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FURTHER GLAUCOLIDES AND OTHER SESQUITERPENE LACTONES FROM *BROCCHIA CINEREA*

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Key Word Index—*Brocchia cinerea*; Compositae; Anthemideae; sesquiterpene lactones; glaucolides; eudesmanolides; germacranolides; guaianolides.

Abstract—The reinvestigation of the aerial parts of *Brocchia cinerea* (= *Cotula cinerea*) afforded in addition to several known sesquiterpene lactones two new eudesmanolides, five guaianolides and seven glaucolides, three of them with an eudesmane skeleton and four with a germacrane skeleton. The structures were elucidated by high field NMR techniques.

INTRODUCTION

Brocchia cinerea (Del.) Vis. is a small annual herb widely distributed in Northern Africa. It represents a monotypic genus originally placed in the genus *Cotula*. However, as it does not fit in with the generic concept of *Cotula* it has been separated [1]. So far spiroketal enolether polyines [1, 2] and coumarins linked with sesquiterpenes [1] as well as several sesquiterpene lactones [2] and some flavones [3] have been reported from this species. As two of the lactones were glaucolides, we have repeated the analysis with somewhat larger amounts of plant material. The results are discussed in this paper.

RESULTS AND DISCUSSION

The extract of the aerial parts afforded in addition to the spiroketal enolethers **9a/b** and the lactones **1a**, **3d**, **5a**, **6d**, **6f** and **6g** reported previously [2], the germacranolides haageanolide [4] and its 1β,10α-epoxides [5], **1b** [6], **3a** [7], **3b** [8], **3c** [7] and **3e** [9], the eudesmanolides **2a** [10], **2b**, **4a** [11] and **4b**, the guaianolides **5b**–**5f** and the glaucolides **6a**–**6d** and **7a**–**7c**. Furthermore camphor and the seco-thujone **8** were isolated.

The ^{1}H NMR spectrum of **2b** (Table 1) showed that the acetate of reynosin [10] was present. Accordingly, the H-1 signal was shifted downfield.

The ^{1}H NMR spectral data of **4b** (Table 1) were in part close to those of **4a** [11]. However, the H-15 olefinic signals were replaced by a methyl singlet at δ1.31 and some signals were shifted. Furthermore, the presence of an angelate followed from the typical signals. Spin decoupling allowed the assignment of all signals which led to the proposed structure. The configuration at C-4 was deduced by comparing chemical shifts with those of similar compounds.

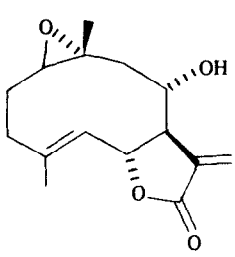
The ^{1}H NMR spectrum of **5b** (Table 2) showed that this guaianolide was the desacetyl derivative of **5a** isolated previously from this plant [2]. This was established by acetylation of **5b** which afforded the acetate **5a**. The ^{1}H NMR spectra of **5c** and **5d** (Table 2) differed only in the chemical shifts of a few signals. In the spectrum of **5d**, a broadened singlet at δ7.59 indicated the presence of hydroperoxide. Triphenylphosphine reduction gave **5c** where all signals could be assigned by spin decoupling. The resulting sequences indicated that **5d** was probably formed by an ene-reaction of **5a** with oxygen while **5c** was formed by reduction of the hydroperoxide. The configuration of the latter was determined by NOE experiments. Clear effects were observed between H-5, H-8 and H-6.

Table 1. ^{1}H NMR spectral data of compounds **2b** and **4b** (400 MHz, $CDCl_3$, δ-values)

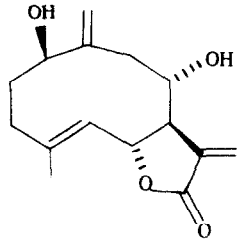
H	**2b** *	**4b** †
1	4.80 *dd*	3.49 *br dd*
2α	1.89 *dddd*	1.76 *dddd*
2β	1.63 *dddd*	1.67 *dddd*
3α	2.19 *br ddd*	1.60 *br ddd*
3β	2.36 *ddd*	1.79 *ddd*
5	2.30 *br d*	1.92 *d*
6	4.01 *dd*	5.79 *dd*
7	2.54 *ddddd*	2.89 *dddd*
8	2.05 *dddd* (α) 1.59 *dddd* (β)	4.05 *ddd*
9α	1.39 *br ddd*	1.50 *br dd*
9β	1.84 *ddd*	2.56 *dd*
13	6.10 *d*	6.09 *d*
13′	5.42 *d*	5.33 *d*
14	0.91 *s*	1.07 *s*
15	5.02 *br s* 4.90 *br s*	1.31 *s*

* OAc: 2.07 *s*; † OAng: 6.21 *qq*, 2.02 *dq*, 1.93 *dq*.

