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EUDESMANOLIDES FROM *ARTEMISIA HERBA-ALBA*

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Key Word Index—*Artemisia herba-alba*; Compositae; Anthemideae; sesquiterpene lactones; eudesmanolides; herbalbin.

Abstract—Extraction of aerials parts of *Artemisia herba-alba* and chromatographic separation yielded, in addition to several known eudesmanolides, a new eudesmanolide which has been named herbalbin. The structure was elucidated by high field NMR techniques. Copyright © 1996 Elsevier Science Ltd

INTRODUCTION

As part of an antitumour screening programme on higher plants [1–5], we are carrying out chemical investigations in North Africa on *Artemisia herba-alba Asso. A. herba-alba* (Anthemideae) is a characteristic plant of the steppes of the Middle East, North Africa and Spain [6]. In Morocco it is widespread in the South Desert and the Atlas Mountains. This plant is used in folk medicine as an antihelminthic by the local population. Several papers have been published on the chemical composition of collections of *A. herba-alba* from Egypt [7–9], Israel [10–13] and Spain [14, 15]. The essential oils have been also widely investigated [14, 16, 17].

In the present paper, we report on the isolation of the new eudesmanolide **6** and the known eudesmanolides (**1–5**) [6–15, 18–21].

RESULTS AND DISCUSSION

Extensive chromatography of a dichloromethane extract of *A. herba-alba* gave six crystalline eudesmanolides (**1–6**). Herbalbin, **6**, was assigned the composition $C_{15}H_{22}O_4$ ([M^+] $m/z = 266$). The IR spectrum indicated the presence of hydroxyl groups (3420 cm^{-1}) and a γ-lactone ring (1770 cm^{-1}). The eudesmanolide structure was assigned to herbalbin (**6**) on the basis of the following data. Its ^1H NMR spectrum (Table 1) showed a three proton singlet at δ 0.9 (C-10 methyl) indicating an angular position of the methyl group. The signal of a second methyl group appeared as a singlet at δ 1.5 (C-4 methyl) and was assigned to the methyl group attached to a carbon bearing an oxygen substituent.

A three-proton doublet centred at δ 1.22 ($J = 7$ Hz) was indicative of the presence of a C-11 methyl group. Furthermore, a doublet at δ 60.87 (C-3) and a singlet at δ 59.09 (C-4) observed in the ^{13}C NMR spectrum suggested that compounds **6** contained a C-3/C-4 epoxide ring.

The patterns for the C-1 and C-3 protons were similar to those observed for the respective signals of the equivalent protons in **7** [22] and **8** [23] and are therefore assigned as shown in **6**. The upfield shift of C-14 (δ 18.2) in the ^{13}C NMR spectrum of compound **6** compared to the signal for C-14 in the spectrums of **9** and **10** (δ 11.77 and δ 11.74) [24], confirmed the α-orientation of the hydroxyl group at C-1. For the

Table 1. ^1H NMR data of compounds **4**, **5*** and **6** (400 MHz, $CDCl_3$, TMS as int. standard)

H	4	5	6
1	3.57 *br s*	3.33 *br s*	3.20 *br s*
2α	2.10 *m*	1.92 *dt* (15.3)	
			1.59 *m*
2β	2.13 *m*	2.07 *m*	
3	3.97 *br s*	4.37 *br s*	3.02 *br s*
5		2.97 *d* (11)	2.32 *d* (11)
6	4.57 *br d* (11.5, 1.5)	4.02 *t* (11)	3.95 *t* (11)
7	1.82 *qd* (12, 3.4)	1.70 *qd* (11, 3.4)	1.90 *m*
8α	1.38 *dt* (14, 4)	1.50 *dt* (13, 3.4)	1.59 *m*
8β	1.55 *m*	1.29 *m*	1.90 *m*
9α	2.17 *m*	1.87 *m*	2.12 *m*
9β	2.01 *m*	2.15 *m*	2.12 *m*
11	2.27 *dq* (12, 6.9)	2.31 *dq* (11, 7)	2.30 *m*
13	1.25 *d* (6.9)	1.20 *d* (7)	1.22 *d* (7)
14	1.07 *s*	0.73 *s*	0.90 *s*
15α	2.05 *br s* (1.5)	5.13 *br s*	1.50 *s*
15β		4.98 *br s*	

*250 MHz in $CDCl_3$.

†Figures in parentheses are coupling constants in Hz.

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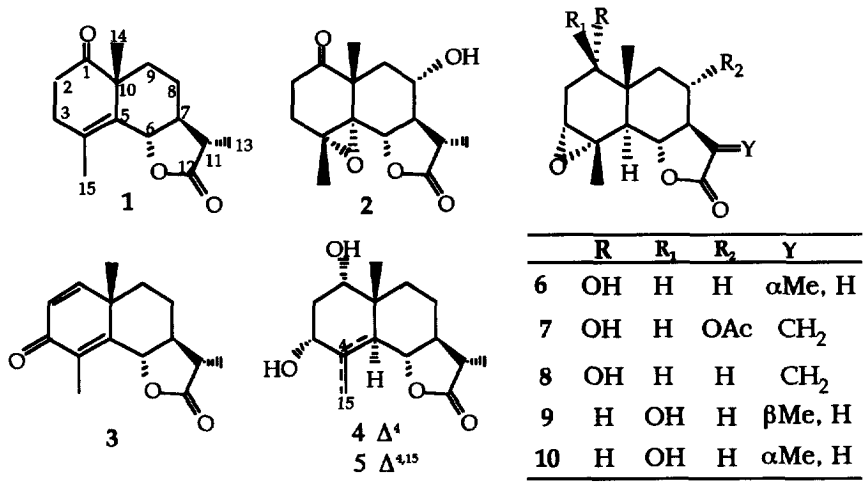

