

New Phenylphenalene Derivatives from Water Hyacinth (*Eichhornia crassipes*)

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Water hyacinth (*Eichhornia crassipes*) is a cause of great concern in terms of environmental and agricultural impacts in many parts of the world. Phytochemical investigation of water hyacinth led to the isolation of six new phenylphenalenes, 2,3-dihydro-3,9-dihydroxy-5-methoxy-4-phenyl-1*H*-phenalen-1-one (**1**), 2,3-dihydro-8-methoxy-9-phenyl-1*H*-phenalene-1,4-diol (**2**), 2,3-dihydro-4,8-dimethoxy-9-phenyl-1*H*-phenalen-1-ol (**3**), 2,3-dihydro-9-(4-hydroxyphenyl)-8-methoxy-1*H*-phenalene-1,4-diol (**4**), 2,6-dimethoxy-9-phenyl-1*H*-phenalen-1-one (**5**), and 7-(4-hydroxyphenyl)-5,6-dimethoxy-1*H*-phenalen-1-one (**6**), together with the four known compounds **7–10**. Their structures were elucidated by spectrometric methods including 1D- and 2D-NMR, and MS analysis. These compounds may be involved in allelopathic interactions of water hyacinth with neighboring plants.

Introduction. – Water hyacinth (*Eichhornia crassipes* (MART.) SOLMS), a plant of the family Pontederiaceae, causes globally annual losses of more than \$100 million to hydroelectricity generation, irrigation schemes, fisheries, riparian communities, and water transport [1]. The free-floating aquatic weed originating from the Amazon basin in South America has been introduced by man in China in the early 20th century [2]. However, water hyacinth possesses the ability to absorb heavy metals which accumulate in the root, making this plant of little use. Previous studies reported the isolation of some phenylphenalenes from water hyacinth [3–5]. We collected the material of water hyacinth in Dianchi Lake at Kunming where the lake is severely polluted. Six new compounds, 2,3-dihydro-3,9-dihydroxy-5-methoxy-4-phenyl-1*H*-phenalen-1-one (**1**), 2,3-dihydro-8-methoxy-9-phenyl-1*H*-phenalene-1,4-diol (**2**), 2,3-dihydro-4,8-dimethoxy-9-phenyl-1*H*-phenalen-1-ol (**3**), 2,3-dihydro-9-(4-hydroxyphenyl)-8-methoxy-1*H*-phenalene-1,4-diol (**4**), 2,6-dimethoxy-9-phenyl-1*H*-phenalen-1-one (**5**), and 7-(4-hydroxyphenyl)-5,6-dimethoxy-1*H*-phenalen-1-one (**6**), together with four known ones, hydroxylanigorufone (= 2-hydroxy-9-(4-hydroxyphenyl)-1*H*-phenalen-1-one; **7**) [3], 5,6-dimethoxy-7-phenyl-1*H*-phenalen-1-one (**8**) [4], 2-hydroxy-8-(4-hydroxyphenyl)-1*H*-phenalen-1-one (**9**) [5], and methyl ferulate (= methyl 3-(4-hydroxy-3-methoxyphenyl)prop-2-enoate; **10**), were isolated from the whole plant of *E. crassipes* (Fig. 1). The known compounds have already been reported in this species.

Results and Discussion. – Compound **1** was found to possess the molecular formula C₂₀H₁₆O₄ according to its ¹H- and ¹³C-NMR and DEPT spectra, which was confirmed

