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Article

Eudesmane Derivatives from *Verbesina turbacensis*

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Quatro sesquiterpenos do tipo eudesmano foram isolados das partes aéreas de *Verbesina turbacensis* e foram caracterizados por métodos espectroscópicos incluindo experimentos bidimensionais de RMN.

Four eudesmane sesquiterpene were isolated from the aerial parts of *Verbesina turbacensis* and were characterised by spectroscopic methods, including 2D NMR experiments.

Keywords: *Verbesina turbacensis*, Asteraceae, sesquiterpenes, eudesmane

Introduction

Species of the genus *Verbesina* (Asteraceae, tribe *Heliantheae*, subtribe *Ecliptinae*) have produced a range of eudesmane sesquiterpenes with cinnamate or a derived ester group,[1-7] of which, α- and β-verbesinolcoumarates were the first reported examples.[1,8] Several species also afforded elemanolides,[9,10] diterpenes,[11] flavonoids,[12] and biological active guanidines as Galegine, the toxic principle of *V. enceloides* Benth.[13,14]

As a continuation of our phytochemical investigation on Venezuelan Compositae,[15-17] in this paper we describe the results of our study of *Verbesina turbacensis* H.B.K., a previously uninvestigated species, which is widely distributed throughout of Central America from Mexico to Colombia and Venezuela.[18] *V. turbacensis* as well as *V. caracasana* Fries, a plant which contains the dose-dependent hypotensive agent Caracasanamide,[19] are frequently used in Venezuela as medicinal plants against a variety of diseases.

Results and Discussion

The dried, ground leaves and stems of *V. turbacensis* were extracted with acetone at room temperature and the obtained extract was purified by standard procedures. Vacuum liquid chromatography separation over silica gel[20] of the fractions obtained from a hexane-EtOAc (3:1, v/v) eluate, gave the sesquiterpenes **1**, **3**, **5** and **9**.

Compound **1** was crystallized from acetone as colourless prisms (mp 221-223º C; $[\alpha]_D$: + 147.2º) and analysed for $C_{24}H_{32}O_4$ ([M^+] *m/z* 384.2189 high resolution EI-MS). Its ^{1}H NMR spectral data were in close agreement with those reported by Bohlmann and Lonitz [2] for the methyl-6β-[cinnamoyloxy]-eudesman-15-oate **2.** However the IR, ^{1}H NMR and ^{13}C NMR spectra of compound **1** revealed the presence of a free carboxyl group [IR: 3600-3200 and 1735 cm^{-1}; δ_H 11.45 broad singlet; δ_C 181.10 (O=C-O-)], instead of the methyl ester present in **2**. In accordance with above assignments, treatment of **1** with CH_2N_2/eter gave **2** and consequently **1** was identified as 6β-[cinnamoyloxy]-eudesman-15-oic acid. Further support to structure **1** was provided by ^{1}H,^{1}H-COSY, HMQC and HMBC experiments, which allowed the assignments in Tables 1 and 2. The NOESY spectrum of **1** showed cross peaks between the signal assigned to H-6 and the signals of H-4 and H-7 respectively, suggesting that all three protons were on the same side of the molecule. Assuming an A/B ring fusion (5αH, 10βMe); these results clearly confirm a β-configuration for C-4 carboxyl-, C-6 "*trans*"-cinnamoyl- and C-7 isopropyl groups.

Compound **3** was obtained as white needles (mp 201-203ºC; $[\alpha]_D$: +176.72º). Its molecular formula $C_{26}H_{34}O_6$ ([M^+] *m/z* 442.3501, high resolution EI-MS) and its NMR spectral data (Tables 1 and 2) indicated that **3** differed from **1** only by the presence of an extra acetoxy group. [IR: 1.740 cm^{-1}; δ_H 2.06; δ_C 21.28 and 170.43]. This group was located in C-1 from the HMBC spectrum, which

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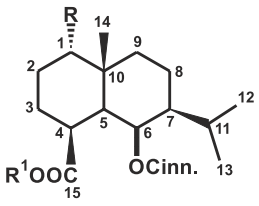

