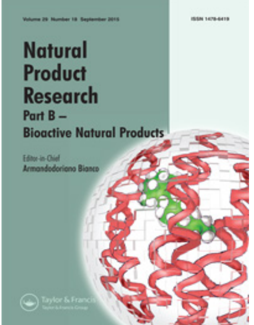

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New cytotoxic and anti-inflammatory compounds isolated from *Morus alba* L.

Jing Qin[ab], Min Fan[b], Juan He[b], Xing-De Wu[b], Li-Yan Peng[b], Jia Su[b], Xiao Cheng[b], Yan Li[b], Ling-Mei Kong[b], Rong-Tao Li[a]* and Qin-Shi Zhao[b]*

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Six Diels–Alder adducts (**1**–**6**) and nine prenylated flavanones (**7**–**15**) were isolated from the root bark of *Morus alba* L. Among them, soroceal B (**1**) and sanggenol Q (**7**) were new compounds. Their structures were elucidated on the basis of extensive spectroscopic methods, including 1D and 2D NMR techniques. Compounds **1**–**3**, **9**, **10**, **12**, **13** and **15** exhibited cytotoxic activity against five human tumour lines and compound **2** inhibited significantly selective cytotoxic activities towards HL-60 and AGS cells with IC_{50} of 3.4 and 3.6 μM. Compounds **3**, **5**, **9** and **12** exhibited moderate inhibitory activity against nitric oxide production in LPS-activated RAW264.7.

Keywords: *Morus alba* L; soroceal B; sanggenol Q; cytotoxic activity; anti-inflammatory activity

1. Introduction

The genus *Morus* (Moraceae) has important economic and medicinal value. Some *Morus* plants are widely cultivated in China and their leaves are indispensable food source for silkworms. The root bark of some species of this genus has been used as traditional medicine ('Mori Cortex') to treat diabetes, arthritis and rheumatism (Pharmacopoeia of P. R. China 2005). Previous phytochemical investigations indicated that this genus is an abundant source of novel phenolic compounds and Diels–Alder type adducts (Dai et al. 2004; Kang et al. 2006), some of which possessed interesting biological activities, such as anti-inflammatory, antioxidant and cytotoxic activities as well as an inhibitory effect on HIF-1 (Dai et al. 2004; Rollinger et al. 2005; Dat et al. 2009; Tan et al. 2009).

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Morus alba L. is widely cultivated in tropical, subtropical and temperate areas (Agarwal & Kanwar 2007). As part of our search for structurally interesting and novel bioactive compounds from traditional Chinese medicines (He et al. 2009; Cheng et al. 2012; Wu et al. 2012), the root bark of *M. alba* has been studied. As a result, two new compounds soroceal B (**1**) and sanggenol Q (**7**), together with five Diels–Alder adducts (**2–6**) and eight prenylated flavonoids (**8–15**) were isolated (Figure S1). These compounds were also screened *in vitro* for their cytotoxic and anti-inflammatory activities. Reported herein are the isolation, structural elucidation and biological evaluation of these compounds.

2. Results and discussion

2.1. *Chemistry*

Soroceal B (**1**), obtained as a brown red amorphous powder, showed a positive reaction to the $FeCl_3$ test, indicating their phenolic nature. Its molecular formula was determined to be $C_{27}H_{22}O_7$ by HR-EI-MS at *m/z* 458.1359 $[M]^+$ (calcd *m/z* 458.1366). The IR spectrum of **1** showed the absorptions for hydroxy moieties, carbonyl group and benzene rings.

