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5,7-DIHYDROXY-3,6,8-TRIMETHOXYFLAVONE FROM THE FLOWERS OF *GNAPHALIUM ELEGANS**

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Key Word Index—*Gnaphalium elegans*; Compositae; 5,7-dihydroxy-3,6,8-trimethoxyflavone; Eu(fod)$_3$ shift reagent.

Abstract—5,7-Dihydroxy-3,6,8-trimethoxyflavone was isolated from the flowers of *Gnaphalium elegans*. The structure elucidation is based mainly on proton resonance studies using Eu(fod)$_3$ shift reagent.

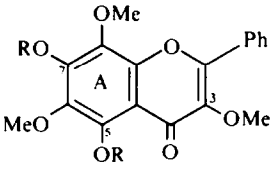
INTRODUCTION

As a part of our current work on the chemistry of some Colombian plants of the genus *Gnaphalium* [1, 2], we have now examined the flowers and leaves of *Gnaphalium elegans* H.B.K. This plant is widely distributed in the cold zone of the Bogotá plain (2500–2700 m altitude) and its surroundings. It is commonly used to remove inflammation of the prostate gland and it is claimed to possess anticancer activity [3].

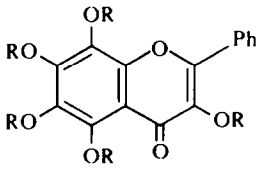
RESULTS AND DISCUSSION

A new dihydroxytrimethoxyflavone **1** was isolated from petrol extracts of the flowers of *Gnaphalium elegans*. The elemental composition (M^+ = 344.0898: $C_{18}H_{16}O_7$) and spectral characteristics indicated that the isolated compound is a pentaoxygenated flavone. Key fragments in the MS at *m/e* 105 (12%) and 77 (8%), as well as ^{1}H NMR signals for 5 protons with chemical shifts and a splitting pattern typical for a phenyl moiety, excluded any substitution at ring B of the flavone skeleton. Since the ^{1}H NMR spectrum exhibits signals for three OMe groups and two OH groups (at δ 12.51 and 6.50, exchangeable upon treatment with D_2O), the isolated flavone must have the basic structure **2**.

Upon methylation with methyl iodide the flavone was converted into its dimethyl ether **3**, a compound having mp 86° and spectral characteristics in agreement with the data described for 3,5,6,7,8-pentamethoxyflavone [4].



1 R = H
3 R = Me
4 R = CD_3



2 3 × R = Me
2 × R = H

From the D_2O-exchangeable signal at δ 12.51 in the ^{1}H NMR spectrum and from the fact that treatment of the flavone with ethereal diazomethane produces only a monomethylated product [^{1}H NMR: δ 12.35 (1 H, *br. s*)], it was concluded that one of the two OH groups must be attached at C-5 and form a hydrogen bridge with the carbonyl group. However, the second OH group cannot be at C-3 because this would give 3,5-dihydroxy-6,7,8-trimethoxyflavone, a substance recently isolated from *Helichrysum* species [4, 5], which has physicochemical data (e.g. mp, MS) different from those of **1**. To deduce the position of the second OH group we prepared, in addition to **3**, the di-trideuteromethyl derivative **4** and recorded ^{1}H NMR spectra of **3** and **4** in the presence of Eu(fod)$_3$ [6]. From corresponding experiments on some polymethoxylated flavones it is known that there is only a very slight influence of the lanthanide shift reagent on a OMe group at C-3 [7], whereas the induced shifts of methoxy groups at ring A decrease in proportion to their distances from the chelating carbonyl carbon atom [7–9].

The δ values observed for the OMe-signals in **3** and **4** are presented in Table 1 together with the corresponding published data for quercetin pentamethyl ether [3,5,7,3′,4′-pentamethoxyflavone (**5**)] [7, 10] and ponkanetin [5,6,7,8,4′-pentamethoxyflavone (**6**)] [8, 9].

Thus these results clearly demonstrate, that the trideuteromethylated flavone **4** has OCD_3 groups at C-5 and C-7 and therefore the flavone from *Gnaphalium elegans* must be 5,7-dihydroxy-3,6,8-trimethoxyflavone (**1**), a new natural product.

EXPERIMENTAL

Plant material of *Gnaphalium elegans* was collected in June 1976 and verified by the Herbario Nacional, Universidad Nacional de Colombia, where a herbarium specimen is deposited under No. 4.81.1820. The flowers (2.5 kg) were extracted with petrol (40–60°) and the conc. extract flocculated with MeOH to remove fats. The filtrate was chromatographed on Si gel using C_6H_6 as eluent. Upon removal of the solvent fractions 20–30 afforded 30 mg of **1**, which was recryst. from MeOH–$CHCl_3$ (9:1) to give orange needles, mp 170°. On TLC with C_6H_6–Me_2CO (9:1) **1** appeared red in UV light, and changed to yellow with I_2 vapour, yellow with

* Part 3 in the series "Colombian Plants of the Genus *Gnaphalium*". For Part 2 see ref. [2].