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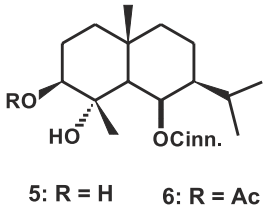
2: R = H, R^1= Me
3: R = OAc, R^1 = H
4: R = OAc, R^1 = Me



5: R = H 6: R = Ac

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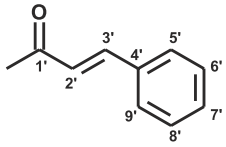

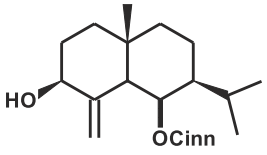

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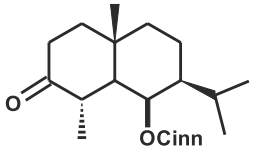

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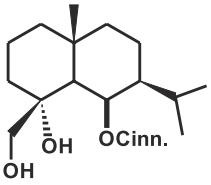

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showed that H-1 (δ 4.54, t J 2.5 Hz) was correlated to C-2/C-3 (δ 22.73), C-5 (δ 43.86), C-10 (δ 37.44), C-14 (δ 20.53) and C-1" (δ 170.43). The α-orientation of acetoxy group already evidenced for the multiplicity of H-1 resonance, also followed from the strong shielding effect on C-3 and C-9 signals, when the chemical shifts of **3** were compared with those of **1** and **2** (Table 2). Thus, **3** was characterised as 1α-acetoxy-6β-[cinnamoyloxy] eudesman-15-oic acid, which had already been reported from *Verbesina eggersii* Hieron,[2] but it was isolated as the methyl ester **4**. Treatment of **3** with CH_2N_2/eter gave a methyl ester with data identical with those ones of **4.** The physical and spectral data of the acid **3** have not been previously published.

Compound **5** was isolated as colourless hexagonal plates (mp185-87º C; $[\alpha]_D$: +65.89º). On the basis of high resolution EI-MS (*m/z* 368.2375 $[M-H_2O]^+$) and ^{13}C-NMR (BB and DEPT) spectral data, the molecular formula of **5** was deduced to be $C_{24}H_{34}O_4$. These data, as well as 2D-NMR experiments, revealed that **5** is a 6β-[cinnamoyloxy]-eudesmane sesquiterpene similar to **1**, with an additional secondary hydroxyl group (δ_H 3.10 (-OH) and 3.44 (H-C-O-); δ_C 78.84), but in which, the C-15 carboxyl group was replaced by a methyl (δ_H 1.14; δ_C 17.92) located on a carbon that supports a tertiary hydroxyl group (δ_C 75.18). The position of secondary hydroxyl group was clarified by the correlation peaks between C-1/H-3; C-2/H-3; C-4/H-3; C-5/H-3; C-15/H-3; C-3/H-1; C-3/H-2; C-3/H-5 and C-3/H-15 in the HMBC spectrum. On acetylation with Ac_2O/Py, compound **5** gave the monoacetate **6**. Treatment of **5** with H_2SO_4 in dry acetone gave the dehydration products **7** and **8**, but did not give an acetonide derivative. In spite of this result, the stereochemistry in C-3 was evidenced on the basis of H-3 multiplicity signal and the magnitude of its coupling constants to H-2α (J 3.5 Hz) and H-2β a (J 11.5 Hz), which were in agreement with a β-equatorial oriented hydroxyl group. All the above data were consistent with the structure proposed for **5**, and these ones also agree well with the spectroscopic data reported for the 6β-[cinnamoyloxy]-3β, 4α-dihydroxyeudesmane, a sesquiterpene previously isolated from *V. persicifolia* D.C.[6] and *V. oncophora* Rob. et Seat.[7]

Compound **9** (oil, $[\alpha]_D$: -13.8º, formula molecular $C_{24}H_{34}O_4$) was also identified as a 6β-[cinnamoyloxy]-eudesmane derivative similar to **5**. The ^{1}H- and ^{13}C-NMR data, as well as the carbon connectivity pattern deduced from ^{1}H,^{1}H-COSY, HMQC and HMBC experiments, indicated that **9** lacks of the C-3 hydroxyl group and that the C-4 methyl has been replaced by a hydroxymethyl group [δ_H 3.35 d (J 11.0 Hz) and 3.75 dd (J 11.0 and J 1.3 Hz); δ_C 62.73]. The W coupling (J= 1.3 Hz) between H-15$_B$ and H-3α revealed a β-axial orientation of hydoxymethyl group. The physical constants and the EIMS and ^{1}H-NMR data of **9**, were in agreement with those published for the 6β-[cinnamoyloxy]-4α, 15-dihydroxyeudesmane, which has been previously isolated from *V. eggersii.*[5] Until now ^{13}C-NMR data of **9** have not been reported in the literature.

