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EUDESMANOLIDES AND OTHER CONSTITUENTS FROM *DIMEROSTEMMA ASPERATUM**

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Key Word Index—*Dimerostemma asperatum*; Compositae; sesquiterpene lactones; eudesmanolides; acetylenic esters; diterpene lactones.

Abstract—The investigation of *Dimerostemma asperatum* afforded four new eudesmanolides, two unusual acetylenic esters and two diterpene lactones related to geranylgeraniol. The structures were elucidated by spectroscopic methods and by some chemical transformations. The chemotaxonomic significance of these findings is discussed briefly.

INTRODUCTION

In the small Brazilian genus *Dimerostemma* (tribe Heliantheae, subtribe Verbesininae [1]) only one species has been investigated chemically so far [2]. In addition to widespread sesquiterpene hydrocarbons and simple derivatives, eudesmanolides were isolated. We now have studied the constituents of a further species, which not only contained similar sesquiterpene lactones, but also some other unusual constituents.

RESULTS AND DISCUSSION

The roots of *Dimerostemma asperatum* Blake afforded germacrene D, bicyclogermacrene, α-humulene, β-selinene, cyperene, the pentaynene **1** and two further acetylenic compounds, the unusual esters **2** and **3**, which could be separated by HPLC only.

The structures followed from the ^{1}H NMR data (Table 1) and the fragmentation pattern in the mass spectrometer. The base peak was fragment A (see scheme), the ion formed by loss of the acetylenic acyloxy radiçals. From the ^{1}H NMR spectra the configurations of the double bonds could be deduced. Structure **3** was an ester of lachnophyllum acid, so far only found as its methyl ester, while **2** was an ester of the rare dihydrolachnophyllum acid. C_{10}-acetylenic compounds are rare in the tribe Heliantheae, but widespread in the Anthemideae and Astereae [3].

The aerial parts also contained germacrene D and bicyclogermacrene. Furthermore, two diterpenes were isolated, the acid lactone **4** and the corresponding methyl ester **5**. Their structures followed from detailed ^{1}H NMR investigations (Table 2). Spin decoupling allowed the assignment of all signals. The configuration of the 14,15-double bond followed from the chemical shift of H-14, while those of 6,7- and 1,11 were deduced also from the chemical shifts of H-6 and H-10 as well as from those of the olefinic methyls, which all had the usual values for geranylgeraniol derivatives with *E* configuration. Due to the small amount of material the ^{13}C NMR spectrum of **5** could be interpreted in part only (see Experimental). The main signals, however, agreed with the proposed structure. **4** we have named dimeroperatic acid. The polar fractions further afforded a complex mixture of sesquiterpene lactones, three of which, **6**, **7** and **9**, had been isolated previously from *D. lippioides* [2]. The ^{1}H NMR data (Table 3) of the other lactones were all very similar indicating the same carbon skeleton and oxidation pattern. The spectrum of **8** clearly showed that the free hydroxyl was at C-8, which supported the proposed relative positions of the ester groups in **6** and **7**. The

Table 1. ^{1}H NMR data of **3** and **4** (270 MHz, $CDCl_3$, TMS as int. stand.)

	3	**4**
H-2	6.22 *d*	6.34 *d*
H-3	6.92 *dd*	6.81 *dt*
H-6	5.66 *brd*	—
H-7	6.31 *dt*	—
H-8	2.15 *brdt*	2.34 *dt*
H-9	1.45 *tq*	1.60 *tq*
H-10	0.93 *t*	1.02 *t*
H-1′	4.82 *brd*	
H-2′	6.22 *dt*	
H-3′	6.60 *d*	
H-5′, 9′	6.63 *s*	
O Me	3.88 *s* (6H)	
	3.85 *s*	

J(Hz): 2,3 = 2′,3′ = 16; 3,6 = 2 (**3**); 3,8 = 1 (**4**); 6,7 = 16 (**3**); compound **3**: 2,3 = 2′,3′ = 6,7 = 16; 3,6 = 2; 7,8 = 8,9 = 9,10 = 7; compound **4**: 2,3 = 2′,3′ = 16; 3,8 = 1; 8,9 = 7; 9,10 = 7.5.

