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Diarylheptanoids from the rhizomes of *Zingiber officinale*

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Abstract

Seven new diarylheptanoids, i.e., (3*S*,5*S*)-3,5-diacetoxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptane, (3*R*,5*S*)-3-acetoxy-5-hydroxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptane, (3*R*,5*S*)-3,5-dihydroxy-1-(4-hydroxy-3,5-dimethoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptane, (5*S*)-5-acetoxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptan-3-one, 5-hydroxy-1-(3,4-dihydroxy-5-methoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptan-3-one, 5-hydroxy-1-(4-hydroxy-3-methoxyphenyl)-7-(3,4-dihydroxy-5-methoxyphenyl)heptan-3-one and 1,5-epoxy-3-hydroxy-1-(4-hydroxy-3,5-dimethoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptane were isolated from the rhizomes of Chinese ginger (*Zingiber officinale* Roscoe), along with 25 known compounds, i.e., 8 diarylheptanoids, 14 gingerol analogs, a diterpene and 2 steroids. Their structures were elucidated by spectroscopic and chemical methods.

Keywords: Ginger; *Zingiber officinale*; Zingiberaceae; Diarylheptanoid

1. Introduction

Ginger, the rhizome of *Zingiber officinale* Roscoe (Zingiberaceae), is one of the most popular spices and has been frequently used in Chinese traditional medicines both in fresh and dried forms (Huang et al., 1997). Numerous chemical investigations of this plant material have led to the isolation and identification of a large number of biologically active compounds, such as gingerols, gingerones and shogaols (Uehara et al., 1987; Kikuzaki et al., 1991a,b; Endo et al., 1990; Kikuzaki et al., 1992; Yu et al., 1998). We wish to report herein the isolation and structural elucidation of seven previously unknown diarylheptanoids from the ethanol extract of the rhizomes of *Z. officinale*. These new compounds are (3*S*,5*S*)-3,5-diacetoxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptane (**1a**), (3*R*,5*S*)-3-acetoxy-5-hydroxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptane (**1b**), (3*R*,5*S*)-3,5-dihydroxy-1-(4-hydroxy-3,5-dimethoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptane (**1c**), (5*S*)-5-acetoxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptan-3-one (**2a**), 5-hydroxy-1-(3,4-dihydroxy-5-methoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptan-3-one (**2b**), 5-hydroxy-1-(4-hydroxy-3-methoxyphenyl)-7-(3,4-dihydroxy-5-methoxyphenyl)heptan-3-one (**2c**) and 1,5-epoxy-3-hydroxy-1-(4-hydroxy-3,5-dimethoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptane (**3**). Also isolated and identified were 25 known compounds, i.e., four acyclic diarylheptanes (**1d**–**g**), two diarylheptanones (**2d** and **2e**), a cyclic diarylheptane (**4**), a diarylheptenone (**5**), a paradol (**6**), three gingerdiols (**7a**–**c**), six gingerols (**8a**–**f**), a dehydrogingerdione (**9**), two shogaols (**10a** and **10b**), a phenylpropanoid (**11**), a diterpene (**12**) and two steroids (**13** and **14**) (Fig. 1). Compound **7a** was reported previously as a synthetic product from reduction of [6]-gingerol (Kikuzaki et al., 1992), and this is the first time it has been isolated from ginger and reported as a natural product. Compounds **2d** and **11** have been identified by GC-MS, but no spectroscopic data were reported previously (Harvey, 1981; Kraus et al., 1990).

2. Results and discussion

The petroleum ether and ethyl acetate fractions of the ethanol extract of the rhizomes of *Z. officinale* were separated by repeated column chromatography on silica

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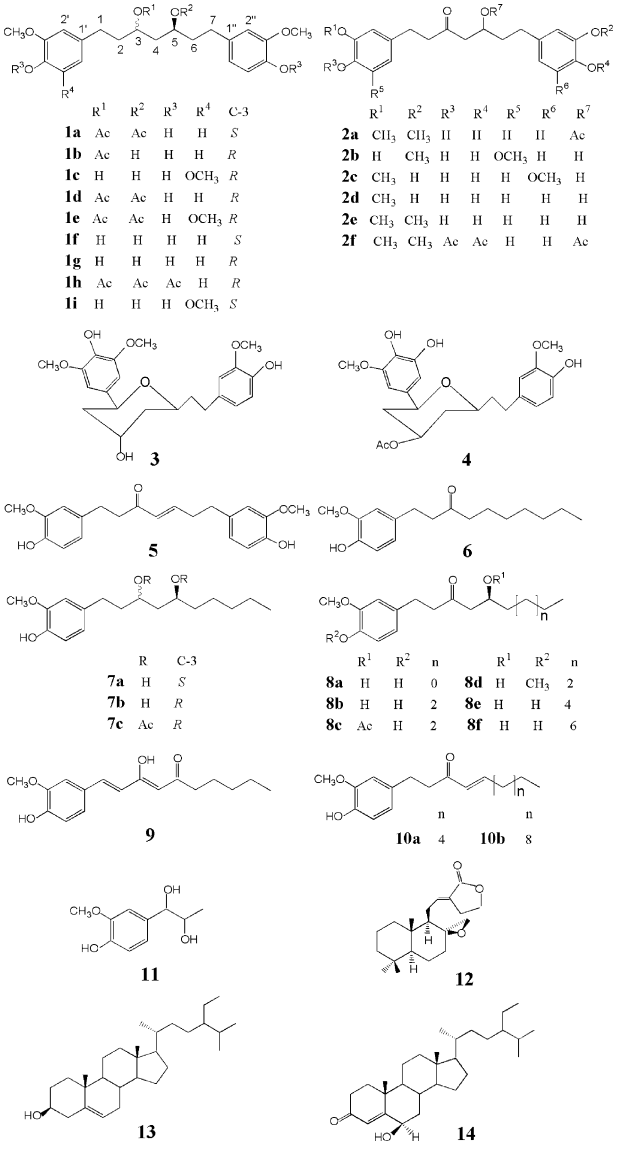

