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CONSTITUENTS OF *HYERONIMA ALCHORNEOIDES*

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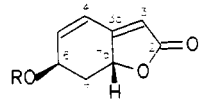
ABSTRACT.—The structures of hyeronimone [**3**] and its acetate **4**, isolated from the roots of *Hyeronima alchorneoides*, have been established, mainly on the basis of 2D nmr spectroscopy. Aquilegiolide [**1**], a rare butenolide, was isolated from the leaves of the plant, which also afforded lupeol, stigmasterol, and a sitosterol.

We have examined the leaves and roots of *Hyeronima alchorneoides* Alkmão (Euphorbiaceae), a canopy tree of the Guyana forest that Fanshawe (1) described previously as *Hieronyma laxiflora*. A decoction of the bark is used locally as an antitussive. The genus appears not to have received previous phytochemical attention. The butenolide **1** was isolated from the leaves. It has been described twice previously, first as a hydrolysis product of menisdaurin, a nitrile glucoside of *Menispermum dauricum* (2), and then as a natural product isolated directly from *Aquilegia atrata* (Ranunculaceae) by Guerriero and Pietra (3), who named it aquilegiolide.

In our isolation procedure, crude **1** was acetylated, and the acetate **2** was investigated. The structure was established by 2D nmr spectroscopy. The acetylation procedure gave, in fact, a mixture of two isomers, **2** and 7a-*epi*-**2**; these were separated, but each reverted to a mixture of the two isomers on standing in solution. The ready epimerization of the parent butenolide has been described by Guerriero and Pietra (3). Lupeol, stigmasterol, and sitosterol were also identified in the leaf extract.

Hyeronimone, $C_{19}H_{31}NO_3$, mp 85–86°, which appears to be a completely new compound, was isolated from an extract of the roots. The same extract also afforded the monoacetate, $C_{21}H_{33}NO_4$, mp 75–77°, of hyeronimone. The ir and nmr spectra of these compounds provided evidence for the presence of a pyridone unit, and a series of 2D nmr experiments, which included ^{1}H-COSY 45 and one-bond and *n*-bond ($n = 2$ or 3) ^{13}C-^{1}H shift correlations, led us to assign tentatively structures **3** and **4** to hyeronimone and its monoacetate. Tables 1 and 2 list assigned chemical shifts and con-



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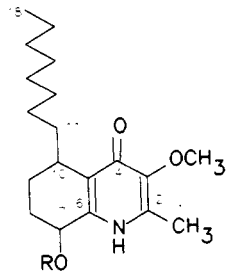

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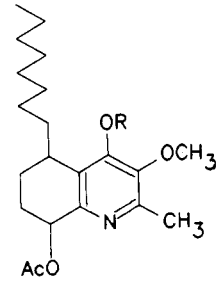
4 R=Ac



5 R=Me
6 R=Ac

TABLE 1. Assigned Chemical Shifts and Connectivities for Hyeronimone [3].

Position	δ_C[a]	δ_H[b]	n-bond connectivities[c]
1	14.05	2.39	
2	140.64	—	2.39
3	144.42	—	2.39, 3.75
4	171.28	—	
5	129.08	—	1.94, 2.93, 4.81
6	146.72	—	1.93, 2.10, 2.93, 4.81
7	66.77	4.81	1.93, 2.10
8	25.85	1.93, 2.10	1.94, 4.81
9	23.21	1.63, 1.94	1.75, 2.93
10	31.89	2.93	1.93
11	32.27	1.39, 1.75	2.93
12	28.37	1.30, 1.46	1.39, 1.75 (w), 2.93 (w)
13	29.86	<1.25>	
14	29.69	<1.23>	1.21
15	29.37	<1.22>	1.26 (w)
16	31.89	<1.21>	0.86, 1.25
17	22.68	<1.26>	0.86, 1.21
18	14.11	0.86	1.21, 1.26
MeO	59.96	3.75	2.39

[a]Chemical shifts measured at 100.6 MHz; $CDCl_3$ solution.

[b]Chemical shifts measured at 400 MHz; $CDCl_3$ solution. Chemical shifts are recorded for each identifiable H (or Me) except that the <average value> is shown for unresolved CH_2 groups.