* Part 336 in the series "Naturally Occurring Terpene Derivatives". For Part 335 see Bohlmann, F., Kramp, W., Robinson, H. and King, R. M. (1981) *Phytochemistry* **20** (in press).

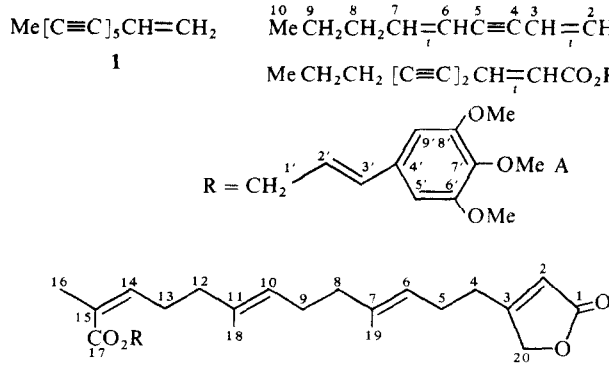
Table 2. ^{1}H NMR data of diterpene lactones **4** and **5** (400 MHz, $CDCl_3$, TMS as int. stand.)

	4	**5**
H-2	5.87 *tt*	5.85 *tt*
H-4	2.47 *brt*	2.46 *brt*
H-5	2.32 *brdt*	
H-6	5.11 *brt*	
H-8	2.06 *m*	
H-9		
H-10	5.10 *brt*	
H-12	2.06 *m*	
H-13	2.61 *brdt*	2.56 *brdt*
H-14	6.05 *tq*	5.92 *tq*
H-16	1.92 *brd*	1.89 *q*
H-18	1.60 *brs*	
H-19	1.62 *brs*	
H-20	4.74 *d*	4.73 *d*
OMe	—	3.73 *s*

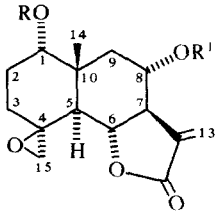
J (Hz): 2,4 = 2,20 = 1.5; 4,5 = 5,6 = 12,13 = 13,14 = 7.5; 14,17 = 1

spectrum of **11a/b** showed that only the ester residues at C-1 were different. These two compounds could not be separated even by HPLC. The ester residues were obviously enantiomeric 2-methyl-2,3-epoxy butyrates. The last lactone was an isomer of **8**. Only a few signals in the spectrum of **12a** differed slightly from those of **8**. Especially the chemical shifts of H-5 and H-15 were different and the *W*-coupling $J_{3,15}$ was missing. Inspection of models indicated that an α-epoxide in **12a** would explain these shifts as H-5 could be deshielded by the epoxide oxygen and H-15 by the lactone oxygen. The mixture of **11a** and **11b** was not acetylated at room temp. in the presence of 4-pyrrolidino pyridine. In boiling chloroform the rearranged acetates **13a** and **13b** were obtained, which, however, also could not be separated. The ^{1}H NMR signals of H-6 and H-8 showed that the 6,12-lactone was transformed to a 8,12-lactone (Table 3). The acetylation of **8** under the same conditions at room temp. gave **10**, while **12a** yielded the diacetate **12b**.

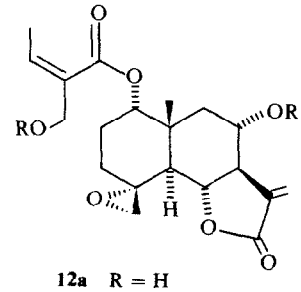
So far the constituents of *Dimerostemma* show a very uniform picture, epoxidized eudesmanolides being typical. The relevance of the acetylenes and the diterpenes so far is not clear. Similar compounds are absent from the other genera of the same subtribe that have been examined. More genera have to be studied to get a clearer phytochemical profile for this subtribe.