Fig. 1. Constituents isolated from ginger.

gel to afford seven new diarylheptanoids (**1a**, **1b**, **1c**, **2a**, **2b**, **2c** and **3**) along with 25 known compounds, **1d**–**g**, **2d**–**e**, **4**–**14** as shown in Fig. 1.

Compound **1a** was obtained as a colorless oil, $[\alpha]_D^{26}$ +7.0° (*c* 0.68, $CHCl_3$). The HR-ESI-MS spectrum exhibited an $[M+NH_4]^+$ ion peak at *m*/*z* 478.2431 corresponding to a molecular formula of $C_{25}H_{32}O_8$ (calc. for $M+NH_4$: 478.2435). The IR, UV and HR-ESI-MS spectra of **1a** were completely identical with those of the known compound (*3R,5S*)-3,5-diacetoxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptane (**1d**) (Kikuzaki et al., 1991b) which was also obtained from the ethanol extract of the rhizomes of *Zingiber officinale*. However, unlike **1d** (a meso compound) **1a** was found to be optically active, suggesting that **1a** was a stereoisomer of **1d**. Comparison of the ^{13}C NMR spectrum of **1a** with that of **1d** indicated that although most signals of both were overlapped, two signals of the two compounds were apparently distinguishable. These were the resonances of C-3/C-5 (δ 69.72 and 70.64 for **1a** and **1d** respectively) and C-2/C-6 (δ 36.66 and 35.91 for **1a** and **1d** respectively). Deacetylation of **1a** and **1d** with KOH/MeOH gave the corresponding 3,5-dihydroxyl derivatives **1f** and **1g** which were identified as (3*S*,5*S*)- and (3*R*,5*S*)-3,5-dihydroxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptanes, respectively, by comparing their ^1H and ^{13}C NMR spectral data and optical rotations with those reported in the literature (Kikuzaki et al., 1991a). Therefore, compound **1a** was assigned as (*3S,5S*)-3,5-diacetoxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptane.

Compound **1b** was obtained as a colorless oil, $[\alpha]_D^{24}$ +6.0° (*c* 0.56, $CHCl_3$). Its HR-ESI-MS spectrum exhibited an $[M+NH_4]^+$ ion peak at *m*/*z* 436.2324, corresponding to the molecular formula of $C_{23}H_{30}O_7$ [calc. for $[M+NH_4]^+$: 436.2330]. The aromatic regions in the ^1H and ^{13}C NMR spectra of **1b** were identical to that of **1a**, suggesting that **1b** possessed also two 4-hydroxy-3-methoxyphenyl groups that was supported by the characteristic base peak at *m*/*z* 137 in its EI-MS spectrum (Uehara et al., 1987). The presence of one acetyl group and the distinctive difference between signals of C-3 and C-5 (δ 66.24 and 71.41 respectively) suggested that **1b** was a monoacetylated derivative of **1f** or **1g**. Acetylation of **1b** with acetic anhydride in pyridine introduced three additional acetyl groups to the molecule producing an optically inactive molecule whose ^1H NMR data were identical to those of **1h** reported in the literature (Kikuzaki et al., 1991a). Therefore, compound **1b** was assigned as (3*R,5S*)-3-acetoxy-5-hydroxy-1,7-bis(4-hydroxy-3-methoxyphenyl)-heptane.

Compound **1c** was obtained as colorless oil, $[\alpha]_D^{24}$ 0 (c 0.55, $CHCl_3$). Its HR-ESI-MS spectrum exhibited an $[M+NH_4]^+$ ion peak at *m*/*z* 424.2326 corresponding to a molecular formula $C_{22}H_{30}O_7$ (calc. for $M+NH_4$: 424.2330). Its ^1H and ^{13}C NMR and IR spectra were similar to those of the known compound (3*S*,5*S*)-3,5-dihydroxy-1-(4-hydroxy-3,5-dimethoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptane (**1i**) (Yamahara et al., 1992). However, appreciable differences in the ^{13}C NMR chemical shifts were observed between both molecules. That is, chemical shifts of C-1, C-4 and C-7 of **1c** were upfield 0.6, 0.5 and 0.5 ppm, respectively, while those of C-2/C-6 and C-3/C-5 were downfield 0.7 and 3.4 ppm, respectively, in comparison with those of **1i**. These facts suggested that **1c** is a stereoisomer of **1i**. Since **1i** was optically active ($[\alpha]_D^{20}$ −24.0°, *c* 1.0, EtOH) (Yamahara et al., 1992) while **1c** was optically inactive, and diarylheptanes isolated from rhizomes of *Zingiber officinale* all possess *S*-configuration at the C-5 position (Uehara et al., 1987; Kikuzaki et al., 1991a,b; Endo et al., 1990; Kikuzaki et al., 1992; Yu et al., 1998), compound **1c** was assigned as (3*R*,5*S*)-dihydroxy-1-(4-

hydroxy-3,5-dimethoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptane.

