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Dihydrocornin, a Novel Natural Iridoid Glucoside

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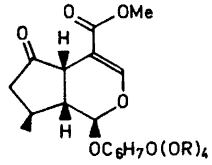
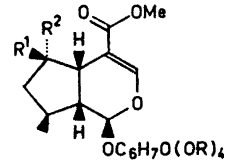
Leaves and twigs of *Cornus Nuttallii* and *C. florida* contain, besides the long-known glucoside cornin *1a*, a minor constituent, dihydrocornin *3*, characterized as the pentaacetate *7*. Reduction of cornin tetraacetate *1b* with $LiAlH(t\text{-}BuO)_3$ yields a 2:1 mixture of two tetraacetates, assigned the structure *4* and *5*, respectively, on the assumption of a more facile approach of the reagent from the convex face of the substrate. The pentaacetate derived from *5* is indistinguishable from that of natural derivation, a conclusion necessitating revision of assignments in the literature. NMR-data for a series of natural and synthetic iridoid glucosides are presented.

In the course of phytochemical studies within the genus *Cornus* (Cornaceae), the species *C. Nuttallii* Aud. and *C. florida* L. have been found to contain a novel, natural iridoid glucoside, additional to cornin *1a*,[1]* a long known constituent of the latter species.[2] The new glucoside, dihydrocornin, $C_{17}H_{26}O_{10}$, was obtained in crystalline form by repeated chromatography of a purified





1a; R = H

1b; R = Ac

2; R=R¹=H, R²=OH

3; R=R²=H, R¹=OH

4; R=Ac, R¹=H, R²=OH

5; R=Ac, R¹=OH, R²=H

6; R=Ac, R¹=H, R²=AcO

7; R=Ac, R²=H, R¹=AcO

* The name cornin applies to a β-glucoside, isolated in 1902 in pure form from *C. florida* L. and characterized by elemental composition, $C_{17}H_{24}O_{10}$, m.p., optical rotation, and a number of chemical reactions (*cf.* Ref. 3). Its identity with 'verbenalin', described as a constituent of *Verbena officinalis* L. in 1908,[4] has been established.[5] In agreement with other authors,[2,5] we propose to delete the name 'verbenalin' as redundant.

iridoid glucoside fraction, isolated from leaves and twigs of the above *Cornus* species. Its structure elucidation rests heavily on ^{1}H NMR data, displaying a considerable overall similarity to those of cornin *1a*, yet with a higher multiplicity of the methine proton at C-5 as a distinctly deviating feature. Successive decouplings from the vinylic proton (at C-3) and the hidden methine proton at C-9, recognized by its coupling to the easily distinguishable acetalic proton at C-1, revealed the existence of residual coupling (J 3 Hz) to a proton at δ 4.17 ppm, obviously residing in a hydroxy-substituted methine group at C-6. In keeping with this conclusion, acetylation of the glucoside afforded a pentaacetate, m.p. 166 – 168°.

Partial synthesis of the 6-epimeric carbinols, (*2*) and (*3*), was accomplished by reduction of cornin (*1a*) with $NaBH_4$ in methanol. The epimer, moving at the slowest rate on chromatography and constituting about 50 % of the mixture, was indistinguishable from dihydrocornin from natural sources. The second epimer did not crystallize, but was characterized as a pentaacetate, m.p. 141 – 142°. Similarly, $NaBH_4$ reduction of cornin tetraacetate (*1b*) yielded a *ca.* 1:1 mixture of the epimeric carbinol tetraacetates *4* and *5*, one crystalline (m.p. 150°), the other amorphous. Reduction with $LiAlH(t\text{-}BuO)_3$, a reputed stereoselective reagent,[6] afforded the same products, yet in the ratio 2:1 in favour of the amorphous product. On the assumption that preferential approach of the bulky reducing agent occurs from the least hindered, *i.e.* the convex face, the crystalline tetraacetate possesses the structure *5*. On acetylation, *5* affords a pentaacetate, *7*, m.p. 166 – 168°, indistinguishable from that of natural derivation, whereas *4* yields the epimeric acetate *6*, m.p. 141 – 142°. Hence, dihydrocornin possesses the structure *3*.