Table 1. ^{1}H NMR resonances (δ) of methoxyl groups of **3**, **4**, **5** and **6**

Position of methoxyl group	^{1}H NMR resonances*							
	Without Eu(fod)$_3$		With addition of Eu(fod)$_3$					
			1 equiv.	2 equiv.	1 equiv.	2 equiv.		
	3	**4**	**3**		**4**		**5**†	**6**‡
3	3.87§	3.86§	4.8§	5.8§	4.8§	5.7§	4.7§	
5	3.96		20.6	22.0	—	—	18.0	17.18
6	3.94§	3.92§	11.2	16.0	10.3	16.1	—	10.37
7	4.09		6.5	9.4		—	5.2	5.75
8	3.98§	3.97§	4.7§	4.4§	4.5§	4.2§		4.40

* 250 MHz in $CDCl_3$.
† Published data [7, 10].
‡ Published data [8, 9].
§ Assignments are tentative.

$CoCl_2$ and remained unchanged with NH_3. $FeCl_3$, Wilson's reagent and Mg/HCl tests were all positive. UV: λ_{max} nm (log ε) [MeOH]: 278 (4.50), 324 (4.07); [$AlCl_3$]: 293, 349; [$AlCl_3$ + HCl]: 293, 394; [NaOMe]: 284, 379; [NaOAc]: 283, 381; [H_3BO_3]: 278, 326; [NaOAc/H_3BO_3]: 278, 377. IR (KBr) cm^{-1}: 3300, 1620, 1540, 1360, 1145, 1010, 770, 745. ^{1}H NMR (Bruker WH 250, $CDCl_3$, TMS): δ 12.51 ppm (1 H, *s*, exchangeable with D_2O), 8.15 8.08 (2 H, *m*), 7.60–7.50 (3 H, *m*), 6.40 (1 H, *s*, exchangeable with D_2O), 4.20 (3 H, *s*), 3.96 (3 H, *s*), 3.86 (3 H, *s*). MS (SM1B, 70 eV): *m/e* > *m/e* 70(% > 5%) = 344.0898 (73, $M^+ = C_{18}H_{16}O_7$), 343 (7), 330 (19), 329 (100), 311 (7), 301 (9), 286 (5), 197 (6), 169 (5), 105 (12), 77 (8).

Methylation of **1**. Methylation of **1** with methyl iodide gave **3**, which was purified on Si gel TLC (C_6H_6–EtOAc, 6:4; R_f 0.52). UV λ_{max} nm (log ε): 309 (4.23), 267 (4.43). IR (KBr) cm^{-1}: 2945, 2845, 1640, 1590, 1460, 1410, 1360, 1270, 1230, 1210, 1175, 1050, 1000. ^{1}H NMR (250 MHz, $CDCl_3$, TMS): δ 8.15–8.09 ppm (2 H, *m*), 7.54 7.46 (3 H, *m*), 4.09 (3 H, *s*), 3.98 (3 H, *s*), 3.96 (3 H, *s*), 3.94 (3 H, *s*), 3.87 (3 H, *s*). MS (MAT 312–GC/MS, 70 eV): *m/e* > *m/e* 70(% > 10%) = 372 (53, M^+), 371 (31), 358 (23), 357 (100), 353 (11), 341 (12), 314 (10), 313 (14), 197 (*ca* 10), 105 (*ca* 10).

Preparation of di-(trideuteromethyl) ether **4**. **4** was prepared using trideuteromethyl iodide as methylating agent. ^{1}H NMR (250 MHz, $CDCl_3$, TMS): δ 8.15–8.09 ppm (2 H, *m*), 7.54–7.46 (3 H, *m*), 3.97 (3 H, *s*), 3.92 (3 H, *s*), 3.86 (3 H, *s*). M^+ = *m/e* 378.

Methylation of **1** *with diazomethane*. To a soln of 1.2 mg **1** in MeOH, 1–2 drops of ethereal diazomethane were added at room temp. After 10 min the soln was evapd. and the residue purified on TLC Si gel (C_6H_6–EtOAc, 6:4; R_f 0.39). ^{1}H NMR (250 MHz, $CDCl_3$, TMS): δ 12.35 ppm (1 H, *br. s*), 8.21–8.08 (2H, *m*), 7.65 7.48 (3 H, *m*), 4.09 (3 H, *s*), 3.94 (6 H, *s*), 3.85 (3 H, *s*). MS (MAT 312 GC/MS, 70 eV): *m/e* > *m/e* 70(% > 10%) = 358 (67, M^+), 357 (12), 344 (22), 343 (100), 327 (10), 105 (*ca* 22), 77 (*ca* 19).

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