

## Constituents from Moghat, the Roots of *Glossostemon bruguieri* (Desf.)

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**Abstract:** The new biflavone moghatin (3''-hydroxycupressuflavone) was isolated from Moghat, the dried peeled roots of *Glossostemon bruguieri* (Desf.), together with five known compounds: 4'-methoxyisoscuteellargin, sesamin, chrysophanol, emodin and methoxyemodin (physcion). The structures of these compounds were assigned on the basis of spectroscopic data. Occurrence of these compounds in Moghat is reported here for the first time.

**Keywords** Moghat, *Glossostemon bruguieri*, Sterculiaceae, biflavone, anthraquinone.

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### Introduction

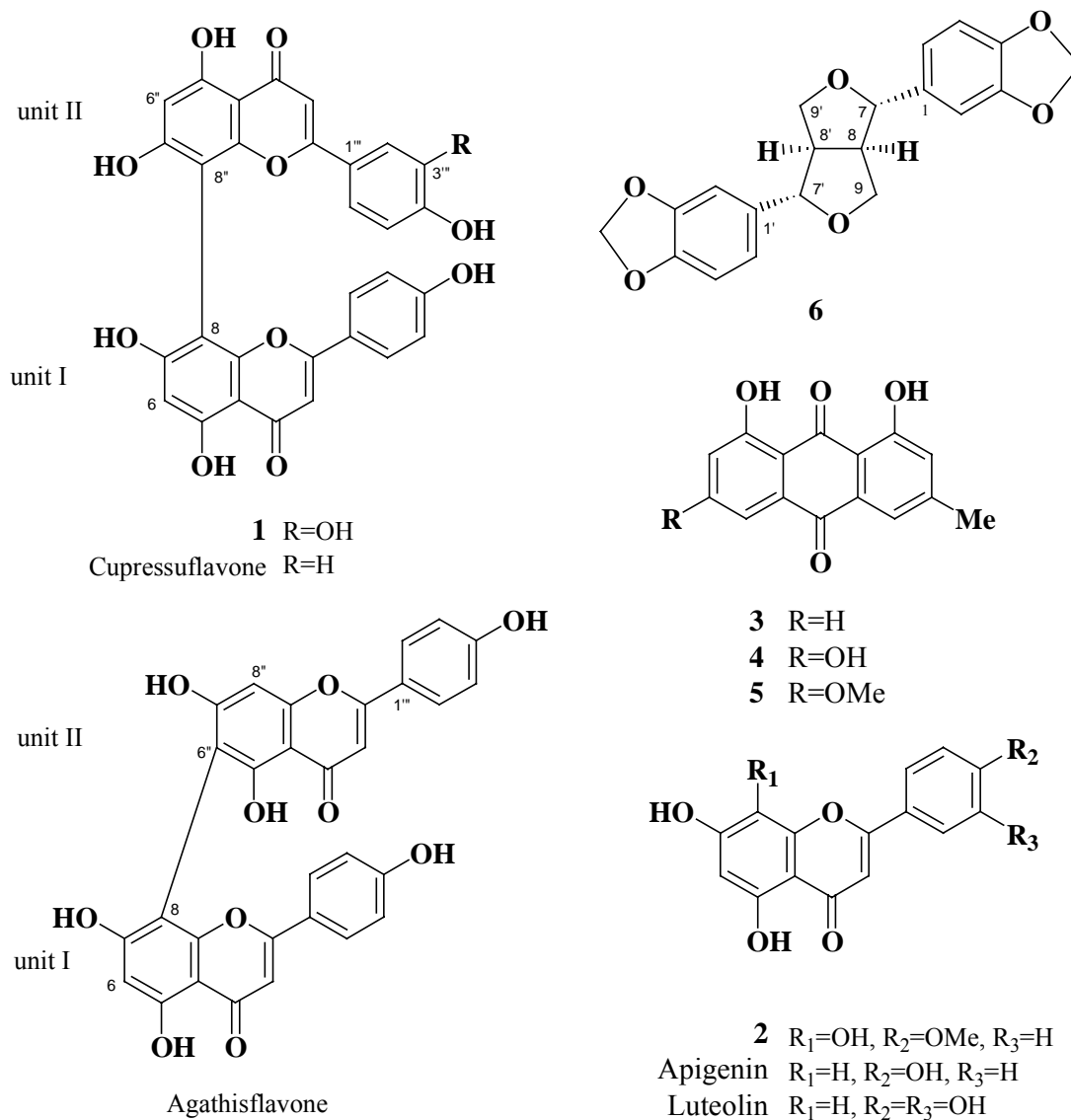
Plants of the cacao family (Sterculiaceae) are trees, shrubs, or herbs comprising about 65 genera and 1,000 species that are further characterized by the presence of stellate hairs and, in most cases, mucilaginous sap [1]. *Glossostemon bruguieri* (Desf.), family Sterculiaceae, is a shrub with thick long tapering dark colored roots (70-100 cm in length and 5-8 cm in breadth) [1, 2]. *G. bruguieri* is native to Iraq and Iran, and was cultivated in Egypt for its edible roots a long time ago. The dried peeled roots of *G. bruguieri* (Desf.) (known in Arabic as Moghat) are used in folk medicine for the treatment of gout and spasms, and as a tonic and nutritive agent [3]. After childbirth, women have especially used hot drinks of powdered Moghat as a general tonic and lactagogue. Due to its high content of mucilage (up to 27% based on dry weight) [2, 4-6], Moghat is also prescribed as a demulcent agent. Previous work