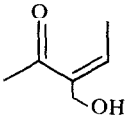
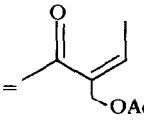
1 **2** **3** **4** R = H **5** R = Me

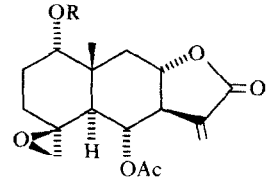


	6	**7**	**8**	**9**	**10**	**11a**†	**11b**†
R	5-OH	Ang†		Ang	5-OAc	Ang†	Epang
R′	Ang	Mebu	H	H	H	H	



12a R = H
12b R = Ac

* 5-OHAng =  ; 5-OAc Ang = 



† Enantiomeric 2-methyl-2,3-epoxybutyrate.

13a/b R = Epang†

Table 3. ^{1}H NMR data of sesquiterpene lactones **8**, **10**, **11a/b**, **12a/b** and **13a/b** (270 MHz, $CDCl_3$, TMS as int. stand.)

	8	**10**	**11a/b**		**12a**	**12b**	**13a/b**	
H-1	4.95 *dd*	4.89 *dd*	4.82 *dd*	4.85 *dd*	4.98 *brs*	4.88 *brs*	4.83 *dd*	4.87 *dd*
H-5	2.41 *d*	2.38 *d*	2.43 *d*	2.41 *d*	2.58 *d*	2.57 *d*	2.59 *d*	
H-6	3.86 *dd*	3.85 *dd*	3.86 *dd*		3.84 *dd*	3.90 *dd*	5.23 *dd*	
H-7	2.52 *dddd*	2.53 *dddd*	2.53 *dddd*	2.57 *dddd*	2.52 *dddd*	2.75 *dddd*	2.67 *dddd*	2.71 *dddd*
H-8	3.99 *ddd*	3.98 *ddd*	3.99 *ddd*		4.11 *ddd*	5.22 *ddd*	4.05 *ddd*	
H-9	2.00 *dd*	2.00 *dd*	} 1.90 *m*		} 1.8 *m*	1.83 *dd*	} 1.90 *m*	
H-9′	1.87 *dd*	1.87 *dd*				1.66 *dd*		
H-13	6.15 *dd*	6.15 *dd*	6.16 *dd*		6.16 *d*	6.13 *d*	6.11 *d*	
H-13′	5.97 *dd*	5.98 *dd*	5.98 *dd*		5.98 *d*	5.52 *d*	5.38 *d*	
H-14	1.13 *s*	1.13 *s*	1.11 *s*	1.10 *s*	1.22 *s*	1.28 *s*	1.16 *s*	1.15 *s*
H-15	3.22 *dd*	3.22 *d*	3.22 *dd*		3.43 *d*	3.42 *d*	2.96 *dd*	
H-15′	2.90 *d*	2.90 *d*	2.90 *d*		2.44 *d*	2.44 *d*	2.60 *d*	
OCOR	6.47 *brq*	6.58 *brq*	3.12 *q*	3.13 *q*	6.43 *brq*	6.57 *brq*	3.12 *q*	3.12 *q*
	4.34 *brs*	4.87 *brd*	1.35 *d*	1.37 *d*	4.35 *brd*	4.82 *brs*	1.36 *d*	1.39 *d*
		4.78 *brd*			4.31 *brd*			
	2.16 *brd*	2.18 *brd*	1.65 *s*	1.14 *s*	2.14 *brd*	2.17 *brd*	1.67 *s*	1.65 *s*
OH	4.12 *s*	4.11 *s*	4.12 *s*				—	—
OAc	—	2.06 *s*	—	—	—	2.08 *s*	2.13 *s*	
						2.06 *s*		

J (Hz): 1,2 = 2; 3,15 = 2; 5,6 = 6,7 = 10; 7,8 = 11; 7,13 = 3.2; 7,13 = 2.8; 8,9 = 11; 8,9′ = 3,5; 9,9′ = 12; 13,13′ = 0.7; 15,15′ = 3.5; Epang: 3′,4′ = 5; 5-OH Ang: 3′,4′ = 7.