[c]Chemical shifts are recorded for protons giving cross peaks to ^{13}C with δ_C shown in second column; $n = 2$ or 3; (w) indicates a weak peak.

TABLE 2. Assigned Chemical Shifts and Connectivities for Acetylhyeronimone [4], and ^{13}C Relaxation Times.

Position	δ_C	T_1[b]	δ_H	n-bond connectivities
1	14.04	0.97	2.30	
2	138.96	—	—	2.30
3	144.95	—	—	2.30, 3.78
4	171.80	—	—	
5	130.42	—	—	1.88, 2.93
6	139.22	—	—	2.10, 2.93, 5.77
7	69.28	0.33	5.77	1.88
8	24.64	0.20	1.90, 2.10	
9	22.97	0.19	1.74, 1.88	
10	31.43	0.33	2.93	1.90, 2.10
11	32.35	0.21	1.35, 1.92	1.37, 2.93
12	27.90	0.31	<1.37>	1.35
13	29.86	0.55	<1.28>	1.26
14	29.75	0.81	<1.29>	1.26
15	29.38	1.20	<1.26>	1.26
16	31.93	1.76	<1.26>	0.87, 1.26
17	22.70	3.08	<1.29>	0.87, 1.26
18	14.13	3.40	0.87	
MeO	59.37	1.10	3.78	
CH_3CO	21.27	1.12	2.10	
MeCO	171.70	—	—	2.10

[a]See footnotes to Table 1 for explanatory details.

[b]^{13}C T_1 measurements used a standard inversion-recovery sequence with delay times appropriate for protonated carbons; the values were determined by exponential fitting with standard Varian software.

nectivities for **3** and **4** respectively. However, several ^{13}C signals were extremely broad, the ^{1}H spectrum was unusually complex in the δ 1.2–1.4 and δ 1.8–2.1 regions, and some crucial 3-bond ^{13}C-^{1}H connectivity peaks were not observed. The pyridone-hydroxypyridine tautomerism was a complicating factor in these experiments, and the structural assignment was not unequivocal. To remove this complication, the methyl ether **5** of the monoacetate **4** was prepared by treatment of **4** with CH_2N_2 in the presence of Si gel (4); to the same end, hyeronimone [**3**] and the monoacetate **4** were treated with Ac_2O and pyridine and converted to the same diacetate **6**.

The one-bond and *n*-bond ^{13}C-^{1}H connectivities of both **5** and **6** were investigated by 2D nmr spectroscopy; our FLOCK pulse sequence (5) was used to generate spectra showing *n*-bond connectivities. The data are summarized in Tables 3 and 4. In these compounds, as in the pyridone precursors, the absence of any protonated carbons on the heterocyclic ring limits the amount of structural information that can be derived from ^{13}C-^{1}H shift correlations. The connectivity of C-3 to its attached OMe protons and to the methyl protons at C-1 is observed in all of the compounds examined, and the connectivity of C-4 to its attached OMe protons is observed in **5** and **6**; in **6** the further important connectivity of C-4 with the C-10 proton is observed. The observed connectivities at C-5 and C-6 show that C-5 is bonded to C-10 and C-6 is bonded to C-7. Both ^{13}C and ^{1}H chemical shifts for methylene groups in the *n*-octyl side chain are observed in a very narrow range; ^{13}C relaxation times (listed in Table 2) were measured for **4** to assist in making chemical shift assignments. Furthermore, 1D nOe difference spectra of **5** showed that irradiation of the protons of the 4-MeO gave positive nOe effects at the C-10 proton and at the protons of the MeO at C-3, while irradiation of the latter MeO protons gave positive nOe effects at the 4-MeO protons and at the Me-1 protons.

