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HIRSUTINOLIDES AND OTHER SESQUITERPENE LACTONES FROM *VERNONIA* SPECIES*

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Key Word Index—*Vernonia compactiflora*; *V. monocephala*; *V. chalybaea*; *V. cotoneaster*; Compositae; sesquiterpene lactones; hirsutinolides; glaucolides.

Abstract—The investigation of *Vernonia compactiflora* afforded in addition to known compounds a hirsutinolide and a glaucolide; the latter is most probably the direct precursor of the former. The aerial parts of *V. monocephala* contain the same hirsutinolide as well as the corresponding acetate, while those of *V. chalybaea* afforded glaucolide B. A reinvestigation of the aerial parts of *V. cotoneaster* gave in addition to the allenic lactone isolated before two further known ones as well as a new glaucolide. The structures of the lactones were elucidated by spectroscopic methods and some chemical transformations. The erroneously assigned stereochemistry at C-10 in the hirsutinolides has been revised. The biogenetic relationship of the *Vernonia* lactones is discussed briefly.

INTRODUCTION

The large genus *Vernonia* with more than 1000 species has been investigated chemically by many groups. Highly oxygenated germacranolides such as glaucolides and hirsutinolides seem to be characteristic for many members of this genus, though many other compounds have also been isolated. In continuation of our investigations of representatives of the tribe Vernonieae we have now studied the constituents of further *Vernonia* species. In addition to already known compounds two new hirsutinolides and two glaucolides were isolated, which support the most likely biogenetic pathway for production of these characteristic lactones.

RESULTS AND DISCUSSION

The aerial parts of *Vernonia compactiflora* Mart afforded large amounts of lupeyl acetate, some lupeol, traces of germacrene D and bicyclogermacrene, and two polar compounds. The main constituent was a colourless gum with an uncharacteristic ^{1}H NMR spectrum (Table 1) showing only broad signals except for those of a caproyl residue and two methyl singlets. The IR spectrum indicated the presence of hydroxyl groups and further showed a broad band at 1755 cm^{-1}, while the mass spectrum gave the molecular formula $C_{21}H_{30}O_7$. The presence of a caproyl residue was supported by the fragments $[M-C_5H_{11}CO_2H]^+$ and $[C_5H_{11}CO]^+$. Acetylation afforded a monoacetate but its ^{1}H NMR spectrum was still uncharacteristic (Table 1). At elevated temperature in deuteriobenzene, however, several clear signals occurred. In particular a broadened doublet at δ 6.29 was significant as spin decoupling showed that it was coupled with two double doublets at 2.38 and 1.83. Furthermore a sharp singlet at 5.73 and two doublets for a CH_2OAc group (5.24 and 5.15) were visible, while the IR spectrum now displayed a γ-lactone band at 1780 cm^{-1} and a hydroxyl band at 3580 cm^{-1}. Acetylation in the presence of 4-pyrrolidino pyridine[1] afforded a diacetate and its ^{1}H NMR spectrum could now be interpreted (Table 1). Spin decoupling allowed the assignment of all signals, which were close to those of hirsutinolides[2–4]. Only structure **9** was in agreement with all data. However, the position of the caproyloxy group, could be assigned only by analogy and by the chemical shift of the proton at the ester group bearing carbon, which both favoured an 8α-position, so far present in all hirsutinolides and related glaucolides[2–6].‡ Furthermore, this position could explain the ^{1}H NMR spectral data of the natural compound, which consequently must be **7**, and those of the monoacetate **8**. In both cases the 10-hydroxyl group most probably formed alternate hydrogen bonds with the ether oxygen and the ester group leading to a mixture of conformers, which would explain the uncharacteristic ^{1}H NMR spectra of **7** and **8**, while **9** had a stable conformation as the disturbing 10β-hydroxyl group was acetylated. Finally, oxidation of **7** afforded the aldehyde **10** and its ^{1}H NMR spectral data clearly established the 8α-position of the caproyloxy group. Though again the signals were mostly broad, the chemical shift differences of H-8β in the spectra of **7** and **10** only agreed with the proposed structure. (Though the absolute configuration of the *Vernonia* lactones is not known, the ones given are most likely from biogentic considerations[2].) The ^{13}C NMR signals of **7** and **9** (see Experimental) were also in good agreement with

*Part 390 in the series "Naturally Occurring Terpene Derivatives". For part 389 see Bohlmann, F., Ahmed M., Jakupovic, J., King, R. M. and Robinson, H. (1982) *Phytochemistry* **21**, 691.

