Journal of Natural Products
Vol. 58, No. 4, pp. 618-621, April 1995

AROMADENDRANE SESQUITERPENES FROM *PHEBALIUM*, *DRUMMONDITA*, AND *ERIOSTEMON* SPECIES

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ABSTRACT.—Two known aromadendrane sesquiterpenes, (+)-spathulenol and (+)-4β,10α-dihydroxyaromadendrane, have been isolated from the aerial parts of *Phebalium tuberculosum* ssp. *megaphyllum* and *P. filifolium*. (−)-Ledol, another sesquiterpene of the same class, was found only in *P. tuberculosum* ssp. *megaphyllum*. Examination of the aerial parts of *Eriostemon brucei* ssp. *brucei* showed it to contain (+)-13-hydroxyspathulenol [**1**], which is novel, while (+)-spathulenol has also been isolated from *Drummondita hassellii* and *D. calida*.

Phebalium, Eriostemon, and *Drummondita* are Australian genera belonging to the family Rutaceae. We recently reported the occurrence of a number of coumarins and triterpene esters from *P. tuberculosum* Benth. ssp. *megaphyllum* (Ewart) P.G. Wilson and *P. filifolium* Turcz. (1), alkaloids, flavonols, and coumarins from *D. hassellii* (F. Muell.) P.G. Wilson and *D. calida* (F. Muell.) P.G. Wilson (2), and alkaloids and some unusual polycyclic geranylcoumarins from *E. brucei* ssp. F. Muell. *brucei* (3,4). In this paper we wish to report the occurrence and distribution of aromadendrane-type sesquiterpenes in these species.

From the petroleum ether extract of *P. tuberculosum* ssp. *megaphyllum* three sesquiterpenes were isolated by a combination of vlc and prep. tlc. They were characterized as (+)-spathulenol (5–7), (+)-4β,10α-dihydroxyaromadendrane (7,8), and (−)-ledol (9,10), by comparison of their physical and spectroscopic data with previously reported values. Similar purification of the petroleum ether extract of the aerial parts of *E. brucei* yielded a novel sesquiterpene [**1**] that was identified by spectroscopic methods as well as by comparison with related compounds.

Compound **1** was isolated as a colorless gum which analyzed for $C_{15}H_{24}O_2$ by hreims. The ^{13}C-nmr spectrum confirmed the presence of 15 carbons, while an HMQC (11) experiment indicated that 22 of the 24 protons in **4** were attached to carbons (Table 1). A broad ir absorption band at 3450 cm^{-1} suggested the presence of one or more hydroxyl substituents. Both the ^{1}H- and ^{13}C-nmr spectra of **1** (Table 1) revealed close correspondence with those of spathulenol and other compounds with the aromadendrane carbon skeleton (6, 7, 12–14). However, one of the methyl groups in spathulenol was replaced by a hydroxymethyl group (δ_C 73.2) with two protons resonating as an AB quartet (J=10.8 Hz) at δ 3.35 and 3.38. This suggested that **1** was a hydroxymethyl analogue of spathulenol.

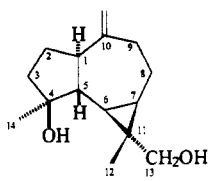

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TABLE 1. Nmr Spectral Data for (+)-13-Hydroxyspathulenol [**4**] in $CDCl_3$.

Position	δ ^{13}C	δ ^{1}H (multiplicity, J [Hz])	COSY[a]	HMBC[b]
1	52.7 d	2.24 dt (6.5, 9.8)	H-2α, H-2β, H-5	C-2
2α	26.3 t	1.65 m	H-1, H-3α, H-3β	
2β		1.90 m	H-1, H-3α, H-3β	
3α	41.7 t	1.60 m	H-2α, H-2β	
3β		1.78 m	H-2α, H-2β	
4	80.9 s			
5	53.1 d	1.36 t (9.8)	H-1, H-6	C-2
6	26.3 d	0.65 dd (11.2, 9.8)	H-5, H-7	
7	24.3 d	0.88 m	H-6, H-8α, H-8β	
8α	24.4 t	1.99 m	H-7, H-9α, H-9β	
8β		1.05 m	H-7, H-9α	
9α	38.6 t	2.06 br t (13.2)	H-8α, H-8β	
9β		2.44 br dd (13.2, 6.2)	H-8α	C-1, C-8, C-10
10	153.1 s			
11	27.1 s			
12	12.0 q	1.15 s		C-6, C-7, C-11, C-13
13a	73.2 t	3.35 ABq (10.8)		C-6, C-7, C-11, C-12
13b		3.38 ABq (10.8)		
14	25.8 q	1.30 s		C-3, C-4, C-5
15	106.7 t	4.69 m		C-1, C-9, C-10
		4.42 m		

[a]Major ^{1}H-^{1}H correlations observed in a COSY-45 experiment (geminal coupling not shown).
[b]Major ^{1}H-^{13}C correlations determined from an HMBC experiment.