Compound **2a** was obtained as a colorless oil, $[\alpha]_D^{24}$ +3.0° (*c* 0.60, $CHCl_3$). Its HR-ESI-MS spectrum exhibited an $[M+NH_4]^+$ ion peak at *m/z* 434.2164 corresponding to a molecular formula of $C_{23}H_{28}O_7$ (calc. for $M+NH_4$: 434.2173). Its IR spectrum showed characteristic absorptions for hydroxyl (3434 cm^{-1}), carbonyl (1717 cm^{-1}) and aromatic (3015, 1606 and 1516 cm^{-1}) functionalities. The ^1H NMR signals at δ 6.81 (dd, *J* = 2.0, 8.0 Hz, 2H), 6.66 (*d*, *J* = 2.0 Hz, 2H) and 6.63 (*d*, *J* = 8.0 Hz, 2H), as well as those at δ 3.85 (*s*, 3H) and 3.87 (*s*, 3H), suggested the presence of two 1,3,4-trisubstituted phenyl groups bearing a methoxyl group that was supported by the characteristic base peak at *m/z* 137 ($[CH_2C_6H_3(OH)(OMe)]^+$) in its EI-MS spectrum for the 4-hydroxy-3-methoxyphenyl moiety in curcumin derivatives (Uehara et al., 1987). The ^1H NMR signal at δ 2.00 (3H, *s*) and the ^{13}C NMR signals at δ 21.04 and 170.41 revealed the presence of an acetyl group that was supported by the fragment ion peak at *m/z* 356 in the EI-MS spectrum from the deacetoxylation of the molecule. Comparison of its ^1H and ^{13}C NMR spectral data with those of hexahydrocurcumin (Uehara et al., 1987; Kikuzaki et al., 1991a,b) suggested that **2a** was an acetylated hexahydrocurcumin with an acetyl group at C-5. This was confirmed by its HMBC spectrum which showed a clear correlation between the acetyl carbonyl carbon (δ 170.41) and H-5 (δ 5.26) (Fig. 2). In order to determine the stereochemistry of C-5, **2a** was acetylated with acetic anhydride in pyridine to yield 5,4′,4″-triacetoxy-hexahydrocurcumin (**2f**) which was identical to the acetylation product of hexahydrocurcumin obtained under the same experimental conditions. Since the configuration of hexahydrocurcumin was known to be 5*S* (Uehara et al., 1987; Kikuzaki et al., 1991a,b), compound **2a** was assigned as (5*S*)-5-acetoxy-1,7-bis(4-hydroxy-3-methoxyphenyl)-heptan-3-one (5-acetyl-hexahydro-curcumin). Significant HMBC correlations are shown in Fig. 2.

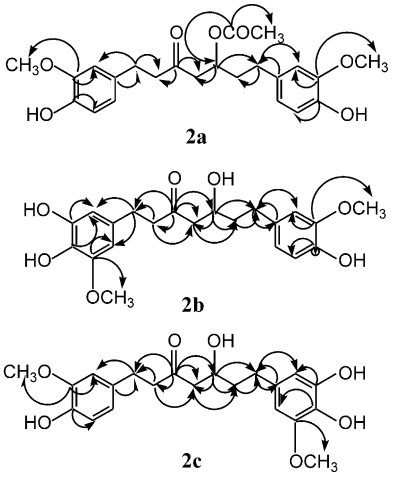

Fig. 2. Significant HMBC (C→H) correlations of **2a**, **2b** and **2c**.

Compounds **2b** and **2c** were obtained as a mixture in a colorless oil and could not be further separated even by HPLC. The HR-MALDI-MS spectrum exhibited a unique M + Na ion peak at *m/z* 413.15708 corresponding to the molecular formula $C_{21}H_{26}O_7$ (calc. for M + Na: 413.15762). The ^1H and ^{13}CNMR spectra of the mixture showed two sets of very similar signals for two hexahydrocurcumin derivatives with almost the same intensity. The EI-MS spectrum of the mixture showed a characteristic base peak at *m/z* 137 ($[CH_2C_6H_3(OH)(OMe)]^+$) for the 4-hydroxy-3-methoxyphenyl moiety in curcumin derivatives (Uehara et al., 1987) and a strong peak at *m/z* 153 (61%) corresponding to a fragment of ($[CH_2C_6H_2(OH)_2(OMe)]^+$). These suggested the presence of two phenyl rings with three and four substituents (MeO, OH, and CH_2, and MeO, OH, OH and CH_2, respectively). Careful analysis of their HMBC and H,H-COSY spectra enabled the two compounds to be distinguished as 5-hydroxy-1-(3,4-dihydroxy-5-methoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptan-3-one (**2b**) and 5-hydroxy-1-(4-hydroxy-3-methoxyphenyl)-7-(3,4-dihydroxy-5-methoxyphenyl)heptan-3-one (**2c**) respectively. Their significant HMBC correlations are shown in Fig. 2.