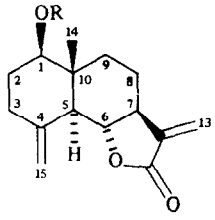
J[Hz]: 1,2α = 2α,3α = 2β,3β = 5; 1,2β = 11; 2α,2β = 2β,3α = 3α,3β = 13; 2α,3β = 2; 5,6 = 6,7 = 7,8β = 10.5; 7,8α = 3; 7,13 = 3.5; 7,13′ = 3; 8α,8β = 8β,9α = 9α,9β = 13; 8α,9α = 4; 8α,9β = 2.5; 8β,9β = 3.5.



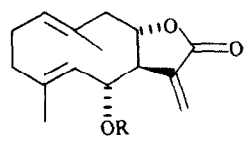
1a



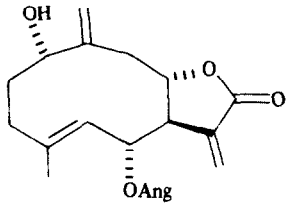
1b



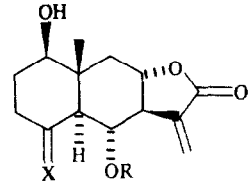
	2a	2b
R	H	Ac



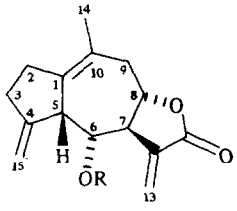
	3a	3b	3c	3d	3e
R	H	H	H	Ang	Ang
		1α,4β epoxide	4β,5α epoxide		4α,5β epoxide

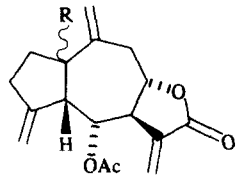


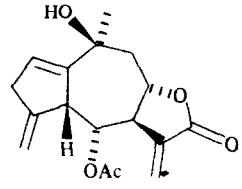
3f



	4a	4b
R	H	Ang
X	CH_2	βMe, αOH







	5a	5b		5c	5d	5e
R	Ac	H	R	βOH	βOOH	αOH

5f

Accordingly, the stereochemistry at C-5–C-8 was settled. The ^{1}H NMR spectrum of **5e** was also close to that of **5c** and showed nearly the same couplings. Also the NOE's on irradiation of H-5 were the same. Thus **5c** and **5e** could only differ in the configuration at C-1. Comparison of the chemical shifts of H-7 indicated that in the case of **5e** the latter was deshielded by the 1α-hydroxy group by 0.57 ppm while in the spectrum of **5c** a downfield shift of H-8 and H-9β was observed. Furthermore, in the case of **5a** a *W*-coupling between H-5 and H-2β also required the proposed stereochemistry. Comparison of the data of **5c** with those of **5a** and **5b** strongly supported identical stereochemistry at C-5–C-8. Accordingly, the proposed stereochemistry at C-5 and C-6 for **5a** (in lit. [2]. compound **5**) had to be revised.

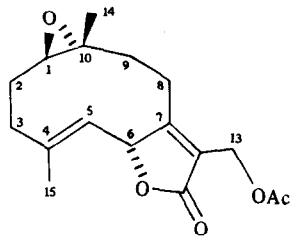
The ^{1}H NMR spectrum of **5f** (Table 2) showed some similarities to that of **5c**. Spin decoupling again allowed the assignment of all signls. The downfield shift of H-5 indicated a position between two double bonds. This was supported by the observed allylic couplings with H-2 and H-15 as well as by the chemical shifts of H-3. The configuration at C-5–C-8 followed from the observed NOE's between H-8, H-5 and H-6. The stereochemistry at C-10 was deduced from the NOE between H-14 and H-9α. The

