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HELIANGOLIDES, KAURANES AND OTHER CONSTITUENTS OF *HELIANTHUS HETEROPHYLLUS*

WERNER HERZ and MAURIZIO BRUNO

Department of Chemistry, The Florida State University, Tallahassee, FL 32306-3006, U.S.A.

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Key Word Index—*Helianthus heterophyllus*; Compositae; Heliantheae; heliangolides; sesquiterpene lactones; kauranes; diterpenes; norsesquiterpenes; 8-methoxyobliquin; coumarin.

Abstract—The chloroform extract of the aerial parts of *Helianthus heterophyllus* afforded the heliangolides 2′,3′-dihydroleptocarpin and 2′,3′-dihydroniveusin C and the new coumarin (−)-8-methoxyobliquin. Other constituents were various *ent*-kaurenoic acids, (+)-vomilifoliol, dehydrovomilifoliol, 9-hydroxy-4-megastigmen-3-one, 9,12,13-trihydroxy-octadeca-10(*E*),15(2*Z*)-dienoic and 9,12,13-trihydroxyoctadec-10(*E*)-enoic acid, 2,6-dimethoxybenzoquinone and vanillin.

INTRODUCTION

Since our most recent article on constituents of *Helianthus* species [1], a spate of reports on this subject, largely by Mabry and coworkers, has appeared [2–15]. We now describe the results of our study of *Helianthus heterophyllus* Nutt. A report from another source [16] has dealt with the flavonoids of *Helianthus* sect. Divaricati, ser. Angustifolii which includes *H. heterophyllus* [17].

RESULTS AND DISCUSSION

Like many other *Helianthus* species, *H. heterophyllus* furnished several *ent*-kaurenoic acids, specifically **1a**–**4a**, which were identified in the form of their methyl esters. Small amounts of two heliangolides, 2′,3′-dihydroleptocarpin (**5a**) [18] and 2′,3′-dihydroniveusin C (**6a**), were also found. These were isolated and purified in the form of their pyrazoline adducts **7** and **8**, analysis of whose ^{1}H NMR spectra (Table 1) permitted their identification. The paramagnetic shift ($\Delta\delta \sim 0.9$) of H-5 of **7**, in comparison with H-5 of leptocarpin (**5b**) and its analogs, indicates that the stereochemistry of the pyrazoline moiety is as shown in the formula and predicted from the model. Likewise the H-5 signal of **8** is shifted downfield ($\Delta\delta \sim 0.3$) compared with H-5 of niveusin C(**6b**) and its analogs [19]. Lactone **6a** appears to be new; its acetate **6c** has been reported from a *Calea* species [20].

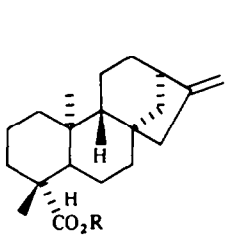
Another new compound was (−)-8-methoxyobliquin (**9a**), an isomer of 5-methoxyobliquin of unspecified rotation from *Helichrysum serpillifolium* [21]. That position C-5 on the coumarin nucleus was unsubstituted was evident from NOE experiments; irradiation at the frequency of H-4 at δ7.57 (Table 2) showed a clear NOE for the doublet at 6.28 (H-3, 32%) and the singlet at 6.79 (H-5, 20%). In the converse experiment, irradiation at the frequency of the H-5 signal enhanced the intensities of the H-4 signal (12%) and the signals of the vinylic protons at δ4.45 and 4.04, thus showing that the methoxy group was not on C-6 but on C-8, and that the five carbon moiety was attached to the coumarin nucleus in the manner of obliquin (**9b**) [22]. The rotation of **9a** was negative, unlike that of (+)-obliquin whose absolute configuration is unknown.