on the chemistry of the compounds produced from Moghat indicated the presence of oestrone [2,7], scopoletin, phytosterols (a mixture of  $\beta$ -sitosterol, stigmasterol and campesterol) [2, 8],  $\alpha$ -amyrin, and glucosides of flavone and chalcone [2]. In addition, amino acids and fatty acids were identified [2,4]. We report herein the isolation and structure elucidation of six compounds from Moghat roots.

## Results and Discussion

Fractionation and repeated chromatography of the MeOH extract of Moghat roots has led to the isolation of six compounds (**1—6**) (Figure 1).

Figure 1



Compounds **2-6** were previously obtained from natural sources and they were identified as 4'-methoxyisoscuteclargin (**2**) [9,10], chrysoferol (**3**) [11], emodin (**4**) [12], methoxyemodin

(physcion) (**5**) [11], and sesamin (**6**) [13,14], respectively, by comparing their physical and spectral data with those reported. Compound **1** is a new natural product and its structure was determined as follows.

Compound **1** was obtained as a yellow amorphous powder that gave a positive magnesium-hydrochloric acid reaction for flavonoids. Its UV spectrum showed absorption bands at 262 and 332 nm, characteristic for flavones. The IR spectrum showed absorptions for hydroxyl ( $3440\text{ cm}^{-1}$ ),  $\alpha,\beta$ -unsaturated C=O ( $1640\text{ cm}^{-1}$ ) and aromatic ( $1610$  and  $1580\text{ cm}^{-1}$ ) functions. Compound **1** was assigned the molecular formula  $\text{C}_{30}\text{H}_{18}\text{O}_{11}$  by EI-MS ( $m/z$  555  $[\text{M}+\text{H}]^+$ ), which suggested the presence of a biflavonoid structure. Examination of the  $^1\text{H-NMR}$  spectrum of **1** (see Experimental) with the aid of  $^1\text{H-}^1\text{H}$  COSY and HMQC experiments indicated that **1** is a biflavone composed of two asymmetric units, an apigenin moiety (**I**) [two doublets (each 2H,  $J=8.7$  Hz) at  $\delta$  6.89 and 8.02, and two singlets at  $\delta$  6.27 and 6.79] and a luteolin moiety (**II**) [three proton signals at  $\delta$  6.93 (d,  $J=8.4$  Hz), 7.44 (d,  $J=2.2$  Hz) and 7.53 (dd,  $J=2.2$  and 8.4 Hz), and two singlets at  $\delta$  6.28 and 6.67].