Compound **3** was obtained as a colorless oil, $[\alpha]_D^{16}$ −24.0° (*c* 0.19, EtOH). The HR-MALDI-MS spectrum exhibited an M + H ion peak at *m/z* 405.19078, corresponding to the molecular formula $C_{22}H_{28}O_7$ (calc. for M + H: 405.19132). The ^1H and ^{13}C NMR spectral data of **3** were very close to those of 1,5-epoxy-3-hydroxy-1-(3,4-dihydroxy-5-methoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptane (Kikuzaki et al., 1996) except that the 3-hydroxyl group of the 1-phenyl moiety in the latter was methylated in **3** as evidenced from its ^1H, ^{13}C and HMBC spectra. The coupling constants of H-3 (δ 4.22, *dddd*, *J* = 2.8, 3.0, 3.0 and 3.2 Hz) demonstrated its equatorial orientation; hence the 3-hydroxyl group is axial oriented. Therefore, **3** was assigned as 1,5-epoxy-3-hydroxy-1-(4-hydroxy-3,5-dimethoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptane. The structure was confirmed by the HMBC and H,H-COSY correlations as shown in Fig. 3.

The structures of the known compounds were identified by comparing their ^1H and ^{13}C NMR, MS and IR spectroscopic data and optical rotations with those reported in the literature as follows: (3*R*,5*S*)-3,5-di-

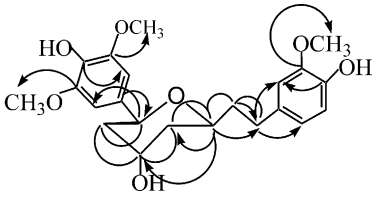

Fig. 3. Significant HMBC (C→H) and H,H-COSY (–) correlations of **3**.

acetoxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptane (**1d**) (Kikuzaki et al., 1991b), (3*R*,5*S*)-3,5-diacetoxy-1-(4-hydroxy-3,5-dimethoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptane (**1e**) (Kikuzaki et al., 1991b), (3*S*,5*S*)-3,5-dihydroxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptane (**1f**) (Kikuzaki et al., 1991a), (3*R*,5*S*)-3,5-dihydroxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptane (**1g**) (Kikuzaki et al., 1991a), 5-hydroxy-1-(4-hydroxy-3-methoxyphenyl)-7-(3,4-dihydroxyphenyl)heptan-3-one (**2d**) (Harvey, 1981), hexahydrocurcumin (**2e**) (Uehara et al., 1987; Kikuzaki et al., 1991b), 3-acetoxy-1,5-epoxy-1-(3,4-dihydroxy-5-methoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptane (**4**) (Kikuzaki et al., 1996), 1,7-bis(4-hydroxy-3-methoxyphenyl)hept-4-en-3-one (**5**) (Kikuzaki et al., 1991b), paradol (**6**) (Tackie et al., 1975), (*3S,5S*)-[6]-gingerdiol (**7a**) (Kikuzaki et al., 1992), (3*R*,5*S*)-[6]-gingerdiol (**7b**) (Kikuzaki et al., 1992), (3*R*,5*S*)-3,5-diacetoxy-[6]-gingerdiol (**7c**) (Kikuzaki et al., 1992), [4]-gingerol (**8a**) (Shoji et al., 1982; Denniff et al., 1980), [6]-gingerol (**8b**) (Shoji et al., 1982; Denniff et al., 1980), 5-acetoxy-[6]-gingerol (**8c**) (Shoji et al., 1982; Denniff et al., 1980), 1-(3,4-dimethoxyphenyl)-5-hydroxy-decan-3-one (**8d**) (Denniff et al., 1980), [8]-gingerol (**8e**) (Shoji et al., 1982; Denniff et al., 1980), [10]-gingerol (**8f**) (Shoji et al., 1982; Denniff et al., 1980), dehydrogingerdione (**9**) (Kiuchi et al., 1982), [6]-shogaol (**10a**) (Connel and Sutherland, 1969), [10]-shogaol (**10b**) (Connel and Sutherland, 1969), 1-(3-methoxy-4-hydroxy-phenyl)-propan-1,2-diol (11) (Kraus et al., 1990), galanolactone (**12**) (Morita and Itokawa, 1988), β-sitosterol (**13**) (Greca et al., 1990) and 6β-hydroxystigmast-4-en-3-one (**15**) (Greca et al., 1990).

Compound **7a** was obtained as colorless needles, mp 71–72 °C. This compound had been synthesized from reduction of [6]-gingerol (**8b**) (Kikuzaki et al., 1992), but not previously been obtained from ginger or other natural products.

Compounds **2d** and **11** had been identified by GC–MS (Harvey, 1981; Kraus et al., 1990), but no spectroscopic data were reported previously. In this paper, their structures were assigned completely by spectroscopic methods.

The in vitro cytotoxicity against human promyelocytic leukemia (HL-60) cells of most of the above mentioned compounds were tested using the [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide] (MTT) colorimetric assay (Price and McMillan, 1990) with etoposide (VP-16) as a positive control. None of them showed significant activity (data not shown).

3. Experimental

3.1. General

^{1}H, ^{13}C, and 2D NMR spectra were recorded on a Bruker AM 400 NMR spectrometer with TMS as internal standard. HR-ESI-MS and EI-MS data were obtained on a Bruker APEX II FT-MS and a HP-5988 MS spectrometers respectively. The IR spectra were taken on a Nicolet 170 SX IR spectrometer. Optical rotations were measured on a Perkin-Elmer 341 polarimeter. Melting points were determined on a Yanagimoto melting point apparatus and uncorrected.

3.2. Plant material

Ginger, the rhizome of *Zingiber officinale* Roscoe was collected in Hui county, Gansu province, China in October 2001. A voucher specimen (No. G011001) was preserved at the National Laboratory of Applied Organic Chemistry, Lanzhou University, Lanzhou, China and the plant sample was identified by Professor Yong-Hong Zhang at the Department of Chemistry, Lanzhou University.