TABLE 3. Assigned Chemical Shifts and Connectivities for **5**, the Methyl Ether of Acetylheronimone.[a]

Position	δ_C	δ_H	*n*-bond connectivities
1	19.14	2.43	
2	152.08	—	2.43
3	145.84	—	2.43, 3.76
4	156.65	—	3.96
5	130.37	—	2.43 (w)[b], 2.93
6	148.85	—	5.80
7	72.15	5.80	1.76, 1.84, 1.97
8	24.66	1.97, 2.10	
9	23.12	1.76, 1.84	
10	32.01	2.93	
11	34.55	1.48, 1.56	
12	27.76	1.35, 1.40	
13	29.58	<1.29>	
14	29.56	<1.29>	
15	29.34	<1.29>	
16	31.92	<1.25>	0.89, 1.29
17	22.69	<1.30>	0.89
18	14.12	0.89	
3-CH_3O	59.98	3.76	
4-CH_3O	60.08	3.96	
CH_3CO	21.56	2.15	
MeCO	170.97	—	2.15

[a]See footnotes to Table 1 for explanatory details.
[b]$n = 5$.

TABLE 4. Assigned Chemical Shifts and Connectivites for Diacetylhyeronimone [6].[a]

Position	δ_C	δ_H	*n*-Bond connectivities
1	19.45	2.49	
2	151.82	—	2.49
3	145.76	—	2.49, 3.75
4	148.49	—	2.36 (w),[b] 2.49 (w)[b], 2.74, 3.75 (w)[b]
5	130.01	—	1.85 (w), 2.49 (w),[c] 2.74, 5.83
6	149.50	—	2.74, 5.83
7	71.71	5.83	1.85, 2.00, 2.14
8	24.51	2.00, 2.14	2.74, 5.83
9	23.00	<1.85>	
10	32.40	2.74	1.56
11	34.15	<1.56>	2.74
12	27.77	1.33, 1.45	1.56
13	29.56	<1.29>	1.33
14	29.54	<1.29>	
15	29.29	<1.28>	
16	31.87	<1.26>	0.89
17	22.66	<1.30>	0.89
18	14.11	0.89	1.30
CH_3O	60.57	3.75	
CH_3COO(C-4)	20.59	2.36	
MeCOO(C-4)	167.60	—	2.36
CH_3COO(C-7)	21.48	2.15	
MeCOO(C-7)	170.79	—	2.15, 5.83 (w)

[a]See footnotes to Table 1 for explanatory details.
[b]$n = 4$.
[c]$n = 5$.

These data establish that the heterocyclic ring in **3** and **4** is a 4-pyridone substituted as shown and that the carbocyclic ring carries an octyl side chain at C-10 and the oxygen function at C-7. A series of ^{1}H-^{1}H decoupling experiments provided evidence confirming the connectivities in **3**. Exact values for many of the coupling constants could not be calculated because of the overlap of signals and broadening caused by tautomerism, but it was possible to determine coupling constants of about 9.8 and 6.7 Hz from H-7 to H-8, and of about 5 and 2 Hz from H-10 to H-9. These values clearly show that the stereochemical environment is different for H-7 and H-10, with the most probable assignment being that the former is pseudoaxial and the latter pseudoequatorial, provided that the ring is in the expected half-chair conformation. It follows then that the OH at C-7 and the octyl group at C-10 have a cis relationship.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Mp's were determined on a micro hot stage. Selected ir absorptions (Ft-ir) are reported (in cm^{-1}). Uv spectra were obtained for MeOH solutions; λ max (ε) values are reported (in nm). Nmr spectra were obtained for $CDCl_3$ solutions; ^{1}H spectra were obtained at 400 MHz and ^{13}C spectra at 100 MHZ. Tables 1–4 provide details for nmr assignments made therein; nmr spectra of **2** are described below by listing δ_H values followed, in parentheses, by apparent multiplicities and coupling constants (in Hz), and the assignment made, and δ_C values are listed with assignments shown in parentheses; the assignments are based on COSY and HETCOR experiments. A VG 70-250S mass spectrometer, operating at 70 eV, was used to obtain mass spectra.

PLANT MATERIAL.—Plants were collected in the Essequibo region of Guyana. Voucher specimens are deposited at the Herbarium of the University of Guyana and at the Institute of Systematic Botany, University of Utrecht, Netherlands.