Table 1. ^{1}H NMR spectral data of compounds **1-9** (400 MHz, $CDCl_3$, ppm from TMS)

H	**1**[a]	**2**[b]	**3**[a]	**4**[b]	**5**[a]	**6**[a]	**7**[b]	**8**[b]	**9**[a]
1α	0.99 td	1.09 m	-	-	1.23 td	1.28 td	1.36 m	1.48 m	1.01 m
1β	1.37 m	1.42 m	4.54 t	4.67 t	1.40 m	1.42 m	1.50 m	1.67 m	1.39 m
2α	1.19 m	1.45 m	1.36 m	1.69 m	1.75 dq	1.76 dq	1.95 m	2.34 ddd	1.19 m
2β	1.85 qt	2.15 qt	2.14 tdd	2.46 tdd	1.54 m	1.53 m	1.53 m	2.48 td	1.92 m
3α	1.07 m	1.47 m	1.05 m	1.63 m	3.44 dd	4.65 dd	3.94 dd	-	2.30 m
3β	1.39 m	1.94 ddt	1.39 m	1.82 ddt	-	-	-	-	NA
4α	2.23 td	2.74 td	2.27 td	2.75 td	-	-	-	2.31 dc	-
5α	1.50 d	1.67 dd	1.97 dd	2.12 dd	1.30 d	1.39 d	1.90 brs	1.34 dd	1.79 d
6α	5.40 brs	5.51 t	5.46 brs	5.56 t	5.87 brs	5.93 brs	5.72 brs	5.54 t	5.96 brs
7α	1.14 m	1.17 m	1.11 m	1.16 m	1.08 m	1.12 m	1.12 m	1.10 m	1.15 m
8α	1.54 m	1.58 m	1.55 m	1.61 m	1.63 m	1.63 m	1.68 m	1.63 m	1.71 m
8β	1.48 m	1.51 m	1.43 m	1.51 m	1.51 m	1.56 m	1.26 m	1.22 m	1.46 m
9α	1.10 m	1.13 m	1.46 m	1.59 m	1.17 m	1.08 m	1.29 m	1.28m	1.05 m
9β	1.51 m	1.56 m	1.20 dt	1.28 dt	1.59 m	1.56 m	1.69 m	1.61 m	1.61 m
11	1.42 m	1.37 m	1.41 m	1.36 m	1.44 m	1.41 m	1.40 m	1.46 m	1.42 m
12	1.02 d	1.00 d	1.02 d	1.01 d	0.90 d	0.89 d	1.04 d	0.96 d	0.88 d
13	0.80 d	0.82 d	0.80 d	0.81 d	0.88 d	0.86 d	0.85 d	0.86 d	0.84 d
14	1.22 s	1.33 s	1.31 s	1.42 s	1.17 s	1.19 s	1.06 s	1.31 s	1.12 s
15	-	-	-	-	1.14 s	1.17 s	5.12 ; 4.81[c]	1.13 d	3.35; 3.75[d]
2'	6.32 d	6.37 d	6.31 d	6.35 d	6.46 d	6.43 d	6.39 d	6.39 d	6.41 d
3'	7.61 d	7.64 d	7.59 d	7.64 d	7.72 d	7.68 d	7.68 d	7.66 d	7.71 d
5', 9'	7.51 m	7.51 m	7.50 m	7.51 m	7.56 m	7.53 m	7.50 m	7.50 m	7.54 m
6', 8'	7.31 m	7.36 m	7.33 m	7.37 m	7.39 m	7.38 m	7.36 m	7.35 m	7.40 m
7'	7.32 m	7.37 m	7.34 m	7.38 m	7.39 m	7.39 m	7.37 m	7.36 m	7.41 m
AcO	-	-	2.06 s	2.00 s	-	2.05 s	-	-	-
-OCH_3	-	3.42 s	-	3.43 s	-	-	-	-	-

