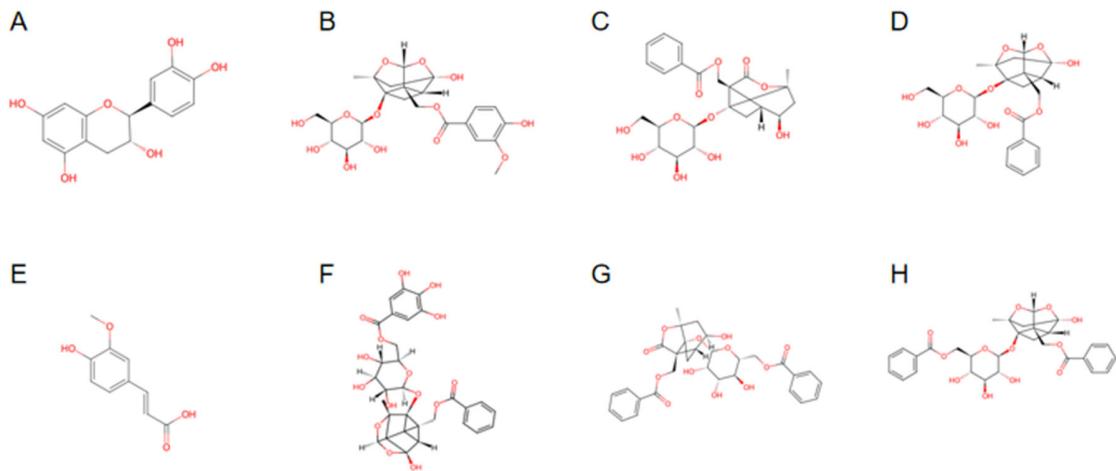
Supplementary Figure 1 The main compounds identified in the herb pair (A-H are respectively catechin, mudanpioside E, albiflorin, paeoniflorin, ferulic acid, galloylpaeoniflorin, benzoylalbiflorin. and benzoylpaeoniflorin).
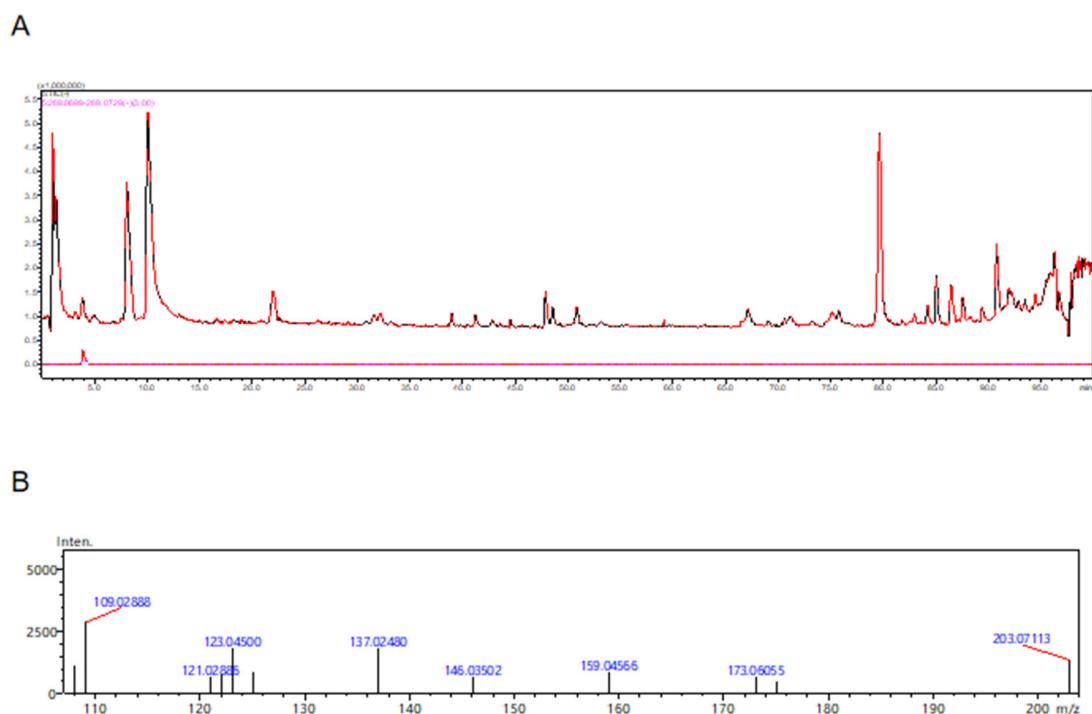