The ^{13}C NMR spectrum exhibited 27 carbon signals, including one methyl, 1 methylene, 12 methines, 12 quaternary carbons and an oxymethylene carbon. The ^{1}H NMR spectrum of **1** displayed two 1,2,4-trisubstituted aromatic systems at δ_H 7.07 (d, $J = 2.4$ Hz, H-17′), 7.02 (dd, $J = 8.3$, 2.4, Hz, H-19′) and 7.33 (br. d, 8.3 Hz, H-20′) for ring B and at δ_H 6.98 (d, $J = 2.4$ Hz, H-11′), 6.65 (dd, $J = 8.6$, 2.4 Hz, H-13′) and 7.87 (d, $J = 8.6$ Hz, H-14′) for ring C. It also exhibited one set of 1,2,4,6-*tetra*-substituted aromatic protons (ring D) at δ_H 7.46 (d, $J = 1.4$ Hz, H-2) and 7.35 (d, $J = 1.4$ Hz, H-6), an aldehyde group at δ_H 10.02 (s), and a methyl group at δ_H 1.74 (s, 3H, H-7′). According to ^{1}H–^{1}H COSY correlations, a partly structure ($CHCHCHCHCH_2$, C-2′ → C-6′) was established. Combined with the HMBC correlations of Me-7′ with C-1′, C-2′ and C-6′ as well as H-2′ with C-6′, ring A was constructed. Furthermore, the position of the three aromatic rings B–D was suggested by the following HMBC correlations. The ring B was connected to C-5′ as deduced from the HMBC correlations of H-5′ with C-15′, C-16′ and C-20′, while ring C was connected to C-4′ through C-8′ as inferred from the HMBC correlations of H-3′, H-4′, H-5′ and H-14′ with C-8′ and of H-4′ with C-9′. The HMBC correlations of H-3′ with C-3, C-4 and C-5 were observed which suggested the connection of ring D to C-3′. Then, to fulfil the MS analysis, the connections of rings B and D to C-8′ through oxygen atom were set up, which can be confirmed by the chemical shift of C-3, C-8′ and C-16′. Finally, the aldehyde group was suggested to be attached to C-1 according to the HMBC correlations of H-2 and H-6 with the aldehyde group. Thus, the planar structure of **1** was established as a ketalised Diels–Alder type adduct, similar to mulberrofuran G (**2**) (Hano, Fukai, et al. 1984; Hano, Itoh, et al.1984). The only difference was that an aldehyde group rather than a benzofuran was attached at C-1 in **1**.

In the ROESY experiment (Figure S2), the correlations of H-6′a/H-20′ and H-6′a/H-5′ were observed, indicating that H-5′ and H-6′a were on the same side (α) which was confirmed by the large coupling constant of H-6′b (dd, 2.16, $J = 16.3$, 11.2 Hz) (Zhang et al. 2013). The correlations of H-3′/H-6′b, H-3′/H-14′ suggested H-3′ and ring C were on the other side (β). Furthermore, H-4′ was confirmed to be in α-orientation by the coupling constant between H-5′ and H-4′ ($J = 5.5$ Hz) (*cis*) (Fukai et al. 1985). Therefore, the structure of compound **1** was elucidated as 3′β, 4′α, 5′α, 8′β-1-oxymethylene-5,10′,12′,18′-tetrahydroxy-3/8′,8′/16′-diketal-7-methyl-cyclohexene one and named as soroceal B.

Sanggenol Q (**7**), a yellow amorphous powder, showed a positive reaction with $FeCl_3$ test, indicating their phenolic nature. Its molecular formula was established as $C_{25}H_{28}O_6$ by HR-EI-MS at *m/z* 424.1895 $[M]^+$ (calcd *m/z* 424.1886). The UV spectrum (λ_{max} 289) and the ABX spin system at δ_H 5.65 (1H, dd, $J = 12.4$, 3.2 Hz, H-2), 3.04 (1H, dd, $J = 17.2$, 12.4 Hz, H-3a) and

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2.71 (1H, dd, $J = 17.2, 3.2$ Hz, H-3b) suggested that **7** should be a flavanone (Kanokmedhakul et al. 2004). The ^{1}H NMR spectrum of **7** contained signals for two dimethylallyl groups and three singlets. On the basis of HMQC and HMBC spectral analysis, all proton and carbon signals were fully assigned, and the positions of the substitution were determined. The aromatic protons at δ_H 5.87 and 5.97 were assigned to H-6 and H-8, respectively, due to the HMBC correlations of H-6 with C-8 (96.3), C-7 (168.4) and C-5 (165.4), as well as H-8 with C-6 (97.1), C-7 and C-8a (165.1) (Figure S3). The remaining aromatic proton at δ_H 6.95 was then located at H-6′ according to the HMBC correlations of H-6′ with C-1′ (119.6), C-2′ (151.8), C-5′ (122.1) and C-1‴ (29.3). The obvious HMBC correlations of H-1″ [3.40 (d, $J = 6.7$ Hz)] with C-3′ (118.8) and of H-1‴ [3.24 (br. d, $J = 7.2$ Hz)] with C-5′ (122.1) indicated that the two prenyl groups was attached to C-3′ and C-5′, respectively. The 1D and 2D NMR spectra of **1** were quite similar to those of sanggenol P (**8**) (Zheng et al. 2012), except for the presence of one prenyl group at C-3′ instead of the geranyl group of **8**. The optical rotation of **7** ($[\alpha]_D^{19.8}$ $-2.84°$) and the *trans*-diaxial coupling constant of H-2 and H-3 ($J = 12.4$ Hz) indicated the *S* configuration of C-2 (Li & Wang 1989; Kanokmedhakul et al. 2004). Thus, the structure of compound **7** was identified as (2*S*)-2′,4′,5,7-tetrahydroxy-3′,5′-diprenylflavone and named sanggenol Q.