	R	R_1	R_2	Y
6	OH	H	H	αMe, H
7	OH	H	OAc	CH_2
8	OH	H	H	CH_2
9	H	OH	H	βMe, H
10	H	OH	H	αMe, H

methyl group on C-11, we proposed the relative configuration α on the basis of the vicinal coupling constant $J_{11,13} = 7$ Hz [25]. In support of this structural assignment was the position of the signal of C-13 (δ 12.45) in the ^{13}C NMR spectrum (Table 2), a typical value in eudesmanolide with α-methyl groups at C-11.

The lactonic proton at C-6 gave rise to a well-defined triplet at δ 3.95 ($J = 11$ Hz), its pattern being consistent with the *trans*-5,6/*trans*-6,7 disposition shown in **6**.

Comparison of the chemical shifts with those of the literature showed that compound **4** was isoerivanin [19] and **5** was erivanin [20]. The compounds taurin (**1**), 8α-hydroxy-4α,5α-epoxytaurin (**2**) and α-santonin (**3**) were characterized by direct comparison. *A. herba-alba* Asso is included in Sect. Seriphidium of the genus *Artemisia* [26]. The absence of compounds with a germacranolide skeleton in the different fractions of the dichloromethane extract may be attributed to seasonal variation of the sesquiterpene lactones of the plant of the extraction procedure used. The presence of an epoxidic eudesmanolide has no precedent in *A. herba-alba*.

Table 2. ^{13}C NMR data of compounds **4**, **5*** and **6** (100 MHz, $CDCl_3$, TMS as int. standard)

C	4	5	6
1	74.63	74.43	73.37
2	32.20†	32.95†	29.20
3	70.75	75.51	60.87
4	131.10	145.63	59.10
5	126.30	42.39	45.69
6	82.77	79.67	80.80
7	52.36	52.38	53.51
8	24.03	22.92	22.75
9	33.76†	34.03†	33.46
10	42.29	43.44	40.08
11	41.08	41.15	40.70
12	178.72	179.45	179.12
13	12.39	12.48	12.45
14	18.08	17.71	18.20
15	25.31	112.75	22.26

*62,5 MHz in $CDCl_3$.
†Assignments may be interchanged.

EXPERIMENTAL

NMR: 400 (^1H) and 100 (^{13}C) MHz; IR: $CHCl_3$; EIMS: 70 eV; CC: silica gel Merck (0.062–0.200 mm); silica gel (Merck plates).

Plant material. Aerial parts of *A. herba-alba* were collected at several geographical locations in the Atlas Mountains, Morocco, in March 1993. A voucher is deposited in the Herbarium of the 'Institut Scientifique Rabat'.

Extraction and chromatography. Air-dried plant material (1 kg) was crushed and soaked overnight in petrol. The petrol extract was discarded. The residue was allowed to stand for several days covered with $CHCl_3$ and then filtered. After removal of the solvent, the tar (80 g) was extracted (×3) with hexane–EtOH–H_2O. The combined aq. phases were extracted with CH_2Cl_2. The CH_2Cl_2 extract, upon evapn, yielded 25 g of a brown residue. One portion of this residue (6 g) was pre frd by coarse CC on silica gel (A: hexane–EtOAc (9:1–B: hexane–EtOAc (7:3)–C: hexane–EtOAc (4:6) D: hexane–EtOAc (3:7)).

Fr. A (0.16 g) was frd by CC on silica gel (CCl_4–CH_2Cl_2, 2:3) to give taurin (**1**) (0.02 g).

Fr. B (0.12 g) was submitted to CC on silica gel (CCl_4–CH_2Cl_2, 1:1 to CH_2Cl_2). The intermediate fractions were further purified by prep. TLC (hexane–CH_2Cl_2, 3:7 × 4). This yielded α-santonin (**3**) (0.012 g).

Fr. C (0.24 g) was separated on silica gel with EtOAC (5–8%) in CCl_4–CH_2Cl_2, 1:2) and yielded three intermediate frs. The first lactone fr (0.08 g) was purified by prep. TLC (hexane–EtOAc, 2:3, ×3) to give erivanin (**5**) (0.036 g). The second lactone fr. (0.074 g) was crystallized from EtOH to give 0.03 g **6**. Crystallization of the third lactone fr. (0.066 g) from EtOH afforded 0.025 g of isoerivanin (**4**).

Fr. D (0.08 g) was sepd by CC on silica gel (CCl_4–CH_2Cl_2–Et_2O, 3:3:4) and yielded 0.015 g lactone, **2**.

1α-Hydroxy-3α,4α-epoxyeudesm-5α,6β,7α,11βH-12,6-olide (*herbalbin*) (**6**). Crystals, mp 195–197° (EtOH); IR ν_{max} (cm^{-1}): 3420 (OH), 1770 (lactone C=O); EIMS *m/z* (rel. int.): 266 $[M]^{+\cdot}$ (2.6), 251 $[M-Me]^+$ (10.8), 248 $[M-H_2O]^+$ (4.8), 193 (15.8), 149 (16.5), 121 (21.3), 107 (14.3), 93 (31.8), 79 (19.8), 55 (48.3), 43 (100). NMR data: Tables 1 and 2.

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