Table 2. ^{1}H NMR spectral data of compounds **5b-5f** (400 MHz, $CDCl_3$, δ-values)

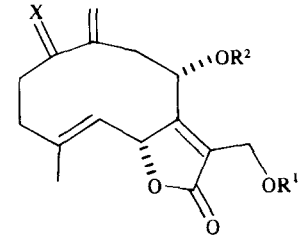
H	5b	5c	5d*	5e	5f†
5	3.34 *br s*	2.75 *br s*	2.61 *br s*	2.67 *br s*	3.78 *br d*
6	4.21 *dd*	5.46 *dd*	5.49 *dd*	5.86 *dd*	5.62 *dd*
7	2.94 *dddd*	3.05 *dddd*	3.06 *dddd*	3.63 *dddd*	2.90 *dddd*
8	4.62 *ddd*	4.60 *ddd*	4.54 *ddd*	4.18 *ddd*	4.56 *ddd*
9	3.08 *br dd*	3.56 *dddd*	3.45 *dddd*	3.05 *br dd*	2.59 *dd*
9′	2.44 *m*	2.50 *dd*	2.53 *dd*	3.01 *dd*	1.76 *dd*
13	6.25 *d*	6.26 *d*	6.26 *d*	6.23 *d*	6.22 *d*
13′	5.96 *d*	5.93 *d*	5.99 *d*	5.75 *d*	5.66 *d*
14	1.72 *br s*	5.34 *br s* 5.12 *br s*	5.43 *br s* 5.36 *br s*	5.17 *br s* 5.08 *br s*	1.59 *s*
15	5.24 *ddd*	5.11 *ddd*	5.09 *ddd*	5.15 *ddd*	5.02 *ddd*
15′	4.93 *ddd*	4.95 *ddd*	4.90 *ddd*	4.73 *ddd*	4.94 *ddd*
OAc	—	2.00 s	2.01 s	2.08 s	2.04 s

* OOH: 7.51 *br s*; † H-2 6.02 *br s*, H-3 3.13 *br d*, 2.87 *br d*.

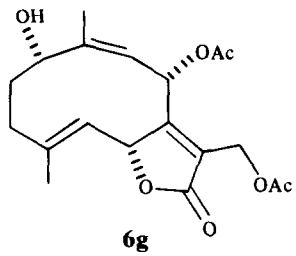
J[Hz]: 3,15 = 3,15′ = 5,15 = 15,15′ ≈ 1.5; 7,13 = 3.5; 7,13′ = 3; compound **5b**: 5,6 = 4.5; 6,7 = 7.5; 7,8 = 8,9′ = 10; 8,9 = 6.5; 9,9′ = 16; compounds **5c** and **5d**: 5,6 = 4.5; 6,7 = 6; 7,8 = 11; 8,9 = 8,9′ = 8; 9,9′ = 14; 9,15 = 9,15′ = 1.5; compound **5e**: 5,6 = 5; 6,7 = 8; 7,8 = 8,9 = 10; 8,9′ = 4; 9,9′ = 13; compound **5f**: 3,3′ = 21; 5,6 = 6.5; 6,7 = 8.5; 7,8 = 9.5; 8,9 = 2; 8,9′ = 12; 9,9′ = 13.



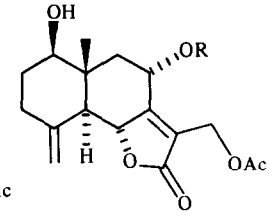
6a



	6b	6c	6d	6e	6f
R^1	Ac	Ac	H	Ac	Ac
R^2	Ac	Ac	H	H	Ac
X	αOH,H	βOH,H	O	O	O

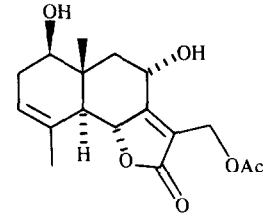


6g

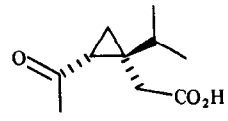


7a **7b**

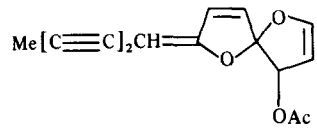
R H Ac



7c



8



9a *Z*

9b *E*

lactones **5e** and **5f** like those of **5d** and **5c** are probably derived from **5a** by ene-reactions with oxygen followed by reduction of the corresponding hydroperoxides.