The known compounds were identified as mulberrofuran G (**2**) (Hano, Fukai, et al. 1984; Hano, Itoh, et al. 1984), mongolicin (**3**) (Kang et al. 2006), mulberrofuran C (**4**) (Hano, Fukai, et al. 1984; Hano, Itoh, et al. 1984), mulberrofuran J (**5**) (Hano, Fukai, et al. 1984; Hano, Itoh, et al. 1984), artonin I (**6**) (Hano et al. 1992), sanggenol P (**8**) (Zheng et al. 2012), sanggenol L (**9**) (Shi et al. 2001), licoflavone C (**10**) (Ngadjui et al. 2002), cyclomulberrin (**11**) (Chen et al. 1993), morusin (**12**) (Kim et al. 2011), morusinol (**13**) (Kim et al. 2011), sanggenon N (**14**) (Hano, Fukai, et al. 1984; Hano, Itoh, et al. 1984) and 3′-geranyl-3-prenyl-2′,4′,5,7-tetrahydroxyflavone (**15**) (Nguyen et al. 2010), by comparing their spectroscopic data with those reported in the literature.

2.2. *Biological activity*

Compounds **1–15** were tested for their cytotoxicity against HL-60, Hela, HepG-2, A-549 and AGS cell lines. As shown in Table 1, compounds **1–3**, **9**, **10**, **12**, **13** and **15** exhibited obvious cytotoxicity against the tested cell lines. As a result, mulberrofuran G (**2**) displayed obvious cytotoxic activities against the HL-60, Hela and AGS cell lines with an IC_{50} value of 3.4, 9.6 and 3.6 μM, respectively. The results suggested that the ketal group maybe a functional group for the cytotoxic activities. And the cytotoxic activity of **2** was stronger than **3**, suggesting that the intensity of cytotoxic activities may show positive correlation with the quantity of ketal groups.

Table 1. Cytotoxicity of compounds **1–3**, **9**, **10**, **12**, **13 and 15** against five tumour cell lines (IC_{50} μM).

Compounds	HL-60	Hela	HepG-2	A-549	AGS
1	17.7 ± 0.2	18.7 ± 0.2	>40	>40	15.8 ± 0.8
2	3.4 ± 0.8	9.6 ± 0.7	14.4 ± 0.8	15.3 ± 0.6	3.5 ± 0.1
3	17.1 ± 0.1	17.1 ± 0.2	17.4 ± 0.8	16.8 ± 0.8	17.5 ± 2.6
4	>40	>40	>40	>40	>40
5	>40	>40	>40	>40	>40
6	>40	>40	>40	>40	>40
9	17.7 ± 0.7	16.4 ± 0.16	16.7 ± 0.2	14.8 ± 0.7	15.7 ± 0.6
10	7.0 ± 1.3	31.8 ± 2.9	10.2 ± 0.9	25.6 ± 2.9	18.3 ± 0.5
12	16.8 ± 0.5	19.0 ± 0.4	>40	39.0 ± 1.9	21.7 ± 1.1
13	18.2 ± 1.0	>40	>40	>40	>40
15	15.8 ± 0.1	17.4 ± 0.1	16.6 ± 1.1	15.8 ± 1.1	16.3 ± 0.2
Cisplatin[a]	3.7 ± 0.3	4.9 ± 1.0	12.5 ± 0.5	12.4 ± 0.2	18.8 ± 2.6

[a]Positive control.

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Furthermore, the cytotoxic activity of **2** was apparently more than **1**, which indicated that the benzofuran group at C-1 of **2** may render the cytotoxic activity (Figure 1).

Considering the traditional use of *Morus* plants, all isolates except compounds **2**, **10**, **14** and **15** were tested for their inhibitory activity against NO production in LPS-stimulated RAW264.7 cells. Compounds **3**, **5**, **9** and **12** exhibited moderate anti-inflammatory activity (Table 2). Among them, **3** showed potent inhibitory activity against NO production with IC_{50} of 8.8 μM.