Supplementary Figure 2 The TIC and EIC (A), and MS/MS spectra (B) of catechin.
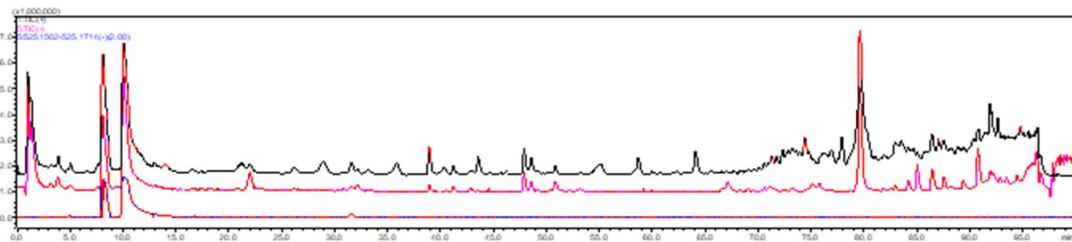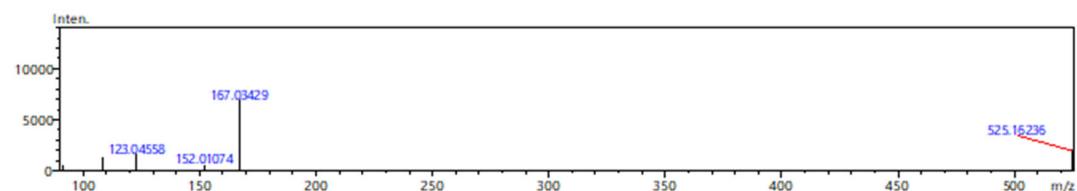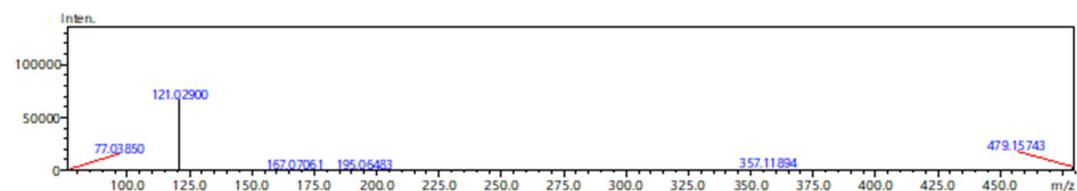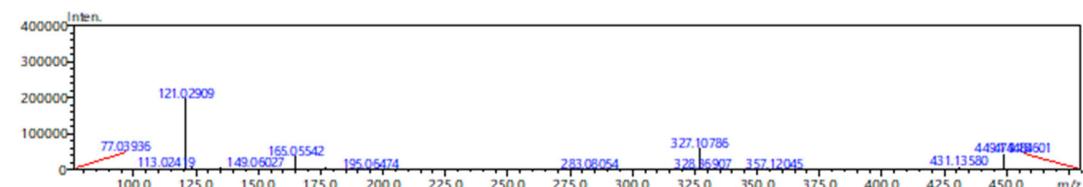
Supplementary Figure 3 The TIC and EIC (A), and MS/MS spectra (B-D) of Mudanpioside E, albiflorin, and paeoniflorin.

A

B
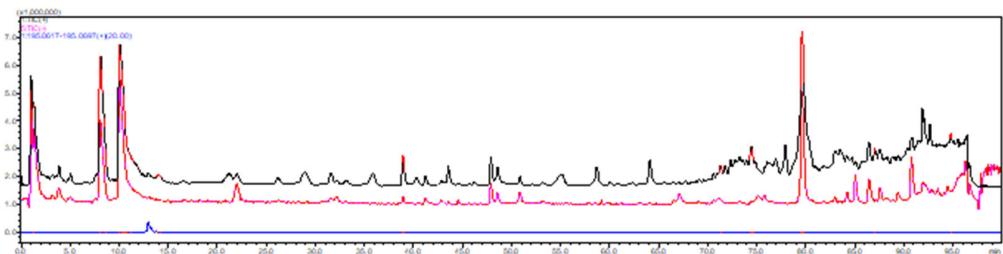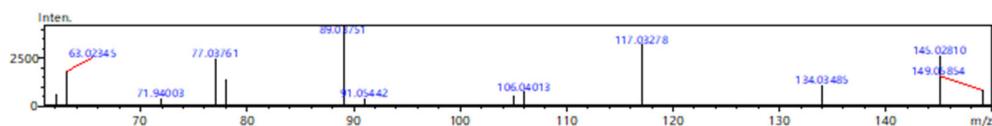
**Supplementary Figure 4** The TIC and EIC (A), and MS/MS spectra (B) of ferulic acid.