‡The stereochemistry at C-10 of the hirsutinolides was erroneously written with a β-methyl group due to the *cis*-orientation of the C-4 and C-10 methyl groups in a model[2].

Table 1. ^{1}H NMR spectral data of **5**, **7**–**10** (400 MHz, TMS as internal standard)

	5($CDCl_3$)	**7**($CDCl_3$)	**8**($CDCl_3$)	C_6D_6(70°)	**9**($CDCl_3$)	**10**(C_6D_6, 70°)
H-1	2.97 *br d*	4.22 *m*	4.18 *m*	4.05 *dd*	4.65 *dd*	3.97 *dd*
H-2	1.63 *m*		1.60 *m*		2.17 *m*	
H-2′	2.12 *br d*			1.96 *m*	2.50 *m*	1.80 *m*
H-3	2.34 *m**		2.1–1.9 *m*	1.60 *m*	2.56 *m*	1.55 *m*
H-3′	2.20 *ddd*			1.20 *m*	2.41 *br d*	1.20 *m*
H-5	6.25 *s*	5.98 *br s*	5.98 *br s*	5.73 *s*	6.10 *s*	5.83
H-8	6.02 *d*			6.29 *br d*	5.71 *dd*	6.74 *br*
H-9	2.46 *d*		2.1–1.9 *m*	2.38 *dd*	2.60 *d*	2.42 *dd*
H-9′	1.83			1.83 *dd*		1.69 *br d*
H-13	5.13 *d*	4.59 *br s*	4.95 *m*	5.24 *d*	5.05 *d*	10.11 *s*
H-13′	4.91 *d*			5.15 *d*	4.87 *d*	
H-14	1.52 *s*	1.16 *br s*	1.13 *br s*	1.05 *s*	1.54 *s*	0.97 *s*
H-15	1.95 *s*	1.49 *s*	1.46 *s*	1.27 *s*	1.43 *s*	1.16 *s*
OCOR	2.36 *dt*	2.36 *t*	2.31 *br t*	2.13 *t*	2.35 *dt*	2.01 *t*
	2.32 *dt*				2.31 *dt*	
	1.63 *tt*	1.64 *tt*	1.60 *tt*	1.55 *tt*	1.61 *tt*	1.55 *tt*
	1.31 *m*	1.30 *m*	1.27 *m*	1.17 *m*	1.30 *m*	1.16 *m*
	0.91	0.90 *t*	0.85 *t*	0.85 *t*	0.90 *t*	0.84 *t*
OAc	2.08 *s*		2.07 *s*	1.75 *s*	2.11 *s*	
	2.04 *s*				2.00 *s*	

*In $CDCl_3/C_6D_6$H-3 2.08 *ddd*.

J(Hz): Compound **5**: 1,2 = 10; 2,2′ = 15; 2,3 = 11.5; 2,3′ = 6; 2′,3′ = 3; 3,3′ = 15; 8,9′ = 10; 9,9′ = 13.5; compound **8** ($CDCl_3$, C_6D_6): 8, 9 = 7.5; 8, 9′ = 2.5; 9, 9′ = 15; 13, 13′ = 12.5; compound **9**: 1, 2 = 10; 1, 2′ = 6; 8, 9 = 3.5; 12, 13′ = 13; OCOR: 2′, 2′ = 15.5; 2′, 3′ = 4′ = 5′, 6′ = 7.

the proposed structures. The minor polar compound was purified as its diactate. The ^{1}H NMR spectral data (Table 1) showed that the epoxide **5** was present. The position of the epoxide followed from the chemical shifts of H-5 and H-15, both being shifted downfield after acetylation of the tertiary hydroxyl. From the coupling $J_{8,9}$ the 8α-orientation of the caproyloxy group could be deduced. The natural lactone therefore was **4**, obviously the direct precursor of **7**. Protonation of the epoxide oxygen and nucleophilic attack by the tertiary hydroxyl at C-1 would lead directly to **7** (Scheme 1). The enol lactone **4** is also most probably formed via an epoxide as outlined before[2]. The isolation of **4** therefore supports the idea that the diepoxide **3** most probably is the common precursor of the hirsutinolides and the glaucolides.