It was possible to trace all of the proton spin systems in **1** with a COSY-45 experiment (Table 1). Proton-detected heteronuclear experiments (HMQC and HMBC) allowed complete assignment of the ^{1}H- and ^{13}C-nmr resonances of **1**. HMBC (15) correlations from the H-12 protons to C-6, C-7, C-11, and C-13 established the hydroxymethyl substituent at C-13 of the aromadendrane nucleus. The relative stereochemistry of the sesquiterpene alcohol was determined by a NOESY experiment, in which the cyclopropane ring protons exhibited strong interactions with the hydroxymethylene protons, indicating their close proximity (Figure 1). On this basis, the structure of compound **1** was established as (+)-13-hydroxyspathulenol. A closely related compound, 8α,13-dihydroxyspathulenol, has been isolated from *Cineraria fruitculorum* (16).

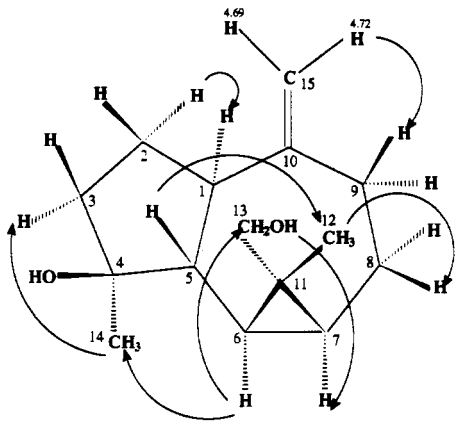

FIGURE 1. Key nOe interactions for **1** observed in a NOESY nmr spectrum.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Mps (uncorrected) were determined with a Reichart sub-stage microscope. The ir spectra were recorded on a Perkin-Elmer 552 spectrophotometer. The ms were obtained on an AEI MS 902 spectrometer. Optical rotations were measured with a Perkin-Elmer 241 polarimeter. Nmr spectra were recorded on a Bruker AMX-400 instrument. The chemical shifts are reported in ppm relative to the residual undeuterated solvent. Inverse detected heteronuclear correlations were measured using HMQC (optimized for $^{1}J_{CH}$=140 Hz) and HMBC

(optimized for $^{n}J_{CH}$=8.3 Hz) pulse sequences. Petroleum ether refers to the bp 60–80° fraction. The following Si gels were used: Si gel (Merck 7749) for vlc and Si gel 60-PF_{254} for tlc.

PLANT MATERIAL.—The aerial parts of *Phebalium tuberculosum* ssp. *megaphyllum* (voucher: Perth 01185373), *Phebalium filifolium* (voucher: Perth 01185365), *Eriostemon brucei* ssp. *brucei* (voucher: Perth 01194356), *Drummondita hassellii* (voucher: Perth 01156713), and *Drummondita calida* (voucher: Perth 01012266) were collected from Western Australia. Voucher specimens are deposited at the Western Australian Herbarium, Perth, Australia.

EXTRACTION AND ISOLATION.—Powdered aerial parts of *P. tuberculosum* ssp. *megaphyllum* (250 g), *P. filifolium* (200 g), *E. brucei* ssp. *brucei* (460 g), *D. hassellii* (500 g), and *D. calida* (350 g) were extracted separately in a Soxhlet apparatus with, successively, petroleum ether (b.p. 60–80°), EtOAc, and MeOH. The concentrated petroleum ether extracts were subjected to vlc, eluting with petroleum ether containing increasing amounts of EtOAc and then with EtOAc/MeOH combinations. Fractions obtained with 10–15% EtOAc in petroleum ether, upon repeated prep. tlc using the solvent toluene-EtOAc (80:10) afforded the sesquiterpenes, spathulenol (75.0 mg) and ledol (5.0 mg) from *P. tuberculosum* ssp. *megaphyllum*, and spathulenol (27.0 mg) and 4β,10α-dihydroxyaromadendrane (2.5 mg) from *P. filifolium*. Fractions obtained with 35–40% EtOAc in petroleum ether upon similar tlc purification using toluene-EtOAc (80:20) gave 2.3 and 2.5 mg of 4β,10α-dihydroxyaromadendrane from *P. tuberculosum* and *P. filifolium*, respectively. Similar treatment of the fractions obtained from 10–12% EtOAc in petroleum ether yielded spathulenol (41.0 mg) from *D. hassellii* and (10.0 mg) from *D. calida*. Compound **1** (7.9 mg) was isolated from *E. brucei* ssp. *brucei* by prep. tlc of the vlc fraction obtained with 5% MeOH in EtOAc, using $CHCl_3$-MeOH (97:3) as the developing solvent.

(+)-*Spathulenol*.—Colorless gum; [α]D +15.6° (*c*=0.1, $CHCl_3$) [lit. (5) +56°]; hreims *m/z* $[M]^+$ 220.1844 (calcd 220.1827 for $C_{15}H_{24}O$); ir, ^{1}H-nmr, ^{13}C-nmr, and eims data in agreement with literature values (6,7).

