Phytochemistry, Vol. 26, No. 1, pp. 195 198, 1987.
Printed in Great Britain

0031 9422/87 $3.00 + 0.00
Pergamon Journals Ltd.

SESQUITERPENE LACTONES FROM *PERITYLE VASEYI*

Richard M. Pfeil, Douglas A. Gage*, Esther F. Lee*, Mahmut Miski*, Tom J. Mabry* and A. Michael Powell

Department of Biology, Sul Ross State University, Alpine, TX 79832, U.S.A.; * Department of Botany, University of Texas at Austin, Austin, TX 78713, U.S.A.

(*Revised received* 8 *November* 1985)

Key Word Index —*Perityle vaseyi*; Asteraceae; chemotaxonomy; sesquiterpene lactones; germacrolides; guaianolide; isorhamnetin.

Abstract The well-known germacrolide, eupatoriopicrin, and two additional sesquiterpene lactones, were isolated from *Perityle vaseyi*. The latter two lactones, one a novel guaianolide and the other a known germacrolide, both contain unusual ten carbon diester side chains attached at C-8. The chemical evidence, in conjunction with morphological data, suggest that *Perityle*, and hence the subtribe Peritylinae, should be placed in the tribe Heliantheae.

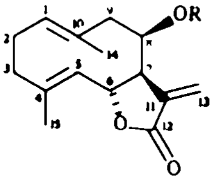
INTRODUCTION

The genus *Perityle* contains approximately 53 species distributed principally in the southwestern United States and northern Mexico, with a single disjunct species in South America [1-6]. Traditionally included in the artificial tribe Helenieae [6], *Perityle* and the subtribe Peritylinae have been placed by later authors in both the Senecioneae [7] and the Heliantheae [8]. Because little is known of the chemistry of this genus [9, 10], we have examined the chemical constituents of *P. vaseyi* Coult. to provide additional data which might be applied to the problem of tribal placement.

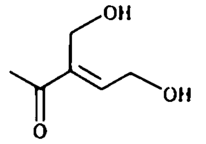
RESULTS AND DISCUSSION

The dichloromethane extract of leaves† of *P. vaseyi* afforded the known germacrolide eupatoriopicrin (**1**) [12, 13] as the major constituent. The second compound (**2**) differed from the first in the nature of the ester side chain at C-8. ^{1}H NMR and mass spectral evidence indicated that in compound **2** the dihydroxytigloyl function found in **1** was further esterified at the 5′-position with a 5′-hydroxytigloyl group. Thus, this compound is the 4′-hydroxy-5′-[5″-hydroxytigloyloxy]-tiglate analogue of eupatoriopicrin which was recently reported under the name 4′-hydroxyliacylindrolide from *Eupatoriastrum nelsonii* [14]. The ^{13}C NMR spectrum of compound **2** (Table 2) displayed signals for the side chain in close agreement with those of provincialin [15], a heliangolide containing the same C_{10} diester function. The remaining resonances were coincident with the skeletal signals reported for both 8β-[5′-hydroxytigloyloxy]costunolide [16] and 2″Z-4′-hydroxyliacylindrolide [14].

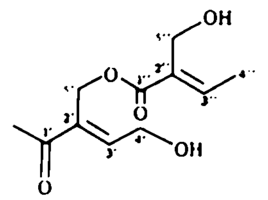
The formula of the new compound (**3**), $C_{25}H_{30}O_9$ (CIMS), suggested that it also contained a C_{10} diester side chain. In the ^{1}H NMR spectrum of compound **3** several signals were nearly identical to those in the spectrum of compound **2** [14]. These were namely a triplet at δ7.12 (H-3′), the AB pair centred at δ4.88 (H-5′a and H-5′b) and a broadened two proton signal at δ4.50 (seen as the AB

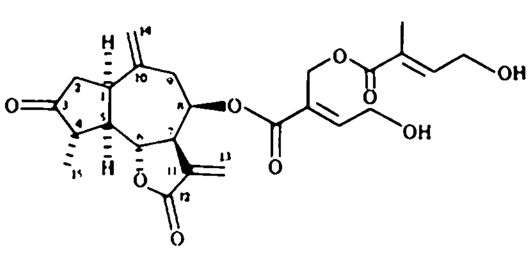


1 R =



2 R =





3

---

†One of us reported [11] that the lactone constituents of *P. vaseyi* are localized in glandular trichomes found on the surface of the leaves and floral parts.