**Table 1.**  $^{13}\text{C-NMR}$  spectral data of **1**, related flavones and biflavones

Carbon No.	<b>1</b>	Apigenin*	Luteolin*	Cupressuflavone <sup>#</sup>	Agathisflavone <sup>#</sup>
2 (2'')	164.4 (164.4)	164.1	164.5 <sup>a</sup>	163.9 (163.9)	163.9 <sup>a</sup> (164.1) <sup>a</sup>
3 (3'')	102.9 (102.8)	102.8	103.3	102.8 (102.8)	102.8 <sup>b</sup> (103.1) <sup>b</sup>
4 (4'')	182.1 (182.0)	181.8	182.2	182.1 (182.1)	182.1 <sup>c</sup> (182.3) <sup>c</sup>
5 (5'')	164.1 (164.1)	157.3	157.9	161.3 (161.3)	160.9 (160.0)
6 (6'')	99.8 (99.7)	98.8	99.2	99.0 (99.0)	98.9 (103.6) <sup>d</sup>
7 (7'')	164.4 (164.4)	163.7	164.7 <sup>a</sup>	162.7 (162.7)	162.7 <sup>e</sup> (162.9) <sup>e</sup>
8 (8'')	104.2 (104.2)	94.0	94.2	98.7 (98.7)	99.4 (93.7)
9 (9'')	157.1 (157.1)	161.5	162.1	155.3 (155.3)	155.1 <sup>f</sup> (157.0) <sup>f</sup>
10 (10'')	104.2 (104.2)	103.7	104.2	104.3 (104.3)	104.0 <sup>d</sup> (103.8) <sup>d</sup>
1' (1''')	121.9 (121.5)	121.3	119.3	121.7 (121.7)	121.7 <sup>g</sup> (121.5) <sup>g</sup>
2' (2''')	128.9 (114.2)	128.4	113.8	127.9 (127.9)	128.2 <sup>h</sup> (128.6) <sup>h</sup>
3' (3''')	115.9 (145.8)	116.0	146.2	116.1 (116.1)	116.2 (116.2)
4' (4''')	161.5 (149.8)	161.1	150.1	161.1 (161.1)	161.2 (161.3)
5' (5''')	115.9 (116.1)	116.0	116.4	116.1 (116.1)	116.2 (116.2)
6' (6''')	128.9 (119.6)	128.4	121.7	127.9 (127.9)	128.2 <sup>k</sup> (128.6) <sup>k</sup>

Data marked <sup>#</sup> are taken from ref. [15] and those marked \* from ref. [16].

Assignments with the same superscript in the same column may be reversed.

When the  $^{13}\text{C-NMR}$  shift values of **1** were compared with those of related biflavones [15] and flavones [16], the involvement of C-8 and C-8'' in the interflavone linkage of the two asymmetric moieties in **1** was suggested (Table 1). Thus, when compared with monomeric flavones with a 5,7-

dihydroxy substitution [15], the C-8 carbons in both moieties were found to be shifted downfield (+ 10.0 ppm), whereas the carbon atoms C-6 and C-6'' in **1** resonate at almost identical chemical shift values ( $\delta$  99.8 and 99.7, respectively) for the respective carbon atoms as seen in monomers. Involvement of C-8 and C-8'' in an interflavonoid linkage was previously reported for cupressuflavone (Table 1), a symmetrical C-8, C-8'' biapigenin, previously isolated from *Cupressus obtusa* [15]. This finding was further supported by a HMBC experiment. Long-range correlation between the  $^1\text{H}$  signals at  $\delta$  6.27 and 6.28 (H-6 and H-6'', respectively) and the  $^{13}\text{C}$  signal at  $\delta$  104.2 was observed. This  $^{13}\text{C}$  signal showed also long range correlation with  $^1\text{H}$  signals at  $\delta$  6.67 and 6.79 (H-3'' and H-3, respectively). The structure of **1** was, therefore, determined to be a 3'''-hydroxy derivative of cupressuflavone and was given the name moghatin.

## Conclusions

In addition to mucilage, oestrone and phytosterol, the present study provides the first report on the presence of biflavones, methoxylated flavones, anthraquinones and lignans in Moghat. These constituents exhibit a variety of biological effects. Besides, sesamin has been found to be good for the liver, and has antihypertensive effects [17]. This broad spectrum of constituents suggests the possible utilization of Moghat as a valuable crude drug.

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## Experimental

### General

Optical rotations were measured with a Jasco DIP-360 automatic polarimeter. UV spectra were measured with a Shimadzu UV-VIS recording spectrophotometer.  $^1\text{H}$ - and  $^{13}\text{C}$ -NMR spectra were measured with a Jeol JNA-LA 400WB-FT spectrometer ( $^1\text{H}$  at 400 MHz,  $^{13}\text{C}$  at 100 MHz), the chemical shifts being reported in ppm with TMS as an internal standard. Electron impact (EI) mass spectra were measured with a Jeol JMS-AX 505 HAD mass spectrometer at an ionization voltage of 70 eV.