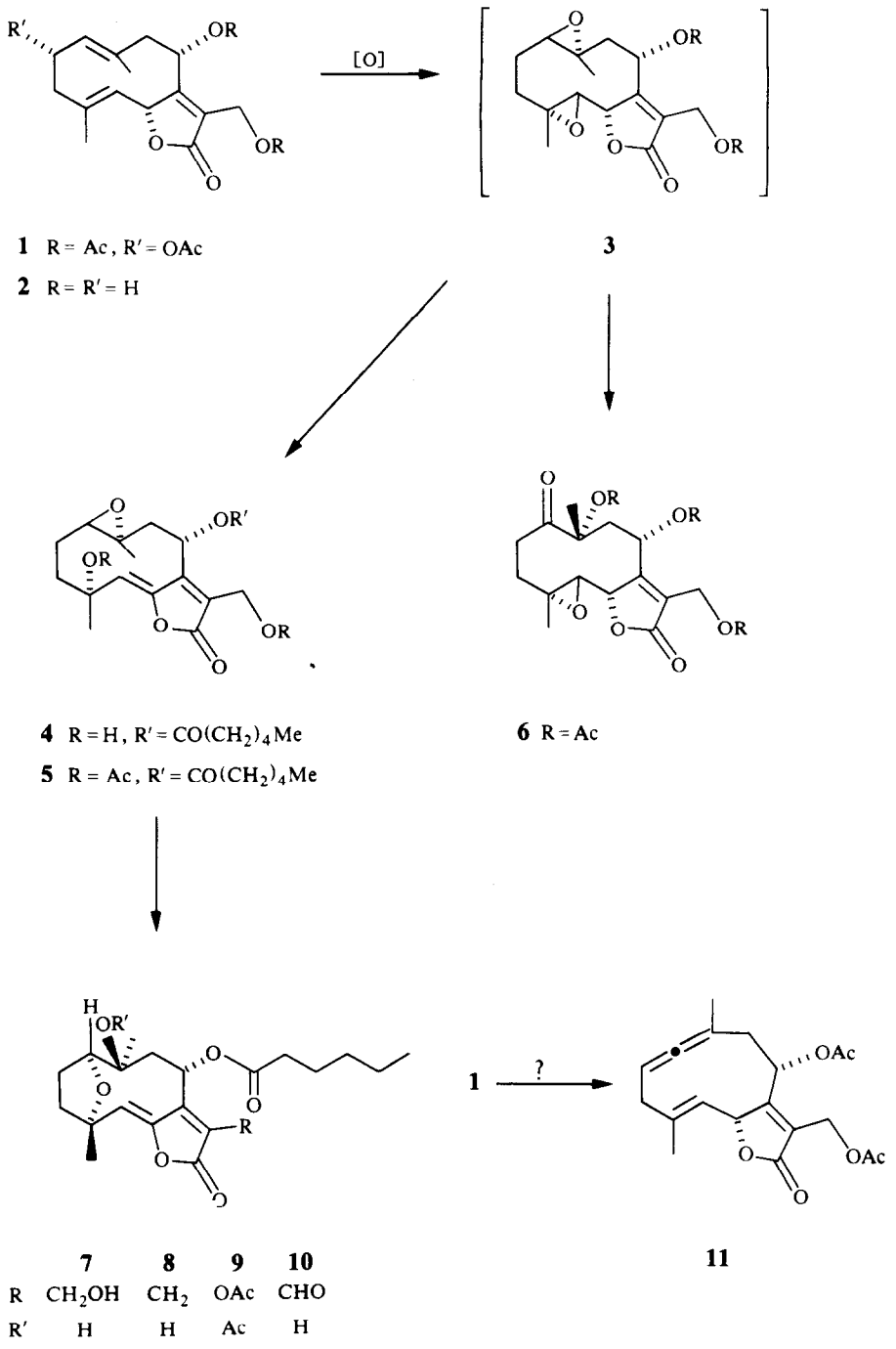
The aerial parts of *Vernonia monocephala* Gardn. in addition to caryophyllene, bicyclogermacrene, lupeyl acetate and lupeol as well as their Δ^{12}-isomers and linolenic acid also afforded **4** and **7**. Furthermore, small amounts of the monoacetate **8** were present, identical with the acetylation product of **7**. The roots only gave lupeol and its acetate as well as their Δ^{12}-isomers. The aerial parts of *V. chalybaea* Mart afforded in addition to lupeyl acetate, its Δ^{12}-isomer, lupeol, its Δ^{12}-isomer, germacrene D, bicyclogermacrene and tridecapentaynene glaucolide B (**6**) [3], while the roots only gave lupeol and its acetate as well as their Δ^{12}-isomers.