This assignment is at variance with statements in the literature. Thus, Büchi and Manning[1] formulated a pentaacetate, m.p. 172 – 174°, obtained in 39 % yield by $NaBH_4$ reduction of cornin (*1a*), followed by acetylation, as *6*. Similarly, reduction of cornin tetraacetate (*1b*), with subsequent epimerization and acetylation, gave a pentaacetate, m.p. 134 – 138°, formulated as *7*.[1] The present evidence suggests that these assignments should be interchanged.

Table 1. Optical rotations, UV-maxima, and analytical data.

Compound	$[\alpha]_D^{20}$ (c in EtOH)	UV-data (EtOH) λ_{max} in nm (ε)	Formula	Analyses Calculated C	Calculated H	Found C	Found H
1b	−135[a] (0.4)	237[b] (9 300)					
3	−126 (0.5)	238 (11 200)	$C_{17}H_{26}O_{10}$	52.30	6.71	52.14	6.68
4	−67 (1.4)	238 (11 200)	$C_{25}H_{34}O_{14}$	53.75	6.15	53.77	6.21
5	−113 (0.4)	236 (10 700)	$C_{25}H_{34}O_{14}$	53.75	6.15	54.05	6.20
6	−86 (0.5)	234[d] (11 700)	$C_{27}H_{36}O_{15}$	54.00	6.04	54.21	6.09
7	−115 (0.5)	233[c] (10 400)					

[a] Reported:[9] −134° (*c* 1.2, EtOH). [b] Reported:[1] 235 nm (9370). [c] Reported:[1] 232 nm (11 200). [d] Reported:[1] 234 nm (11 500).

Table 2. ^{1}H NMR-data (*cf.* Experimental).

Compound (solvent)		δ-Values[a] H-1	H-3	H-5	H-6	OCH_3	10-CH_3	Coupling constants[b] $J_{1,9}$	$J_{3,5}$	$J_{5,6}$	$J_{5,9}$
1a	(D_2O)[c]	5.38	7.58	~3.5	–	3.78	1.22	5.5	1.2	–	–
1b	($CDCl_3$)	~5.2	7.40	3.50	–	3.77	1.22	–	1.5	–	8
2	(D_2O)[c]	5.21	7.70	2.99	4.52	3.81	1.16	8.5	1.3	4	9.5
3	(D_2O)[c]	5.52	7.48	2.88	4.17	3.81	1.15	2.5	1.0	3	8
4	($CDCl_3$)	~5.1	7.60	2.88	4.48	3.75	1.12	–	1.2	4	9
5	($CDCl_3$)	5.22	7.36	2.49	4.01	3.78	1.12	3.0	1.0	4	9
6	($CDCl_3$)	5.06	7.51	3.08	5.52	3.75	1.13	8	1.2	4	9
7	($CDCl_3$)	5.37	7.38	3.00	5.25	3.73	1.13	2.0	1.0	2	9

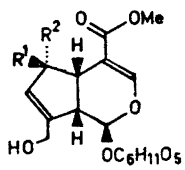
[a] In ppm ±0.02 ppm. [b] $J_{3,5}$ ±0.1 Hz; others ±0.5 Hz. [c] DDS = 0 ppm.

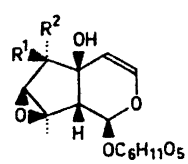
Analytical data and physical constants are presented in Tables 1 and 2. Certain consistencies in the ^{1}H NMR data are apparent: (*i*) in members of the 6α-series, the methine proton at C-6 is more deshielded than in the 6β-series; (*ii*) the proton at C-1 appears at lower field in the 6β- than in the 6α-series (*cf.* Table 3); (*iii*) the vinylic proton at C-3 is most deshielded in the 6α-

Table 3. Differences in δ-values of epimeric 6-OH-iridoids.

Compound	δH-1 (α) − δH-1 (β)	δH-3 (α) − δH-3 (β)
2 and *3*	−0.31	0.22
4 and *5*	–	0.24
6 and *7*	−0.31	0.13
9 and *8*	−0.36	0.20
11 and *10*	−0.10	0.06

series (*cf.* Table 3). Two related pairs of epimers: scandoside methyl ester-daphylloside (*8,9*),[7,8] and anthirrinoside-procumbide (*10,11*),[9,10] conform to these regularities (*cf.* Table 3).