The ^{13}C NMR spectrum of **9a** is listed in Table 3; assignments are based on chemical shifts and on analysis of the fully coupled ^{13}C–^{1}H NMR spectrum with NOE enhancement. Thus the signal at δ114.46 was a sharp doublet with a C–H coupling constant of 173 Hz, whihe $^1J_{C-H}$ of the signal at δ108.92 which also exhibited a long

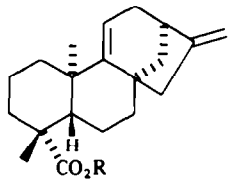
Table 1. ^{1}H NMR spectra of compounds **7** and **8** [270 MHz, $CDCl_3$; *J* (Hz) in parentheses]

H	7	8
1	2.86 *dd* (10, 5)	4.01 *dd* (8, 4.5)
2a	1.80 *ddd* (15, 10, 2.5)	2.56 *dd* (15, 4.5)
2b	2.50 *dt* (15, 5)	2.39 *d* (15)
3	4.54 *dd* (5, 2.5)	—
5	6.28 *dd* (12, 1)	5.97 *dd* (3, 1.5)
6	6.88 *br d* (12)	5.56 *ddd* (3, 2, 1.5)
7	2.45 *br s*	3.75 *dd* (4, 2)
8	5.03 *dd* (4.5, 2)	5.26 *ddd* (10, 6, 4)
9a	2.75 *dd* (15, 4.5)	2.07 *dd* (14, 10)
9b	1.20 *dd* (15, 2)	1.74 *dd* (14, 6)
13a	1.95 *m*	1.96 *m*
13b	1.65 *m*	1.40 *m*
14*	1.50 *s*	1.44 *s*
15*	1.89 *d* (1)	1.89 *t* (1.5)
16a	4.82 *m*	4.80 *ddd* (18, 10, 5)
16b	4.68 *m*	4.68 *ddd* (18, 10, 8)
2′	2.40 *tq* (7)	2.26 *tq* (7)
3′a	1.65 *m*	1.63 *m*
3′b	1.45 *m*	1.40 *m*
4′*	0.90 *t* (7)	0.87 *t* (7)
5′*	1.16 *d* (7)	1.07 *d* (7)
OH	2.18 *s*	2.95 *d* (8)
		3.26 *s*

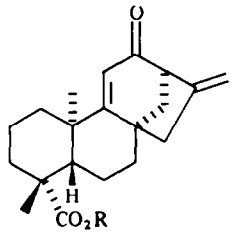
*Intensity three protons.



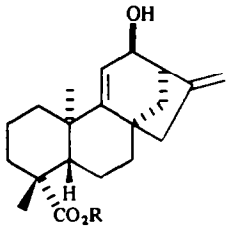
1a R = H
1b R = Me



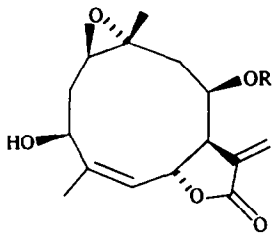
2a R = H
2b R = Me



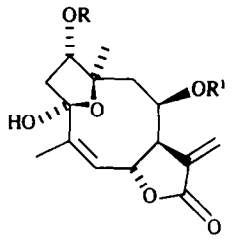
3a R = H
3b R = Me



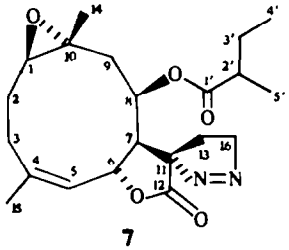
4a R = H
4b R = Me



5a R = 2-MeBu
5b R = Ang



6a R = H, R^1 = 2-MeBu
6b R = H, R^1 = Ang
6c R = Ac, R^1 = 2-MeBu



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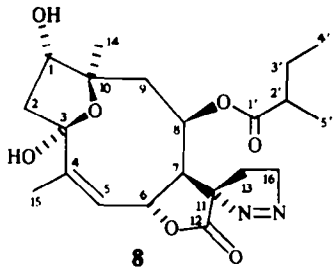

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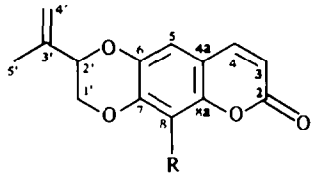
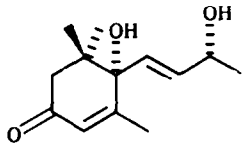


9a R = OMe
9b R = H



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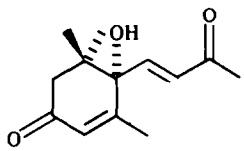

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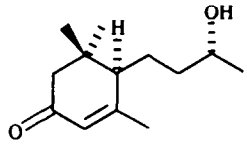

