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Article

Flavonoids and a New Polyacetylene from *Bidens parviflora* Willd.

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Abstract: Fifteen flavonoids, **1**-**7** and **9**-**16**, and a polyacetylene, **8**, were isolated from the ethanol extract of the dried whole plant of *Bidens parviflora* Willd. by various chromatographic techniques. Their structures have been elucidated on the basis of spectroscopic analyses and chemical studies. Compound **8** is new and was identified as 3-(*R*),8(*E*)-decene-4,6-diyne-1,3,10-triol. All the flavonoid compounds were isolated for the first time from this plant species.

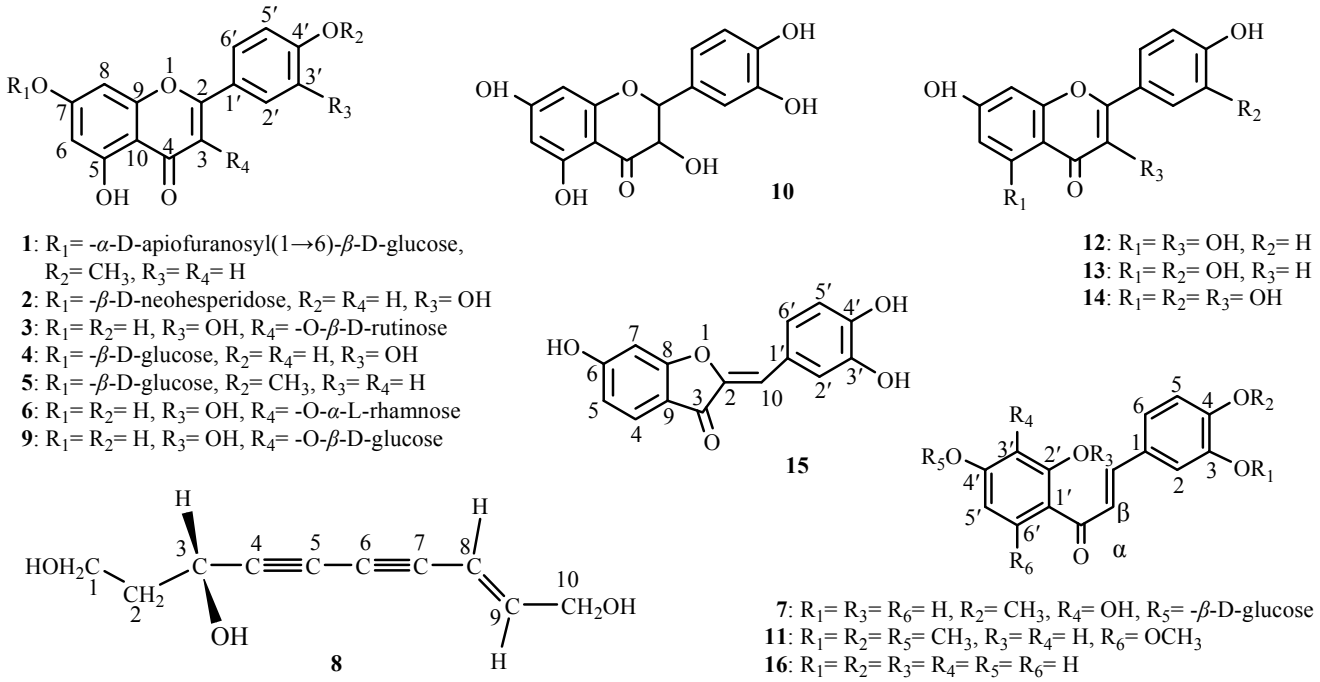
Keywords: *Bidens parviflora* Willd.; Polyacetylene; Flavonoids.

Introduction

The plant *Bidens parviflora* Willd. is used in Chinese folk medicine as an antipyretic, anti-inflammatory and antirheumatic [1, 2]. Flavones [3], flavonones [4], flavonoid glycosides [3, 5], flavonol glycosides [6, 7], chalcones [8, 9], aurones [7, 8], sterols [10], polyacetylene glucosides [4, 9] and monoterpenes [4, 10] have all been previously reported in this species. In our previous studies, sucrose esters [11], phenolic acids [12], polyacetylene glucosides [13], monoterpene glycosides [14],

neolignan glucosides [15], phenolic glucosides [16] and caffeoylquinic acid derivatives [17] were isolated from this plant. As a part of an ongoing research program, this paper describes the isolation and structural determination of a new ployacetylene, **8**, and 15 known flavonoids from *B. parviflora* Willd. (Figure 1).

Figure 1. The structures of compounds **1**-**16**.



Results and Discussion

Compounds **1**-**7** and **9**-**16** were identified as acacetin 7-*O*-(α-D-apio-furanosyl)(1→6)-β-D-glucoside (**1**), luteolin 7-*O*-β-D-neohesperidoside (**2**), quercetin 3-*O*-β-D-rutinoside (**3**), luteolin 7-*O*-β-D-glucoside (**4**), acacetin 7-*O*-β-D-glucoside (**5**), quercitrin (**6**), 4-methoxyl-3,2′,3′-trihydroxychalcone 4′-*O*-β-D-glucoside (**7**), quercetin 3-*O*-β-D-glucoside (**9**), taxifolin (**10**), 2′-hydroxy-3,4,4′,6′-tetramethoxychalcone (**11**), kaempferol (**12**), luteolin (**13**), quercetin (**14**), sulfuretin (**15**) and 3,4,2′,4′-tetrahydroxychalcone (**16**), respectively, by spectroscopic analysis (^{1}H-NMR, ^{13}C-NMR, UV, IR and MS) and comparisons with literature data. All these compounds have been isolated from this plant for the first time.