A

B
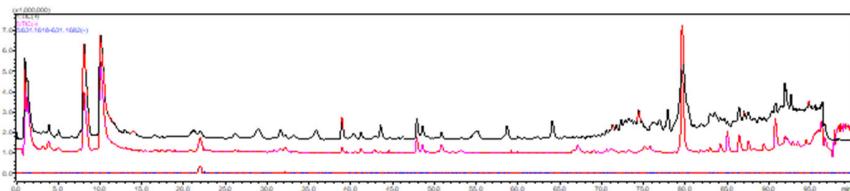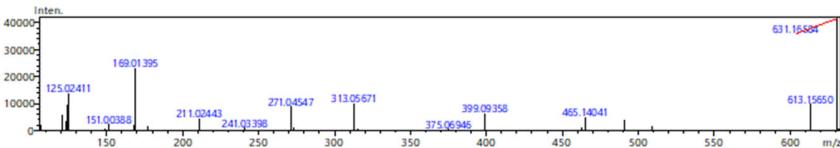
Supplementary Figure 5 The TIC and EIC (A), and MS/MS spectra (B) of galloylpaeoniflorin.
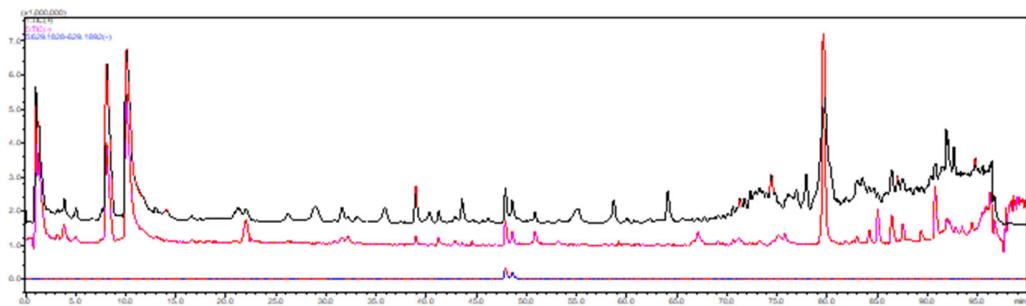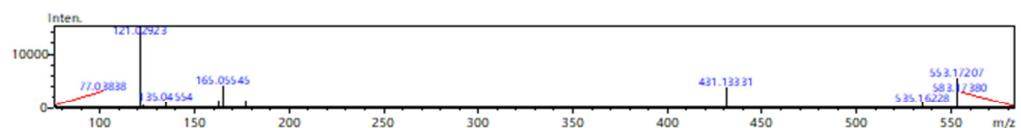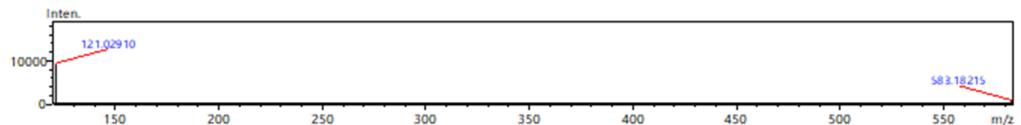
Supplementary Figure 6 The TIC and EIC (A), and MS/MS spectra (B) of benzoylalbiflorin and benzoylpaeoniflorin.