3.3. Extraction and isolation

Slices of ginger were dried in the shade. The ground ginger (800 g) was extracted repeatedly (5 times, 3 days each time) with ethanol at room temperature. The ethanol extract (150 g) was suspended in water and successively extracted with petroleum ether, AcOEt and *n*-BuOH. The petroleum ether and AcOEt extracts (35 g and 30 g respectively) were subjected to column chromatography on silica gel (200–300 mesh) with a gradient system of petroleum ether–acetone (20:1, 15:1, 10:1, 5:1, 3:1, 1:1 and 0:1). Repeated chromatography of each fraction afforded **1a** (4 mg), **1b** (2 mg), **1c** (2 mg), **1d** (3 mg), **1e** (2 mg), **1f** (2 mg), **1g** (3 mg), **2a** (4 mg), **2b** and **2c** (2 mg), **2d** (3 mg), **2e** (3 mg), **3** (1 mg), **4** (1 mg), **5** (3 mg), **6** (50 mg), **7a** (20 mg), **7b** (15 mg), **7c** (50 mg), **8a** (10 mg), 8b (80 mg), **8c** (20 mg), **8d** (6 mg), **8e** (9 mg), **8f** (20 mg), **9** (8 mg), **10a** (10 mg), **10b** (6 mg), **11** (2 mg), **12** (10 mg), **13** (400 mg) and **14** (6 mg).

3.4. (3S,5S)-diacetoxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptane (*1a*)

Colorless oil. $[\alpha]_D^{26}$ +7.0° (*c* 0.68, $CHCl_3$); $[M+NH_4]^+$ *m*/*z* 478.2431 for $C_{25}H_{36}NO_8$ (Calc. 478.2435). IR ν_{max}^{KBr} cm^{-1}: 3438 (OH), 1731 (CO), 1607, 1516 (Ar). EI-MS *m*/*z* (rel. int.): 460 (15) $[M]^+$, 400 (4), 340 (3), 204 (5), 190 (17), 175 (7), 163 (13), 150 (12), 137 (100). For ^{1}H and ^{13}C NMR spectral data, see Table 1.

3.5. 3R-acetoxy-5S-hydroxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptane (*1b*)

Colorless oil; $[\alpha]_D^{24}$ +6.0° (*c* 0.56, $CHCl_3$). $[M+NH_4]^+$ *m*/*z* 436.2324 for $C_{23}H_{34}NO_7$ (calc. 436.2330). IR ν_{max}^{KBr} cm^{-1}: 3431 (OH), 1714 (CO), 1606, 1516 (Ar). EI-MS *m*/*z* (rel. int.): 418 (8) $[M]^+$, 358 (7), 190 (9), 163 (9), 150

Table 1
^{1}H and ^{13}C NMR chemical shifts for the new diarylheptanoids **1a**, **1b**, **1c**, **2a**, **2b**, **2c and 3**[a]