The ^{1}H NMR spectrum of **6a** (Table 3) showed the typical signals of a glaucolide (δ5.44 *br d*, H-6, 4.89 and 4.84 *br d*, H-13). Furthermore, the spectrum was very close to that of the 1β,10α-epoxide of haageanolide [5]. Accordingly, the configurations at C-1, C-9 and C-10 were assumed to be identical in these two lactones.

The ^{1}H NMR spectra of **6b** and **6c** (Table 3) were close to the corresponding glaucolides from *Artemisia afra* [12] which only differ in the position of the acetoxy group. Spin decoupling clearly showed that in the case of **6b** and **6c** an 8α-acetoxy was present while the configuration at C-1 was deduced from the observed couplings of H-1 which were identical with those of the corresponding *Artemisia* lactones where the configuration was determined by NOE experiments. The ^{1}H NMR spectrum of **6d** (Table 3) showed that this lactone was a bis-desacetyl derivative of **6b** where the 1-hydroxy group was oxidized. Accordingly, the H-14 signals were shifted downfield.

The ^{1}H NMR spectra of **7a**–**7c** (Table 3) showed that again glaucolides were present as followed from the typical signals of H-6 and H-13. Spin decoupling showed that eudesmanolides were present with oxygen functions at C-1 and C-8. The spectrum of **7a** was close to that of dentatin A [6] but of course the H-6 and H-8 signals were shifted downfield. In agreement with the presence of *trans*-decalin systems, a *W*-coupling between H-14 and H-9 was observed in the spectra of **7a**–**7c**. The spectrum of **7b** showed that this lactone was the 8-O-acetate of **7a** while that of **7c** indicated an isomeric situation of the 4(15)-double bond. Accordingly, the spectrum of the latter was in part close to that of balchanin [13].

The ^{1}H and ^{13}C NMR data of **8** (see Experimental) indicated the presence of a seco-thujene. Accordingly, the data were in part close to those of α-thujene. However, a methyl singlet at δ2.33 and a pair of doublets at δ2.50 and 2.66 indicated a changed situation which led to the structure **8**. In the mass spectrum the highest peak corresponds to $C_{10}H_{14}O_2$ obviously formed by loss of water and *m/z* 124 was the result of a McLafferty fragmentation $[M - HOAc]^+$. Also $[M - COMe]^+$ (*m/z* 141) was present. These results agreed with the structure.

The chemistry of this species supports the separation from the genus *Cotula*. Most compounds show relationships to *Artemisia* where similar acetylenes, coumarins and lactones have been isolated. The presence of glaucolides is of interest as this type of lactone usually only occurs in the tribe Vernonieae. However, a few of these compounds were isolated from *Artemisia* species [12, 14, 15]. Most likely, all lactones are derived either from the 12,6α- or 12,8α-olides. Typical is the high degree of oxygenation. The formation of eudesmanolides and guaianolides is probably induced by epoxidation of the 1(10)-double bond. This kind of biogenetic pathway has never been observed in representatives of *Cotula* where C_{17}-acetylenes are widespread and only a few, very simple sesquiterpene lactones have been isolated [16].

EXPERIMENTAL

The air-dried aerial parts (2600 g, collected in the Eastern desert, 50 km South of Ismaelia, Egypt, in April 1987, voucher deposited in the Faculty of Pharmacy, Zagazig University, Egypt) were extracted and worked-up as reported previously [17]. The CC fractions were combined to give four fractions (1:Et_2O–petrol, 1:1; 2:Et_2O; 3:Et_2O–MeOH, 9:1 and 4:Et_2O–MeOH, 3:1). Fraction 1 gave 8 g camphor and one-fifth of fraction 2 afforded by TLC (Et_2O–petrol, 1:1) 10 mg **3d**, 7 mg **9a** and 10 mg **5a**. HPLC of one-third of fraction 3 (MeOH–H_2O, 7:3, always RP *ca* 100 bar) gave six fractions (3/1–3/6). Repeated HPLC (MeOH–H_2O, 3:2) of 3/1 gave 6 mg **8** (R_t 6.5 min), 5 mg **5c** (R_t 7.2 min) and 4 mg **5d**, purified by TLC (Et_2O–petrol, 7:3, R_f 0.6). TLC of 3/2 (Et_2O–petrol, 1:1) gave 15 mg **3a** and 3/3 contained 30 mg **5b** (R_t 6.5 min). TLC of 3/4 (Et_2O–petrol, 3:2)