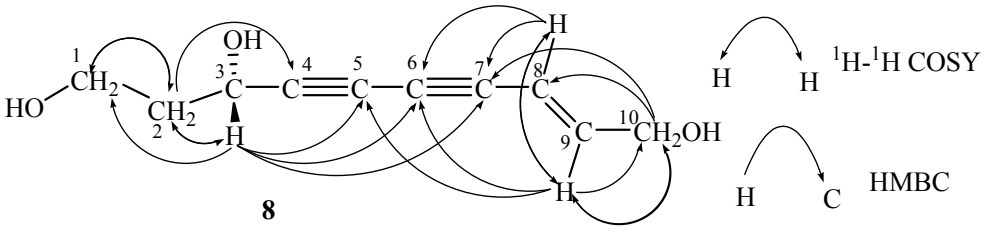
Compound **8** was obtained as a brown powder with optical rotation $[\alpha]_D^{25}$: –16.9° (MeOH, *c* = 0.1), and its molecular formula was determined to be $C_{10}H_{12}O_3$, as indicated by the $[M+Na]^+$ ion at *m/z* 203.0679 (calcd. for 203.0687 $[M+Na]^+$) in the HRESIMS spectrum. In the IR spectrum, absorption bands attributable to acetylene (2341 cm^{-1}, 2360 cm^{-1}), hydroxyl (3182 cm^{-1}) and ethylene (1627cm^{-1}) groups were observed. The UV spectrum of **8** was typical for an ene-diyne chromophore (λ_{max} = 228, 240, 252, 266, 282 nm) [18]. The ^{13}C-NMR (Table 1) and DEPT spectra of **8** present 10 carbon signals, including three methylene groups at δ 41.4, 59.1 and 62.6, one methine group at δ 60.4, two olefinic carbons at δ 108.6 and 148.2, and four quaternary carbons at δ 69.5, 74.1, 77.5 and 84.2 which were confirmed to be ethynyl carbons. Extensive analysis of the ^{1}H-NMR spectrum, together with ^{1}H-

^{1}H COSY and HMQC spectra, presented a methylene proton at δ 4.13 (2H, dd, J = 2.1, 4.7 Hz, H-10), coupled with two *E*-configured olefinic protons at δ 6.39 (1H, td, J = 4.7, 15.9 Hz, H-8) and δ 5.79 (1H, dtd, J = 0.7, 2.1, 15.9 Hz, H-9) indicating a methylene allyl moiety, two methylene protons at δ 3.69 (2H, m, H-1) and δ 1.88 (2H, m, J = 6.8 Hz, H-2), one methine proton at δ 4.57 (1H, t, J = 6.8 Hz, H-3). In the COSY spectrum, the correlations between δ 3.69 (H-1) and δ 1.88 (H-2), δ 1.88 (H-2) and δ 4.57 (H-3) suggested the presence of a CH_2CH_2CH moiety. In the HMBC spectrum, heteronuclear multiple-bond connectivity between the following: δ_H 5.79 (H-9)/δ_C 77.5, δ_H 6.39 (H-8)/δ_C 77.5 and δ_C 74.1, δ_H 4.13 (H-10)/δ_C 77.5 (C-7) could be observed; furthermore, the intensity of correlations between δ_H 6.39/δ_C 77.5 was weaker than that between δ_H 6.39/δ_C 74.1, suggesting that δ_C 74.1 and δ_C 77.5 form a alkynyl group and δ_C 77.5 directly connected with δ_C (148.2) of the $CH{=}CHCH_2$ moiety, while δ_C 69.5 and δ_C 84.7 form another alkynyl. The peak at δ_H 4.57 (H-3) correlates simultaneously with δ_C 84.7, 69.5, 77.5 and 74.1, and together with δ_H 5.37 (H-9) presents a correlation with δ_C 69.5, suggesting two adjacent alkynyls, and δ_C 60.4 of the CH_2CH_2CH moiety is connected to δ_C 84.7. Thus, based on the chemical shifts of protons and carbons, the planar structure of compound **8** was determined to be 8-(*E*)-decene-4,6-diyne-1,3,10-triol. All ^{1}H- and ^{13}C-NMR signals as shown in Table 1 were assigned according to DEPT, HMQC, HMBC and ^{1}H-^{1}H COSY experiments. Figure 2 shows the key correlations presented in the ^{1}H-^{1}H COSY and HMBC spectra of **8**.

Table 1. ^{13}C-NMR (100 MHz, in CD_3OD) and ^{1}H-NMR (400 MHz) data of compound **8**.

Position	δ_C (ppm)	δ_H (ppm)	HMBC (H to C)
1	59.1	3.69 (2H, m)	C-2, 3
2	41.4	1.88 (2H, m)	C-1, 3, 4
3	60.4	4.57 (1H, t, J = 6.8 Hz)	C-1, 2, 4, 5, 6, 7
4	84.2		
5	69.5		
6	74.1		
7	77.5		
8	148.2	6.39 (1H, td, J = 4.7, 15.9 Hz)	C-6, 7
9	108.6	5.79 (1H, dtd, J = 0.7, 2.1, 15.9 Hz)	C-5, 6, 8, 9
10	62.6	4.13 (2H, dd, J = 2.1, 4.7 Hz)	C-7, 8, 9

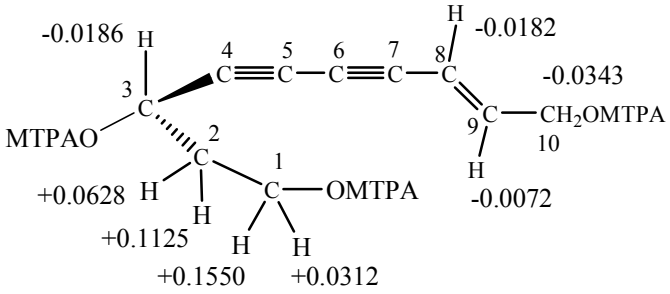
Figure 2. The key HMBC and ^{1}H-^{1}H COSY correlations of compound **8**.



8

The stereochemistry at the chiral center (C-3) in compound **8** was determined using the modified Mosher method on the *R*-(+)-*α*-methoxy-*α*-trifluoromethylphenylacytyl (MTPA) and *S*-(-)-MTPA esters of **8**. In the ^{1}H-NMR spectrum of the *R*-(+)-MTPA ester, the H-1 and H-2 protons appeared upfield, suggesting an effect of the MTPA pheyl ring [19]. In contrast, H-8, H-9 and H-10 were downfield from the corresponding *S*-(-)-MTPA ester. The result was shown in Figure 3. Thus, the absolute configuration at C-3 of **8** was determined to be *R*.

Figure 3. $\delta = \delta_S - \delta_R$ values (ppm) obtained from the MTPA ester of **8** in $CDCl_3$ at 25 °C.