### Plant Material

The air-dried peeled roots of *G. bruguieri* (Desf.), family Sterculiaceae, were purchased from Harraz drugstore in Cairo in March 2001. The plant material was kindly identified by Prof. Dr. El-Sayed A. Aboutabl, Professor of Pharmacognosy, Faculty of Pharmacy, Cairo University, and a voucher specimen was deposited in the museum of the Department of Pharmacognosy of this university.

### Extraction and Isolation

Commercially available Moghat (900 g) was pulverized and extracted with portions of MeOH under reflux (2L x 4, 2h each). The MeOH solutions were combined and evaporated under reduced pressure to give 75 g of a viscous residue. This residue was again dissolved in MeOH (500 mL) under sonication and H<sub>2</sub>O (500 mL) was added. The organic solvent was evaporated under reduced pressure and the suspension was applied to a column of Diaion HP-20 (1 L). The column was washed with H<sub>2</sub>O (4 L), MeOH (3 L) and finally with CHCl<sub>3</sub> (3 L). The MeOH and CHCl<sub>3</sub> eluates were separately evaporated to dryness to give 37.5 g and 20 g of oily residues, respectively. The residue from the MeOH eluate was suspended in MeCN-H<sub>2</sub>O (9:1 v/v, 500 mL) and partitioned with n-hexane (500 mL x 4) to give an MeCN-sol. fraction and a hexane-sol. fraction. The MeCN-soluble fraction was evaporated to give 9.4 g of a dry residue. This residue was chromatographed on a column of reversed phase silica gel (RP-18, 32 x 4 cm, i.d.). Elution was started with MeOH-H<sub>2</sub>O (3:7 v/v, 200 mL), and then with 10% increments of MeOH [100 fractions (15 mL each) were collected]. Fractions 3 and 4 were pooled and evaporated to give a residue (670 mg). This residue was chromatographed on a Sephadex LH-20 column (20 x 2 cm, i.d.) with EtOH-H<sub>2</sub>O (3:7 v/v) elution and 20 mL subfractions were collected to give compound **1** (35 mg) from subfractions 9 and 10.

Compound **2** (83 mg) was obtained as a light yellow amorphous powder from fractions 5-11 after crystallization of the crude isolate (120 mg) from MeOH.

The residue from the CHCl<sub>3</sub> eluate was similarly suspended in MeCN-H<sub>2</sub>O (9:1 v/v, 400 mL), and partitioned with n-hexane (400 mL x 4). After evaporation, an MeCN-sol. fraction (2.8 g) of the CHCl<sub>3</sub> eluate was obtained. This residue was chromatographed on a column of silica gel (30 x 3.5 cm, i.d.). After the column was washed with n-hexane (1 L), elution was started with 2% increment of acetone in n-hexane (250 mL each), and 60 fractions (each of 15 mL) were obtained. Fractions 2-7 (1.2 g, eluted with 2% acetone in n-hexane) was chromatographed on a column of silica gel (20 x 2 cm, i.d.) eluting with n-hexane (50 mL), and then successively with 200 mL of n-hexane-EtOAc (8:2 and then 6:4 v/v). The fractions eluted with n-hexane-EtOAc (20 mg) were pooled and applied to a column of reversed phase silica gel (RP-18, 20 x 1.5 cm, i.d.). Elution with MeCN-H<sub>2</sub>O (9:1 v/v) gave **5** (physcion, 8 mg) as fine orange needles (MeOH) from subfractions 4 and 5, while subfraction 7 gave **3** (chrysophanol, 7 mg) as orange needles from a hexane-acetone mixture. On concentration, fractions 36-39 (eluted with 6% acetone in n-hexane) gave **6** (sesamin, 21 mg) as colorless prisms from n-hexane-acetone mixture. Fractions 49-56 (eluted with 6-8% acetone in n-hexane, 20 mg) was further purified by MPLC

[column: RP-18 (Merck, size A); mobile phase: MeOH-H<sub>2</sub>O (8:2 v/v), flow rate: 10 mL/min, and fractions of 10 mL were collected] to give **4** (emodin, 8 mg) from subfractions 9-12 as fine orange crystals from n-hexane.