Table 3. ^{1}H NMR spectral data of compounds **6a**–**6d** and **7a**–**7c** (400 MHz, $CDCl_3$, δ-values)

H	6a	6b	6c	6d	7a	7b	7c *
1	2.72 *dd*	3.93 *br dd*	3.90 *br d*	—	3.46 *dd*	3.46 *dd*	3.61 *dd*
5	4.68 *br d*	4.86 *br d*	4.64 *br d*	4.64 *br d*	1.76 *br d*	1.84 *br d*	1.95 *br d*
6	5.44 *br d*	5.61 *br d*	5.66 *br d*	5.46 *br d*	4.96 *br d*	4.98 *br d*	4.81 *br d*
8	3.06 *br d* 2.47 *br dd*	5.38 *br dd*	5.17 *br dd*	4.26 *br dd*	4.81 *br dd*	5.59 *dd*	4.87 *br dd*
9	3.23 *br d*	2.93 *br d*	2.90 *br d*	3.26 *br dd*	2.62 *dd*	2.61 *dd*	2.59 *dd*
9′		2.43 *br dd*	2.80 *br dd*	2.73 *dd*	1.39 *br dd*	1.44 *br dd*	1.33 *br dd*
13	4.89 *br d*	4.99 *br d*	5.00 *br d*	4.68 *dd*	5.11 *br d*	5.05 *br d*	5.15 *br d*
13′	4.84 *br d*	4.84 *br d*	4.84 *br d*	4.62 *dd*	5.07 *br d*	4.84 *br d*	5.10 *br d*
14	1.28 *s*	5.38 *br s* 5.25 *br s*	5.13 *br s* 5.00 *br s*	5.76 *d* 5.73 *d*	0.89 *s*	0.96 *s*	0.97 *s*
15	1.88 *d*	1.78 *br s*	1.87 *br s*	1.75 *d*	5.05 *br s* 4.95 *br s*	5.09 *br s* 4.99 *br s*	1.89 *br s*
OAc	2.09 *s*	2.11 *s* 2.09 *s*	2.11 *s* 2.08 *s*	—	2.10 *s*	2.14 *s* 2.06 *s*	2.00 *s*

* H-2 2.39 *br dd* and 1.99 *br dd*; H-3 5.42 *br s*.

J [Hz]: 13,13′ = 13; compound **6a**: 1,2 = 2; 1,2′ = 11; 5,6 = 10.5; 8,8′ = 14; 8′,9 = 9.5; compound **6b**: 1,2 = 3; 1,2′ = 9; 5,6 = 10; 8,9 = 2; 8,9′ = 7.5; 9,9′ = 16; compound **6c**: 1,2 = 5,6 = 8,9′ = 10; 8,9 = 3; 9,9′ = 13.5; compound **6d**: 5,15 = 1.5; 8,9 = 3; 8,9′ = 11; 9,9′ = 13; 9,14 = 1.5;9,14′ = 13, OH = 13′, OH = 2; compounds **7a** and **7b**: 1,2 = 4.5; 1,2′ = 12; 5,6 = 10; 8,9 = 6.5; 8,9 = 6.5; 8,9′ = 9,9′ = 12.5; compound **7c**: 1,2 = 6.5; 1,2′ = 10; 2,2′ = 16; 5,6 = 11.5; 8,9 = 6; 8,9′ = 9,9′ = 12.