Coupling constants: **(1)-(9)**: $J_{5\alpha,6\alpha} = J_{6\alpha,7\alpha} \cong 1.5$ Hz; $J_{11,12} = J_{11,13} \cong 7$ Hz ; $J_{2',3'} = 16$ Hz; **(1)-(2)**: $J_{1\alpha,1\beta} = J_{2\alpha,2\beta} = J_{3\alpha,3\beta} \cong 13.5$ Hz; $J_{1\alpha,2\alpha} \cong 7.8$ Hz; $J_{1\alpha,2\beta} \cong 12.5$ Hz; $J_{1\beta,2\alpha} = J_{1\beta,2\beta} = J_{2\beta,3\beta} \cong 3.4$ Hz; $J_{2\alpha,3\beta} = J_{3\beta,4\alpha} \cong 1.8$; $J_{3\alpha,4\alpha} = J_{4\alpha,5\alpha} \cong 5.3$ Hz; (**3**)-(**4**): $J_{1\beta,2\alpha} = J_{1\beta,2\beta} \cong 2.5$ Hz; $J_{2\alpha,2\beta} = J_{3\alpha,3\beta} \cong 13.5$; $J_{2\beta,1\alpha} = J_{9\alpha,9\beta} = 12.5$; $J_{2\beta,3\beta} \cong 4.4$ Hz; $J_{3\alpha,4\alpha} = J_{4\alpha,5\alpha} \cong 5.5$ Hz; $J_{3\beta,4\alpha} = J_{8\alpha,9\beta} = J_{8\beta,9\beta} \cong 1.8$; **(5)-(6)**: $J_{1\alpha,1\beta} = J_{1\alpha,2\beta} = J_{2\alpha,2\beta} \cong 13.8$ Hz; $J_{1\alpha,2\alpha} = J_{1\beta,2\alpha} = J_{1\beta,2\beta} \cong 4.0$ Hz; $J_{2\alpha,3\alpha} \cong 3.5$ Hz; $J_{2\beta,3\alpha} \cong 11.5$ Hz; **(8)**: $J_{1\alpha,2\beta} = J_{2\alpha,2\beta} \cong 14.9$ Hz; $J_{1\alpha,2\alpha} \cong 4.7$ Hz; $J_{1\beta,2\alpha} \cong 2.1$ Hz; $J_{1\beta,2\beta} \cong 6.3$ Hz; $J_{4\alpha,15} \cong 7.0$ Hz.
[a] Assigned by ^{1}H,^{1}H-COSY, HMQC and HMBC experiments. [b] Assigned by ^{1}H,^{1}H-COSY and HMQC experiments. (*m*): Signals either overlapping or obscured; *J* unresolved. [c] (**8**):H-15_A and H-15_B both broad singlets. [d] (**9**): H-15_A: δ 3.75, d; H-15_B: 3.75 dd [$J_{15A,15B} \cong 11.0$ Hz, $J_{3\alpha,15B} \cong 1.3$ Hz].

Table 2. ^{13}C NMR spectral data of compounds **1-9** (100 MHz, $CDCl_3$, ppm from TMS)