The reinvestigation of the aerial parts of *V. cotoneaster* Less. afforded in addition to germacrene D, bicyclogermacrene, α-humulene, lupeyl acetate, the allenic lactone **11** and also **12** and **13**[7] as well as small amounts of the lactone **1**. The ^{1}H NMR spectrum of **1** (Table 2) indicated the presence of a triacetate. Two broadened singlets at δ 1.85 and 1.54 and a pair of doublets at 5.00 and 4.82 showed that a germacranolide was present with a 7,11-double bond. Spin decoupling allowed the assignment of all signals. Starting with the three-fold doublet at 5.74, which was

Table 2. ^{1}H NMR spectral data of compound **1** (400 MHz, $CDCl_3$, TMS as internal standard)

H-1	5.22 *br d*	H-9β	2.89 *br dd*
H-2β	5.74 *ddd*	H-13	5.00 *d*
H-3α	2.78 *dd*	H-13′	4.82 *br d*
H-3β	2.23 *dd*	H-14	1.54 *br s*
H-5	4.48 *br d*	H-15	1.85 *br s*
H-6	5.83 *d*	OAc	2.10 *s*
H-8β	4.97 *dd*		2.08 *s*
H-9α	2.40 *dd*		2.06 *s*

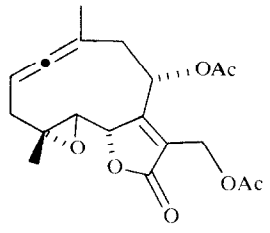
J(Hz): 1,2β = 2β, 3α = 8.5; 2β, 3β = 2; 3α, 3β = 14.5; 5, 6 = 10; 8β, 9α = 12; 8β, 9β = 5; 9α, 9β = 13; 13, 13′ = 13.



Scheme 1.

coupled with the olefinic proton at C-1, and therefore was the signal of the proton under the acetoxyl group at C-2, collapsed the double doublets at 2.78 and 2.23 to doublets. As the double doublet at 4.95 was coupled with the broadened double doublet at 2.89 and the double doublet at 2.40, the former was the signal of H-8β. Irradiation at 1.85 caused a sharpening of the broadened doublet at 4.48, which therefore was the signal of H-5, which further was coupled with the downfield doublet of H-6. Inspection of a model showed that the proposed structure and stereochemistry of **1** agreed with the couplings observed.

The corresponding 4,5-epoxide has been isolated before from a *Vernonia* sp. [7]. The biogenetic relationship of **1** to **3** and **4** is obvious. The additional 2-acetoxyl group may be an indication, that the allenic lactones could be formed by elimination of a 2-oxygen function, while the so far unknown diacetate **2** would be the direct precursor of **3**. The roots gave lupeol and its acetate, **11**, **13**, 15-desoxygoyazensolide



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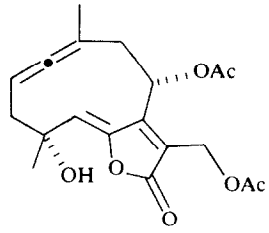
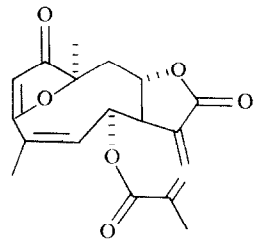
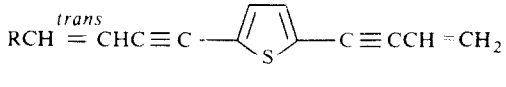


13

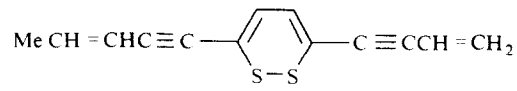


14



15 R = Me

16 R = CHO



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(**14**) [8], the thiophenes **15** and **16** [9] as well as the dithio compound **17** [9]. These results for *Vernonia* sp. again show that highly oxygenated germacranolides are characteristic of this genus. Furthermore, the isolation of additional lactones supports the proposed biogenetic relationships of the main sesquiterpene lactones isolated from this genus (Scheme 1).