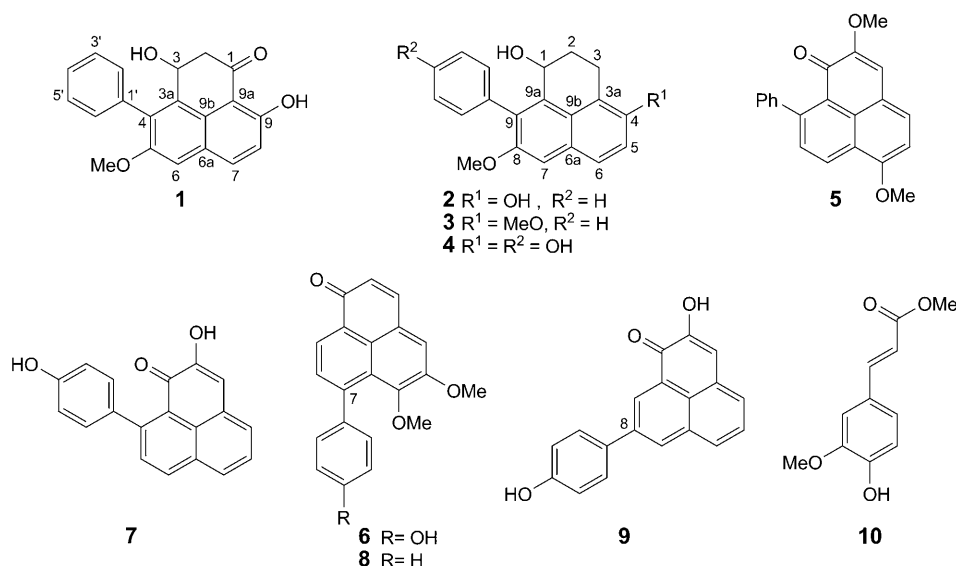


Fig. 1. Compounds **1**–**10** isolated from water hyacinth (*Eichhornia crassipes*)

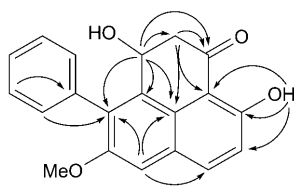
by the HR-ESI-MS (m/z 321.1131 ($[M+H]^+$), corresponding to 13 degrees of unsaturation in the molecule. The ^{13}C -NMR (DEPT) spectra showed 20 C-atom signals, including one MeO ($\delta(\text{C})$ 56.0 (q)), one CH_2 ($\delta(\text{C})$ 43.9 (t)), and nine CH groups ($\delta(\text{C})$ 136.8, 105.6, 128.6 (2 CH), 126.9 (2 CH), 126.7, 116.5, and 37.5), and nine quaternary C-atoms ($\delta(\text{C})$ 202.5, 161.2, 144.7, 145.4, 143.2, 128.0, 121.6, 115.8, and 110.1). Comparison of the ^1H - and ^{13}C -NMR data of **1** with those of known compounds from this species (Tables 1 and 2), in combination with the HMBC data, suggested that **1** had a phenalene skeleton substituted by a phenyl group [5][6].

In the HMBC spectrum of **1**, the correlations between $\delta(\text{H})$ 5.01 (d , $J = 7.5$ Hz) and the C-atom signals at $\delta(\text{C})$ 43.9 (t , C(2)), 202.5 (s , C(1)), 143.2 (s), 115.8 (s), and 128.0 (s) enabled us to assign this H-atom signal to H–C(3). From the correlations between CH_2 (2) ($\delta(\text{H})$ 3.06 (d , $J = 15.0$ Hz)) and 3.36 (dd , $J = 7.5$ Hz, 15.0)) and $\delta(\text{C})$ 37.5 (d , C(3)), 143.2 (s), 115.8 (s), 110.1 (s), and 128.0 (s), the signals at $\delta(\text{C})$ 143.2, 115.8, 110.1, and 128.0 were attributed to C(4), C(3a), C(9a), and C(9b), respectively. The HMBCs from $\delta(\text{H})$ 12.80 (s) to $\delta(\text{C})$ 110.1 (C(9a)) and 116.5 (d) indicated the OH group to be at C(9), and the latter C-atom signal consequently to be C(8). So, $\delta(\text{H})$ 7.01 (d , $J = 9.0$ Hz) was attributed to H–C(8) from the HSQC spectrum, and its coupled signal at $\delta(\text{H})$ 7.82 (d , $J = 9.0$ Hz) should be H–C(7), which was supported in the $^1\text{H},^1\text{H}$ -COSY plot. In the HBMBC experiment, the H-atom at $\delta(\text{H})$ 7.05 (s , 1 H) correlated with $\delta(\text{C})$ 136.8 (d , C(7)), 143.2 (s , C(4)), and 128.0 (s , C(9b)) and thus should be H–C(6). The H-atoms at $\delta(\text{H})$ 7.16 (d , $J = 7.5$ Hz, H–C(2',6')) showed correlation with C(4), which indicated that the phenyl substituent was attached to C(4) (Fig. 2). Finally, the MeO group should be attached at the left quaternary C-atom ($\delta(\text{C})$ 145.4 (s , C(5))) which was supported by HMBC cross-peaks between C(5) and