3. Experimental

3.1. *General*

Melting points were obtained on an X-4 micro melting point apparatus; UV spectra were obtained using a Shimadzu UV-2401A spectrophotometer (Shimadzu, Kyoto, Japan). IR spectra were obtained by using a Bruker Tensor 27 spectrophotometer (Bruker Optics, Ettlingen, Germany) with KBr pellets. Optical rotations were measured on a JASCO-20C digital polarimeter (JASCO, Tokyo, Japan). 1D and 2D NMR were performed on Bruker AM-400, DRX-500 or AVANCE III-600 spectrometers with TMS as an internal standard (Bruker Optics, Ettlingen, Germany). ESI-MS was performed on an API QSTAR time-of-flight spectrometer (Agilent, California, USA). HR-EI-MS was recorded on a Waters AutoSpec Premier P776 spectrometer (Waters, Milford, MA, USA). Silica gel and Sephadex LH-20 were used for column chromatography (CC). MPLC was performed on a Lisui EZ Purify III System including pump manager P03, detector modules P02 and fraction collector P01 and columns packed with MCI gel. Semi-preparative HPLC was performed on an Agilent 1260 apparatus equipped with a UV detector and a Zorbax SB-C-18 column. Fractions were monitored by TLC and spots were visualised by heating silica gel plates sprayed with 10% H_2SO_4 in EtOH.

3.2. *Plant material*

The root bark of *M. alba* was purchased from the Herb Material Market of Juhuacun, Kunming, Yunnan Province, P. R. China, in April 2013, and identified by Prof. Xiao Cheng, Kunming Institute of Botany. A voucher specimen (KIB20130401c01) has been deposited at the State Key Laboratory of Phytochemistry and Plant Resources in West China, Kunming Institute of Botany, Chinese Academy of Sciences.

3.3. *Extraction and isolation*

The root bark of *M. alba* (8 kg) was extracted with MeOH and then concentrated under reduced pressure to yield a residue, which partitioned between H_2O and EtOAc. The EtOAc fraction

Figure 1. Structures of compounds **1** and **7**.

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Table 2. Inhibitory effects of compounds from *M. alba* L. on LPS-activated NO production in RAW264.7 cells.

Compounds	IC_{50} (μM)
1	>25
3	8.8 ± 0.8
4	>25
5	21.4 ± 2.3
6	>25
7	>25
8	>25
9	12.5 ± 1.6
11	>25
12	18.1 ± 0.7
13	>25
MG-132[a]	0.1 ± 0.1

[a]Positive control substance.

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(800 g) which was chromatographed on a silica gel column with a gradient elution of petroleum ether–acetone (1:0 to 0:1) to yield five fractions (1–5). Fraction 3 (110 g) was subjected to reversed-phase MPLC (MCI) ($MeOH/H_2O$, 60%→100%) to yield fractions (3.1–3.5). Fraction 3.1 was purified by recrystallisation to obtain **12** (1 g) and **13** (900 mg). Fraction 3.2 was subjected to silica gel CC ($CHCl_3$–MeOH, 40:1) to afford four subfractions (3.2.1–3.2.4). Subfraction 3.2.2 was subjected to Sephadex LH-20 (MeOH) and repeated silica gel CC to yield **7** (40 mg) and **9** (30 mg). Subfraction 3.2.3 was recrystallised to obtain **11** (1.2 g) and a mixture, which was further purified by semi-preparative HPLC (MeCN–H_2O, 55:45) to afford **10** (5 mg). Fraction 3.3 was subjected to silica gel CC ($CHCl_3$–MeOH, 60:1) to afford five subfractions (3.3.1–3.3.5). Subfraction 3.3.3 was subjected to Sephadex LH-20 (MeOH) to yield three subfractions (3.3.3.1–3.3.3.3). Compounds **14** (15 mg) and **15** (20 mg) were obtained from subfraction 3.3.3.2 by semi-preparative HPLC (MeOH–H_2O, 60:40). Subfraction 3.3.3.4 was subjected to repeated silica gel CC and followed by Sephadex LH-20 (MeOH) to yield **8** (52 mg). Fraction 5 (150 g) was subjected to reversed-phase MPLC (MCI) ($MeOH/H_2O$, 50% → 100%) to obtain fractions 5.1–5.4. Fr. 5.1 was subjected to Sephadex LH-20 (MeOH), followed by silica gel CC ($CHCl_3$–MeOH, 30:1) to obtain **1** (7.5 mg). Fr. 5.2 was subjected to silica gel CC ($CHCl_3$–MeOH, 60:1) to afford three subfractions (5.2.1–5.2.3). Subfraction 5.2.1 was subjected to Sephadex LH-20 (MeOH) and repeated silica gel CC to obtain **2** (16 mg), **3** (17.8 mg) and **6** (7 mg). Subfraction 5.2.2 was subjected to Sephadex LH-20 (MeOH) and further purified by semi-preparative HPLC (MeCN–H_2O, 56:44) to afford **4** (15 mg) and **5** (14 mg).

