## Flavonoids from the Resin of Dracaena cochinchinensis

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Chemical investigation of the red herbal resin of *Dracaena cochinchinensis* resulted in the isolation of three new configurationally isomeric flavonoids: 6,4'-dihydroxy-7-methoxy-8-methylflavane (= 3,4-dihydro-2-(4-hydroxyphenyl)-7-methoxy-8-methyl-2*H*-[1]benzopyran-6-ol; **1**), 5,4'-dihydroxy-7-methoxy-6-methylflavane (= 3,4-dihydro-2-(4-hydroxyphenyl)-7-methoxy-6-methyl-2*H*-[1]benzopyran-5-ol; **2**), and 7,4'-dihydroxy-5-methoxyhomoisoflavane (= 3,4-dihydro-3-[(4-hydroxyphenyl)methyl]-5-methoxy-2*H*-[1]benzopyran-7-ol; **3**). Their structures were identified by means of detailed spectral analysis.

In addition, thirteen known compounds were isolated from *D. cochinchinensis*: 7-hydroxy-4'-methoxyflavane (4), 2,4,6-trimethoxy-4'-hydroxydihydrochalcone (5), 2,4-dimethoxy-4'-hydroxydihydrochalcone (6), 7,8-(methylenedioxy)-4'-hydroxyhomoisoflavane (7), 4',7-dihydroxy-8-methylflavane (8), 2,6-dimethoxy-4,4'-dihydroxydihydrochalcone (9), 2-methoxy-4,4'-dihydroxydihydrochalcone (10), 7-methoxy-6,4'-dihydroxyhomoisoflavane (11), 2-methoxy-4,4'-dihydroxychalcone (12), 4',7-dihydroxyflavane (13), 7,4'-dihydroxyhomoisoflavane (14), 7,4'-dihydroxyhomoisoflavone (15), and 7,4'-dihydroxyflavone (16). Compounds 7, 8, 9, 14, and 15 have been isolated for the first time from this type of herbal source.

**Introduction.** – The red resin of *Dracaena cochinchinensis* S. C. CHEN (Agavaceae), a raw material of Chinese Dragon's Blood, is commonly used in traditional Chinese medicine for the treatment of traumatic and visceral hemorrhages [1]. The original plant of this herb material is native to the tropic region of Southwest China, from which steroidal glycosides and flavonoids have been isolated [2-5]. Chemical studies have revealed that the resin mainly contains phenolic compounds, as well as some steroids and aliphatic acids [6-11]. In the case of the red resin of *D. cochinchinensis*, phenol derivatives are the main constituents.

As a part of our continuing studies on Chinese Dragon's Blood, a total of 16 flavonoids (1-16) have been isolated, including three new ones: 6,4'-dihydroxy-7-methoxy-8-methylflavane (1), 5,4'-dihydroxy-7-methoxy-6-methylflavane (2), and 7,4'-dihydroxy-5-methoxyhomoisoflavane<sup>1</sup>) (3). Here, we describe the isolation and characterization of the new compounds 1-3, which were identified by means of spectroscopic methods.

**Results and Discussion.** – The CHCl<sub>3</sub> extract of the resin of *D. cochinchinensis* was subjected to repeated column chromatography (CC) on *a*) silica gel, *b*) Sephadex LH-20 gel, *c*) CHP-20 gel, and *d*) RP-18 gel, affording, besides 1-3, the known flavonoids 4-16. The latter were identified, based on comparison with literature data, as 7-

<sup>1)</sup> For systematic names, see the Exper. Part.



hydroxy-4'-methoxyflavane (4) [7], 2,4,6-trimethoxy-4'-hydroxydihydrochalcone (5) [12], 2,4-dimethoxy-4'-hydroxydihydrochalcone (6) [13], 7,8-(methylenedioxy)-4'-hydroxyhomoisoflavane (7) [14], 4',7-dihydroxy-8-methylflavane (8) [13], 2,6-dimethoxy-4,4'-dihydroxydihydrochalcone (9) [15], 2-methoxy-4,4'-dihydroxydihydrochalcone (10) [12], 7-methoxy-6,4'-dihydroxyhomoisoflavane (11) [7], 2-methoxy-4,4'-dihydroxychalcone (12) [11], 4',7-dihydroxyflavane (13) [9], 7,4'-dihydroxyhomoisoflavane (14) [13], 7,4'-dihydroxyhomoisoflavone (15) [13], and 7,4'-dihydroxyflavone (16) [14].