Experimental

General

UV spectra were measured in MeOH using a Shimadzu UV2401PC spectrophotometer (Shimadzu Co., Japan) and optical rotations were determined on a JASCO P-1020 polarimeter in MeOH. IR spectra were recorded on a Shimadau FTIR8400 spectrophotometer (Shimadzu Co., Japan) using KBr discs as stated. ESI-MS were obtained using a Bruker Esquire 2000 mass spectrometer (Bruker Co., Germany) and HR-ESI-MS were recorded on a Micromass Q-TOF mass spectrometer. NMR spectra were recorded on a Bruker AVANCE 400 NMR spectrometer (^{1}H at 400 MHz, ^{13}C at 100 MHz, Bruker Co., Germany). Chemical shifts (δ) are shown in ppm relative to TMS as internal standard and coupling constants (J) are given in Hz. The solvent used was DMSO-d_6, unless otherwise stated. Preparative HPLC was carried out on a Shimadzu Pak equipped with a UV-Vis detector (Shimadzu Co., Japan) using a Shim-pack PREP-ODS column (5 μ, 10×250 mm, Shimadzu Co., Japan). Open column chromatography was carried out on D101 macroporous adsorption resin (Tianjin Nankai Daxue Chemical Plant, P. R. China), silica gel H60 (200-300 mesh, Qingdao Haiyang Chemical Group Co., P. R. China), Sephadex LH-20 (Amersham Pharmacia Biotech Co., UK) and ODS (60-80 μm, Merck Co., Germany) as packing materials. Thin-layer chromatography was performed on silica gel H60 plates (Qingdao Haiyang Chemical Group Co., P.R. China) and RP-18 plate (Merck Co., Germany).

Plant material

The whole plant material of *Bidens parviflora* Willd. was collected in July 2003 in Liaoning province, P.R. China, and was identified by Prof. Weichun Wu (Department of Medical Plants, Shenyang Pharmaceutical University, Shenyang, P.R. China). A voucher specimen (99-DHS-953) was

deposited in the herbarium of the Department of Natural Products Chemistry, Shenyang Pharmaceutical University, Shenyang, P. R. China.

Extraction and Isolation

The dried whole plant of *B. parviflora* Willd. (6.0 kg) was extracted three times with 60% EtOH (48 L) under reflux. The resulting EtOH extract (704 g) was dissolved in water (1.0 L) and applied to a D101 macroporous adsorption resin column, then eluted with water, 30% EtOH, 50% EtOH and 95% EtOH, respectively, to yield four fractions. The 30% EtOH eluate (BPB, 72 g) was subjected to silica gel column chromatography using a gradient solvent system ($CHCl_3$-MeOH = 100: 0→ 98: 2→ 95: 5→ 9: 1→ 8: 2→ 7: 3→ 6: 4→ 0: 1) to give twelve fractions BPB-1 to BPB-12. The BPB-3 fraction (5.45 g) was subjected to ODS column chromatography, eluted with gradient of increasing MeOH (MeOH-H_2O = 20: 80→ 40: 60→ 60: 40→ 90: 10), to give six fractions BPB-3a to BPB-3e; BPB-3c (940 mg) was purified by silica gel chromatography (gradient elution separation with a $CHCl_3$-MeOH system: 95: 5→ 9: 1→ 85: 15→ 8: 2), followed by preparative HPLC (ODS, MeOH-H_2O = 48:52) to yield compounds **1** (10 mg) and **2** (13 mg). BPB-3d (1.85 g) was subjected to Sephadex LH-20 column chromatography (MeOH-H_2O = 50:50), followed by preparative HPLC (ODS, MeOH-H_2O = 55:45) to obtained **3** (68 mg) and **4** (25 mg). The BPB-6 fraction (7.24 g) was subjected to ODS column chromatography, eluted with an increasing gradient of MeOH (MeOH-H_2O = 10:90→ 30:70→ 50:50→ 70:30) to give six fractions BPB-6a to BPB-6e. BPB-6b (2.40 g) and BPB-6c (2.02 g) were subjected to Sephadex LH-20 column chromatography (MeOH- H_2O = 1:1), followed by ODS column chromatography and preparative HPLC (ODS, MeOH-H_2O = 35: 65) to yield **5** (12 mg), **6** (45 mg), **7** (13 mg), **8** (30 mg) and **9** (22 mg).

The 60% EtOH eluate (BPC, 48 g) was subjected to silica gel column chromatography using a gradient solvent system (hexane-acetone =100: 0→ 98: 2→ 95: 5→ 9: 1→ 8: 2→ 7: 3→ 6: 4→ 0: 1) to give twelve fractions BPC-1 to BPC-12. Fraction BPC-1 (3.34 g) was subjected to silica gel column chromatography, eluted with hexane and ethyl acetate in increasing order of polarity, to give eight fractions BPC-1a to BPC-1h. BPC-1g (107 mg) was purified with Sephadex LH-20 column chromatography ($CHCl_3$-MeOH = 1:1) to yield **10** (23 mg). BPC-3 (5.43 g) and BPC-6 (3.06 g) were subjected to Sephadex LH-20 column chromatography ($CHCl_3$-MeOH = 1:1) followed by silica gel column chromatography (gradient elution separation with $CHCl_3$-acetone system) to obtain **11** (30 mg), **12** (24 mg), **13** (15 mg), **14** (20 mg), **15** (16 mg) and **16** (10 mg).

Acacetin 7-O-(α-D-apio-furanosyl) (1→6)-β-D-glucoside (**1**) [20]: $C_{27}H_{30}O_{14}$; yellow needle (MeOH); mp 244-245 °C; $[\alpha]_D$ 23.0° (MeOH, *c* = 0.23, 24 °C); ESI-MS (positive) *m/z* 579 $[M+H]^+$; UV (MeOH) λ_{max} nm (logε): 268 (4.34), 325 (4.40); IR ν_{max} (KBr): 3424, 2927, 1652, 1610, 1500, 1432, 1303, 1257, 1174, 1058, 831 cm^{-1}; ^{1}H-NMR δ: 8.06 (2H, d, *J* = 9.1 Hz, H-2′, 6'), 7.13 (2H, d, *J* = 9.1 Hz, H-3′, 5′), 6.95 (1H, s, H-3), 6.46 (1H, d, *J* = 2.3 Hz, H-6), 6.82 (1H, d, *J* = 2.3 Hz, H-8), , 3.87 (3H, s, 4′-OCH_3), 5.07 (1H, d, *J* = 7.5 Hz, glc-H-1), 4.82 (1H, d, *J* = 3.2 Hz, api-H-1); ^{13}C-NMR δ: 163.8 (s, C-2), 103.8 (d, C-3), 182.0 (s, C-4), 161.0 (s, C-5), 99.6 (d, C-6), 162.9 (s, C-7), 94.8 (d, C-8), 156.9 (s, C-9), 105.4 (s, C-10), 122.7 (s, C-1′), 114.6 (d, C-2′, 6′), 128.4 (d, C-3′, 5′), 162.4 (s, C-4′), 55.6 (q, 4′-OCH_3), 99.8 (d, glc-C-1), 72.9 (d, glc-C-2), 76.2 (d, glc-C-3), 69.6 (d, glc-C-4), 75.5 (d, glc-C-5), 67.3