*Soroceal B (**1**):* Yellow amorphous powder; $[\alpha]_D^{19.6} + 560.44°$ ($c = 0.15$, MeOH); UV (MeOH) λ_{max} (log ϵ): 281 (4.24), 221 (4.67), 203 (4.95) nm; IR (KBr) ν_{max} 3440, 2923, 1626, 1510, 1435, 1128 cm^{-1}; ESI-MS: *m/z* 457 $[M-H]^-$; HR-EI-MS $[M]^+$ *m/z* 458.1359 (calcd for $C_{27}H_{22}O_7$, 458.1366). ^1H NMR (C_5D_5N, 600 MHz) δ 10.02 (1H, s, H-CHO), 7.87 (1H, d, $J = 8.6$, H-14′), 7.46 (1H, d, $J = 1.4$, H-2), 7.35 (1H, d, $J = 1.4$, H-6), 7.33 (1H, br. d, $J = 8.3$, H-20′), 7.07 (1H, d, $J = 2.4$, H-17′), 7.02 (1H, dd, $J = 8.3, 2.4$, H-19′), 7.01 (1H, d, $J = 2.4$, H-2′), 6.98 (1H, d, $J = 2.4$, H-11′), 6.65 (1H, dd, $J = 8.6, 2.4$, H-13′), 4.32 (1H, t, $J = 5.5$, H-4′), 4.30 (1H, m, H-3′), 3.47 (1H, td, $J = 11.2, 5.5$, H-5′), 2.72 (1H, dd, $J = 16.3, 6.2$, H-6′α), 2.16 (1H, dd, $J = 16.3, 11.2$, H-6′β), 1.74 (3H, s, H-7′); ^{13}C NMR (C_5D_5N, 150 MHz) δ 192.6 (d, CHO), 161.4 (s, C-12′), 159.8 (s, C-5), 159.2 (s, C-18′), 159.1 (s, C-10′), 155.6 (s, C-3), 154.1 (s, C-16′), 137.6 (s, C-1), 134.5 (s, C-1′), 131.1(d, C-14′), 128.5 (d, C-20′), 122.8 (s, C-4), 122.8 (d, C-2′), 117.5 (s, C-15′), 117.0 (s, C-9′), 111.5 (d, C-2), 110.8 (d, C-19′), 108.8 (d, C-6), 107.6

(d, C-13′), 105.4 (d, C-11′), 104.8 (d, C-17′), 103.4 (s, C-8′), 37.6 (d, C-4′), 36.6 (d, C-6′), 36.3 (d, C-3′), 29.0 (d, C-5′), 24.4 (q, C-7′).