Table 1. ^{1}H NMR spectral data of compound **3** (TMS as internal standard)

H	CDCl$_3$ 360 MHz	Me$_2$CO-d_6 200 MHz
1	3.10 *ddd*	3.32 *ddd*
2	2.62 *dd*	2.68 *dd*
2β	2.54 *dd*	2.42 *dd*
4	2.39 *br dq**	2.33 *br dq*
5	2.32 *ddd*	2.48 *ddd*
6	4.52 *t*	4.55 *t*
7	3.33 *dddd*	3.67 *dddd*
8	5.79 *m*	5.81 *m*
9a	2.70 *dd*	2.77 *dd*
9b	2.52 *dd*	2.69 *dd*
13a	6.36 *d*	6.21 *d*
13b	5.71 *d*	5.70 *d*
14a	4.86 *br s*	4.81 *br s*
14b	5.01 *br s*	5.02 *br s*
15	1.28 *d* (3H)	1.21 *d*
3'	7.12 *t*	7.10 *t*
4'ab	4.52† (2H)	4.46 *br d*
5'a	4.93 *d*	4.91 *d*
5'b	4.83 *d*	4.83 *d*
3"	6.64 *br t*	6.71 *br t*
4"ab	4.31† (2H)	4.28 *br d*
5"	1.77 *br s* (3H)	1.75 *br s*

J (Hz): 1, 2α = 1, 5 = 9.5; 1, 2β = 3; 2α, 2β = 19; 4, 5 = 10.5; 4, 15 = 6.5; 5, 6 = 6, 7 = 10; 7, 8 = 2; 7, 13a = 3.8; 7, 13b = 3.1; 8, 9a = 3.5; 8, 9b = 4; 9a, 9b = 14.4; 3', 4' = 6; 5'a, 5'b = 11.5; 3", 4" = 6; 3", 5" = 1.9; 4", 5" = 1.

*Signal partially overlapped, multiplicity determined by spin decoupling.

†Centre of AB part of ABX pattern.

Table 2. ^{13}C NMR spectral data* of compounds **2** and **3** (22.6 MHz, TMS as internal standard)

C	**2** CDCl$_3$	**3** CDCl$_3$	Me$_2$CO-d_6
1	127.5 *d*	39.8 *d*	40.2
2	26.3 *t*	43.9† *t*	44.2†
3	39.5 *t*	219.5 *s*	219.1
4	142.8 *s*	47.1§ *d*	47.5‖
5	131.1 *d*	50.1 *d*	50.7
6	75.7‡ *d*	82.7 *d*	83.2
7	52.8 *d*	47.1§ *d*	47.5‖
8	72.6 *d*	67.9 *d*	69.2
9	44.1 *t*	44.3† *t*	44.5†
10	134.2 *s*	143.6 *s*	145.6
11	136.8 *s*	134.5 *s*	136.6
12	169.8 *s*	169.9 *s*	169.7
13	121.2 *t*	123.4 *t*	122.1
14	19.1 *q*	116.3 *t*	115.9
15	17.6 *q*	14.2 *q*	14.4
1'	165.2 *s*	165.2 *s*	165.7
2'	127.1 *s*	126.7 *s*	127.4¶
3'	148.1 *d*	148.9 *d*	150.1
4'	59.5 *t*	59.5 *t*	59.6
5'	58.3 *t*	58.1 *t*	58.7
1"	167.4 *s*	167.4 *s*	167.6
2"	131.6 *s*	127.3 *s*	127.4¶
3"	142.6 *d*	142.3 *d*	143.6
4"	14.4 *q*	59.2 *t*	59.3
5"	56.6 *t*	12.5 *q*	12.6

*Assigned by analogy to known compounds [37, 38].

†Interchangeable within a column.

‡Overlapping solvent signal.

§‖¶ Overlapping signals.

part of an ABX pattern at 360 MHz), which indicated that a 5'-esterified dihydroxytigloyl residue was also present in compound **3**. However, the quartet at δ6.91 in the spectrum of **2** was replaced by a broadened triplet at 6.64 (*J* = 6.0, 1.9 Hz), which was long range coupled to a methyl doublet at 1.77 (*J* = 1.9 Hz).