EXPERIMENTAL

The air dried plant material, collected in February 1981 in north-eastern Brazil, was extracted with Et_2O–petrol (1:2) and the resulting extract was separated by CC (Si gel) and further by repeated TLC (Si gel). Known compounds were identified by comparing their ^1H NMR spectra with those of authentic material. Vouchers are deposited in the United States National Herbarium.

Vernonia compactiflora (*voucher RMK* 8978). The roots (230 g) afforded 200 mg lupeol, 20 mg of its Δ^{12}-isomer, 1.5 g lupeyl acetate and 300 mg of its Δ^{12}-isomer, while the aerial parts (320 g) gave 2 g lupeyl acetate, 100 mg lupeol, 10 mg linolenic acid, 1 mg germacrene D, 1 mg bicyclogermacrene, 10 mg **4** (Et_2O–petrol, 3:1) and 80 mg **7** (Et_2O–petrol, 3:1).

Vernonia monocephala (*voucher RMK* 8794). The roots (160 g) afforded 80 mg lupeol, 40 mg of its Δ^{12}-isomer, 1 g lupeyl acetate and 300 mg of its Δ^{12}-isomer, while the aerial parts (400 g) gave 4 mg caryophyllene, 1 mg bicyclogermacrene, 200 mg lupeol, 50 mg of its Δ^{12}-isomer, 2 g lupeyl acetate, 200 mg of its Δ^{12}-isomer, 5 mg **4**, 50 mg **7** and 3 mg **8** (Et_2O–petrol, 3:1), identical with the monoacetate prepared from **7**.

Vernonia chalybaea (*voucher RMK* 8710). The roots (100 g) afforded 40 mg lupeyl acetate, 10 mg of its Δ^{12}-isomer, 10 mg lupeol and 3 mg of its Δ^{12}-isomer, while the aerial parts (315g) gave 1 mg tridecapentaynene, 5 mg germacrene D, 3 mg bicyclogermacrene, 400 mg lupeyl acetate, 100 mg of its Δ^{12}-isomer, 20 mg lupeol, 5 mg of its Δ^{12}-isomer and 80 mg **6**.

Vernonia cotoneaster (*voucher RMK* 8681). The aerial parts (210 g) afforded 5 mg germacrene D, 10 mg bicyclogermacrene, 2 mg α-humulene, 500 mg lupeyl acetate, 5 mg **1** (Et_2O–petrol, 3:1), 40 mg **11**, 20 mg **13** and 2 mg **14**, while the roots (70 g) gave 15 mg lupeol, 20 mg of its acetate, 4 mg **11**, 2 mg **13**, 8 mg **14**, 3 mg **15**, 2 mg **16** and 2 mg **17**.

2α-8α-13 *Tri-acetoxygermacra*-1(10), 5, 7(11)-*trien*-6,12-*olide* (**1**). Colourless gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 1780 (γ-lactone), 1755, 1240 (OAc); MS *m/s* (rel. int.): 406.163 [M]$^+$ (1) ($C_{21}H_{26}O_8$), 346 [M − HOAc]$^+$ (11), 304 [346 − ketene]$^+$ (5), 286 [346 − HOAc]$^+$ (12), 244 [286 − ketone]$^+$ (32), 226 [286 − HOAc]$^+$ (35), 211 [226 − Me]$^+$ (25), 178 (100), 161 (91).

$$[\alpha]_{24°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436 \text{ nm}}{-81 \quad -85 \quad -98 \quad -176} \text{ (CHCl}_3\text{; } c0.41\text{)}.$$

8β-*Caproyloxycompactifloride* (**4**). Colourless gum, which was purified as its diacetate **2**, colourless gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 1775 (γ-lactone), 1750, 1235 (CO_2R); MS *m/z* (rel. int.): 478.220 [M]$^+$ (4)($C_{25}H_{34}O_9$), 419 [M − OAc]$^+$ (2), 376 [M − HOAc − ketone]$^+$ (0.5), 260 [376 − HO_2CR]$^+$ (14), 99 [$C_5H_{11}CO$]$^+$ (100), 71 [99 − CO]$^+$ (98).

$$[\alpha]_{24°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436 \quad 365 \text{ nm}}{-102.5 \quad -107.9 \quad -125.6 \quad -251.7 \quad -531.6}$$

(CHCl$_3$; *c*0.89).