Sanggenol Q (7): Yellow amorphous powder; m.p. 143–144°C; $[\alpha]_D^{19.8}$ $-2.84°$ ($c = 0.25$, MeOH); UV (MeOH) λ_{max} (log ϵ): 289 (4.17), 208 (4.75) nm; IR (KBr) ν_{max} 3428, 2920, 1635, 1475, 1443, 1258, 1163, 1092, 1067 cm^{-1}; ESI-MS: *m/z* 423 $[M-H]^-$; HR-EI-MS $[M]^+$*m/z* 424.1895 (calcd for $C_{25}H_{28}O_6$, 424.1886); ^{1}H NMR (CD_3OD, 400 MHz) δ 6.95 (1H, s, H-6′), 5.97 (1H, br. s, H-8), 5.87 (1H, br. s, H-6), 5.65 (1H, dd, $J = 12.4$, 3.2, H-2), 5.27 (1H, t, $J = 7.2$, H-2‴), 5.17 (1H, t, $J = 6.7$, H-2″), 3.40 (1H, d, $J = 6.7$, H-1″), 3.24 (1H, br. d, $J = 7.2$, H-1‴), 3.04 (1H, dd, $J = 17.2$, 12.4, H-3a), 2.71 (1H, dd, $J = 17.2$, 3.2, H-3b), 1.77 (1H, br. s, H-4″), 1.73 (1H, br. s, H-5‴), 1.68 (1H, br. s, H-4‴), 1.67 (1H, br. s, H-5″); ^{13}C NMR (CD_3OD, 100 MHz) δ 198.3 (s, C-4), 168.4 (s, C-7), 165.4 (s, C-5), 165.1 (s, C-8a), 154.3 (s, C-4′), 151.8 (s, C-2′), 133.5 (s, C-3‴), 132.6 (s, C-3″), 125.5 (d, C-6′), 124.0 (d, C-2″), 124.0 (d, C-2‴), 122.1 (s, C-5′), 119.6 (s, C-1′), 118.8 (s, C-3′), 103.4 (s, C-4a), 97.1 (d, C-6), 96.3 (d, C-8), 76.7 (d, C-2), 43.1 (t, C-3), 29.3 (t, C-1‴), 26.0 (q, C-5″), 23.8 (t, C-1″), 18.1 (q, C-4), 17.9 (q, C-4‴).

3.4. *Cytotoxicity assay*

The cytotoxicity of compounds **1–15** against human myeloid leukaemia (HL-60), human cervical cancer (Hela), human hepatoma (HepG-2), human non-small cell lung cancer (A-549) and human gastric cancer (AGS) cell lines was assessed using the MTT method (Mosmann 1983). Cells were plated in 96-well plates 12 h before treatment and continuously exposed to different concentrations of compounds. After 48 h, 20 μL of 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) solution was added to each well, which was incubated for another 4 h. Then, 20% SDS (100 μL) was added to each well. After 12 h at room temperature, the OD value of each well was recorded at 595 nm. The IC_{50} value of each compound was calculated by using the Reed and Muench method (Reed & Muench 1938).

3.5. *Anti-inflammatory assay*

Murine monocytic RAW264.7 macrophages were dispensed into 96-well plates (2×10^5 cells/well) containing RPMI-1640 medium (Hyclone) with 10% FBS under a humidified atmosphere of 5% CO_2 at 37°C. After 24 h preincubation, cells were treated with serial dilutions of the compounds, with the maximum concentration of 25 μM in the presence of 1 μg/mL LPS for 18 h. Each compound was dissolved in DMSO and further diluted in medium to obtain different concentrations. NO production in each well was assessed by adding 100 μL of Griess reagent (reagent A and reagent B, respectively; Sigma, Missouri, USA) to 100 μL of each supernatant from LPS (Sigma)-treated or LPS- and compound-treated cells in triplicate. After 5 min incubation, the absorbance was measured at 570 nm with a 2104 Envision Multilabel Plate Reader (Perkin-Elmer Life Sciences, Inc., Boston, MA, USA). MG-132 was used as a positive control (Fan et al. 2010).

The cytotoxicity of the tested compounds was evaluated using an MTS assay (Monks et al. 1991). Briefly, RAW264.7 cells, 2×10^5 cells/well, were seeded in 96-well plates. After 24 h incubation, cells were treated with or without test compounds at given concentrations for 18 h. Then, MTS was added to each well and the plates were incubated for another 4 h. Test compounds were dissolved in DMSO, and the absorbance was read at 490 nm. Cytotoxicity was calculated with respect to the cell viability, which was considered to be 100% without test compounds.

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4. Conclusion

In summary, two new compounds soroceal B (**1**) and sanggenol Q (**7**), together with five Diels–Alder adducts (**2–6**) and eight prenylated flavonoids (**8–15**) were isolated. The cytotoxic activity of all isolates against HL-60, Hela, HepG-2, A-549 and AGS cell lines was evaluated. Compounds **1–3**, **9**, **10**, **12**, **13** and **15** exhibited cytotoxic activity against five human tumour lines and compound **2** inhibited significantly selective cytotoxic activities towards HL-60 and AGS cells with IC_{50} of 3.4 and 3.6 μM, respectively. Meanwhile, compounds **3**, **5**, **9** and **12** possessed moderate anti-inflammatory activity with IC_{50} of 8.8, 21.4, 12.5 and 18.1 μM, respectively.

Supplementary material

Supplementary material relating to this paper is available online, alongside Figures S1–S29 and Tables S1 and S2.

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