(t, glc-C-6), 109.1 (d, api-C-1), 75.9 (d, api-C-2), 78.7 (s, api-C-3), 73.3 (t, api-C-4), 63.3 (t, api-C-5). Resonance assignments were based on ^{1}H-^{1}H COSY, HMQC and HMBC spectra.

Luteolin 7-O-β-D-neohesperidoside (**2**) [21, 22]: $C_{28}H_{32}O_{15}$; yellow needles; mp 249-251 °C; ESI-MS (positive) *m/z* 609 $[M+H]^+$; ^{1}H-NMR *δ*: 7.44 (1H, dd, *J* = 8.3, 2.1 Hz, H-6′), 7.40 (1H, d, *J* = 2.1 Hz, H-2′), 6.90 (1H, *J* = 8.3 Hz, H-5′), 6.75 (1H, s, H-3), 6.74 (1H, d, *J* = 2.1 Hz, H-8), 6.38 (1H, d, *J* = 2.1 Hz, H-6), 5.25 (1H, d, *J* = 7.3 Hz, glc-H-1), 5.14 (1H, d, *J* = 1.2 Hz, rha-H-1), 1.20 (3H, d, *J* = 6.2 Hz, rha-CH_3); ^{13}C-NMR *δ*: 166.3 (s, C-2), 105.1 (d, C-3), 183.7 (s, C-4), 163.0 (s, C-5), 99.6 (d, C-6), 164.4 (s, C-7), 92.6 (d, C-8), 158.8 (s, C-9), 107.3 (s, C-10), 123.2 (s, C-1′), 115.4 (d, C-2′), 147.6 (s, C-3′),151.8 (s, C-4′), 117.9 (d, C-5′), 121.0 (d, C-6′), 102.3 (d, glc-C-1), 101.2 (d, rha-C-1), 78.8 (d, glc-C-2), 79.0 (d, glc-C-3), 72.2 (d, glc-C-4), 78.2 (d, glc-C-5), 62.4 (d, glc-C-6), 72.4 (d, rha-C-2), 71.5 (d, rha-C-3), 73.7 (d, rha-C-4), 70.2 (d, rha-C-5), 19.9 (q, rha-CH_3).

Quercetin 3-O-β-D-rutinoside (**3**) [23]: $C_{27}H_{30}O_{16}$; yellow powder; $[\alpha]_D^{26}$ -2.8° (*c* = 0.1, MeOH); ESI-MS^n (positive and negative) *m/z* 633 $[M+Na]^+$, 487$[M+Na-146]^+$, 609$[M-H]^-$, 301$[M-H-(146+162)]^-$; UV (MeOH) λ_{max} nm (logε): 355 (4.09), 256 (4.20); ^{1}H-NMR *δ*: 6.21 (1H, d, *J* = 2.2 Hz, H-6), 6.40 (1H, d, *J* = 2.2 Hz, H-8), 7.67 (1H, d, *J* = 2.5 Hz, H-2′), 6.87 (H, d, *J* = 8.3 Hz, H- 5′), 7.63 (1H, dd, *J* = 8.6, 2.2 Hz, H-6′), 5.11(1H, d, *J* = 7.7 Hz, glc-H-1) 4.52 (1H, d, *J* = 1.5 Hz, rha-H-1), 1.12 (3H, d, *J* = 6.1 Hz, rha-H-6); ^{13}C-NMR *δ*: 159.4 (C-2), 135.7 (s, C-3), 179.5 (s, C-4), 163.0 (s, C-5), 99.9 (d, C-6), 166.1 (s, C-7), 94.9 (d, C-8), 158.6 (s, C-9), 105.7 (s, C-10), 123.2 (s, C-1′), 116.1 (d, C-2′), 145.9 (s, C-3′), 149.9 (s, C-4′), 117.7 (d, C-5′), 123.6 (d, C-6′), 104.8 (d, glc-C-1), 75.8 (d, glc-C-2), 78.2 (d, glc-C-3), 71.4 (d, glc-C-4), 77.3 (d, glc-C-5), 68.6 (t, glc-C-6), 102.5 (d, rha-C-1), 72.3 (d, rha-C-2), 72.1 (d, rha-C-3), 74.0 (d, rha-C-4), 69.8 (d, rha-C-5), 17.9 (q, rha-C-6).

Luteolin 7-O-β-D-glucoside (**4**) [24]: $C_{21}H_{20}O_{11}$; yellow needles; mp 257-259 °C; ESI-MS (positive) *m/z* 449 $[M+H]^+$; ^{1}H-NMR (DMSO-d_6) *δ*: 13.0 (1H, s, br, 5-OH), 6.74 (1H, s, H-3), 6.44 (1H, d, *J* = 2.1 Hz, H-6), 6.78 (1H, d, *J* = 2.1 Hz, H-8), 7.42 (1H, d, *J* = 2.2 Hz, H-2′), 6.90 (1H, d, *J* = 8.2 Hz, H-5′), 7.44 (1H, dd, *J* = 8.2, 2.2 Hz, H-6′), 5.08 (1H, d, *J* = 7.3 Hz, glc-H-1); ^{13}C-NMR (DMSO-d_6) *δ*: 163.7 (s, C-2), 102.4 (d, C-3), 181.1 (s, C-4), 160.3 (s, C-5), 98.8 (d, C-6), 162.1 (s, C-7), 93.9 (d, C-8), 156.2 (s, C-9), 104.5 (s, C-10), 120.6 (s, C-1′), 112.8 (d, C-2′), 145.0 (s, C-3′), 149.1 (s, C-4′), 115.2 (d, H-5′), 118.4 (d, H-6′), 99.1 (d, glc-C-1), 72.3 (d, glc-C-2), 76.4 (d, glc-C-3), 68.8 (d, glc-C-4), 75.6 (d, glc-C-5), 59.8 (t, glc-C-6).