8β - *Caproyloxy* - 10β - *hydroxy* - 1 - *desoxyhirsutinolide* (**7**). Colourless gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3560, 3460 (OH), 1755 (lactone, CO_2R); MS *m/z* (rel. int.): 394.199 [M]$^+$ ($C_{21}H_{30}O_7$), 376 [M − Me]$^+$ (0.5), 376 [M − HOAc]$^+$ (8), 334 [M − ketene]$^+$ 218 [260 − C_2H_2O]$^+$ (100), 99 [$C_5H_{11}CO$]$^+$ (60), 71 [99 − CO]$^+$ (61); ^{13}C NMR ($CDCl_3$) (C-1 through C-15): 87.9 *s*, 31.1 *t*, 39.6 *t*, 81.5 *s*, 125.9 *d*, 150.3 *s*, 146.0 *s*, 66.2 *d*, 34.3 *t*, 76.3 *s*, 132.9 *s*, 168.3 *s*, 54.4 *t*, 25.3 *q*, 28.6 *q*; OCOR: 173.0 *s*, 34.3 *t*, 24.3 *t*, 31.1 *t*, 22.1 *t*, 13.7 *q* (a few signals may be interchangeable).

Compound **7** (20 mg) was heated with 0.1 ml Ac_2O for 2 hr at 70°. TCL (Et_2O–petrol, 3:1) afforded 20 mg **8**, colourless gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3580 (OH), 1780 (γ-lactone), 1750, 1240 (CO_2R); MS *m/z* (rel. int.): 436.210 [M]$^+$ (1) ($C_{23}H_{32}O_8$), 421 [M − Me]$^+$ (0.5), 376 [M − HOAc]$^+$ (8), 334 [M − ketene]$^+$ (0.5), 320 [M − HO_2CR]$^+$ (2), 278 [M − ketene]$^+$ (6), 260 [M − HOAc]$^+$ (33), 218 [260 − C_2H_2O]$^+$ (100), 99 [$C_5H_{11}CO$]$^+$ (74), 70 [99 − CO]$^+$ (76).

Compound **8** (20 mg) in 1 ml $CHCl_3$ and 0.1 ml Ac_2O were heated for 1 hr with 50 mg 4-pyrrolidino pyridine at 60°. TLC (Et_2O–petrol, 1:1) afforded 15 mg **9**, colourless gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 1775 (γ-lactone), 1750, 1255, 1245 (OAc, CO_2R); MS *m/z* (rel. int.): 463 [M − Me]$^+$ (0.5), 419 [M − OAc]$^+$ (0.5), 376 [M − HOAc, ketene]$^+$ (12), 302.115 [M − HOAc, $HO_2CC_5H_{11}$]$^+$ (28) ($C_{17}H_{18}O_5$), 260 [302 − ketene] (80), 242 [302 − HOAc]$^+$ (86), 260 [M − C_2H_2O]$^+$ (82), 99 [$C_5H_{12}CO$]$^+$ (100), 71 [99 − CO]$^+$ (93); ^{13}C NMR ($CDCl_3$)(C-1

through C-15): 87.5 *s*, 29.5 *t*, 38.4 *t*, 82.4 *t*, 124.2 *d*, 153.8 *s*, 148.3 *s*, 65.9 *d*, 43.2 *t*, 83.7 *s*, 126.4 *s*, 167.0 *s*, 29.5 *q*; OCOR: 173.2 *s*, 33.9 *t*, 24.5 *t*, 31.3 *t*, 22.3 *t*, 13.8 *q* (some signals may be interchangeable).

$$[\alpha]^{\lambda}_{24^\circ} = \begin{array}{ccccc} 589 & 578 & 546 & 436 & 365\ \text{nm} \\ \hline +5.2 & +4.7 & +3.9 & -14.9 & -120.4 \end{array}$$

($CHCl_3$; *c*0.81).

Compound **7** (10 mg) in 2 ml Et_2O was stirred with 150 mg MnO_2 for 2 hr. TLC (Et_2O–petrol, 3:1) afforded 2 mg **10** and 5 mg unreacted **7**, colourless gum, ^1H NMR see Table 1.

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