Compound **1**, obtained as a colorless amorphous powder, with an  $[\alpha]_{20}^{26}$  value of 0.00 (MeOH), had the molecular formula  $C_{17}H_{18}O_4$ , as determined by EI-MS ( $M^+$  signal at m/z 286) and HR-ESI-MS ( $[M + Na]^+$  signal at m/z 309.1099 ( $C_{17}H_{18}NaO_4^+$ ; calc. 309.1102)). The structure of **1** was established by <sup>1</sup>H-NMR, <sup>13</sup>C-NMR (*Tables 1* and 2, resp.), HMQC, HMBC, <sup>1</sup>H,<sup>1</sup>H-COSY, and NOESY experiments as 6,4'-dihydroxy-7-methoxy-8-methylflavane (= 3,4-dihydro-2-(4-hydroxyphenyl))-7-methoxy-8-methyl-2H-[1]benzopyran-6-ol).

Table 1. <sup>1</sup>*H*-*NMR Data for Compounds* **1**–**3**. At 400 MHz in CD<sub>3</sub>OD;  $\delta$  in ppm, *J* in Hz.

	1	2	3
H-C(2)	4.82 (br. $d, J = 8.4, 1$ H)	4.80 (dd, J = 2.2, 13.2, 1  H)	3.99 (dd, J = 10.4, 1.6, 1 H),
			3.59 (dd, J = 10.4, 8.1, 1 H)
H-C(3)	2.07 (m, 1 H),	1.71 ( <i>m</i> , 1 H),	2.06 ( <i>m</i> , 2 H)
	1.89 (m, 1 H)	1.94 ( <i>m</i> , 1 H)	
H-C(4)	2.85 (m, 1 H),	2.44 ( <i>m</i> , 1 H),	2.56 ( <i>m</i> , 1 H),
	2.61 (m, 1 H)	2.51 ( <i>m</i> , 1 H)	2.09 (m, 1 H)
H-C(5)	6.40 (s, 1 H)	_	_
H-C(6)	_	-	5.95 (d, J = 2.0)
H-C(8)	_	6.02 (s, 1 H)	5.85(d, J = 2.0)
H-C(9)	_	_	2.48(m)
H - C(2', 6')	7.20 (d, J = 8.6)	7.21 (d, J = 8.5)	6.70(d, J = 8.6)
H - C(3', 5')	6.77 (d, J = 8.6)	6.77 (d, J = 8.5)	6.97 (d, J = 8.6)
Me	2.07(s)	1.96 (s)	_
MeO	3.69 (s)	3.70 (s)	3.68(s)

A total of 16 signals were observed in the <sup>13</sup>C-NMR (DEPT) spectrum of **1** (*Table 2*), including five quaternary C-atoms, seven CH, two CH<sub>2</sub>, one MeO, and one Me group. The spectrum showed the presence of twelve aromatic C-atoms ( $\delta_C$  100–165), together with an oxygenated CH ( $\delta_C$  78.9) and two CH<sub>2</sub> ( $\delta_C$  31.7 and 26.6) groups, indicating a flavane skeleton. In addition, Me and MeO signals ( $\delta_C$  9.7 and 61.1, resp.) were observed.

	1	2	3
C(2)	78.9	78.5	70.2
C(3)	31.7	30.7	34.8
C(4)	26.6	20.5	25.6
C(4a)	120.6	103.3	102.6
C(5)	114.6	155.0	159.6
C(6)	146.5	104.9	92.0
C(7)	148.2	157.1	157.1
C(8)	118.7	92.0	96.0
C(8a)	144.5	155.5	156.0
C(9)	-	-	37.8
C(1')	135.1	134.7	134.1
C(2')	128.6	128.2	130.5
C(3')	116.5	116.0	115.7
C(4')	158.3	157.8	156.6
C(5')	116.5	116.0	115.7
C(6')	128.6	128.2	130.5
Me	9.7	8.1	-
MeO	61.1	55.6	55.3

Table 2. <sup>13</sup>C-NMR Data for Compounds 1–3. Assignments are based on HMBC and ROESY experiments. At 100.5 MHz in CD<sub>3</sub>OD;  $\delta$  in ppm.

In the <sup>1</sup>H-NMR spectrum, the *s* at  $\delta_{\rm H}$  6.40, assigned to H–C(5) by means of a HMBC interaction with C(4) at  $\delta_{\rm C}$  26.6 (*Figure*), indicated that the flavane ring *A* had three neighboring substituents. HMBC Correlations of H–C(5) with three oxygenated C-atoms ( $\delta_{\rm C}$  148.2, 146.5, and 144.5) indicated oxygenations in both 6- and 7-positions, so that the Me group ( $\delta_{\rm C}$  9.7) was at C(8). In the ROESY spectrum of **1**, the upfield Me H-atoms ( $\delta_{\rm H}$  2.07) were correlated with the MeO H-atoms ( $\delta_{\rm H}$  3.69), indicating that the MeO group was attached at C(7) (*Figure*). The *AA'BB'* spin system at  $\delta_{\rm H}$  7.20 (*d*, *J* = 8.60 Hz, 2 H) and 6.77 (*d*, *J* = 8.60 Hz, 2 H) indicated that the remaining OH group was in 4'-position (ring *B*).