Posit.	**1a**[b]		**1b**[b]		**1c**[b]		**2a**[b]		**2b**[c]		**2c**[c]		**3**[c]	
	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C
1a	2.52 *dt* (15.6,7.8)		2.53 *dt* (14.0,6.8)		2.63 *dt* (13.6,6.8)						2.51 *dt* (14.4,7.6)			
1b	2.54 *dt* (15.9,7.8)	31.2	2.54 *dt* (13.2,6.4)	31.7	2.70 *dt* (13.6,6.4)	31.9	2.80 *t* (6.9)	29.3	2.68 *t* (6.8)	32.1	2.60 *dt* (13.2,7.6)	32.3	4.73 *d* (9.6)	74.4
2a			1.80 *ddd* (14.4,7.6, 4.0)										1.67 *ddd* (2.8,2.8,2.8)	
2b	1.84 *quart* (7.2)	36.7	1.91 *ddd* (14.4,8.8, 3.2)	36.9	1.77 *dt* (13.2,6.0)	40.1	2.69 *t* (6.3)	45.2	2.76 *m*	45.8	2.76 *m*	45.9	1.85 *ddd* (11.2,2.8,2.4)	41.7
3	5.00 *quint* (6.0)	69.7	5.11 *m*	71.4	3.89 *m*	72.5		206.9		210.2		210.2	4.22 *dddd* (3.2,3.0,3.0, 2.8)	63.6
4a	1.76 *dt* (11.7,5.7)		1.60 *m*		1.62 *dt* (13.6,6.4)		2.56 *dd* (16.5,6.9)						1.51 *ddd* (13.2,2.8,2.0)	
4b	1.92 *dt* (14.1,6.9)	38.6	1.62 *m*	43.1	1.77 *dt* (13.2,6.0)	43.2	2.68 *dd* (16.8,6.3,)	47.4	2.57 *dd* (16.4,7.6)	50.8	2.57 *dd* (16.4,7.6)	50.8	1.68 *m*	39.3
5	5.00 *quint* (6.0)	69.7	3.44 *m*	66.2	3.89 *m*	72.4	5.26 *quint* (6.6)	70.0	4.02 *quint* (6.0)	67.6	4.02 *quint* (6.0)	67.6	3.92 *m*	71.8
6a			1.65 *dt* (10.0,6.8)										1.68 *m*	
6b	1.84 *quart* (7.2)	36.7	1.74 *dt* (9.2,5.2)	38.9	1.77 *dt* (13.2,6.0)	40.1	1.86 *dt* (12.9,5.4)	36.0	1.67 *dt* (15.2,7.2)	40.2	1.67 *dt* (15.2,7.2)	40.1	1.77 *dddd* (13.6,5.6,4.8,3.2)	39.2
7a	2.52 *dt* (15.6,7.8)		2.61 *dt* (14.0,7.2)		2.63 *dt* (13.6,6.8)				2.51 *dt* (14.4,7.6)					
7b	2.54 *dt* (15.9,7.8)	31.2	2.73 *ddd* (14.4,9.2, 5.6)	31.8	2.70 *dt* (13.6,6.4)	31.4	2.56 *dt* (16.5,6.9)	31.3	2.60 *dt* (13.2,7.6)	32.3	2.74 *m*	32.1	2.67 *dt* (16.0,8.0)	31.9
1′		133.2		133.1		132.9		132.8		134.0		133.6		135.4
2′	6.67 *d* (1.8)	110.9	6.64 *d* (1.6)	111.0	6.42 *s*	105.0	6.66 *s*	111.1	6.34 *s*	109.6	6.80 *s*	112.9	6.68 *s*	104.5
3′		146.3		146.4		147.0		146.4		148.2		146.0		148.5
4′		143.7		143.9		132.9		143.9		133.1		134.5		135.8
5′	6.81 *d* (8.1)	114.2	6.81 *d* (8.0)	114.2		147.0	6.82 *d* (7.8)	114.3		148.9	6.71 *d* (8.0)	115.6		148.5
6′	6.62 *dd* (7.5,1.8)	120.8	6.64 *dd* (7.2,2.0)	120.9	6.42 *s*	105.0	6.64 *d* (7.8)	120.8	6.34 *s*	104.7	6.62 *d* (8.0)	121.4	6.68 *s*	104.5
1″		133.2		134.1		133.6		133.0		133.6		134.0		134.6
2″	6.67 *d* (1.8)	110.9	6.70 *d* (1.6)	111.1	6.70 *s*	111.0	6.66 *s*	111.0	6.80 *s*	113.0	6.34 *s*	109.7	6.79 *s*	113.0
3″		146.3		146.4		146.5		146.4		146.0		148.2		148.5
4″		143.7		143.7		143.8		144.0		134.5		133.1		145.4
5″	6.81 *d* (8.1)	114.2	6.82 *d* (8.8)	114.4	6.84 *d* (7.6)	114.3	6.82 *d* (7.8)	114.3	6.71 *d* (8.0)	115.6		148.9	6.71 *d* (8.4)	115.6
6″	6.62 *dd* (7.5,1.8)	120.8	6.66 *dd* (8.0,2.0)	120.9	6.69 *dd* (9.2,2.0)	120.9	6.65 *d* (7.8)	120.8	6.62 *d* (8.0)	121.5	6.34 *s*	104.6	6.63 *d* (8.0)	121.6
3-OAc	2.00 *s*	21.1 170.7	2.06 *s*	21.0,										
172.5														
5-OAc	2.01 *s*	21.1 170.7					2.00 *s*	21.0 170.4						
3′-OMe	3.87 *s*	55.8	3.87 *s*	55.9	3.87 *s*	56.3	3.85 *s*	55.9			3.79 *s*	56.2	3.82 *s*	56.7
5′-OMe					3.87 *s*	56.3			3.77 *s*	56.4			3.82 *s*	56.7
3″-OMe	3.87 *s*	55.8	3.87 *s*	55.9	3.87 *s*	55.9	3.87 *s*	55.9	3.80 *s*	56.2			3.78 *s*	56.2
5″-OMe											3.76 *s*	56.4		

[a] Coupling constants (Hz) in parentheses.
[b] Determined in $CDCl_3$.
[c] Determined in $(CD_3)_2CO$.

(7), 137 (100). For ^{1}H and ^{13}C NMR spectral data see Table 1.

3.6. (3R,5S)-3,5-dihydroxy-1-(4-hydroxy-3,5-dimethoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptane (**1c**):

Colorless oil; $[\alpha]_D^{24}$ 0 (c 0.55, $CHCl_3$); $[M+NH_4]^+$ *m/z* 424.2326 for $C_{22}H_{34}NO_7$ (Calc. 424.2330). IR ν_{max}^{KBr} cm^{-1}: 3412 (OH), 1726 (CO), 1612, 1517 (Ar). EI-MS *m/z* (rel. int.): 406 (15) $[M]^+$, 388 (3), 181 (12), 168 (86), 137 (83), 43 (100). For ^{1}H and ^{13}C NMR spectral data see Table 1.

3.7. (5S)-5-acetoxy-1,7-bis(4-hydroxy-3-methoxyphenyl)heptan-3-one (**2a**)

Colorless oil; $[\alpha]_D^{24}$ +3.0° (c 0.60, $CHCl_3$); $[M+NH_4]^+$ *m/z* 434.2164 for $C_{23}H_{32}NO_7$ (Calc. 434.2173). IR ν_{max}^{KBr} cm^{-1}: 3434 (OH), 1717 (CO), 1606, 1516 (Ar). EI-MS *m/z* (rel. int.): 416 (6) $[M]^+$, 356 (6), 177 (3), 163 (4), 150 (9), 137 (100). For HMBC correlations see Fig. 2. For ^{1}H and ^{13}C NMR spectral data see Table 1.