*Moghatin (1)*: Yellow amorphous powder;  $[\alpha]_D + 2.0^\circ$  (MeOH, *c* 0.1); UV  $\lambda_{\max}$  (log  $\epsilon$ ): 262 (4.2), 330 (4.1) nm; IR  $\nu_{\max}$  (KBr): 3440, 1640, 1610, 1580, 1165  $\text{cm}^{-1}$ ; EI-MS: *m/z* 555  $[\text{M}+\text{H}]^+$ , 428, 354, 280, 206; <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>)  $\delta$ : 6.27 (1H, s, H-6), 6.28 (1H, s, H-6''), 6.67 (1H, s, H-3''), 6.79 (1H, s, H-3), 6.89 (2H, d, *J* = 8.7 Hz, H-3' and H-5'), 6.93 (1H, d, *J* = 8.4 Hz, H-5'''), 7.44 (1H, d, *J* = 2.2 Hz, H-2'''), 7.53 (1H, dd, *J* = 2.2 and 8.4 Hz, H-6'''), and 8.02 (2H, d, *J* = 8.7 Hz, H-2' and H-6'); <sup>13</sup>C-NMR (DMSO-*d*<sub>6</sub>)  $\delta$ : 99.7 (C-6''), 99.8 (C-6), 102.8 (C-3''), 102.9 (C-3), 104.2 (C-8, C-8'', C-10, and C-10''), 114.2 (C-2'''), 115.9 (C-3'), 116.1 (C-5'''), 119.6 (C-6'''), 121.5 (C-1'''), 121.9 (C-1'), 128.9 (C-2'), 145.8 (C-3'''), 149.8 (C-4'''), 157.1 (C-9, C-9''), 161.5 (C-4'), 164.1 (C-5, C-5''), 164.4 (C-2, C-2'', C-7, C-7''), 182.0 (C-4''), and 182.1 (C-4).

*4'-Methoxyisoscuteallargin (2)*: Light yellow amorphous powder; UV  $\lambda_{\max}$  (log  $\epsilon$ ): 270 (2.8), 300 (2.1, sh), 326 (3.2), 360 (2.3, sh) nm; IR  $\nu_{\max}$  (KBr): 3420, 1660, 1610, 1580  $\text{cm}^{-1}$ ; EI-MS: *m/z* 300  $[\text{M}]^+$ , 285  $[\text{M}-\text{CH}_3]^+$ , 206, 193, 177, 133  $[(\text{M}-\text{C}_9\text{H}_9\text{O})+\text{H}]^+$ , 94; <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>)  $\delta$ : 3.84 (3H, s, -OCH<sub>3</sub>), 6.28 (1H, s, H-6), 6.84 (1H, s, H-3), 7.10 (2H, d, *J* = 8.7 Hz, H-3' and H-5'), 8.09 (2H, d, *J* = 8.7 Hz, H-2' and H-6'), 9.93 (1H, br s, -OH), and 12.68 (1H, br s, OH); <sup>13</sup>C-NMR (DMSO-*d*<sub>6</sub>)  $\delta$ : 55.5 (-OCH<sub>3</sub>), 99.5 (C-6), 103.2 (C-3), 103.9 (C-10), 114.5 (C-3' and C-5'), 121.1 (C-8), 122.9 (C-1'), 128.5 (C-2' and C-6'), 149.7 (C-9), 156.9 (C-5), 157.0 (C-7), 162.4 (C-4'), 163.4 (C-2), and 181.9 (C-4).

*Chrysophanol (3)*: Orange needles, m.p. 197-199°C (lit.[18] m.p. 196°C); EI-MS: *m/z* 254  $[\text{M}]^+$ ; <sup>1</sup>H-NMR (CDCl<sub>3</sub>)  $\delta$ : 2.46 (3H, s, CH<sub>3</sub>), 7.08 (1H, br s, H-2), 7.27 (1H, dd, *J* = 1.1 and 8.4 Hz, H-7), 7.64 (1H, br s, H-4), 7.67 (1H, d, *J* = 8.4 Hz, H-6), 7.81 (1H, br d, *J* = 8.4 Hz, H-5), 11.99 (1H, s, -OH), and 12.10 (1H, s, -OH); <sup>13</sup>C-NMR (CDCl<sub>3</sub>)  $\delta$ : 22.3 (CH<sub>3</sub>), 113.8 (C-12), 115.9 (C-13), 119.9 (C-7), 121.4 (C-4), 124.4 (C-5), 133.3 (C-14), 136.9 (C-6), 149.3 (C-3), 162.4 (C-1), 162.7 (C-8), 181.9 (C-10), and 192.5 (C-9).