This, together with the absence of the C-4" methyl doublet found in **3**, suggested that the side chain in **1** terminated with either the isomeric 4"-hydroxyangelate (*Z* isomer) or 4"-hydroxytiglate (*E* isomer) [18], rather than 5"-hydroxytiglate as in **3**. Curiously, both of the 4'-hydroxy isomers (*Z* and *E*) are reported to exhibit nearly identical ^{1}H and ^{13}C NMR signals [17, 18]. The CI mass spectrum supported the structure of the double ester by displaying prominent peaks at *m/z* 359, corresponding to the loss of hydroxytiglic (or hydroxyangelic acid) and *m/z* 245, for the loss of the entire diester side chain. Also, the base peak at *m/z* 99 provided additional evidence that the hydroxytigloyl (or hydroxyangeloyl) ester was in the terminal position of the diester side chain. The ^{13}C NMR data supported the presence of this novel diester with signals for C-1',C-5' in close accord with the analogous signals in **2**, and an additional five resonances in agreement with either those of a 4-hydroxyangelate or a 4-hydroxytiglate residue [16].

NOE difference experiments were conducted to establish the configuration of the terminal ester. Initial NOE experiments indicated the *Z* configuration of the double bond (i.e. 4"-hydroxyangelate) in terminal ester by demonstrating a weak enhancement of the vinyl methyl signal (H-5") when the H-3" triplet was irradiated, and no NOE between the methyl and the hydroxymethylene signal (H-4"a and H-4"b, AB part of ABX pattern centred at δ4.31). Further experiments using different parameters (see Experimental) did demonstrate that a weak H-3"–4" NOE was present, suggesting that the terminal ester was indeed the 4"-hydroxytiglate. This conflict was finally resolved by comparison of difference NOE. Studies were also conducted with the guaianolide graminiliatrin*, which has a 4'-acetoxyangeloyloxy ester [19]. The results with both difference and 2-D NOE experiments using this model compound were clear. Strong reciprocal NOEs were observed between H-3' and the *cis* H-5' methyl, but not between the H-5' methyl and the H-4' acetoxymethylene. In comparison to these results, the interaction between H-3" and the H-5" methyl group in **1** was insignificant. Additional evidence also suggested that the esters in **1** and graminiliatrin differed in the configuration of the 2',3'-double bond. The chemical shifts of H-3' in graminiliatrin (δ6.03) and H-3" in **1** (δ6.64) were rather

---

*Kindly provided by Prof. W. Herz.

different as predicted by comparison of model esters; in the corresponding (4-desoxy) tigloyloxy and angeloyloxy esters H-3′ is found at *ca* δ6.8 and 6.2, respectively. On the basis of these observations, the terminal ester in **1** is the *E* isomer, 4″-hydroxytiglate. We are unable to account for a previous report [17] of a '4′-hydroxyangelate' with similar chemical shift values to our 4″-hydroxytiglate.

Compound **3** was an α-methylene-γ-lactone (^{1}H NMR doublets at δ6.36 and 5.71; IR absorbance at 1750 cm^{-1}; and ^{13}C NMR resonances at δ169.9 *s*, 123.4 *t* and 134.5 *s*), but the skeleton was clearly unlike that of **2**. One notable difference in the ^{13}C NMR spectrum of **3** was the singlet at δ219.5, indicative of a cyclopentanone moiety. Also, the triplet at δ116.3 and singlet at 143.6, in conjunction with the two broadened singlets in the ^{1}H NMR at δ5.01 and 4.86, suggested that an exocyclic methylene function was present in **3** in place of a vinyl methyl group. In addition, a methyl doublet (δ1.28) replaced the other olefinic methyl found in **2**. Spin decoupling experiments, as well as correlation of the ^{1}H and ^{13}C NMR spectra with known compounds, established that **3** was an 8β-acyloxy analogue of 3-dehydro-4β,15-dihydrozaluzanin C [18 25]. The configuration of the C-4 methyl group was confirmed by NOE difference spectroscopy as α, since irradiation of the H-15 doublet increased the intensity of the H-5 signal, and to a lesser extent those of H-1 and H-7. Thus, compound **3** is 8β-4′-hydroxy-5′-[4″-hydroxytigloyloxy]-tigloyloxy-3-dehydro-4β,15-dihydrozaluzanin C.

From the methanol extract of *P. vaseyi* we isolated the known flavonoid isorhamnetin [26, 27]. A previous study of this species afforded penduletin 4′-*O*-methyl ether [10].