3.8. 5-Hydroxy-1-(3,4-dihydroxy-5-methoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptan-3- one (**2b**) and 5-hydroxy-1-(4-hydroxy-3-methoxyphenyl)-7-(3,4-dihydroxy-5-methoxyphenyl)heptan-3-one (**2c**)

Colorless oil. $[M+Na]^+$ *m/z* 413.15708 for $C_{21}H_{26}O_7Na$ (calc. 413.15762). IR ν_{max}^{KBr} cm^{-1}: 3411 (OH), 1701 (CO), 1607, 1518 (Ar). EI-MS *m/z* (rel. int.): 390 (11) $[M]^+$, 372 (3), 179 (8), 167 (11), 153 (61), 137 (100); For HMBC correlations, see Fig. 2. For ^{1}H and ^{13}C NMR spectral data see Table 1.

3.9. Acetylation of **1b**

Compound **1b** (1 mg) was acetylated with Ac_2O–pyridine at room temperature to give **1h** whose spectral data were identical to those reported in the literature (Kikuzaki et al., 1991a). $[\alpha]_D^{25}$ 0° (c 0.25, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$, TMS): δ 1.90–1.81 (6H, *m*, H-2, H-4 and H-6), 2.02 (6H, *s*, 3-OAc, 5-OAc), 2.30 (6H, *s*, 4′-OAc and 4″-OAc), 2.60 (4H, *t*, *J* = 7.8 Hz, H-1 and H-7), 3.82 (6H, *s*, 3′-OMe and 3″-OMe), 5.03 (2H, *m*, H-3 and H-5), 6.74 (2H, *d*, *J* = 7.5 Hz, H-5′ and H-5″), 6.78 (2H, *s*, H-2′ and H-2″), 6.93 (2H, *dd*, *J* = 8.1 and 1.8 Hz, H-6′ and H-6″). EIMS *m/z* (rel. int.): 544 (0.3) $[M]^+$, 502 (6), 442 (6), 400 (7), 340 (7), 190 (15), 163 (13), 150 (9), 137 (87), 43 (100).

3.10. Hydrolysis of **1a**

Compound **1a** (2 mg) was hydrolyzed with KOH/MeOH to give **1f** whose spectral data were identical to those reported in the literature (Kikuzaki et al., 1991a). $[\alpha]_D^{27}$ −7.0° (*c* 0.18, $CHCl_3$). ^{1}H NMR (400 MHz, $(CD_3)_2CO$, TMS): δ 1.57 (2H, *m*, H-4), 1.70 (4H, *m*, H-2 and H-6), 2.55 (2H, *dt*, *J* = 13.6 and 7.6 Hz, H-1a and H-7a), 2.68 (2H, *dt*, *J* = 13.6 and 8.0 Hz, H-1b and H-7b), 3.80 (6H, *s*, 3′-OMe and 3″-OMe), 3.88 (2H, *m*, H-3 and H-5), 6.63 (2H, *d*, *J* = 8.0 Hz, H-6′ and H-6″), 6.71 (2H, *d*, *J* = 8.0 Hz, H-5′ and H-5″), 6.80 (2H, *s*, H-2′ and H-2″). ^{13}C NMR (100 MHz, $(CD_3)_2CO$, TMS): δ 32.4 (C-1 and C-7), 40.9 (C-2 and C-6), 44.7 (C-4), 56.2 (3′-OMe and 3″-OMe), 68.3 (C-3 and C-5), 112.9 (C-2′ and C-2″), 115.6 (C-5′ and C-5″), 121.5 (C-6′ and C-6″), 134.8 (C-1′ and C-1″), 145.3 (C-4′ and C-4″), 148.1 (C-3′ and C-3″). EIMS *m/z* (rel. int.): 376 (10) $[M]^+$, 358 (7), 190 (6), 179 (5), 163 (6), 150 (8), 137 (100).

3.11. Acetylation of **2a**

Compound **2a** (2 mg) was acetylated with Ac_2O–pyridine at room temperature to give **2f** whose spectral data were identical to those of the acetylation product of hexahydrocurcumin (**2e**) under the same experimental conditions. (5*S*)-5-acetoxy-1,7-bis(4-acetoxy-3- methoxyphenyl)-heptan-3-one (**2f**): Colorless oil. $[\alpha]_D^{25}$ −2.0° (*c* 0.18, $CHCl_3$). $[M+NH_4]^+$ *m/z* 518.2387 for $C_{27}H_{32}O_9$ (calc. 518.2385). ^{1}H NMR (400 MHz, $CDCl_3$, TMS): δ 1.90 (2H, *dt*, *J* = 14.1 and 7.5 Hz, H-6), 1.99 (3H, *s*, 5-OAc), 2.30 (6H, *s*, 4′-OAc and 4″-OAc), 2.59 (2H, *dt*, *J* = 11.7 and 5.7 Hz, H-7), 2.63 (1H, *dd*, *J* = 13.5 and 7.5 Hz, H-4a), 2.73 (2H, *t*, *J* = 6.9 Hz, H-2), 2.74 (1H, *dd*, *J* = 11.7 and 4.8 Hz, H-4b), 2.86 (2H, *t*, *J* = 6.6 Hz, H-1), 3.80 (3H, *s*, 3′-OMe), 3.82 (3H, *s*, 3″-OMe), 5.28 (1H, *quint*, *J* = 6.3 Hz, H-5), 6.73 (2H, *d*, *J* = 7.5 Hz, H-5′ and H-5″), 6.78 (2H, *s*, H-2′ and H-2″), 6.92 (2H, *d*, *J* = 6.6 Hz, H-6′ and H-6″). EI-MS *m/z* (rel. int.): 500 (2) M^+, 458 (18), 398 (16), 356 (22), 137 (100).