Table 1. ^{13}C -NMR Data of Compounds **1**–**6** in CDCl_3

	1	2	3	4^{a)}	5	6^{b)}
C(1)	202.5 (s)	37.8 (d)	38.0 (d)	37.8 (d)	179.8 (d)	184.2 (s)
C(2)	43.9 (t)	28.9 (t)	29.0 (t)	29.8 (t)	152.1 (s)	127.5 (d)
C(3)	37.5 (d)	17.9 (t)	18.0 (t)	18.8 (t)	111.9 (d)	142.3 (d)
C(3a)	115.8 (s)	116.1 (s)	120.1 (s)	117.0 (s)	121.4 (s)	125.8 (s)
C(4)	143.2 (s)	148.2 (s)	152.1 (s)	150.2 (s)	130.6 (d)	121.5
C(5)	145.4 (s)	114.9 (d)	110.8 (d)	115.6 (d)	104.6 (d)	150.4 (s)
C(6)	105.6 (d)	125.6 (d)	125.2 (d)	126.1 (d)	157.4 (s)	147.3 (s)
C(6a)	121.6 (s)	123.7 (s)	123.7 (s)	124.1 (s)	124.3 (s)	127.8 (s)
C(7)	136.8 (d)	104.5 (d)	104.1 (d)	105.4 (d)	128.3 (d)	145.6 (s)
C(8)	116.5 (d)	145.2 (s)	145.4 (d)	146.6 (s)	130.6 (d)	130.9 (d)
C(9)	161.2 (s)	142.2 (s)	142.1 (s)	143.5 (s)	148.4 (s)	126.8 (d)
C(9a)	110.1 (s)	117.6 (s)	117.7 (s)	118.9 (s)	126.3 (s)	133.4 (s)
C(9b)	128.0 (s)	127.4 (s)	127.4 (s)	128.5 (s)	125.7 (s)	123.7 (s)
C(1')	144.7 (s)	144.5 (s)	144.7 (s)	136.7 (s)	142.9 (s)	138.7 (s)
C(2')	128.6 (d)	127.8 (d)	127.8 (d)	115.4 (d)	127.8 (d)	129.6 (d)
C(3')	126.9 (d)	128.0 (d)	128.0 (d)	129.6 (d)	128.1 (d)	113.8 (d)
C(4')	126.7 (d)	125.9 (d)	125.7 (d)	156.1 (d)	126.8 (d)	156.3 (s)
C(5')	126.9 (d)	128.0 (d)	128.0 (d)	129.6 (d)	128.1 (d)	113.8 (d)
C(6')	128.6 (d)	127.8 (d)	127.8 (d)	115.4 (d)	127.8 (d)	129.6 (d)
MeO–C(2)					55.4 (q)	
MeO–C(4)			56.3 (q)			
MeO–C(5)	56.0 (q)					60.2 (q)
MeO–C(6)					55.8 (q)	56.7 (q)
MeO–C(7)		55.7 (q)	55.7 (q)	55.9 (q)		

^{a)} Measured in (D_6)acetone. ^{b)} Measured in (D_6)DMSO.

Fig. 2. Key HMBCs (H \rightarrow C) of **1**

H–C(6) and the MeO group ($\delta(\text{H})$ 3.98 (s)). So, **1** was structurally elucidated to be 2,3-dihydro-3,9-dihydroxy-5-methoxy-4-phenyl-1*H*-phenalen-1-one.

The ^1H - and ^{13}C -NMR (DEPT) spectra of **2** indicated its molecular formula as $\text{C}_{20}\text{H}_{18}\text{O}_3$, which was supported by the HR-ESI-MS at m/z 307.1325 ($[M + \text{H}]^+$), corresponding to twelve degrees of unsaturation. Comparison of the ^1H - and ^{13}C -NMR spectra of **2** with those of **1** indicated that **2** was similar to **1**, except that the C=O C-atom at $\delta(\text{C})$ 202.5 (s, C(1)) and the chelated OH group ($\delta(\text{H})$ 12.8 (OH–C(9))) of **1** were replaced in **2** by a CH_2 group ($\delta(\text{C})$ 17.9 (t, C(3))) and an OH group appearing at higher field ($\delta(\text{H})$ 4.87 (s)) which indicated the absence of the ketone function. So, the structure of **2** was 2,3-dihydro-8-methoxy-9-phenyl-1*H*-phenalene-1,4-diol.

Compound **3** had the molecular formula $\text{C}_{21}\text{H}_{20}\text{O}_3$ on the basis of the HR-ESI-MS (m/z 321.1493 ($[M + \text{H}]^+$)). Comparison of the ^1H - and ^{13}C -NMR spectra of **3** with

Table 2. $^1\text{H-NMR}$ Data of Compounds **1–6** in CDCl_3 , δ in ppm, J in Hz.