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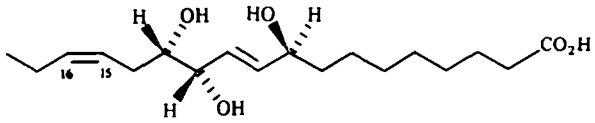

13a
13b 15,16-dihydro

range $^3J_{C-H}$ of *ca* 5 Hz was 163 Hz. Hence the signal at lower field was that of C-3, while the signal at higher field was that of C-5 [23], a conclusion which could be confirmed by selective decoupling. Among the quaternary carbons the signal at δ112.69 which showed only one $^3J_{C-H}$ of ~ 8 Hz could be assigned to C-4a because of its chemical shift and the complex signal at δ136.02 which displayed several long range coupling constants could be identified as that of C-3'. Among the remaining three signals, that at δ139.16, which exhibited two $^3J_{C-H}$s of about 8 Hz each, was therefore that of C-8a, the signal at δ140.78 with one long range coupling constant of *ca* 8 Hz was that of C-7 and the signal at δ141.12 with one long range coupling constant of 3.5 Hz ($^2J_{C-H}$) was that of C-6.

Table 2. ^{1}H NMR spectrum of compound **9a** [*J* (Hz) in parentheses]

H	δ
3	6.29 *d* (9.5)
4	7.57 *d* (9.5)
5	6.79 *s*
1'a	4.45 *dd* (11.5, 2.3)
1'b	4.04 *dd* (11.5, 8.2)
2'	4.54 *br dd* (8.2, 2.3)
4'a	5.19 *br s*
4'b	5.14 *br s*
5'*	1.80 *br s*
OMe*	4.03 *s*

*Intensity three protons.

Table 3. ^{13}C NMR spectrum of compound **9a** (67.89 MHz, $CDCl_3$)

C	δ
2	160.50 *s*
3	114.46 *d*
4	143.39 *d*
4a	112.69 *s*
5	108.92 *d*
6	141.12 *s*
7	140.28 *s*
8	142.48 *s*
8a	139.16 *s*
1'	67.60 *t*
2'	75.63 *d*
3'	136.02 *s*
4'	114.88 *t*
5'	18.77 *q*
OMe	61.69 *q*

Other compounds identified in the *H. heterophyllus* extract were the bisnorsesquiterpenoids (+)-vomilifoliol (blumenol A, **10**) [24, 25], dehydrovomifoliol (**11**) [26] and 9-hydroxy-4-megastigmen-3-one (**12**) [27], 2,6-dimethoxybenzoquinone, vanillin and two trihydroxy-C_{18} acids **13a**, **13b** first isolated by us from *Rudbeckia fulgida* [28] and later found in *Wedelia pinetorum* [29]. Two antifungal acids apparently identical with **13a**, **13b** have been isolated from a rice cultivar infected with rice blast disease [30] and are apparently present as such in resistant rice varieties [31]. Thus at least **13a** and **13b**, if not the other metabolites, appear to be part of the defense system of *H. heterophyllus*.

EXPERIMENTAL

Helianthus heterophyllus Nutt. was collected by Dr. John B. Nelson within the Apalachicol National Forest along Forest Service Road # 344 1.5 miles So. of Hoxham, Leon County in October 1983. The dried aerial parts (2.27 kg) were extracted with $CHCl_3$ and worked up in the usual fashion [32]. The crude gum (3.2 g) was adsorbed on 6 g of silicic acid (Mallinckrodt 100 mesh) and chromatographed over 200 g of the same adsorbent packed in hexane, 125 ml fractions being collected as follows: 1–4 (hexane), 5–8 (hexane–EtOAc, 9:1), 9–12 (hexane–EtOAc, 4:1), 13–16 (hexane–EtOAc, 2:1), 17–20 (hexane–EtOAc, 1:1), 21–24 (hexane–EtOAc, 2:3), 25–28 (hexane–EtOAc, 1:4), 29–32 (EtOAc) and 33–36 (EtOAc–MeOH, 9:1).