Figure. Selected HMBC and ROESY correlations observed for flavane 1

Compound **2** showed the same molecular formula  $(C_{17}H_{18}O_4)$  as **1**, as deduced by EI-MS ( $M^+$  at m/z 286) and HR-ESI-MS ( $[M + Na]^+$  signal at m/z 309.1113 ( $C_{17}H_{18}NaO_4^+$ ; calc. 309.1102)), and also gave rise to similar MS fragmentation patterns. From the spectral data, compound **2** – a regioisomer of **1** – was identified as 5,4'-dihydroxy-7-methoxy-6-methylflavane (= 3,4-dihydro-2-(4-hydroxyphenyl)-7-methoxy-6-methyl-2*H*-[1]benzopyran-5-ol).

The <sup>1</sup>H- and <sup>13</sup>C-NMR spectral features (*Tables 1* and 2) of **2** were closely related to those of **1**, showing the occurrence of a flavane skeleton with a 4'-OH substituted ring *B*, and ring *A* containing a Me ( $\delta_C 8.1, \delta_H 1.96$ ) and a MeO group ( $\delta_C 55.6, \delta_H 3.70$ ). However, significant differences were observed for ring-*A* <sup>13</sup>C-NMR

chemical shifts,  $\delta_{\rm C}$  values of 155.0, 157.1, and 155.5 (three oxygenated quaternary C-atoms) indicating that **2** had two oxy groups at C(5) and C(7), and a methine group ( $\delta_{\rm C}$  92.0) at C(8). Thus, the upfield Me group at  $\delta_{\rm C}$  8.1 was deduced to be at C(6). In the HMBC spectrum (data no shown), correlations of H–C(4) ( $\delta_{\rm H}$  2.51) with C(8a), of H–C(8) ( $\delta_{\rm H}$  6.02) with C(8a) and C(7), and of the MeO H-atoms at  $\delta_{\rm H}$  3.70 with C(7) were observed.

Compound **3** was also shown to have the molecular formula  $C_{17}H_{18}O_4$  according to HR-ESI-MS ( $[M + Na]^+$  signal at m/z 309.1101 (calc. 309.1102)). From the spectral data, the structure of **3** was determined to be 7,4'-dihydroxy-5-methoxy-homoiso-flavane (= 3,4-dihydro-3-[(4-hydroxyphenyl)methyl]-5-methoxy-2*H*-[1]benzopyran-7-ol).

The homoisoflavane skeleton of compound **3** was indicated by the <sup>13</sup>C-NMR signals of two CH<sub>2</sub> ( $\delta_C$  25.6, 37.8), one OCH<sub>2</sub> ( $\delta_C$  70.2), one CH ( $\delta_C$  34.8), and twelve aromatic C-atoms. In addition, there was a MeO group ( $\delta_C$  55.3) present in ring *A*. In the <sup>1</sup>H-NMR spectrum, an *AA'BB'* spin system ( $\delta_H$  6.97, 6.70 (2*d*, *J* = 8.6 Hz each, 2 × 2 H) indicated a 4'-OH substituent in ring *B*. In addition, two *meta*-coupling H-atom resonances ( $\delta_H$  5.95 and 5.85 (2*d*, *J* = 2.0 Hz each, 2 × 1 H), together with the typical <sup>13</sup>C-NMR chemical shifts of flavonoids ( $\delta_C$  159.6, 157.1, and 156.0) suggested a 5,7-dioxy-substituted ring *A*. In the HMBC spectrum of **3** (data not shown), the MeO ( $\delta_H$  3.68) and H–C(4) ( $\delta_C$  2.09, 2.56) resonances were both correlated with C(5) ( $\delta_C$  159.6).

## **Experimental Part**

General. Column chromatography (CC): silica gel (200–300 mesh; Qingdao), CHP-20P gel (MCI), Sephadex LH-20 and FUJI (ODS) gel (Mitsubishi Chemical Co.). TLC: precoated plates (Qingdao), eluent: MeOH/CHCl<sub>3</sub> 1:9. M.p.: XRC-1 apparatus. UV Spectra: UV-210A spectrophotometer (company apparatus);  $\lambda_{max}$  in nm (log  $\varepsilon$ ). IR Spectra: Bio-Rad FTS-135 spectrometer; in cm<sup>-1</sup>. NMR Spectra: Bruker AM-400 or DRX-500 spectrometers, in CD<sub>3</sub>OD;  $\delta$  values in ppm (rel. to the residual solvent signal), J in Hz. EI-MS: Autospec-3000 mass spectrometer; in m/z (rel. %). HR-ESI-MS: API Qstar Pulsa spectrometer.