	1 ^{a)}	2	3	4 ^{b)}	5	6
H–C(1)		4.83 (br. s)	4.80 (br. s)	4.72 (s)		
CH_2 (2) or H–C(2)	3.06 (d, $J=15.0$), 3.36 (dd, $J=7.5, 15.0$)	2.16–2.19 (m), 2.30 (br. d, $J=16$)	2.14–2.18 (m), 2.24 (dd, $J=5, 10$)	2.02–2.05 (m), 2.18 (dd, $J=4.5, 10$)		6.70 (d, $J=9.6$)
H–C(3) or CH_2 (3)	5.01 (d, $J=7.5$)	2.52 (dt, $J=5, 16$), 2.93 (br. d, $J=16$)	2.47 (dt, $J=5, 15$), 3.11 (br. d, $J=15$)	2.40 (dt, $J=4.5, 15.0$), 3.05 (br. d, $J=15.0$)	6.82 (s)	7.71 (d, $J=9.6$)
H–C(4)					7.60 (d, $J=8.0$) 6.89 (d, $J=8.0$)	7.57 (s)
H–C(5)		6.96 (d, $J=9.0$)	7.12 (d, $J=8.8$)	6.97 (d, $J=7.5$)		
H–C(6)	7.05 (s)	7.52 (d, $J=9.0$)	7.60 (d, $J=8.8$)	7.47 (d, $J=7.5$)		
H–C(7)	7.82 (d, $J=9.0$)	7.06 (s)	7.05 (s)	7.14 (s)		
H–C(8)	7.01 (d, $J=9.0$)				8.63 (d, $J=8.4$) 7.57 (d, $J=8.4$)	7.56 (d, $J=7.6$) 8.54 (d, $J=7.6$)
H–C(9)						7.28 (d, $J=7.5$) 6.89 (d, $J=7.5$)
H–C(2',6')	7.16 (d, $J=7.5$)	7.00 (d, $J=7.5$)	7.00 (d, $J=7.5$)	6.76 (d, $J=7.5$)		
H–C(3',5')	7.10 (t, $J=7.5$)	7.20 (t, $J=7.5$)	7.20 (t, $J=7.5$)	6.63 (t, $J=7.5$)		
H–C(4')	7.13 (t, $J=7.5$)	7.15 (t, $J=7.5$)	7.15 (t, $J=7.5$)			
OH–C(3) or OH–C(1)	6.20 (s)	5.90 (s)	5.89 (s)	7.42 (s)		
MeO–C(2)					3.86 (s)	
OH–C(9) or OH–C(4)	12.80 (s)	4.87 (s)		7.92 (s)		
MeO–C(4)			3.90 (s)			
MeO–C(5)	3.98 (s)					4.01 (s)
MeO–C(6)					4.08 (s)	3.30 (s)
MeO–C(8)		3.97 (s)	3.97 (s)	3.91 (s)		
OH–C(4')				7.97 (s)		

^{a)} Measured in (D_6)acetone.

those of **2** indicated that the two compounds were very similar, except for an MeO group in **3** instead of an OH group at C(4) in **2**. The location of this MeO group at C(4) of **3** was supported by an HMBC between $\delta(\text{H})$ 3.90 (*s*, MeO) and $\delta(\text{C})$ 152.1 (*s*, C(4)). So, the structure of **3** was elucidated to be 2,3-dihydro-4,8-dimethoxy-9-phenyl-1*H*-phenalen-1-ol.

The HR-ESI-MS (m/z 323.1282 ($[M + H]^+$)) of **4** indicated its molecular formula as $\text{C}_{20}\text{H}_{18}\text{O}_4$. Comparison of the ^1H - and ^{13}C -NMR spectra of **4** with those of **2** established that the two compounds were similar, with the exception of the presence of one more OH group in **4**. This additional OH group was deduced to be connected to C(4') from the coupling pattern of the H-atoms of the aryl substituent. So, the structure of **4** was 2,3-dihydro-9-(4-hydroxyphenyl)-8-methoxy-1*H*-phenalene-1,4-diol.