EXPERIMENTAL

The air-dried plant material (voucher RMK 8230) was extracted with Et_2O–petrol, 1:2, and the resulting extracts were separated first by column chromatography (Si gel) and further by repeated TLC (Si gel) or HPLC (reversed phase, RP 18). Known compounds were identified by comparing the IR and ^{1}H NMR spectra with those of authentic material. The roots (300 g) afforded traces of **1**, 10 mg germacrene D, 5 mg bicyclogermacrene, 3 mg α-humulene, 5 mg β-selinene, 20 mg cyperene, 3 mg **2** and 5 mg **3** (separated by HPLC, MeOH–H_2O, 4:1). The aerial parts gave 250 mg germacrene D, 120 mg bicyclogermacrene, 2 mg **4**, 20 mg **5** (Et_2O–petrol, 3:1), 5 mg **6**, 7 mg **7**, 6 mg **8**, 2 mg **9**, 13 mg **11a** and **11b** (not separable) and 3 mg **13** (the lactones **6**, **7** and **9** were separated by repeated TLC using first Et_2O–petrol 4:1, then CH_2Cl_2–C_6H_6–Et_2O, 1:1:1, while for the others first Et_2O and then CH_2Cl_2–C_6H_6–Et_2O, 1:1:10 were used as solvents).

6,7-Dihydrolachnophyllum acid[*4-O-methyl siringenin*]*-ester* (**2**). Colourless gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 2245 (C≡C); 1725, 1620, 1590 (C=CCO_2R), 970 (*trans* CH=CH); MS: *m/z* (rel. int.) 370.178 (M^+, 22) ($C_{22}H_{26}O_5$), 223 (M − RCO$^\cdot$, 20), 207 (M − RCO$_2^\cdot$, 100).

Lachnophyllum acid-[*4-O-methylsiringenin*]*-ester* (**3**). Colourless gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 2200 (C≡C); 1720, 1620, 1590, (C=CCO), 970 (*trans* CH=CH); MS: *m/z* (rel. int.) 368.162 (M^+, 28), 207 (M − RCO$_2^\cdot$, 100).

Dimeroperatic acid (**4**). Colourless gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3330–2600, 1690, 1640 (C=C CO_2H), 1788, 1750 (lactone); MS: *m/z* (rel. int.) 332.199 (M^+, 3), 314 (M − H_2O, 21), 386 (314 − CO, 11), 98 ($C_5H_6O_2$, 100, McLafferty).

Methyl dimeroperatate (**5**). Colourless gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 1720, 1650 (C=CCO_2R), 1790, 1760 (lactone); MS: *m/z* (rel. int.) 346.214 (M^+, 4), 314 (M − MeOH, 18), 286 (314 − CO, 13), 149 ($C_{10}H_{13}O$, 100), 98 ($C_5H_6O_2$, 58); ^{13}C NMR ($CDCl_3$): C-2 115.7 *d*, C-6 122.1 *d*, C-10 124.6 *d*, C-14 145.7 *d*, C-16 20.5 *q*, C-18, 16.2 *q*, C-19 16.0 *q*, C-20 73.2 *t* (C-7 and C-11 137.5, 134.6; C-4, -5, -8, -9, -12, -13 39.1, 28.8, 28.3, 28.1, 26.5, 25.8).