Fr. 6 (0.04 g) was a mixture of **1a** and **2a** which was separated after methylation with CH_2N_2 and subsequent chromatography to give 7 mg of **1b** and 6 mg of **2b** identified by comparison with authentic material [33]. Fr. 9–10 (0.15 g) after purification by CC (C_6H_6–EtOAc, 17:3) gave 35 mg of **9a** as a gum, $[\alpha]_{546}$ −29.5° ($CHCl_3$, *c* 0.420); MS *m/z* (rel. int.): 274 $[M]^+$ (100), 259 (1.4), 243 (20.4), 219 (19.11), 207 (3.4), 192 (10.4); ^{1}H and ^{13}C NMR spectra: Tables 2 and 3. [Calc. for $C_{15}H_{14}O_5$: M_r 274.0841 Found: M_r (MS) 274.0838].

Fr. 11–12 (0.25 g) were rechromatographed over 40 g of silica gel (70–230 mesh) packed in hexane. Elution with hexane–EtOAc (4:1) gave 5 mg of **9a**, 5 mg of vanillin and a mixture which was methylated with CH_2N_2. Acetylation (Ac_2O–pyridine, overnight) and purification by TLC (hexane–EtOAc, 9:1, two developments) gave 4 mg of **3b** [34]. Fr. 13–14 (0.25 g) on initial purification by CC furnished impure **4a** which was methylated and then rechromatographed to give 10 mg of **4b** [35].

Reaction of fr. 15–16 (0.2 g) with CH_2N_2 and CC ($CHCl_3$) gave crude **7** ane **8**. After rechromatography (hexane–EtOAc, 4:1), **7** was induced to crystallize (hexane–EtOAc, 9:1, −20°), mp 122° (dec); $[\alpha]_{546}$ −288° ($CHCl_3$, *c* 0.102); MS *m/z* (rel. int.): 406 $[M]^+$ (0.07), 378 (0.9), 360 (0.2), 276 (0.7), 258 (3.0), 233 (7.0), 232 (7.2), 177 (12.0), 163 (13.8), 149 (11.1), 135 (24.1), 95 (22.9), 85 (33.4), 57 (100); MS (CI) *m/z* (rel. int.): 407 $[M+1]^+$ (0.45), 379 (100), 361 (2.2), 277 (33.3) znd 259 (6.8). The ^{1}H NMR spectrum is listed in Table 1.

Crude **8** was purified by CC (hexane–EtOAc, 7:3) and crystallized from hexane–EtOAc (overnight, −20°), mp 149–150°; $[\alpha]_{546}$ −253° ($CHCl_3$, *c* 0.072); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3508, 1770, 1724 and 1600; MS *m/z* (rel. int.): 422 $[M]^+$ (0.4), 394 (0.6), 376 (0.7), 292 (4.7), 274 (3.6), 279 (20.8), 85 (21.5), 57 (100); MS (CI) *m/z* (rel. int.): 423 $[M+1]^+$ (12.8), 395 (100), 377 (27.4), 293 (32.8) and 275 (46.8). The ^{1}H NMR spectrum is listed in Table 1.

Fr. 17–19 (0.28 g) on CC (hexane–EtOAc, 7:3) gave 5 mg of 2,6-dimethoxybenzoquinone and a mixture containing **11** and **12**. Separation of the mixture by prep. TLC (C_6H_6–EtOAc, three developments) gave in the upper band 8 mg of **11** [25] and in the lower band 10 mg of **12** [26]. The ^{1}H NMR spectra of these substances coincided with those reported in the literature. Fr. 20–22 (0.2 g) containing one major component were purified by radial chromatography (hexane–EtOAc, 7:3) to give 60 mg of **10**, mp 110–112° (hexane–EtOAc), $[\alpha]_{546}$ +245° ($CDCl_3$; *c* 0.08); ^{1}H NMR (270 MHz, $CDCl_3$): δ 5.90 (*J* = 1 Hz, H-4), 5.86 (*dd*, *J* = 17.5 Hz, H-8), 5.78 (*d*, *J* = 17 Hz, H-7), 4.41 (*dq*, *J* = 6.5, 5 Hz, H-9), 2.44 and 2.25 (AB system, *J* = 17 Hz, H-2a, b), 1.89 (*d*, *J* = 1 Hz, vinyl Me), 1.30 (*d*, *J* = 6.5 Hz, H-10), 1.08 (Me) and 1.01 (Me). Fr. 23–24 (0.2 g) on purification by CC (hexane–EtOAc, 3:7) gave 40 mg of a 1:1 mixture of **13a** and **13b** [27].

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