The sesquiterpene lactones found in *P. vaseyi* are not characteristic of the tribe Senecioneae, but they do indicate a relationship to the Heliantheae. Previously, eupatoriopicrin (**1**) was isolated from several genera now in the tribe Heliantheae that were formerly placed in the Helenieae [12, 28]. The diester compounds **2** and **3**, on the other hand, appear to be more restricted to some members of the tribe Eupatorieae [14, 29 33], where **1** has also been isolated [12, 16]. Recently however, a similar C_{10} diester guaianolide was isolated from *Hymenopappus newberryi* (Gray) I. M. Johnston [34], a species long thought to belong in the Anthemideae now included in the Heliantheae [34]. Although *Perityle* is not allied to the Eupatorieae on morphological grounds, this line of evidence does support its placement in the Heliantheae. SEM studies [11] have shown that the ligule surface is papilloid in several members of the Peritylinae, including *P. vaseyi*, *P. emoryi*, *Amauria rotundifolia* Benth. and *A. carterae* Powell, consistent with the placement of this subtribe in the Heliantheae, rather than the Senecioneae [35].

EXPERIMENTAL

Plant material. Leaves of *P. vaseyi* (400 g) were collected *ca* 70 miles S of Alpine, Texas (Brewster Co.) on Hwy. 118 in the summer of 1983. Voucher specimens (A. M. Powell and S. Powell no. 4117 and R. M. Pfeil no. 56) are deposited in the Sul Ross University Herbarium.

Extraction and isolation of compounds. The air dried unground leaves were extracted overnight in CH_2Cl_2 and worked up in the usual manner [36]. The crude extract was then taken up in CH_2Cl_2 and partitioned against a 5% aq. Na_2CO_3 soln. The CH_2Cl_2 layer was then extracted with distilled H_2O, dried with $MgSO_4$ and concd to dryness. Redissolved in 70% aq. MeOH, the purified CH_2Cl_2 extract was injected in aliquots through Sep-pak cartridges (Waters). The eluant was filtered through a 0.45 μm membrane filter prior to separation by HPLC (RP-18, semi-prep. 25 cm × 10 mm ID, 3 ml/min). Eupatoriopicrin (1 g, **1**), R_t 0.6 min, was isolated from two additional peaks, R_t 3.8 min and R_t 7.2 min with 70% aq. MeOH. The more rapidly eluting peak was rechromatographed in 40% aq. MeCN to yield **2** (100 mg, R_t 14.8 min). The second peak (R_t 7.2 min in 70% aq. MeOH) was rechromatographed using a H_2O MeCN–MeOH (55:15:30) mobile phase to give **3** (200 mg, R_t 12 min). The flavonoid isorhamnetin (200 mg) was isolated by standard procedures [26] and identified by its mass, UV and ^{1}H NMR spectra [26, 27].

8β-4′-Hydroxy-5′-[4″-hydroxytigloyloxy]-tigloyloxy-3-dehydro-4β,15-dihydrozaluzanin C (**3**). Colourless gum; IR ν_{max}^{KBr} cm^{-1}: 3420, 1750, 1725, 1705, 1640, 1238. CIMS (CH_4, 0.5 torr; direct probe) *m/z* (rel. int.): 475 $[M+1]^+$ (5.3), 457 $[M+1-H_2O]^+$ (9.9), 359 $[M+1-C_5H_7O_3+H]^+$ (12.9), 263 (10.1), 245 $[M+1-C_{10}H_{13}O_6+H]^+$ (9.6), 117 (55.9), 99 [hydroxytigloyl acylium]$^+$ (100), 71 [acylium (99) − CO]$^+$ (15.9).

Acknowledgements—This work was supported by the Chihuahuan Desert Research Funds granted to Sul Ross University by the State of Texas. Financial support at the University of Texas was provided by the National Science Foundation (Grant BSR-8402017) and Robert A. Welch Foundation (Grant F-130). We wish to thank Dr. Ben Shoulders for high field ^{1}H NMR and ^{13}C NMR spectra, Steve Sorey for running the NOE experiment and Prof. B. L. Turner for helpful comments on the manuscript.