Acacetin 7-O-β-D-glucoside (**5**) [25]: $C_{22}H_{22}O_{10}$; yellow powder; ESI-MS (positive) *m/z* 447 $[M+H]^+$; ^{1}H-NMR *δ*: 12.92 (1H, s, 5-OH), 6.96 (1H, s, H-3), 6.46 (1H, d, *J* = 2.2 Hz, H-6), 6.86 (1H, d, *J* = 2.2 Hz, H-8), 8.07 (2H, d, *J* = 9.0 Hz, H-2′, H-6′), 7.14 (2H, d, *J* = 9.0 Hz, H-3′, H-5′), 5.07 (1H, d, *J* = 7.7 Hz, glc-H-1), 3.89 (3H, s, 4′-OCH_3); ^{13}C-NMR *δ*: 163.8 (s, C-2), 103.7 (d, C-3) , 181.7 (s, C-4), 161.0 (s, C-5), 99.5 (d, C-6), 162.9 (s, C-7), 94.8 (d, C-8), 156.8 (s, C-9), 105.3 (s, C-10), 122.6 (s, C-1′), 128.4 (d, C-2′), 114.6 (s, C-3′), 162.4 (s, C-4′), 114.6 (d, H-5'), 128.4 (d, H-6'), 99.8 (d, glc-C-1), 72.9 (d, glc-C-2), 77.1 (d, glc-C-3), 69.5 (d, glc-C-4), 76.3 (d, glc-C-5), 60.5 (t, glc-C-6), 55.5 (q, 4′-OCH_3).

Quercitrin (**6**) [26]: $C_{21}H_{20}O_{11}$; yellow powder; ESI-MS (positive) *m/z* 449 $[M+H]^+$; ^{1}H-NMR *δ*: 12.65 (1H, s, 5-OH), 6.86 (1H, d, *J* = 8.3 Hz, H-5′), 6.39 (1H, d, *J* = 2.2 Hz, H-8), 7.30 (1H, d, *J* = 2.1 Hz, H-

2′), 7.25 (1H, dd, *J* = 2.1, 8.3 Hz, H-6′), 6.20 (1H, d, *J* = 2.2 Hz, H-6), 5.25 (1H, d, *J* = 1.4 Hz, rha-H-1), 3.97 (1H, t, *J* = 1,4 Hz, rha-H-2), 0.81 (3H, d, *J* = 6.0 Hz, rha-CH_3); ^{13}C-NMR *δ*: 157.2 (s, C-2), 134.2 (s, C-3), 177.7 (s, C-4), 161.3 (s, C-5), 98.7 (d, C-6), 164.2 (s, C-7), 93.6 (d, C-8), 156.4 (s, C-9), 104.0 (s, C-10), 120.7 (s, C-1′), 115,4 (d, C-2′), 145.2 (s, C-3′), 148.4 (s, C-4′), 115.6 (d, C-5′), 121.1 (d, C-6′), 101.8 (d, rha-C-1), 70.5 (d, rha-C-2), 70.3 (d, rha-C-3), 71.2 (d, rha-C-4), 70.0 (d, rha-C-5), 17.4 (q, rha-CH_3).

4-Methoxy-3,2′,3′-trihydroxychalcone 4′-O-β-D-glucoside (**7**) [27, 28]: $C_{22}H_{24}O_{11}$; yellow powder; ESI-MS (positive) *m/z* 465 $[M+H]^+$, 302 $[M-162]^+$; UV (MeOH) λ_{max} nm (logε): 229 (3.65), 312 (3.22); IR ν_{max} (KBr) cm^{-1}: 3367, 2925, 1637, 1567, 1511, 1448, 1367, 1272, 1087; ^{1}H-NMR (CD_3OD) *δ*: 7.79 (1H, d, *J* = 15.4Hz, *β*-H), 7.62 (1H, d, *J* = 15.4 Hz, *α*-H), 7.25 (1H, d, *J* = 2.2 Hz, H-2), 7.00 (1H, d, *J* = 8.3 Hz, H-5), 7.22 (1H, dd, *J* = 8.3, 2.2Hz, H-6), 7.65 (1H, d, *J* = 9.2Hz, H-6′), 6.86 (1H, d, *J* = 9.2 Hz, H-5′), 3.91(3H, s, 4-OCH_3), 4.98 (H, d, *J* = 7.3 Hz, glc-H-1); ^{13}C-NMR (CD_3OD) *δ*: 119.3 (d, C-*α*), 146.5 (d, C-*β*), 194.6 (s, C=O), 129.4 (s, C-1), 115.3 (d, C-2), 152.0 (s, C-3), 148.1 (s, C-4), 119.3 (d, C-5), 123.8 (d, C-6), 117.4 (s, C-1′), 146.5 (s, C-2′), 143.3 (s, C-3′), 165.1 (s, C-4′), 108.2 (d, H-5′), 122.7 (d, H-6′), 102.7 (d, glc-C-1), 74.8 (d, glc-C-2), 78.5 (d, glc-C-3), 71.3 (d, glc-C-4), 77.6 (d, glc-C-5), 62.5 (t, glc-C-6), 56.5 (q, 4′-OCH_3).