(−)-*4β,10α-Dihydroxyaromadendrane*.—Colorless plates from *n*-hexane/EtOAc, mp 129–131° [lit. (8) 132°]; [α]D −12.4° (*c*=0.2, $CHCl_3$) [lit. (8) −25°]; hreims *m/z* $[M]^+$ 238.1939 (calcd 238.1933 for $C_{15}H_{26}O_2$); ir, ^{1}H-nmr, and eims data in agreement with literature values (7,8).

(−)-*Ledol*.—Fine white needles from *n*-hexane/EtOAc, mp 86–88° [lit. (10) 103–104°]; [α]D −7.2° (*c*=0.2, $CHCl_3$) [lit. (11) −5.6°]; hreims *m/z* $[M]^+$ 222.1996 (calcd 222.1983 for $C_{15}H_{26}O$), $[C_7H_{11}]^+$ (22); ir, ^{1}H-nmr, ^{13}C-nmr, and eims data in agreement with literature values (9,10).

(+)-*13-Hydroxyspathulenol* [**1**].—Colorless gum; [α]D +5.5° (*c*=0.7, $CHCl_3$); ir ν max (film) 3450, 3150, 2920, 1630, 1450, 1380 cm^{-1}; ^{1}H- and ^{13}C-nmr data, see Table 1; hreims *m/z* $[M]^+$ 236.1764 (calcd 236.1776 for $C_{15}H_{24}O_2$) (32), 218 $[M-H_2O]^+$ (91), 203 [*m/z* 218−CH_3]$^+$ (51), 188 [203−CH_3]$^+$ (28), 178 [M−$C(CH_3)CH_2OH$]$^+$ (19), 160 [*m/z* 218−$C(CH_3)CH_2OH$]$^+$ (90), 145 [*m/z* 160−CH_3]$^+$ (85), 105 [*m/z* 160−C_4H_7]$^+$ (100), 55 $[C_4H_7]^+$ (44).

ACKNOWLEDGMENTS

M.A. Rashid thanks the Association of Commonwealth Universities, UK for the award of a scholarship. Nmr spectra were obtained from the Strathclyde University Nmr laboratory. The authors thank the Western Australian Herbarium, Perth, Australia, for making the plant collections used in this study.

LITERATURE CITED

1. M.A. Rashid, A.I. Gray, P.G. Waterman, and J.A. Armstrong, *J. Nat. Prod.*, **55**, 851 (1992).
2. M.A. Rashid, J.A. Armstrong, A.I. Gray, and P.G. Waterman, *Phytochemistry*, **31**, 1265 (1992).
3. M.A. Rashid, J.A. Armstrong, A.I. Gray, and P.G. Waterman, *Nat. Prod. Lett.*, **1**, 79 (1992).
4. M.A. Rashid, J.A. Armstrong, A.I. Gray, and P.G. Waterman, *Phytochemistry*, **31**, 3583 (1992).
5. S.M. Juell, R. Hansen, and H. Jork, *Arch. Pharm.*, **309**, 458 (1976).
6. F. Inagaki and A. Abe, *J. Chem. Soc., Perkin Trans. II*, 1773 (1985).
7. H. Iwabuchi, M. Yoshikara, and W. Kamisaki, *Chem. Pharm. Bull.*, **37**, 509 (1989).
8. F. Bohlmann, M. Grenz, J. Jakupovic, and R.M. Robinson, *Phytochemistry*, **22**, 1213 (1983).
9. S.C. Pakrashi, P.P. Ghosh Dastidar, S. Chakrabarty, and B. Achari, *J. Org. Chem.*, **45**, 4765 (1980).
10. H.J.M. Gijsen, J.B.P.A. Wijnberg, G.A. Stork, A. de Groot, M.A. de Waard, and J.G.M. van Nistelrooy, *Tetrahedron*, **48**, 2465 (1992).
11. M.F. Summers, L.G. Marzilli, and A. Bax, *J. Am. Chem. Soc.*, **108**, 199 (1986).
12. J. Streith, P. Pesnelle, and G. Ourisson, *Bull. Chim. Soc. Fr.*, 518 (1963).

13. A.S. Feliciano, M. Medarde, M. Gordaliza, E.D. Olmo, and J.M.M.D. Corral, *Phytochemistry,* **28**, 2117 (1989).
14. G.M. Cabrera, J.C. Oberti, E.G. Gros, and A.M. Seldes, *Phytochemistry,* **34**, 1642 (1993).
15. A. Bax and M.F. Summers, *J. Am. Chem. Soc.*, **108**, 2093 (1986).
16. F. Bohlmann, P. Sing, and J. Jakupovic, *Phytochemistry,* **21**, 2531 (1982).

Received 12 October 1994