The NMR and DEPT spectra of **5** suggested its molecular formula to be $\text{C}_{21}\text{H}_{16}\text{O}_3$, which was supported by the HR-ESI-MS. Comparison of 1D- and 2D-NMR spectra of **5** with those of the known compound **7** suggested that there were two MeO groups in **5** instead of an OH group in **7**. One was located at C(2) on the basis of an HMBC between $\delta(\text{H})$ 3.86 (*s*, MeO) and $\delta(\text{C})$ 152.1 (*s*, C(2)). The other MeO group was deduced to be at C(6) from HMBCs of $\delta(\text{C})$ 157.4 (*s*, C(6)) with $\delta(\text{H})$ 4.08 (*s*, MeO), 8.63 (*d*, $J = 8.4$ Hz, H–C(7)), 6.89 (*d*, $J = 8.0$ Hz, H–C(5)), and 7.60 (*d*, $J = 8.0$ Hz, H–C(4)) in **5**. Therefore, **5** was identified as 2,6-dimethoxy-9-phenyl-1*H*-phenalen-1-one.

The NMR (DEPT) spectra of **6** indicated that its molecular formula was $\text{C}_{21}\text{H}_{16}\text{O}_4$, consistent with the HR-ESI-MS (m/z 333.1133 ($[M + H]^+$)). Comparison of the NMR and MS data of **6** with those of the known compound **8** suggested that both compounds had the same structure except for an additional OH group appearing in **6** [4]. Further ^1H - and ^{13}C -NMR data ($\delta(\text{H})$ 7.28 (*d*, $J = 7.5$ Hz, 2 H) and 6.89 (*d*, $J = 7.5$ Hz, 2 H); $\delta(\text{C})$ 138.7 (*s*, C(1')), 129.6 (*d*, C(2',6')), 113.8 (*d*, C(3',5')), and 156.3 (*s*, C(4'))) placed the OH group at C(4') of the aryl substituent. So, **6** was 7-(4-hydroxyphenyl)-5,6-dimethoxy-1*H*-phenalen-1-one.

Recent studies have shown that allelopathic interactions might contribute more to the success of invasive plant species than previously thought [7][8]. Allelopathic substances could inhibit the growth of other organisms around through eluviation, root exudation, and volatilization. Like in *Musa acuminata*, phenylphenalenes in water hyacinth might be phytoalexins and phytoanticipins [9–11].

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Experimental Part

General. Column chromatography (CC): silica gel (SiO_2 ; 200–300 mesh; *Qingdao Marine Chemical Factory*, Qingdao, P. R. China) and *RP-18* SiO_2 (20–45 μm ; *Fuji Silysia Chemical Ltd.*). Optical rotations: *Horiba-SEAP-300* spectropolarimeter. UV Spectra: *Shimadzu-UV-2401PC* spectrophotometer; λ_{max} ($\log \epsilon$) in nm. IR Spectra: *Tensor-27* spectrometer using KBr pellets; $\tilde{\nu}$ in cm^{-1} . NMR Spectra: *Bruker-AM-400* and *-DRX-500* spectrometers; δ in ppm rel. to Me_4Si as internal standard, J in Hz. MS and HR-MS: *Finnigan-Trace-DSQ* and *API-QuStar-Pulsar-I* spectrometer, resp.; in m/z (rel. %).

Plant Material. The whole plant of *E. crassipes* was collected in March, 2006, in Dianchi Lake at Kunming, Yunnan Province, P. R. China, and identified by *X. L.* A voucher specimen (WMZ20060301) is

deposited with the herbarium of the Kunming Institute of Botany, Chinese Academy of Sciences (KUN), P. R. China.

Extraction and Isolation. The air-dried and powdered plant (10 kg) was extracted with MeOH (4 × 25 l) at r.t., and the extract concentrated *in vacuo* to give a crude residue (500 g), which was suspended in H₂O (2.5 l) and extracted with AcOEt (3 × 2.5 l). The residue of the AcOEt layer (70 g) was subjected to CC (SiO₂, CHCl₃/acetone 10:0, 9:1, 8:2, 7:3, 6:4, 1:1, and 0:10): *Fractions 1–9*. *Fr. 4* (4.4 g) was subjected to CC (*RP-18*, MeOH/H₂O 1:1; then SiO₂, petroleum ether/AcOEt 30:1): **1** (1.7 mg) and **2** (20 mg). *Fr. 2* (15 g) was subjected to CC (SiO₂, petroleum ether/AcOEt 50:1; then *RP-18*, MeOH/H₂O 6:4): **3** (15 mg). *Fr. 6* (2 g) was separated by CC (*RP-18*, MeOH/H₂O 1:1; then SiO₂, petroleum ether/AcOEt 20:1): **7** (1 mg). *Fr. 7* (4 g) was subjected to CC (SiO₂, petroleum ether/AcOEt 20:1): **9** (11 mg). *Fr. 9* (7 g) was subjected to CC (*RP-18*, MeOH/H₂O 1:1; then SiO₂, petroleum ether/AcOEt 5:1): **4** (10 mg), **8** (2 mg), **5** (21 mg), **6** (9 mg), and **10** (10 mg).