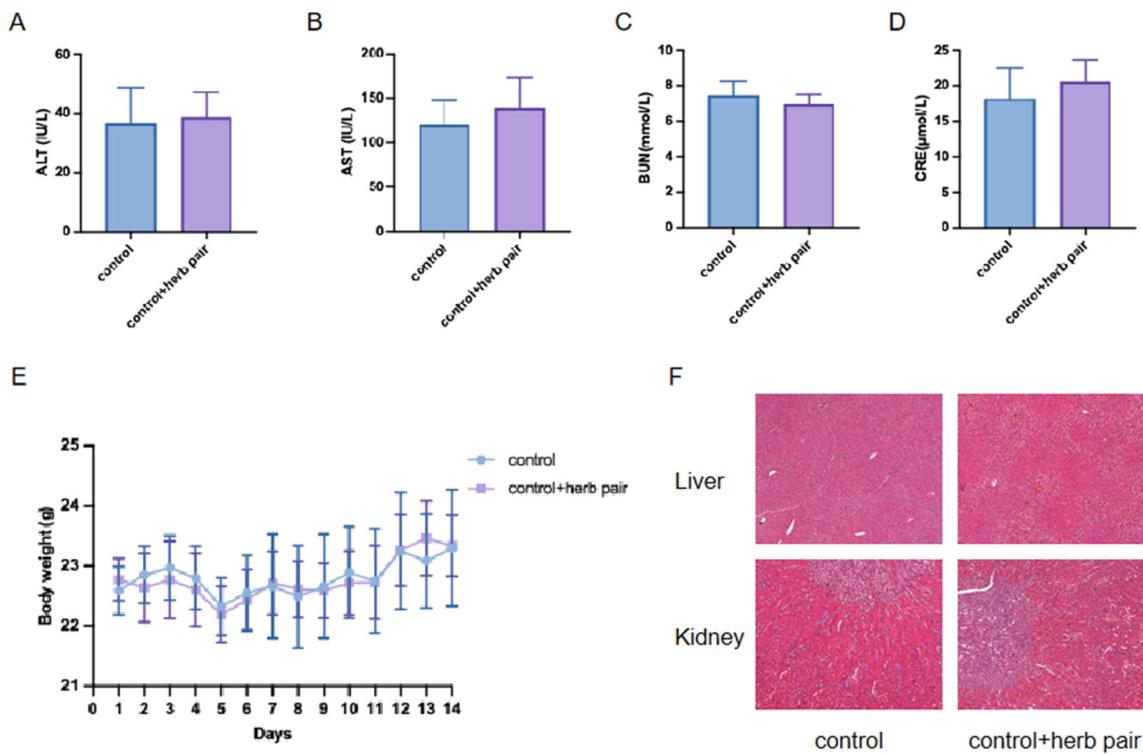
Supplementary Figure 7 Herb pair had no toxicity or side-effects on viscera. A: Serum ALT level, B: Serum AST level, C: Serum BUN level, D: Serum CRE level, E: Body weight, F: HE staining results of liver and kidney. For all the experiments: $n = 10$. ALT: Alanine Aminotransferase, AST: Aspartate aminotransferase, BUN: Blood urea nitrogen, CRE: Serum creatinine.