3.12. 5-Hydroxy-1-(4-hydroxy-3-methoxyphenyl)-7-(3,4-dihydroxyphenyl)heptan-3-one (**2d**)

Colorless needles; m.p. 163–164 °C; $[\alpha]_D^{16}$ 0 (*c* 0.74, EtOH). $[M+NH_4]^+$ *m/z* 378.1923 for $C_{20}H_{24}O_6NH_4$ (calc. 378.1911). IR ν_{max}^{KBr} cm^{-1}: 3392 (OH), 1703 (CO), 1606, 1518 (Ar). ^{1}H NMR (400 MHz, $(CD_3)_2CO$,TMS): δ 1.66 (2H, *dt*, *J* = 14.4 and 7.2 Hz, H-6), 2.48 (1H, *dt*, *J* = 13.6 and 8.1 Hz, H-7a), 2.56 (2H, *d*, *J* = 6.0 Hz, H-4), 2.61 (1H, *dt*, *J* = 14.4 and 8.0 Hz, H-7b), 2.74 (2H, *t*, *J* = 6.8 Hz, H-1), 2.76 (2H, *t*, *J* = 6.8 Hz, H-2), 3.79 (3H, *s*, 3′-OMe), 4.01 (1H, *quint*, *J* = 6.0 Hz, H-5), 6.50 (1H, *d*, *J* = 8.8 Hz, H-6″), 6.62 (1H, *d*, *J* = 8.4 Hz, H-6′), 6.67 (1H, *s*, H-2″), 6.70 (2H, *d*, *J* = 7.6 Hz, H-5′ and H-5″), 6.80 (1H, *s*, H-2′). ^{13}C NMR (100 MHz, $(CD_3)_2CO$, TMS): δ 29.4 (C-1), 31.8 (C-7), 40.2 (C-6), 45.9 (C-2), 50.8 (C-4), 56.2 (3′-OMe), 67.6 (C-5), 112.9 (C-2′), 115.6 (C-5″), 115.8 (C-5′), 116.2 (C-2″), 120.3 (C-6″), 121.4 (C-6′), 133.6 (C-1′), 134.7 (C-1″), 143.7 (C-3″), 145.5 (C-4′), 145.6 (C-4″), 148.2 (C-3′), 210.3 (C-3). EI-MS *m/z* (rel. int.): 360 (6) $[M]^+$, 248 (8), 150 (16), 137 (100).

3.13. 1,5-Epoxy-3-hydroxy-1-(4-hydroxy-3,5-dimethoxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)heptane (**3**)

Colorless oil; $[\alpha]_D^{16}$ −24° (*c* 0.19, EtOH). $[M+H]^+$ *m/z* 405.19078 for $C_{22}H_{29}O_7$ (calc. 405.19132. IR ν_{max}^{KBr} cm^{-1}: 3399 (OH), 1731 (CO), 1615, 1519 (Ar). EI-MS *m/z* (rel. int.) 404 (4) $[M]^+$, 181 (19), 168 (36), 137 (100). For HMBC and H,H-COSY correlations see Fig. 3. For ^1H and ^{13}C NMR data see Table 1.

3.14. 1-(4-Hydroxy-3-methoxyphenyl)propan-1,2-diol (**11**)

Colorless oil. ^1H NMR (400 MHz, $CDCl_3$, TMS): δ 1.06 (3H, *d*, *J* = 6.4 Hz, 3-Me), 3.84 (1H, *dq*, *J* = 7.6 and 6.4 Hz, H-2), 3.90 (3H, *s*, 3′-OMe), 4.31 (1H, *d*, *J* = 7.6 Hz, H-1), 6.82 (1H, *dd*, *J* = 8.4 and 2.0 Hz, H-6′), 6.88 (1H, *d*, *J* = 2.4 Hz, H-2′), 6.89 (1H, *d*, *J* = 8.0 Hz, H-5′). ^{13}C NMR (100 MHz, $CDCl_3$, TMS): δ 18.8 (C-3), 56.0 (3′-OMe), 72.3 (C-1), 79.5 (C-2), 109.0 (C-2′), 114.3 (C-5′), 120.0 (C-6′), 133.1 (C-1′), 145.6 (C-4′), 146.7 (C-3′). EI-MS *m/z* (rel. int.): 198 (16) $[M]^+$, 153 (100), 125 (25), 93 (68), 65 (38).

3.15. Cytotoxicity assay

Human promyelocytic leukemia (HL-60) cell lines were obtained from Shanghai Institute of Biochemistry and Cell Biology, Chinese Academy of Sciences. The cells were cultured at 37 °C under a humidified atmosphere of 5% CO_2 in RPMI 1640 medium supplemented with 10% fetal serum and dispersed in a 96-well replicate plate with 1×10^4 cells/well. The cells were then incubated with 10–100 μM of compounds **1**–**11** or etoposide (VP-16) which was used as a positive control. After 48 h exposure to the toxins cell viability was determined using the [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide] (MTT) colorimetric assay (Price and McMillan, 1990) by measuring the absorbance at 570 nm with a Bio-Rad 550 ELISA microplate Reader. Each test was performed in triplicate.

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