Carbon	**1**[a]	**2**[b]	**3**[a]	**4**[b]	**5**[a]	**6**[a]	**7**[b]	**8**[b]	**9**[a]
1	44.06	44.23	77.39	77.53	40.83	40.72	41.99	42.46	45.00
2	18.53	18.64	22.73	22.84	26.59	25.22	32.29	37.69	19.60
3	29.60	30.56	22.73	23.79	78.84	80.27	73.35	212.51	36.65
4	42.49	42.60	41.68	41.76	75.18	73.63	149.30	42.65	74.85
5	50.40	50.29	43.86	43.73	55.09	55.49	51.92	53.63	56.92
6	74.04	74.45	74.16	74.47	69.53	69.12	71.66	70.96	69.12
7	51.04	51.18	50.63	50.78	49.75	49.88	50.65	48.72	50.22
8	20.68	20.89	20.13	20.35	21.15	20.96	20.57	20.47	20.57
9	44.39	44.51	36.41	36.52	45.04	44.73	41.10	40.69	43.38
10	34.24	34.27	37.44	37.47	34.65	34.40	35.96	33.52	34.98
11	28.18	28.26	28.06	28.13	28.73	28.53	28.07	28.64	28.67
12	20.13	20.21	20.07	20.15	20.76	20.55	20.38	20.26	19.95
13	22.18	22.07	22.20	22.04	21.59	21.20	22.01	21.60	20.99
14	20.60	20.66	20.53	20.61	21.24	21.35	20.69	19.71	21.47
15	181.10	175.92	181.17	175.98	17.92	18.63	104.27	11.45	62.73
1'	166.75	166.36	166.54	166.21	168.26	167.62	166.77	165.97	168.05
2'	118.63	118.92	118.32	118.62	118.17	118.20	118.58	117.91	117.96
3'	144.18	144.40	144.35	144.67	145.83	145.48	144.84	144.98	146.02
4'	134.84	134.45	134.60	134.29	134.15	134.12	134.42	134.14	134.05
5', 9'	128.19	128.07	128.14	128.06	128.27	128.12	128.09	127.97	128.31
6', 8'	128.58	128.83	128.72	128.84	128.91	128.81	128.83	128.76	128.93
7'	129.74	130.18	130.02	130.29	130.58	130.42	130.24	130.26	130.64
1"	-	-	170.43	170.30	-	170.76	-	-	-
2"	-	-	21.28	21.25	-	21.66	-	-	-
OMe	-	51.64	-	51.77	-	-	-	-	-

[a] Assigned by DEPT, HMQC and HMBC experiments. [b] Assigned by DEPT and HMQC experiments.

From a chemotaxonomic aspect the isolated eudesmane sesquiterpenes have to be regarded as typical constituents of *Verbesina* species, even though similar eudesmane cinnamates has sporadically been isolated from *Ambrosia*, *Brintonia* and *Solidago* species.[21-23] From a biological point of view, it is also important to highlight that many eudesmane derivatives, with structures related to **1**-**9**, have shown antifeedant,[24] antibacterial[25] or cytotoxic[26] properties.

Experimental

General experimental procedures

Melting points were determined with a Fisher-Johns apparatus and they have not been corrected. Optical activities were measured in $CHCl_3$ on a Rudolph Research Autopol III polarimeter. IR spectra were taken on a Perkin-Elmer FT-1725X spectrophotometer as film or KBr pellets. ^1H, ^{13}C and two-dimensional NMR spectra were measured on a Bruker-Avance DRX400 instrument, using $CDCl_3$ as solvent with TMS as internal standard. EI-MS and HREI-MS were run on a Hewlett-Packard 5930A and on an Autospec VG spectrometer, respectively; direct inlet, 70 eV. TLC was carried out on 0.25 mm layers of silica gel PF 254 (Merck). VCC was performated with silica gel 60 (70-230 mesh.).

Plant material

Verbesina turbacensis H.B.K. was collected at La Hechicera, Municipio Autónomo Libertador (Mérida, Venezuela) in November. A *voucher specimen* (JMA 1511) was deposited in the MERF Herbarium (Faculty of Pharmacy, ULA).

Extraction and isolation of the constituents

Leaves and stems of *V. turbacensis* (*ca* 7.5 kg) were air-dried, ground and exhaustively extracted with acetone at room temperature for 1 week. The dissolution obtained was concentrated *in vacuo* to afford a dark brown residue (450 g), which was preadsorbed on silica gel and subjected to VCC over silica gel,[20] using hexane with increasing amounts of EtOAc as eluent. Fractions of 1 L were collected and combined based upon TLC monitoring. From fractions eluted with hexane-EtOAc (7:3, v/v), stigmasterol (55 mg) was purified by crystallisation and identified by mp, IR, ^1H NMR and TLC comparison. Fractions eluted with hexane-EtOAc (3:2, v/v) were purified by repeated flash chromatography or preparative TLC yielding four compounds, which were obtained in the following sequence: **1** (2.83 g), **3** (870 mg), **5** (6.25 g) and **9** (35 mg).