afforded 4 mg **3e** and 7 mg **3f**. TLC of 3/5 gave 10 mg **2b** (R_f 0.5) and 3/6 afforded 20 mg **5a** (R_t 11.0 min). CC fraction 4 was separated first by flash chromatography (silica gel, ϕ 30–60 μ, using Et_2O–petrol and Et_2O–MeOH mixtures). The fractions were combined into four (4/1–4/4). HPLC of 4/1 (MeOH–H_2O, 3:2) gave three fractions (4/1/1–4/1/3). TLC of 4/1/1 (Et_2O–petrol, 1:1) afforded 3 mg **5c** (R_f 0.35). 5 mg **6f** and 4 mg haageanolide. TLC of 4/1/2 (Et_2O–petrol, 3:2) gave 3 mg **5d** (R_f 0.6), 5 mg **3c**, 2 mg **5f** (R_f 0.4) and 6 mg **2a**. TLC of 4/1/3 (Et_2O–petrol, 7:3) gave 1 mg **5b** (R_f 0.7), 3 mg **3a** and 2 mg **9b**. HPLC of 4/2 (MeOH–H_2O, 7:3) gave two fractions. The first one was separated by TLC (CH_2Cl_2–Et_2O, 4:1) affording 6 mg **1a** and 3 mg **6e**. TLC of the second fraction (same solvent) gave 5 mg **6f** and 4 mg haageanolide. TLC of 4/3 (Et_2O) gave two bands (4/3/1 and 4/3/2). HPLC of 4/3/1 gave two fractions. The first one gave by TLC ($CHCl_3$–MeOH, 9:1) 4 mg **6e** and 6 mg **1b** while the second one ($CHCl_3$–MeOH, 19:1) afforded 1 mg **6g**, 3 mg **7b** (R_f 0.75) and 5 mg **4a**. HPLC of 4/3/2 (MeOH–H_2O, 7:3) gave three fractions (4/3/2/1–4/3/2/3). TLC of 4/3/2/1 (CH_2Cl_2–MeOH, 19:1) gave 4 mg **6d** (R_f 0.7), of 4/3/2/2 (same solvent) 5 mg 1β,10α-epoxyhaageanolide and 3 mg **1b**. TLC of 4/3/2/3 (CH_2Cl_2–Et_2O–petrol, 1:1:1) gave 4 mg **6c** (R_f 0.57 and 3 mg **6b** (R_f 0.45). TLC of 4/4 (Et_2O) gave three bands (4/4/1–4/4/3). TLC of 4/4/1 (CH_2Cl_2–MeOH, 30:1) afforded 2 mg **5e** (R_f 0.7) and 3 mg **3b**. HPLC 4/4/2 (MeOH–H_2O, 13:7) gave three fractions (4/4/2/1–3). 4/4/2/1 gave by TLC (Et_2O) 1 mg **6a** (R_f 0.5). 4/4/2/2 contained 3 mg **7c** (R_t 6.0 min) and TLC of 4/4/2/3 (Et_2O) afforded 1 mg **4b** (R_f 0.6). HPLC of 4/4/3 (MeOH–H_2O, 13:7) afforded 3 mg **7a** (R_t 7.5 min). Due to the complicated separation considerable losses of material must have occurred. Known compounds were identified by comparing their 400 MHz ^{1}H NMR spectra with those of authentic material or by rigourous NMR techniques.

Reynosin acetate (**2b**). Colourless oil; IR $\nu_{max}^{CCl_4}$ cm^{-1}: 1770 (γ-lactone), 1730 (OAc); MS *m/z* (rel. int.): 290 $[M]^+$ (3), 230.131 $[M-HOAc]^+$ (34) (calc. for $C_{15}H_{18}O_2$: 230.131), 215 (10), 173 (48), 119 (100).

6α-*Angeloyloxy*-1β,4α-*dihydroxyeudesm*-11(13)-*en*-8α,12-*olide* (**4b**). Colourless oil; IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3630 (OH), 1760 (γ-lactone); 1720 (C=CCO_2R); MS *m/z* (rel. int.): 346.178 $[M-H_2O]^+$ (3) (calc. for $C_{20}H_{26}O_5$: 346.178), 247 $[346-OAng]^+$ (6), 246 $[346-AngOH]^+$ (3), 229 $[247-H_2O]^+$ (6), 83 $[RCO]^+$ (100), 55 $[83-CO]^+$ (74).