Dimerostemmolide-1-O-[*5-hydroxy angelate*] (**8**). Colourless gum, IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1775 (γ-lactone), 1720 (C=CCO_2R); MS: *m/z* (rel. int.) 360.157 (M − H_2O, 1) ($C_{20}H_{24}O_6$), 262 (M − RCO_2H, 11), 244 (360 − RCO_2H, 31), 229 (244 − $^\cdot$Me, 16), 99 (RCO$^+$, 100), 81 (99 − H_2O, 95).

$$[\alpha]_{24°}^{\lambda} = \begin{array}{ccccc} 589 & 578 & 546 & 436 & 365\ nm \\ \hline -31 & -32 & -36 & -66 & -111 \end{array}$$

(c = 0.57, $CHCl_3$)

To 6 mg **8** in 1 ml $CHCl_3$ 10 mg 4-pyrrolidinopyridine [4] and 0.1 ml Ac_2O were added. After 20 hr at room temp. usual work-up and TLC (Et_2O) afforded 5 mg **10**, colourless gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 1790 (γ-lactone), 1750 (OAc), 1730 (C=CCO_2R); MS (CI, isobutane): *m/z* (rel. int.) 421 (M + 1, 60), 403 (421 − H_2O, 40), 343 (403 − AcOH, 11), 263 (421 − RCO_2H, 29) 245 (263 − H_2O, 71), 159 (RCOOH + 1), 100), 117 (159 − ketene, 91), 99 (159–AcOH, 73).

Dimerostemmolide-1-O-[*2-methyl-2,3-epoxy butyrate*] (**11a/b**). Inseparable colourless gum, IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3620 (OH), 1775 (γ-lactone), 1750 (CO_2R); MS: *m/z* (rel. int.) 378.168 (M^+, 0.5) ($C_{20}H_{26}O_7$), 360 (M − H_2O, 4), 262 (M − RCO_2H, 52), 244 (262 − H_2O, 81), 229 (244 − $^\cdot$Me, 83), 116 (RCO_2H, 100).

$$[\alpha]_{24°}^{\lambda} = \begin{array}{ccccc} 589 & 578 & 546 & 436 & 365\ nm \\ \hline -42 & -43 & -50 & -94 & -169 \end{array}$$

(c = 1.25, $CHCl_3$)

10 mg 4-pyrrolidinopyridine and 0.1 ml Ac_2O were added to 12 mg **11a/b**. After 20 hr no reaction was observed. After 3 hr reflux, usual work-up and TLC (Et_2O) afforded 4 mg unchanged **11a/b** and 4 mg **13a/b**, which could not be separated, colourless

gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 1790 (γ-lactone) 1760 (OAc), 1740 (CO_2R); MS (CI, isobutane) 421 (M + 1, 100) 391 (421 − CH_2O, 20), 361 (421 − AcOH, 8), 305 (421 − RCO_2H, 20), 245 (305 − AcOH, 25), 117 (RCO_2H + 1, 10).

4-*Iso-dimerostemmolide*-1-O-[5-*hydroxy angelate*] (**12a**). Colourless gum, IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3610 (OH), 1775 (γ-lactone), 1725 (C=C CO_2R): MS: *m/z* (rel. int.) 360.157 (M − H_2O, 0.5) ($C_{20}H_{24}O_6$), 262 (M − RCO_2H, 18), 244 (262 − H_2O, 14), 99 (RCO^+, 71) 55 (100).

$$[\alpha]_{24°}^{\lambda} = \frac{\begin{array}{ccccc} 589 & 578 & 546 & 436 & 365\ nm \end{array}}{\begin{array}{ccccc} +85 & +90 & +103 & +177 & +276 \end{array}}$$

(c = 0.23, $CHCl_3$)

2 mg **12a** were reacted as above at room temp. with 4-pyrrolidinopyridine. TLC afforded 2 mg **12b**, colourless gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 1790 (γ-lactone), 1750 (OAc), 1730 (C=CCO_2R); MS (CI, isobutane) *m/z* (rel. int.) 463 (M + 1, 6), 403 (463 − AcOH, 9), 343 (403 − AcOH, 4), 305 (463 − RCO_2H, 16), 245 (305 − AcOH, 52), 159 (RCO_2 H + 1, 100), 141 (159 − H_2O, 48), 117 (159 − ketene, 47), 99 (159 − AcOH, 46).

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