*6β-[cinnamoyloxy]-eudesman-15-oic acid (**1**)*

Colourless prisms (Me_2CO), mp 221-223º C (lit.[2] mp 193º C); $[\alpha]_D$: + 147.2º (*c*, 0.43 $CHCl_3$).; IR ν_{max}/ cm^{-1} 3600-3200, 2950, 1735, 1690, 1665, 1220, 1180, 790, 700 (KBr); UV (MeOH) λ_{max}: 275 nm; HR-EIMS, [*m/z*, (% rel. int.)]: 384.2189 (M^+, calcd. for $C_{24}H_{32}O_4$: 384.2300) (4.32); LR-EIMS [*m/z*, (% rel. int.)]: 384 [M^+] (4.35), 366 [M^+-H_2O] (3.80), 356 [M^+-CO] (2.89), 338 [M^+-HCOOH] (46.48), 253 [M^+-PhCH=CHCO] (9.07), 236 [M^+-PhCH=CHCOOH] (39.60), 235 (48.34), 221 [M^+-PhCH=CHCOOH-CH_3] (7.74), 193 [M^+-PhCH=CHCOOH-C_3H_7] (54.51), 190 (30.48), 175 (22.15), 148 (20.83), 147 (20.88), 131 [PhCH=CHCO]$^+$ (100), 103 [PhCH=CH]$^+$ (22.45), 91 (8.59), 77 (9.62).

*Methyl-6β-[cinnamoyloxy]-eudesman-15-oate (**2**)*

Compound **1** (320 mg) was treated with excess ethereal CH_2N_2 and solution was left standing overnight in a refrigerator at 4°. Evaporation of ether yielded the methyl ester **2** (325 mg): White needles (EtOAc), mp 109-111º C; $[\alpha]_D$: + 45.1º (*c*, 0.32 $CHCl_3$); IR ν_{max}/ cm^{-1} 1723, 1700, 1667, 1211, 1018, 859, 727 (KBr); HR-EIMS [*m/z*, (% rel. int.)]: 398.2808 (M^+, calcd. for $C_{25}H_{34}O_4$: 398.2497) (1.52); LR-EIMS [*m/z*, (% rel. int.)]: 398 [M^+] (1.54), 370 (0.53), 338 [M^+-$HCOOCH_3$] (9.89), 293 (1.23), 267 [M^+-PhCH=CHCO] (6.66), 250 [M^+-PhCH=CHCOOH] (5.89), 235 [M^+-PhCH=CHCOOH-CH_3] (20.88), 207 [M^+-PhCH=CHCOOH-C_3H_7] (9.11), 190 [M^+-PhCH=CHCOOH-$HCOOCH_3$] (16.47), 175 (4.01), 131 [PhCH=CHCO]$^+$ (100), 103 (20.36), 91 (4.58), 77 (7.98), 55 (6.43).

*1α-Acetoxy-6β-[cinnamoyloxy]-eudesman-15-oic (**3**)*

White needles (EtOAc), mp 201-203ºC; $[\alpha]_D$: +176.72º (*c*, 0.55 $CHCl_3$).; IR ν_{max}/ cm^{-1} 3500-3250, 2950, 1740,1735, 1700, 1650, 1280, 1180, 750, 690 (KBr); UV (MeOH) λ_{max}: 275 nm; HR-EIMS [*m/z*, (% rel. int.)]: 442.3501 (M^+, calcd. for $C_{26}H_{34}O_6$: 442.2355) (0.56); LR-EIMS [*m/z*, (% rel. int.)]: 442 [M^+] (0.60), 424 [M^+-H_2O] (2.52), 414 [M^+-CO] (2.76), 396 [M^+-HCOOH] (6.06), 364 [M^+-H_2O-AcOH] (1.83), 336 [M^+-HCOOH-AcOH] (13.85), 311 [M^+-PhCH=CHCO] (2.73), 294 [M^+-PhCH=CHCOOH] (3.30), 293 (9.27), 251 [M^+-PhCH=CHCOOH-C_3H_7] (1.08), 234 [M^+-PhCH=CHCOOH-AcOH] (29.97), 233 (12.03), 219 [M^+-PhCH=CHCOOH-AcOH-CH_3] (6.93), 191[M^+-PhCH=CHCOOH-AcOH-C_3H_7] (22.41), 148 (6.34), 147

(16.02), 145 (18.71), 131 [PhCH=CHCO]$^+$ (100), 103 [PhCH=CH]$^+$ (22.10), 91 (7.45), 77 (8.37).