Quercetin 3-O-β-D-glucoside (**9**) [29]: $C_{21}H_{20}O_{12}$; yellow powder; ESI-MS (positive and negative) *m/z* 487 $[M+Na]^+$, 325 $[M+Na-162]^+$, 463 $[M-H]^-$, 301 $[M-H-162]^-$; UV (MeOH) λ_{max} nm (logε): 356 (4.05), 297 (3.83), 256 (4.13); ^{1}H-NMR *δ*: 12.62 (1H, s, 5-OH), 10.85 (1H, s, br, 7-OH), 9.72 (1H, s, br, 4′-OH), 9.17 (1H, s, br, 3′-OH), 6.20 (1H, d, *J* = 2.0 Hz, H-6), 6.41 (1H, d, *J* = 2.0 Hz, H-8), 7.53 (1H, d, *J* = 1.9 Hz, H-2′), 6.82 (1H, d, *J* = 8.3 Hz, H-5′), 7.66 (1H, dd, *J* = 8.3, 1.9 Hz, H-6′), 5.37 (1H, d, *J* = 7.6 Hz, glc-H-1); ^{13}C-NMR *δ*: 156.2 (s, C-2), 133.4 (s, C-3), 177.4 (s, C-4), 161.2 (s, C-5), 98.6 (d, C-6), 164.1 (s, C-7), 93.4 (d, C-8), 156.2 (s, C-9), 103.9 (s, C-10), 121.0 (s, C-1′), 115.1 (d, C-2′), 144.8 (s, C-3′), 148.4 (s, C-4′), 115.9 (d, C-5′), 121.9 (d, C-6′), 101.8 (d, glc-C-1), 74.0 (d, glc-C-2), 77.5 (d, glc-C-3), 69.8 (d, glc-C-4), 76.4 (d, glc-C-5), 60.8 (t, glc-C-6).

Taxifolin (**10**) [30]: $C_{15}H_{12}O_7$; white powder; ESI-MS (positive) *m/z* 327 $[M+Na]^+$; ^{1}H-NMR *δ*: 11.87 (1H, s, 5-OH), 10.79 (1H, s, 7-OH), 8.99 (1H, s, 4′-OH), 8.94 (1H, s, 3′-OH), 4.96 (1H, d, *J* = 11.2 Hz, H-2), 4.48 (1H, d, *J* = 11.2 Hz, H-3), 5.84 (1H, d, *J* = 2.1 Hz, H-6), 5.89 (1H, d, *J* = 2.1 Hz, H-8), 6.85 (1H, s, br, H-2′) , 6.71 (1H, d, *J* = 8.0 Hz, H-5′), 6.73 (1H, d, *J* = 8.0 Hz, H-6′); ^{13}C-NMR *δ*: 83.5 (d, C-2), 72.0 (d, C-3), 198.2 (s, C-4), 163.8 (s, C-5), 96.4 (d, C-6), 167.2 (s, C-7), 95.4 (d, C-8), 163.0 (s, C-9), 101.0 (s, C-10), 128.5 (s, C-1′), 115.6 (d, C-2′), 145.4 (s, C-3′), 146.2 (s, C-4′), 115.8 (d, C-5′), 119.8 (d, C-6′).

2′-Hydroxy-3,4,4′,6′-tetramethoxychalcone (**11**) [31]: $C_{19}H_{20}O_6$; yellow powder; ESI-MS (positive and negative) *m/z* 345 $[M+H]^+$, 343 $[M-H]^-$; ^{1}H-NMR (acetone-d_6) *δ*: 7.91 (1H, d, *J* = 15.4 Hz, H-*α*), 7.75 (1H, d, *J* = 15.4 Hz, H-*β*), 7.33 (1H, d, *J* = 2.0 Hz, H-2), 7.03 (1H, d, *J* = 8.2 Hz, H-5), 7.30 (1H, dd, *J* = 8.2, 2.0 Hz, H-6), 6.13 (1H, d, *J* = 2.4 Hz, H-3′), 6.09 (1H, d, *J* = 2.4 Hz, H-5′), 4.01 (3H, s, 4′-OCH_3), 3.91 (3H, s, 3-OCH_3), 3.88 (6H, 4, 6′-OCH_3); ^{13}C-NMR (acetone-d_6) *δ*: 193.4 (s, C=O), 126.0 (d, C-*α*), 143.6 (d, C-*β*), 129.3 (s, C-1), 111.7 (d, C-2), 150.6 (s, C-3), 152.7 (s, C-4), 112.6 (d, C-5),

123.7 (d, C-6), 106.9 (s, C-1′), 169.1 (s, C-2′), 91.8 (d, C-3′), 163.7 (s, C-4′), 94.7 (d, C-5′), 167.4 (s, C-6′), 56.2 (q, 3-OCH_3), 56.1 (q, 4-OCH_3), 56.5 (q, 4′-OCH_3), 56.1 (q, 6′-OCH_3).

Kaempferol (**12**) [32]: $C_{15}H_{10}O_6$; yellow powder; ESI-MS (negative) *m/z* 285 $[M-H]^-$; UV (MeOH) λ_{max} nm (logε): 254 (4.15), 366 (4.02); ^{1}H-NMR (acetone-d_6) δ: 6.27 (1H, d, J = 2.2 Hz, H-6), 6.54 (1H, d, J = 2.2 Hz, H-8), 8.14 (2H, d, J = 8.0 Hz, H-2′, 6′), 7.02 (2H, d, J = 8.0 Hz, H-3′, 5′); ^{13}C-NMR (acetone-d_6) 147.1 (s, C-2), 136.7 (s, C-3), 176.6 (s, C-4), 162.4 (s, C-5), 99.2 (d, C-6), 165.0 (s, C-7), 94.5 (d, C-8), 157.8 (s, C-9), 104.2 (s, C-10), 123.4 (s, C-1′), 130.5 (d, C-2′, 6′), 116.4 (d, C-3′, 5′), 160.2 (s, C-4′).

Luteolin (**13**) [25]: $C_{15}H_{10}O_6$; yellow needles; mp 328-330 °C; ESI-MS (positive) *m/z* 287 $[M+H]^+$; ^{1}H-NMR δ: 6.74 (1H, s, H-3), 6.44 (1H, d, J = 1.9 Hz, H-6), 6.79 (1H, d, J = 1.9 Hz, H-8), 7.42 (1H, d, J = 2.2, H-2′), 6.90 (1H, d, J = 8.2, H-5′), 7.45 (1H, dd, J = 8.2, 2.2, H-6′); ^{13}C-NMR δ: 164.5 (s, C-2), 103.1 (d, C-3), 181.9 (s, C-4), 161.1 (s, C-5), 99.1 (d, C-6), 163.9 (s, C-7), 94.2 (d, C-8), 156.9 (s, C-9), 104.3 (s, C-10), 121.3 (s, C-1′), 113.5 (d, C-2′), 145.8 (s, C-3′), 150.3 (s, C-4′), 115.9 (d, C-5′), 119.2 (d, C-6′).