8 ; R^1=OH, R^2=H

9 ; R^1=H, R^2=OH

10; R^1=OH, R^2=H

11 ; R^1=H, R^2=OH

EXPERIMENTAL

When not otherwise indicated, melting points are uncorrected and determined in capillary tubes in a heated bath. NMR-Spectra are recorded on a Varian HA-100 instrument, with TMS as an internal or external standard in $CDCl_3$ or D_2O, respectively. Preparative thin layer chromatography (TLC) was performed on 20×40 cm plates covered with a 1 mm thick layer of silica gel PF_{254} (Merck); detection: UV-light. Analyses were performed at Dr. A. Bernhardt, Mikroanal. Labor., Germany.

Isolation of glucosides. Frozen foliage (50 g) of *C. Nuttallii* or *C. florida*, collected in the summer of 1972 and stored at $-28°$ in polyethylene bags, was homogenized in EtOH (150 ml). After filtration, extraction was repeated with another 150 ml-portion of EtOH. The combined filtrates were concentrated *in vacuo*, and the residue was partitioned between water (50 ml) and ether (150 ml). The aqueous phase was extracted with an additional amount of ether (150 ml) and the organic phases were discarded. The aqueous solution was filtered through a column of neutral alumina (50 g); the column was eluted with water (500 ml). After evaporation to dryness, the residue was dissolved in the minimum amount of MeOH:H_2O and applied to a column of silica gel. Acetone (400 ml) eluted a glycoside fraction which was further subjected to separation into two UV-absorbing zones on preparative TLC in $CHCl_3$:MeOH (4:1). A faster, major zone afforded pure cornin *1a* after recrystallization from EtOH, m.p. 178°, $[\alpha]_D^{20}$ $-170°$ (*c* 0.4, EtOH) [reported: m.p. $182-183°$ (corr.);[1,11] $[\alpha]_D^{24}$ $(-165°)-(-166°)$ (EtOH),[11] $(-171°)-(-173°)$ (H_2O);[1,11] others quote numerically higher rotation values, *viz.* $[\alpha]_D^{25}$ $-198°$ (*c* 1.41, H_2O) [12] and $(-184°)-(-185°)$ (H_2O) [13]]. A slightly slower-moving, minor zone contained dihydrocornin. Rechromatography, under the same conditions, of the combined material from several isolations, afforded a yellowish, sirupy fraction which was adsorbed on charcoal. After rinsing with water, the glucoside was eluted with MeOH; it separated from water-saturated EtOAc as colourless crystals, m.p. $90-100°$, containing *ca.* 0.75 mol of H_2O. A specimen dried at room temperature over P_2O_5 at 2 mm for 24 h was employed for analysis (Table 1). Rotation and UV-absorption are likewise reported in Table 1.

Leaves and twigs of *C. Nuttallii* contained $0.2-0.3$ % and 0.5 % of cornin, respectively. The corresponding contents of dihydrocornin were 0.03 and 0.04 %, respectively. In *C. florida*, leaves were found to contain $0.12-0.13$ % of cornin and 0.03 % of dihydrocornin, twigs 0.14 % and 0.08 %, respectively.

Dihydrocornin pentaacetate 7. A solution of dihydrocornin (35 mg) in pyridine (0.5 ml), containing Ac_2O (0.3 ml), afforded, after work up, a crude pentaacetate (55 mg) which was recrystallized twice from EtOH to give colourless crystals of *7*, m.p. $166-168°$ (on Kofler Micro Hot Stage, $172-173°$) [reported [1] m.p. $172-174°$ (Kofler)]. For other data, *cf.* Tables $1-3$.

Reduction of cornin 1a. Cornin (300 mg) was dissolved in MeOH (10 ml) and $NaBH_4$ (100 mg) was added. After stirring for 10 min, 2 drops of AcOH were added. The reaction mixture was concentrated *in vacuo* and the residue subjected to two developments on TLC plates in Bz:EtOH:EtOAc, 1:1:4, resulting in the separation of two zones. The fastest moving of these, containing *2* (143 mg), was separated from the slower moving fraction, chiefly *3*, contaminated with traces of *2*. Both fractions were freed of AcONa by adsorption on charcoal, washing with H_2O, and elution with MeOH. The α-epimer, *2*, could not be induced to crystallize from a number of solvents tried, and apart from recording of its NMR-spectrum (Table 2), further characterization was abandoned. Recrystallization of the β-epimer, *3*, from wet EtOAc, gave colourless needles, indistinguishable from dihydrocornin (m.p., m.m.p., NMR-spectrum).

epi-*Dihydrocornin pentaacetate* 6. On acetylation with Ac_2O in pyridine solution, the amorphous α-epimer from the above reduction yielded a crystalline pentaacetate, m.p. (from MeOH) $141-142°$, (Kofler, m.p. $141.5-142.5°$), assigned the structure *6* (reported:[1] $134-138°$, Kofler). For further data, *cf.* Tables $1-3$.