Supplementary Table 1 Primer sequences used for RT-quantitative polymerase chain reaction analysis

Primer	Forward primer 5′-3′	Reverse primer 5′-3′
Tumor necrosis factor	GGAACTGGCAGAAGAGGCACTC	AATGAGAAGAGGCTGAGACATAGGC
Il-6	GAGAGGAGACTTCACAGAGGATACC	TCATTTCCACGATTTCCCAGAGAAC
Il-1b	CTCGCAGCAGCACATCAACAAG	CCACGGGAAAGACACAGGTAGC
Il-18	GAGACCTGGAATCAGACAACTTTGG	CTGGGGGTTCACTGGCACTTTG
Interferon-gamma	CTGGAGGAACTGGCAAAAGGATG	CAGGTGTGATTCAATGACGCTTATG
Il-13	CTTGCTTGCCTTGGTGGTCTC	GGAGTCTGGTCTTGTGTGATGTTG
Il-10	GGACAACATACTGCTAACCGACTC	TGGATCATTTCCGATAAGGCTTGG
Toll-like receptor 4	CGCTCTGGCATCATCTTCATTGTC	CCTCCCATTCCAGGTAGGTGTTTC
V-rel reticuloendotheliosis viral oncogene homolog A	ATGGGAAACCGTATGAGCCTGTG	AGTTGTAGCCTCGTGTCTTCTGTC
Porcupine	CTGGCTGCTGTGCTGCTGTC	AAGCGATGCCGATGTGAACAGTC
Wnt 5b	GTCCTGACTACTGCCTGCGTAATG	AGCCGTCCATGCCCTCTGAG
Wnt6	GCAGCCGATTCACCCGACTTC	GCGACCGCAGCACAACAGG
Wnt 9a	GACTGCTTTCCTCTACGCCATCTC	TTCCAGGTCGGGTGCCTCATC
Wnt 10a	GAGTGCTTTCGCCTACGCCATAG	TCGCAACCGCAAGCCTTCAG
Wnt 4	GCGTAGCCTTCTCACAGTCCTTTG	CTTCCTGCCAGCCTCGTTGTTG
Leucine-rich repeat-containing G-protein coupled receptor 5	ACGATGTTGAGAAACGGTCCTGTG	CGGAAGTGGAAGGCAAGTCATAGG
SRY-box transcription factor 9	ATGACCGACGAGCAGGAGAAGG	CCGAGGGACAGGGCGAACC
Homeodomain-only protein homeobox	CACCACGCTGTGCCTCATCG	TCTGACCGCCGCCACTCTG
Achaete scute-like 2	GCCGCACCAGAACTCGTAGC	AGGCTCCAGCGACTCCAGAC
Telomerase reverse transcriptase	AAGACGAGCCTCACCTTCCAGAG	CTTCAACCGCAAGACCGACAGG
Mucin 2	CTGACGAGTGGTTGGTGAATGAC	ATGATGAGGTGGCAGACAGGAG
Lysozyme 1	ACTCCTCCTGCTTTCTGTCACTG	CACGGTAGCCATCCATTCCATTC
Chromogranin A	CCACCAATACCCAATCACCAACC	AGCCTCCTCTTCCTCCTCCTC
Occludin	GGCGGCTATGGAGGCTATGG	CTAAGGAAGCGATGAAGCAGAAGG
Nuclear receptor subfamily 1 group H member 4	ACAGCGAAGGGCGTGACTTG	GGTCTGTTGGTCTGCCGTGAG
Fibroblast growth factor 15	CTCTGTGGCTGGCTGTGTCTG	ATTCGGAGGAAGCAGTTGGAGAC
Nuclear receptor subfamily 0 group B member 2	GCCAAGACAGTAGCCTTCCTCAG	GTACCGCTGCTGGCTTCCTC

Supplementary Table 2 The identification of main compounds of herb pair by ultra high performance liquid chromatography-quadrupole time of flight mass spectrometry /MS

No.	Compound	t_R (min)	Formula	Ion mode	Calculated (*m/z*)	Determined (*m/z*)	Error (ppm)	MS/MS fragments ions (*m/z*)
1	Catechin	3.838	$C_{15}H_{14}O_6$	Negative	289.07121 [M-H]$^-$	289.07164	1.4875	203.07113, 151.04047, 137.02438, 123.04357, 109.02923
2	Mudanpioside E	5.002	$C_{24}H_{30}O_{13}$	Negative	525.16082 [M-H]$^-$	525.16236	2.9324	167.03429, 152.01074, 123.04558, 108.02197, 91.01872
3	Albiflorin	8.258	$C_{23}H_{28}O_{11}$	Negative	525.16082 [M+HCOO]$^-$	525.16132	0.9521	479.15743, 357.11894, 195.06483, 121.02900, 77.03850
4	Paeoniflorin	10.072	$C_{23}H_{28}O_{11}$	Negative	525.16082 [M+HCOO]$^-$	525.16038	-0.8378	479.15601, 449.14484, 327.10786, 165.05542, 121.02909
5	Ferulic acid	13.000	$C_{10}H_{10}O_4$	Positive	195.06573 [M+H]$^+$	195.06567	-0.3076	145.02810, 134.03485, 117.03278, 89.03751, 77.03761
6	Galloylpaeoniflorin	22.007	$C_{30}H_{32}O_{15}$	Negative	631.16630 [M-H]$^-$	631.16584	-0.7288	613.15650, 313.05671, 271.04547, 169.01395, 125.02411
7	Benzoylalbiflorin	47.907	$C_{30}H_{32}O_{12}$	Negative	629.18703 [M+HCOO]$^-$	629.18760	0.9059	583.17380, 553.17207, 165.05545, 121.02923, 77.03838
8	Benzoylpaeoniflorin	48.631	$C_{30}H_{32}O_{12}$	Negative	629.18703 [M+HCOO]$^-$	629.18696	-0.1113	583.18215, 121.02910

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