Quercetin (**14**) [25, 33]: $C_{15}H_{10}O_7$; yellow needles; mp 310-312 °C; ESI-MS (positive and negative) *m/z* 303 $[M+H]^+$, 301 $[M-H]^-$; UV (MeOH) λ_{max} nm (logε): 255 (4.26), 370 (4.20); ^{1}H-NMR δ: 12.48 (1H, s, 5-OH), 10.75 (1H, s, br, 3-OH), 9.56 (1H, s, br, 7-OH), 9.32 (2H, s, br, 2×-OH), 6.19 (1H, d, J = 2.0 Hz, H-6), 6.41 (1H, d, J = 2.0 Hz, H-8), 7.68 (1H, d, J = 2.2 Hz, H-2′), 6.89 (1H, d, J = 8.5 Hz, H-5′), 7.54 (1H, dd, J = 2.2, 8.5 Hz, H-6′); ^{13}C-NMR δ: 147.6 (s, C-2), 135.6 (s, C-3), 175.8 (s, C-4), 160.6 (s, C-5), 98.1 (d, C-6), 163.8 (s, C-7), 93.3 (d, C-8), 156.1 (s, C-9), 103.0 (s, C-10), 121.9 (s, C-1′), 115.0 (d, C-2′), 145.0 (s, C-3′), 146.7 (s, C-4′), 115.5 (d, C-5′), 120.0 (d, C-6′).

Sulfuretin (**15**) [34]: $C_{15}H_{10}O_5$; yellow powder; ESI-MS (positive and negative) *m/z* 271 $[M+H]^+$, 269.0 $[M-H]^-$; ^{1}H-NMR δ: 6.84 (1H, d, J = 8.3 Hz, H-4), 6.70 (1H, dd, J = 8.3, 2.0 Hz, H-5), 6.74 (1H, d, J = 2.0 Hz, H-7), 6.63 (1H, s, H-10), 7.45 (1H, d, J = 1.2 Hz, H-2′), 7.59 (1H, d, J = 8.3 Hz, H-5′), 7.24 (1H, dd, J = 8.3, 1.2 Hz, H-6′); ^{13}C-NMR δ: 145.6 (s, C-2), 181.2 (s, C-3), 125.8 (d, C-4), 113.0 (d, C-5), 166.3 (s, C-6), 98.4 (d, C-7), 167.5 (s, C-8), 113.2 (s, C-9), 111.9 (d, C-10), 123.4 (s, C-1′), 118.0 (d, C-2′), 145.7 (s, C-3′), 148.0 (s, C-4′), 116.1 (d, C-5′), 124.6 (d, C-6′).

3,4,2′,4′-Tetrahydroxychalcone (**16**) [35, 36]: $C_{15}H_{12}O_4$; yellowish oily substance; ESI-MS (positive and negative) *m/z* 287 $[M+H]^+$, 285 $[M-H]^-$; UV (MeOH) λ_{max} nm (logε): 218 (3.50), 250 (2.91), 289 (0.54); IR ν_{max} (KBr) cm^{-1}: 2554, 1693, 1597, 1570, 1504, 1420, 1377, 1327; ^{1}H-NMR (CD_3OD) δ: 6.70 (1H, d, J = 15.3 Hz, H-α), 6.90 (1H, d, J = 15.3 Hz, H-β), 6.37 (1H, s, H-2), 6.01 (1H, d, J = 8.0 Hz, H-5), 6.28 (1H, d, J = 8.0 Hz, H-6), 5.48 (1H, d, J = 2.0 Hz, H-3′), 5.67 (1H, dd, J = 8.9, 2.0 Hz, H-5′), 7.11 (1H, d, J = 8.9 Hz, H-6′); ^{13}C-NMR (CD_3OD) δ: 193.5 (s, C=O), 146.0 (d, C-α), 118.3 (d, C-β), 128.4 (s, C-1), 115.8 (d, C-2), 146.8 (s, C-3), 149,8 (s, C-4), 116.6 (d, C-5), 123.6 (d, C-6), 114.7 (s, C-1′), 167.4 (s, C-2′), 103.8 (d, C-3′) 166.2 (s, C-4′), 109.1 (d, C-5′), 133.2 (d, C-6′).

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References

1. Jiangsu New Medical College. *Dictionary of Chinese Crude Drugs*; Shanghai Technological Publisher: Shanghai, P.R. China, 1986; pp. 1413, 1694, 2239.
2. Xia, G.; Zhang, J.; Chen, X.; Ma, L. The resource utilization of *Bidens parviflora* Willd. in Baoding district. *Chin. Trad. Herb. Drugs* **1985**, *16*, 37-40.
3. Ma, T. B.; Li, J. L.; Yuan, J. R. Isolation and identification of flavonoid glycosides from the leaf of smallflower beggarticks (*Bidens parviflora*). *Chin. Trad. Herb. Drugs* **1991**, *22*, 531-533.
4. Hoffman, B.; Hozel, J. Weitere acylierte Chalcone aus *Bidens pilosa*. *Planta Med*. **1988**, *54*, 450-451.
5. Hoffman, B.; Hozel, J. A methylated chalcone glucoside from *Bidens pilosa*. *Phytochemistry* **1988**, *27*, 3700-3703.
6. Tommasi, N. De; Piacente, S.; Pizza, C. Flavonol and Chalcone Ester Glycosides from *Bidens andicola*. *J. Nat. Prod*. **1998**, *61*, 973-977.
7. Sashida, Y.; Ogawa, K.; Kitada, M.; Karikome, H.; Mimaki, Y.; Shimomura, H. New aurone glucosides and phenylpropanoid glucosides from *Bidens pilosa*. *Chem. Pharm. Bull*. **1991**, *39*, 709-711.
8. Redl, K.; Davis, B.; Bauer, R. Chalcone glycosides from *Bidens campylotheca*. *Phytochemistry* **1993**, *32*, 218-220.
9. Bauer, R.; Redl, K.; Davis, B. Four polyacetylene glucosides from *Bidens campyplotheca*. *Phytochemistry* **1992**, *31*, 2035-2037.
10. Chen, A. H.; Lin, S. R.; Hong, C. H. *Hua Hsueh* **1975**, *9*, 42-48.
11. Wang, N. L.; Yao, X. S.; Ishii, R.; Kitanaka, S. Bioactive sucrose esters from *Bidens parviflora* willd. *Phytochemistry* **2003**, *62*, 741-746.
12. Wang, J.; Wang, N. L.; Yao, X. S.; Kitanaka, S. Structures and anti-histamine activities of phenolic acid derivatives from *Bidens parviflora* Willd. *Chin. J. Med. Chem.* **2006**, *16*, 168-171.
13. Wang, N. L.; Yao, X. S.; Ishii, R.; Kitanaka, S. Antiallergic agents from natural sources. 3.[1)] Structures and inhibitory effects on nitric oxide production and histamine release of five novel polyacetylene glucosides from *Bidens parviflora* Willd. *Chem. Pharm. Bull*. **2001**, *49*, 938-942.
14. Wang, N. L.; Wang, J.; Yao, X. S.; Kitanaka, S. Two new monoterpene glycosides and a new (+)-jasmololone glucoside from *Bidens parviflora* Willd. *J. Asian Nat. Prod. Res.* **2007**, *9*, 449-455.
15. Wang, N. L.; Wang, J.; Yao, X. S.; Kitanaka, S. Two neolignan glucosides and antihistamine release activities from *Bidens parviflora* Willd. *Chem. Pharm. Bull.* **2006**, *54*, 1190-1192.