2,3-Dihydro-3,9-dihydroxy-5-methoxy-4-phenyl-1H-phenalen-1-one (1): Colorless oil. $[\alpha]_D^{24} = +120$ ($c = 0.08$, CHCl₃). UV (CHCl₃): 240 (4.25). IR: 3497, 3483, 3441, 2921, 1708, 1627. ¹H- (500 MHz, CDCl₃) and ¹³C-NMR (100 MHz, CDCl₃): *Tables 1* and *2*. EI-MS: 320 (100, *M*⁺), 305 (10). HR-ESI-MS: 321.1131 ($[M + H]^+$, C₂₀H₁₇O₃⁺; calc. 321.1126).

2,3-Dihydro-8-methoxy-9-phenyl-1H-phenalene-1,4-diol (2): Colorless oil. $[\alpha]_D^{24} = +53$ ($c = 0.5$, CHCl₃). UV (CHCl₃): 241 (4.62). IR: 3492, 3485, 3441, 2921, 1604. ¹H- (500 MHz, CDCl₃) and ¹³C-NMR (125 MHz, CDCl₃): *Tables 1* and *2*. EI-MS: 306 (100, *M*⁺), 289 (7). HR-ESI-MS: 307.1325 ($[M + H]^+$, C₂₀H₁₉O₃⁺; calc. 307.1334).

2,3-Dihydro-4,8-dimethoxy-9-phenyl-1H-phenalen-1-ol (3): Colorless oil. $[\alpha]_D^{24} = +38.0$ ($c = 0.4$, CHCl₃). UV (CHCl₃): 241 (4.67). IR: 3490, 3481, 2925, 1601. ¹H- (400 MHz, CDCl₃) and ¹³C-NMR (100 MHz, CDCl₃): *Tables 1* and *2*. EI-MS: 320 (100, *M*⁺), 305 (2). HR-ESI-MS: 321.1493 ($[M + H]^+$, C₂₁H₂₁O₃⁺; calc. 321.1490).

2,3-Dihydro-9-(4-hydroxyphenyl)-8-methoxy-1H-phenalene-1,4-diol (4): Colorless oil. $[\alpha]_D^{24} = +18$ ($c = 0.4$, MeOH). UV (MeOH): 236 (4.72), 285 (3.78). IR (KBr): 3489, 3478, 2921, 1602. ¹H- (500 MHz, (D₆)acetone) and ¹³C-NMR (100 MHz, (D₆)acetone): *Tables 1* and *2*. EI-MS: 322 (100, *M*⁺), 228 (40). HR-ESI-MS: 323.1282 ($[M + H]^+$, C₂₀H₁₉O₄⁺; calc. 323.1283).

2,6-Dimethoxy-9-phenyl-1H-phenalen-1-one (5): Orange powder. UV (CHCl₃): 243 (4.36), 262 (4.32), 306 (4.18), 450 (3.95). IR: 3487, 3481, 2922, 1605. ¹H- (400 MHz, CDCl₃) and ¹³C-NMR (125 MHz, CDCl₃): *Tables 1* and *2*. EI-MS: 316 (50, *M*⁺), 315 (100). HR-ESI-MS: 317.1174 ($[M + H]^+$, C₂₁H₁₆O₃⁺; calc. 317.1177).

7-(4-Hydroxyphenyl)-5,6-dimethoxy-1H-phenalen-1-one (6): Orange powder. UV (MeOH): 204 (4.44), 270 (4.14), 456 (3.84). IR: 3482, 2925, 1716, 1609. ¹H- (400 MHz, CDCl₃) and ¹³C-NMR (125 MHz, (D₆)DMSO): *Tables 1* and *2*. EI-MS: 332 (100, *M*⁺), 317 (30), 302 (15). HR-ESI-MS: 333.1133 ($[M + H]^+$, C₂₁H₁₇O₄⁺; calc. 333.1126).

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