*Methyl-1α-acetoxy-6β-[cinnamoyloxy]-eudesman-15-oate (**4**)*

Compound **3** (200 mg) was methylated following the procedure above indicated. The methyl ester **4** (195 mg) was obtained as white needles (hexane), mp 172-75ºC; $[\alpha]_D$: +68.25º (*c*, 0.18 $CHCl_3$).; IR ν_{max}/ cm^{-1} 1735, 1730, 1715, 1635, 1280, 1243, 1018, 750 (KBr); HR-EIMS [*m/z*, (% rel. int.)]: 456.2491 (M^+, calcd. for $C_{27}H_{36}O_6$: 456.2511) (0.86); LR-EIMS [*m/z*, (% rel. int.)]: 456 [M^+] (0.70), 428 (0.67), 396 [M^+-AcOH] (7.67), 350 (7.85), 336 [M^+-AcOH-$HCOOCH_3$] (7.81), 325 [M^+-PhCH=CHCO] (6.47), 308 [M^+-PhCH=CHCOOH] (3.22), 293 [M^+-PhCH=CHCOOH-CH_3] (25.06), 248 [M^+-PhCH=CHCOOH-AcOH] (23.33), 233 (14.08), 205 [M^+-PhCH=CHCOOH-AcOH-C_3H_7] (27.26), 188 (13.56), 147 (15.38), 145 (40.11), 131 [PhCH=CHCO]$^+$ (100), 103 (37.60), 91 (7.28), 77 (15.09), 55 (18.08).

*6β-[cinnamoyloxy]-3β, 4α-dihydroxyeudesmane (**5**)*

Colourless hexagonal plates (EtOAc), mp 185-87º C; $[\alpha]_D$: +65.89º (*c*, 0.39 $CHCl_3$); IR ν_{max}/ cm^{-1} 3430, 2950, 1730, 1647, 1200, 1100, 700 (KBr); UV (MeOH) λ_{max}: 280 nm; HR-EIMS [*m/z*, (% rel. int.)]: 368.2375 ([M-H_2O]$^+$, calcd. for $C_{24}H_{32}O_3$: 368.2351) (2.82); LR-EIMS [*m/z*, (% rel. int.)]: 368 [M^+-H_2O] (2.79), 350 [M^+-2H_2O] (0.79), 307 [M^+-2H_2O-C_3H_7] (1.49), 238 [M^+-PhCH=CHCOOH] (48.76), 220 [M^+-PhCH=CHCOOH-H_2O] (20.72), 205 [M^+-PhCH=CHCOOH-H_2O-CH_3] (7.86), 202 [M^+-PhCH=CHCOOH-2H_2O] (8.91), 195 [M^+-PhCH=CHCOOH-C_3H_7] (20.61), 177 [M^+-PhCH=CHCOOH-H_2O-C_3H_7] (38.20), 159 [M^+-PhCH=CHCOOH-2H_2O-C_3H_7] (10.45), 149 (12.68), 137 (20.14), 131 [PhCH=CHCO]$^+$ (100), 121 (10.14), 103 [PhCH=CH]$^+$ (27.84), 95 (11.68), 91 (10.75), 81 (18.10), 77 (11.79).