*Plant Material.* The red resin of *Dracaena cochinchinensis* was purchased from *Weihe Pharmaceutical Factory* (Yunnan, China). A sample was deposited in our laboratory. Identification of the extract was supported by an HPLC comparison with an authentic sample.

*Extraction and Isolation.* The red resin (1.0 kg) of *D. cochinchinensis* was ground and successively extracted with CHCl<sub>3</sub>, AcOEt, and MeOH. The CHCl<sub>3</sub> extract (90 g) was subjected to CC (SiO<sub>2</sub>; CHCl<sub>3</sub>, CHCl<sub>3</sub>/MeOH 20:1, 10:1, 10:2, then MeOH): Fractions *Fr.* 1-6 (based on TLC evaluation). *Fr.* 1 (20.0 g) was subjected to repeated CC (SiO<sub>2</sub>; CHCl<sub>3</sub>/MeOH) to yield the pure compounds **4** (2.0 g), **5** (3.0 g), **6** (14.0 g), and **7** (56 mg). *Fr.* 2 (3.0 g) was subjected to CC (1. SiO<sub>2</sub>; CHCl<sub>3</sub>/MeOH; 2. *Sephadex LH-20* gel) to yield the pure compounds **1** (50 mg), **2** (20 mg), **8** (10 mg), and **9** (25 mg). *Fr.* 3 (2.0 g) was also subjected to repeated CC (SiO<sub>2</sub>, *Sephadex LH-20* gel, *ODS* gel), affording the pure compounds **10** (140 mg), **3** (100 mg), **11** (15 mg), and **12** (50 mg). Finally, *Fr.* 4 (4.5 g) was purified by CC (SiO<sub>2</sub>, *CHP-20P* gel, *Sephadex LH-20* gel) to yield the pure compounds **13** (200 mg), **14** (1.0 g), **15** [8] (520 mg), and **16** (90 mg).

3,4-Dihydro-2-(4-hydroxyphenyl)-7-methoxy-8-methyl-2H-[1]benzopyran-6-ol (1). Amorphous solid.  $[\alpha]_{26}^{26} = 0.00 \ (c = 0.2, \text{ MeOH}). \text{ UV (MeOH}): 207 \ (2.6), 284 \ (0.4), 396 \ (0.006). \text{ IR (KBr): } 3433 \ (br., \text{ OH}), 2933 \ (CH), 1374, 1041, 887. ^{1}\text{H- and }^{13}\text{C-NMR}: \text{see Tables 1 and 2, resp. EI-MS: } 286 \ (65, M^+), 269 \ (25), 253 \ (16), 180 \ (73), 167 \ (100), 151 \ (33), 137 \ (55), 120 \ (70), 107 \ (51), 91 \ (25), 83 \ (10), 67 \ (11). \text{ HR-ESI-MS: } 309.1099 \ ([M+\text{Na}]^+, \text{C}_{17}\text{H}_{18}\text{NaO}_4^+; \text{ calc. } 309.1102).$ 

3,4-Dihydro-2-(4-hydroxyphenyl)-7-methoxy-6-methyl-2H-[1]benzopyran-5-ol (2). Colorless crystals (MeOH). M.p. 155–156°. [a]<sup>26</sup> $_{D}$ =0.00 (c=0.2, MeOH). UV (MeOH): 211 (3.07), 275 (0.24). IR (KBr): 3433 (br., OH), 2933 (CH), 1374, 1041, 887. <sup>1</sup>H- and <sup>13</sup>C-NMR: see *Tables 1* and 2, resp. EI-MS: 286 (48,  $M^+$ ), 269 (2), 255 (1), 180 (3), 166 (15), 138 (50), 121 (54), 109 (21), 91 (100), 78 (35), 69 (30). HR-ESI-MS: 309.1113 ([M + Na]<sup>+</sup>, C<sub>17</sub>H<sub>18</sub>NaO<sup>‡</sup>; calc. 309.1102).

3,4-Dihydro-3-[(4-hydroxyphenyl)methyl]-5-methoxy-2H-[1]benzopyran-7-ol (**3**). Amorphous solid. [a]<sub>D</sub><sup>26</sup> = +24.45 (c = 0.2, MeOH). UV (MeOH): 209 (2.56), 224 (1.77), 281 (0.54). IR (KBr): 3587 (br., OH), 3287, 2916, 1600, 1512, 1237, 1151, 1029, 852. <sup>1</sup>H- and <sup>13</sup>C-NMR: see *Tables 1* and 2, resp. EI-MS: 286 (92,  $M^+$ ), 161 (12), 148 (67), 133 (26), 123 (35), 107 (100), 73 (6), 55 (13). HR-ESI-MS: 309.1101 ([M + Na]<sup>+</sup>, C<sub>17</sub>H<sub>18</sub>NaO<sup>+</sup><sub>4</sub>; calc. 309.1102).

## Helvetica Chimica Acta - Vol. 87 (2004)

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