6α-*Hydroxyguaia*-1(10),4(15),11(13)-*trien*-8α,12-*olide* (**5b**). Colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1760 (γ-lactone); MS *m/z* (rel. int.): 246.126 $[M]^+$ (12) (calc. for $C_{15}H_{18}O_3$: 246.126), 228 $[M-H_2O]^+$ (5), 121 (100), 99 (52). Acetylation of **5b** (Ac_2O, 1 hr, 70°) afforded **5a**, identical with the natural compound (^{1}H NMR, TLC).

6α-*Acetoxy*-1β-*hydroxyguaia*-4 (15),10(14),11(13)-*trien*-8α,12-*olide* (**5c**). Colourless gum; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1770 (γ-lactone), 1730 (OAc), MS *m/z* (rel. int.): 304 $[M]^+$ (2), 244.110 $[M-HOAc]^+$ (48) (calc. for $C_{15}H_{16}O_3$: 244.110), 226 $[244-H_2O]^+$ (43), 216 $[244-CO]^+$ (38), 201 $[216-Me]^+$ (30), 120 (100), 91 (74).

6α-*Acetoxy*-1β-*hydroxyperoxyguaia*-4 (15),10(14),11(13)-*trien*-8α,12-*olide* (**5d**). Colourless gum; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3630 (OH), 1770 (γ-lactone), 1740 (OAc); MS *m/z* (rel. int.): 287.128 $[M-OOH]^+$ (52) (calc. for $C_{17}H_{19}O_4$: 287.128), 261 $[M-OAc]^+$ (7), 245 $[287-ketene]^+$ (54), 227 $[287-HOAc]^+$ (84), 91 (74), 55 (100). Addition of triphenylphosphine in $CHCl_3$ afforded **5c**, identical with the natural compound (^{1}H NMR, TLC).

6α-*Acetoxy*-1α-*hydroxyguaia*-4(15), 10 (14),11(13)-*trien*-8α,12-*olide* (**5e**). Colourless gum; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1770 (γ-lactone), 1740 (OAc); MS *m/z* (rel. int.): 304.131 $[M]^+$ (5) (calc. for $C_{17}H_{20}O_5$: 304.131), 244 $[M-HOAc]^+$ (14), 226 $[244-H_2O]^+$ (12), 216 $[244-CO]^+$ (16), 61 (100).

6α-*Acetoxy*-10β-*hydroxyguaia*-1, 4 (15), 11 (13)-*trien*-8α,12-*olide* (**5f**). Colourless gum; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1770 (γ-lactone), 1745 (OAc); MS *m/z* (rel. int.): 304.131 $[M]^+$ (4) (calc. for $C_{17}H_{20}O_5$: 304.131), 244 $[M-HOAc]^+$ (56), 226 (12), 123 (82), 61 (100).

13-*Acetoxy*-9β-*hydroxy*-1β,10α-*epoxygermacra*-4,7(11)-*dien*-6α,12-*olide* (**6a**). Colourless gum; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH) 1770 (γ-lactone), 1740 (OAc); MS *m/z* (rel. int.): 322.141 $[M]^+$ (1.4) (calc. for $C_{17}H_{22}O_6$: 322.142), 262 $[M-HOAc]^+$ (4), 244 $[262-H_2O]^+$ (3), 216 $[244-CO]^+$ (8), 81 (88), 61 (100).

8α,13-*Diacetoxy*-1α-*hydroxygermacra*-4,7(11),10(14)-*trien*-6α,12-*olide* (**6b**). Colourless gum; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3500 (OH), 1760 (γ-lactone, OAc); MS *m/z* (rel. int.): 304.131 $[M-HOAc]^+$ (2) (calc. for $C_{17}H_{20}O_5$: 304.131), 286 $[304-H_2O]^+$ (4), 244 $[304-HOAc]^+$ (70), 226 $[244-H_2O]^+$ (64), 211 $[226-Me]^+$ (42), 55 (100).

8α,13-*Diacetoxy*-1β-*hydroxygermacra*-4,7(11),10(14)-*trien*-6α,12-*olide* (**6c**). Colourless gum; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1760 (γ-lactone, OAc); MS *m/z* (rel. int.): 304.131 $[M-HOAc]^+$ (2) (calc. for $C_{17}H_{20}O_5$: 304.121), 286 $[304-H_2O]^+$ (3), 244[304 $-HOAc]^+$ (100), 226 $[244-H_2O]^+$ (68), 211 $[226-Me]^+$ (46).