REFERENCES

1. Shinners, L. H. (1959) *Southwest. Nat.* **4**, 204.
2. Niles, W. E. (1970) *Mem. N.Y. Bot. Gard.* **21**, 1.
3. Powell, A. M. (1969) *Rhodora* **71**, 785.
4. Powell, A. M. (1973) *Sida* **5**, 61.
5. Powell, A. M. (1974) *Rhodora* **76**, 806.
6. Turner, B. L. and Powell, A. M. (1977) in *The Biology and Chemistry of the Compositae* (Heywood, V. H., Harborne, J. B. and Turner, B. L., eds) p. 699. Academic Press, London.
7. Powell, A. M. and Turner, B. L. (1974) *Am. J. Botany* **61**, 87.
8. Robinson, H. (1981) *Smithson. Contrib. Botany* **51**, 1.
9. Powell, A. M. and Tsang, H. S. (1966) *Southwest. Nat.* **11**, 190.
10. Southwick, L., Mabry, T. J., Averett, J. and Powell, A. M. (1972) *Phytochemistry* **11**, 2351.
11. Pfeil, R. M. (1984) Masters Thesis. Sul Ross State University.
12. Dolejs, M. and Herout, V. (1962) *Coll. Czech. Chem. Commun.* **27**, 2654.
13. Herz, W. and Govindan, S. V. (1980) *Phytochemistry* **19**, 1234.
14. Bohlmann, F., Zdero, C. and Turner, B. L. (1985) *Phytochemistry* **24**, 1263.
15. Melek, F. R., Gage, D. A. and Mabry, T. J. (1985) *J. Nat. Prod.* **48**, 331.
16. Herz, W., de Groote, R., Murari, R. and Kumar, N. (1979) *J. Org. Chem.* **44**, 2785.
17. Ito, K., Sakakibara, Y., Haruna, M. and Lee, K. (1979) *Chem. Letters* 1469.
18. Bohlmann, F. and Fiedler, L. (1978) *Chem. Ber.* **111**, 408.
19. Herz, W., Poplawski, J. and Sharma, R. P. (1975) *J. Org. Chem.* **40**, 199.
20. Vichnewski, W., Welbaneide, L., Machado, F., Rabi, J. A., Murari, R. and Herz, W. (1977) *J. Org. Chem.* **42**, 3910.

21. Herz, W., Kumar, N., Vichnewski, W. and Blout, J. F. (1980) *J. Org. Chem.* **45**, 2503.
22. Bohlmann, F. and LeVan, N. (1977) *Phytochemistry* **16**, 487.
23. Halim, A. F., Zaghloul, A. M. and Bohlmann, F. (1980) *Phytochemistry* **19**, 2767.
24. Rustaiyan, A., Niknejad, A., Zdero, C. and Bohlmann, F. (1981) *Phytochemistry* **20**, 2427.
25. Gershenzon, J. and Mabry, T. J. (1984) *Phytochemistry* **23**, 1959.
26. Wilkins, C. K. and Bohm, B. A. (1976) *Can. J. Botany* **54**, 2177.
27. Mabry, T. J., Markham, K. R. and Thomas, M. B. (1970) *The Systematic Identification of Flavonoids*. Springer, New York.
28. Geissman, T. A. and Atala, S. (1971) *Phytochemistry* **10**, 1075.
29. Bohlmann, F., Ahmed, M., King, R. M. and Robinson, H. (1981) *Phytochemistry* **20**, 2027.
30. Bohlmann, F. and Datta, L. N. (1979) *Phytochemistry* **18**, 847.
31. Herz, W. and Wahlberg, I. (1973) *J. Org. Chem.* **38**, 2485.
32. Herz, W. and Kumar, N. (1980) *Phytochemistry* **19**, 2387.
33. Bohlmann, F., Banerjee, S., Wolfrum, C., Jakupovic, J., King, R. M. and Robinson, H. (1985) *Phytochemistry* **24**, 1319.
34. Bohlmann, F., Zdero, C. and Turner, B. L. (1984) *Phytochemistry* **23**, 1055.
35. Baagoe, J. (1977) in *The Biology and Chemistry of the Compositae* (Heywood, V. H., Harborne, J. B. and Turner, B. L., eds) p. 119. Academic Press, New York.
36. Mabry, T. J., Miller, M. E., Kagan, H. B. and Renold, W. (1966) *Tetrahedron* **22**, 1139.
37. da Silva, A. J. R., Garcia, M., Baker, P. M. and Rabi, J. A. (1981) *Org. Magn. Reson.* **16**, 230.
38. Ahmed, A. A., Whittemore, A. T. and Mabry, T. J., *J. Nat. Prod.* (in press).