16. Wang, J.; Wang, N. L.; Yao, X. S.; Kitanaka, S. Phenolic glucosides from *Bidens parviflora* and their anti-histamine activities. *Chin. Trad. Herb. Drugs* **2007**, *38*, 647-649.
17. Wang, J.; Wang, N. L.; Yao, X. S.; Kitanaka, S. Caffeoylquinic acid derivatives from *Bidens parviflora* and their antihistamine release activites. *Chin. Trad. Herb. Drugs* **2006**, *37*, 966.
18. Bohlman, E.; Burkhardt, T.; Zdero, C. *Naturally Occuring Acetylenes*; Academic Press: London, UK, 1973.
19. Ohtani, I.; Kusumi, T.; Kashman, Y.; Kakisawa, H. High-field FT-IR application of Mosher's method-the absoluteconfiguration of marine terpenoids. *J. Am. Chem. Soc*. **1991**, *113*, 4092-4096.
20. Wanjala, C. W.; Majinda, R. T. Flavonoid glycosides from *Crotalaria podocarpa*. *Phytochemistry* **1999**, *51*, 705-707.
21. Kawashty, S. A.; El-Garf, I. A. The flavonoid chemosystematics of Egyptian Verbena species. *Biochem. System. Ecol.* **2000**, *28*, 919-921.
22. Zhang, Q. Y.; Wang, X. Y.; Ying, H. P.; Cheng, T. M.; Zhao, Y. Y. Studies on the Chemical Constituents of *Carduus crispus* L. *China J. Chin. Mat. Med.* **2001**, *26*, 837-839.
23. Wang, J. H.; Wang, Y. L.; Luo, F. C. Study on the chemical constituents from seeds *Sophora japonica*. *Chin. Trad. Herb. Drugs* **2001**, *32*, 471-473.
24. Harborne, J. B. *The Flavonoids*: *Advances in Reseach since 1986*; Chapman & Hall: London, UK, 1994; pp. 450-451.
25. Jia, L. Y.; Sun, Q. S.; Huang, S. W. Isolation and identification of flavonoids from *chrysanthemum moriflolium* Ramat. *Chin. J. Med. Chem.* **2003**, *13*, 159-161.
26. Markham, K. R.; Ternai, B.; Stanley, R.; Geiger, K.; Mabry, T. J. Carbon^{-13} NMR studies of flavonoids-Ⅲ naturally occurring flavonoid glycosides and their acylated derivatives. *Tetrahedron* **1978**, *34*, 1389-1397.
27. Nunziatina, D. T.; Cosimo, P. Flavonol and Chalcone Ester Glycosides from *Bidens leucantha*. *J. Nat. Prod.* **1997**, *60*, 270-273.
28. Nunziatina, D. T.; Sonia, P.; Cosimo, P. Flavonol and Chalcone Ester Glycosides from *Bidens andicola*. *J. Nat. Prod.* **1998**, *61*, 973-977.
29. Jia, Z. J.; Gong, N. C.; Du, M. Chemical constituents of *Saussurea medusa* Maxim. *Chem. J. Chin. Univ.* **1990**, *11*, 202-204.
30. HU, H. Y.; Yang, Y.; Yu, N. J.; Zhao, Y. M. Study on chemical constituents of bark of *Paeonia suffruticos*. *China J. Chin. Mat. Med.* **2006**, *31*, 1795-1797.
31. Phrutivorapongkul, A.; Lipipun, V.; Ruangrungsi, N.; Kirtikara, K.; Nishikkawa, K.; Maruyama, S.; Watanabe, T.; Ishikawa, T. Studies on the chemical constituents of stem bark of *Millettia leucantha*: Isolation of new chalcones with cytotoxic, anti-herpes simplex virus and anti-inflammatory activities. *Chem. Pharm. Bull.* **2003**, *51*, 187-190.
32. Wei, Y. H.; Wu, X. A.; Zhang, C. Z.; Li, C.; Song, L. Studies on Chemical Constituents of *Rheum glabricaule* Sam.(II). *Chin. Pharm. J.* **2006**, *41*, 253-254.
33. Li, X.; Shi, R. B.; Liu, B.; Chen, Y. P. Study on chemical components from effective fraction of Qingnaoxuanqiao Formula (II). *J. Beijing Univ. Trad. Chin. Med.* **2006**, *29*, 545-550.
34. Venkateswarlu, S.; Panchagnula, G. K.; Subbaraju, G. V. Synthensis and antioxidative activity of 3', 4', 6, 7-tetrahydroxyaurone, a metabolite of *Bidens frondosa*. *Biosci. Biotechnol. Biochem*. **2004**, *68*, 2183-2185.

35. Zhang, Y.; Shi, S. P.; Zhao, M. B.; Jiang, Y.; Tu, P. F. A novel chalcone from *Coreopsis tinctoria* Nutt. *Biochem. System. Ecol.* **2006**, *34*, 766-769.
36. Tian, G. l.; Zhang, B.; Zhang, T. Y.; Yang, F. Q.; Yoichiro, I. Separation of flavonoids from the seeds of *Vernonia anthelmintica* Willd by high-speed counter-current chromatography. *J. Chromatogr. A* **2004**, *1049*, 219-222.

Sample Availability: Samples of the compounds **1**, **3**, **4**, **6**, **8**, **9**, **11-15** are available from the authors.