*3β-Acetoxy-6β-[cinnamoyloxy]-4α-hydroxyeudesmane (**6**)*

Compound **5** (340 mg) was dissolved in pyridine (5 ml) and treated with Ac_2O (10 ml) at room temperature overnight. Ice was added to the reaction mixture and immediately was extracted with EtOAc. The EtOAc layer was dried on $MgSO_4$ and evaporated to yield the acetate **6** (295 mg): Colourless oil; $[\alpha]_D$: +24.5º (*c*, 0.20 $CHCl_3$); ν_{max}/ cm^{-1} 3515, 1712, 1633, 1264, 1076, 700 (nujol); HR-EIMS [*m/z*, (% rel. int.)]: 428.2556 ([M^+], calcd. for $C_{26}H_{36}O_5$: 428.2562) (0.32); LR-EIMS [*m/z*, (% rel. int.)]: 428 [M^+] (0.18), 410 [M^+-H_2O] (0.15), 368 [M^+-AcOH] (2.34), 350 (0.91), 280 [M^+-PhCH=CHCOOH] (5.27), 262 [M^+-PhCH=CHCOOH-H_2O] (12.94), 238 (7.15), 220 [M^+-PhCH=CHCOOH-AcOH] (33.98), 202 (32.72), 187 (9.00), 177 (26.21), 159 (22.73), 148 (9.75), 137 (14.34), 131 [PhCH=CHCO]$^+$ (100), 107 (10.41), 103 [PhCH=CH]$^+$ (46.61), 91 (12.67), 77 (25.52), 69 (11.37), 55 (14.84).

*Treatment of compound **5** with H_2SO_4/Me_2CO*

Compound **5** (210 mg) was dissolved in dry acetone (20 ml) and a few drops of H_2SO_4 were added. The solution was stirred for 24 h. at room temperature, neutralized with 10% aq. HCO_3Na and extracted with EtAcO. The EtAcO layer was dried on Na_2SO_4 and concentrated "*in vacuo*" yielding an oily residue, which after chromatography gave **7** (95 mg) and **8** (45 mg).

6β-[cinnamoyloxy]-3β-hydroxyeudesm-4,15-ene (**7**): mp140-42º C; IR ν_{max}/cm^{-1} 3510, 1735, 1630, 1255 1085, 890, 710 (KBr); HR-EIMS [*m/z*, (% rel. int.)]: 368.22321 ([M^+], calcd. for $C_{24}H_{32}O_3$: 368.2351) (3.72); LR-EIMS [*m/z*, (% rel. int.)]: 368 [M^+] (4.07), 350 [M^+-H_2O] (2.25), 307 [M^+-H_2O-C_3H_7] (4.69), 220 [M^+-PhCH=CHCOOH] (15.87), 202 [M^+-PhCH=CHCOOH-H_2O] (41.31), 187 (9.39), 177 [M^+-PhCH=CHCOOH- C_3H_7] (15.25), 159 (44.99), 145 (6.37), 131 [PhCH=CHCO]$^+$ (100), 103 [PhCH=CH]$^+$ (38.03), 91 (13.29), 77 (20.43), 67 (6.63), 55 (9.46).

6β-[cinnamoyloxy]-3-oxo-4βH-eudesmane (**8**): Colourless oil; IR ν_{max}/cm^{-1} 1732, 1715, 1633, 1220, 740, 710 (nujol).

*6β-[cinnamoyloxy]-4α,15-dihydroxyeudesmane (**9**)*

Colourless oil; $[\alpha]_D$: -13.8º (*c*, 0.25 $CHCl_3$).; IR ν_{max}/cm^{-1} 3590, 2960, 1720, 1642, 1130, 710 (nujol); UV (MeOH) λ_{max}: 278 nm; HR-EIMS [*m/z*, (% rel. int.)]: 368.2369 ([M-H_2O]$^+$, calcd. for $C_{24}H_{32}O_3$: 368.2351) (7.98); LR-EIMS [*m/z*, (% rel. int.)]: 368 [M^+-H_2O] (8.45), 355 [M^+-CH_2OH] (11.23), 337 [M^+-H_2O-CH_2OH] (2.81), 325 [M^+-H_2O-C_3H_7] (1.37), 294 (0.42), 238 [M^+-PhCH=CHCOOH] (12.35), 220 [M^+-PhCH=CHCOOH-H_2O] (32.18), 207 [M^+-PhCH=CHCOOH-CH_2OH] (73.18), 205 (7.53), 195 [M^+-PhCH=CHCOOH-C_3H_7] (19.15), 189 (27.16), 177 [M^+-PhCH=CHCOOH-H_2O-C_3H_7] (32.71), 146 (11.36), 131 [PhCH=CHCO]$^+$ (100), 103 [PhCH=CH]$^+$ (38.15), 91 (12.41), 77 (11.10).

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