8α,13-*Dihydroxy*-1-*oxo*-*germacra*-4, 7 (11), 10 (14)-*trien*-6α,12-*olide* (**6d**). Colourless gum; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1770 (γ-lactone), 1680 (C=CC=O); MS *m/z* (rel. int.): 260.105 $[M-H_2O]^+$ (1.3) (calc. for $C_{15}H_{16}O_4$: 260.105), 245 $[260-Me]^+$ (1), 216 (4.5), 196 (12), 95 (44), 44 (100).

13-*Acetoxy*-1β,8α-*dihydroxyeudesma*-4 (15), 7 (13)-*dien*-6α,12-*olide* (**7a**). Colourless gum; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1760 (γ-lactone, OAc); MS *m/z* (rel. int.): 304.131 $[M-H_2O]^+$ (10) (calc. for $C_{17}H_{20}O_5$: 304.131), 262 $[M-HOAc]^+$ (100), 244 $[262-H_2O]^+$ (92), 226 $[244-H_2O]^+$ (92), 211 $[226-Me]^+$ (36).

8α,13-*Diacetoxy*-1β-*hydroxyeudesma*-4 (15), 7 (13)-*dien*-6α,12-*olide* (**7b**). Colourless gum; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1760 (γ-lactone, OAc); MS *m/z* (rel. int.): 262.120 $[M-HOAc, ketene]^+$ (58) (calc. for $C_{15}H_{18}O_4$: 262.120), 244 $[262-H_2O]^+$ (82), 226 $[244-H_2O]^+$ (100), 211 $[226-Me]^+$ (53), 124 (81), 105 (78).

13-*Acetoxy*-1β,8α-*dihydroxyeudesma*-3,7(13)-*dien*-6α,12-*olide* (**7c**). Colourless gum; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3630 (OH), 1770 (γ-lactone), 1740 (OAc); MS *m/z* (rel. int.): 304.131 $[M-H_2O]^+$ (0.6) (calc. for $C_{17}H_{20}O_5$: 304.131), 262 $[M-HOAc]^+$ (12), 244 $[262-H_2O]^+$ (28), 216 $[244-CO]^+$ (60), 109 (76), 60 (51), 55 (100).

Seco-thujene (**8**). Colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3500–2500, 1715 (CO_2H), 1715 (C=O); MS *m/z* (rel. int.): 166.100 $[M-H_2O]^+$ (32) (calc. for $C_{10}H_{14}O_2$: 166.099), 151 $[166-Me]^+$ (17), 141 $[M-COMe]^+$ (26), 124 [McLafferty]$^+$ (100), 109 $[124-Me]^+$ (96), 95 (84), 71 (92), 69 (93): ^{1}H NMR ($CDCl_3$): δ2.03 (*dd*, H-1), 2.50 and 2.66 (*d*, H-4), 1.28 and 0.92 (*dd*, H-6), 2.33 (*s*, H-7), 1.35 (*qq*, H-8), 0.97 (*d*, H-9, 10) (J [Hz]: 1,6 = 5.5; 1,6′ = 8; 4,4′ = 17; 6,6′ = 4.5; 8,9 = 8,10 = 7); ^{13}C NMR ($CDCl_3$, C-1–C-10): δ36.0 *d*, 207.4 *s*, 177.5 *s*, 31.3 *t*, 35.7 *s*, 21.0 *t*, 32.0 *q*, 32.6 *d*, 19.7 *q*, 19.2 *q*.

Addition of CH_2N_2 gave the methyl ester, colourless oil; MS *m/z* (rel. int.): 198 $[M]^+$ (3), 166.099 $[M-MeOH]^+$ (35) (calc. for $C_{10}H_{14}O_2$: 166.099), 155 $[M-COMe]^+$ (13), 124 [McLafferty]$^+$ (94), 109 $[124-Me]^+$ (100); ^{1}H NMR ($CDCl_3$): 2.02 (*dd*, H-1), 2.43 and 2.84 (*d*, H-4), 0.87 and 1.24 (*dd*, H-6), 2.36 (*s*, H-7), 1.31 (*qq*, H-7), 0.91 and 0.94 (*d*, H-9, 10).

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