EXTRACTION.—Dried, ground leaves were extracted with CH_2Cl_2, and the crude extract (69 g) was dissolved in MeOH-H_2O (9:1), extracted with hexane, diluted with H_2O until 40% aqueous, and re-extracted with CH_2Cl_2. The hexane extract afforded lupeol, mp 193–194°, and a 1:1 stigmasterol-sitosterol mixture, mp 129–131°. Identification was made by spectroscopic comparison with reference samples; in the latter case, the ^{1}H-nmr spectrum of a mixture prepared from commercial stigmasterol and β-sitosterol was used. The CH_2Cl_2 extract afforded 26 g of material on Si gel chromatography, a portion (13 g) gave a fraction (5.1 g) eluted with hexane-Me_2CO (3:1). Rechromatography of this fraction with hexane-EtOAc (19:1) elution gave aquilegiolide [**1**] (1.34 g) as an oil. Treatment of the oil (300 mg) with Ac_2O/pyridine followed by fractional crystallization of the product provided the acetate **2**: mp 100–102°, [α]D +60.9° (c = 0.11, $CHCl_3$), uv 254 (18,500), ^{1}H nmr 6.70 (d, J = 9.7, H-4), 6.30 (dd, J = 9.7, 5.2, H-5), 5.86 (very narrow m, H-3), 5.55 ('t'd, J = 4.8, 2.0, H-6), 5.19 (ddd, J = 12.8, 5.1, 1.8, H-7a), 2.62 (complex m, H-7α), 2.08 (s, acetyl Me), 1.86 ('t'd, J = 13.2, 4.4, H-7β); ^{13}C nmr 172.7 (C-2), 169.9 (acetyl C=O), 161.6 (C-3a), 133.6 (C-5), 123.9 (C-4), 113.2 (C-3), 75.9 (C-7a), 66.2 (C-6), 34.3 (C-7), 20.8 (acetyl Me); eims 194 (13), 152 (72), 134 (100), 106 (50), 78 (46).

Dried, ground roots (880 g) were extracted with $CHCl_3$-MeOH (1:1) to provide a gum (63 g) that was separated, by the procedure described above, into a hexane-soluble fraction (20 g) and a CH_2Cl_2-soluble fraction (31 g). A portion (15 g) of the latter fraction was chromatographed on Si gel with elution by ($CHCl_3$-MeOH (49:1) to give material (10.8 g) that was rechromatographed with elution by hexane/$CHCl_3$ mixtures of increasing polarity. A 3:1 mixtute eluted the acetylated pyridone (145 mg), and a 2:3 mixture eluted the pyridone (358 mg).

Hyeronimone [**3**].—Mp 85–86°; [α]D +115° (c = 0.06, $CHCl_3$); ir 3343, 1620; eims 321 (46), 293 (46), 265 (52), 209 (60), 192 (100), 166 (52), 130 (33); hreims 321.2304, calcd for $C_{19}H_{31}NO_3$, 321.2304.

O-*Acetylhyeronimone* [**4**].—Mp 75–77°; [α]D +178° (c = 0.06, $CHCl_3$), ir 3367, 3275, 1746, 1637; uv 216 (20,000); eims 363 (12), 320 (100), 292 (10), 251 (11), 191 (47); hreims 363.2407, calcd for $C_{21}H_{33}NO_4$, 363.2410.

O-*Acetylhyeronimone methyl ether* [**5**].—An ethereal solution of the pyridone **4** (132 mg) was stirred with tlc-grade SiO_2 while CH_2N_2 was passed into the solution. The product was a colorless oil (72 mg): [α]D 0° (c = 0.34, $CHCl_3$); ir 1727, uv 216 (8600), 268 (4000); eims 377 (6), 348 (7), 334 (100), 320 (9), 306 (30), 204 (32); hreims 377.2563, calcd for $C_{22}H_{35}NO_4$, 377.2566.

O,O-*Diacetylhyeronimone* [**6**].—Hyeronimone [**3**] (75 mg) treated overnight with Ac_2O (1 ml) and pyridine (1 ml) provided **6** (72 mg); **6** was also prepared by treating acetylhyeronimone [**4**] in the same way. Diacetate **6** was obtained as crystals: mp 51–52°; [α]D −4° (c = 0.45, $CHCl_3$); ir 1765, 1739, 1242, 1229; uv 210 (8500), 270 (5400); eims 405 (7), 362 (43), 320 (100), 292 (9), 207 (8), 190 (28); hreims 405.2519, calcd for $C_{23}H_{35}NO_5$, 405.2515.

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