*Emodin (4)*: Fine orange needles, m.p. 263-265°C (lit.[11] m.p. 264-265°C); EI-MS: *m/z* 270  $[\text{M}]^+$ ; <sup>1</sup>H-NMR (2:1 CDCl<sub>3</sub>+ MeOH-*d*<sub>4</sub>)  $\delta$ : 2.44 (3H, s, CH<sub>3</sub>), 6.58 (1H, d, *J* = 2.2 Hz, H-7), 7.06 (1H, s, H-2), 7.21 (1H, d, *J* = 2.5 Hz, H-5), and 7.56 (1H, br s, H-4); <sup>13</sup>C-NMR (2:1 CDCl<sub>3</sub>-*d*<sub>1</sub>+ MeOH-*d*<sub>4</sub>)  $\delta$ : 20.9 (-CH<sub>3</sub>), 107.6 (C-7), 108.7 (C-5), 109.0 (C-12), 113.1 (s, C-13), 120.3 (C-4), 123.6 (C-2), 132.7 (C-14), 134.8 (C-11), 147.5 (C-3), 161.6 (C-1), 164.7 (C-8), 165.6 (C-6), 181.9 (C-10), and 189.8 (C-9).

*Methoxyemodin (Physcion) (5)*: Orange needles, m.p. 210-212°C (lit. [18] 207°C); EI-MS: *m/z* 284  $[\text{M}]^+$ ; <sup>1</sup>H-NMR (CDCl<sub>3</sub>)  $\delta$ : 2.45 (3H, s, CH<sub>3</sub>), 3.50 (3H, s, -OCH<sub>3</sub>), 6.69 (1H, d, *J* = 2.6 Hz, H-7), 7.08 (1H, br s, H-2), 7.36 (1H, d, *J* = 2.6 Hz, H-5), 7.62 (1H, br s, H-4), 12.10 (1H, s, -OH), and 12.31 (1H,

s, -OH);  $^{13}\text{C}$ -NMR ( $\text{CDCl}_3$ )  $\delta$ : 22.2 ( $\underline{\text{C}}\text{H}_3$ ), 56.1 ( $-\text{O}\underline{\text{C}}\text{H}_3$ ), 106.8 (C-7), 108.2 (C-5), 113.7 (C-12 and C-13), 121.3 (C-4), 124.5 (C-2), 133.3 (C-14), 135.3 (C-11), 148.5 (C-3), 162.5 (C-8), 165.2 (C-1), 166.6 (C-6), 182.0 (C-10), and 190.8 (C-9).

*Sesamin* (**6**): Colorless prisms, m.p. 127-129°C; EI-MS:  $m/z$  354  $[\text{M}]^+$ , 323, 203, 161, 149, 135, 103;  $^1\text{H}$ -NMR ( $\text{CDCl}_3$ )  $\delta$ : 3.04 (2H, m, H-8 and H-8'), 3.84 (2H, m, H<sub>b</sub>-9 and H<sub>b</sub>-9'), 4.23 (2H, m, H<sub>a</sub>-9 and H<sub>a</sub>-9'), 4.71 (2H, m, H-7 and H-7'), 5.92 [4H, s,  $(-\text{O}-\underline{\text{C}}\text{H}_2-\text{O}-)_2$ ], 6.76-6.84 (6H, m, aromatic protons);  $^{13}\text{C}$ -NMR ( $\text{CDCl}_3$ )  $\delta$ : 54.4 (C-8 and C-8'), 71.7 (C-9 and C-9'), 85.8 (C-7 and C-7'), 101.1 [ $(-\text{O}-\underline{\text{C}}\text{H}_2-\text{O}-)_2$ ], 106.5 (C-6 and C-6'), 108.2 (C-3 and C-3'), 119.4 (C-2 and C-2'), 135.1 (C-1 and C-1'), 147.1 (C-4 and C-4'), and 148.0 (C-5 and C-5').

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*Sample Availability:* Available from the author.

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