Reduction of cornin tetraacetate 1b *with* $NaBH_4$. Cornin tetraacetate *1b* (190 mg), obtained by acetylation of cornin,[1] was dissolved in MeOH (10 ml), and a mixture of $NaBH_4$ (35 mg) and H_3BO_3 (50 mg) was added with stirring. After 5 min at ambient temperature, AcOH (6 drops) and H_2O (40 ml) were added. The solution was concentrated to 20 ml and extracted with two 40-ml portions of ether. The ether-soluble residue (178 mg), containing the two epimeric dihydro-derivatives *4* and *5* and some unreduced *1b*,

in the ratio 2:2:1 according to NMR-analysis (in CCl_4; low-field region), was subjected to TLC-separation (3 plates) with Bz:Et_2O (1:2) as the eluant (two developments). The fastest migrating zone contained unchanged *1b* (35 mg). The next zone afforded the pure *β*-carbinol *5* (44 mg), recrystallized from Et_2O to give an analytical specimen, m.p. 150° (*cf.* Tables 1 – 3). The slowest moving band, not completely separated, gave the *α*-carbinol *4*, contaminated with the epimer *5*. All attempts to induce rechromatographed *4* to crystallize proved abortive. An analytical specimen was produced by filtering a CH_2Cl_2-solution through charcoal, followed by high-vacuum drying of the residue. For further data, *cf.* Tables 1 – 3.

On acetylation with Ac_2O in pyridine solution, *5* yielded a pentaacetate, m.p. 166 – 168°, indistinguishable from *7*, deriving from dihydrocornin. Under the same conditions, *4* afforded a crystalline pentaacetate, m.p. 141 – 142°, identical with *6* described above.

Reduction of cornin tetraacetate 1b *with LiAlH*(t-*BuO*)$_3$. A solution of $LiAlH_4$ (150 mg) in anhydrous ether (25 ml) was refluxed under N_2 for 1 h and then siphoned, through a plug of glass wool, into another flask, containing *t*-BuOH (900 mg) in ether (25 ml). After 1 h, the solution was decanted from the precipitate, which was dissolved in freshly distilled THF (25 ml). Under stirring, the tetraacetate (70 mg) was added to the clear solution, and after 30 min the reaction was stopped by adding AcOH (10 drops) and water (40 ml). In this case the reduction was complete, the epimeric carbinols *4* and *5* being present, according to NMR-analysis, in the ratio 2:1.

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REFERENCES

1. Büchi, G. and Manning, R. E. *Tetrahedron* **18** (1962) 1049.
2. Hegnauer, R. In *Beiträge zur Biochemie und Physiologie von Naturstoffen*, Festschrift für K. Mothes, G. Fischer, Jena 1965, p. 235.
3. Miller, E. R. *J. Am. Pharm. Assoc.* **17** (1928) 744.
4. Bourdier, L. *Arch. Pharm.* **246** (1908) 272.
5. Reichert, B. *Arch. Pharm.* **273** (1935) 357.
6. Brown, H. C. and McFarlin, R. F. *J. Am. Chem. Soc.* **78** (1956) 252.
7. Inouye, H., Inouye, S., Shimokawa, N. and Okigawa, M. *Chem. Pharm. Bull.* **17** (1969) 1942.
8. Inouye, H., Okigawa, M. and Shimokawa, N. *Chem. Pharm. Bull.* **17** (1969) 1949.
9. Cheymoll, J. *Bull. Soc. Chim. France* **1938** 642.
10. Bianco, A., Esposito, P., Guiso, M. and Scarpati, M. L. *Gazz. Chim. Ital.* **101** (1971) 764.
11. Chatterjee, A. and Parks, L. M. *J. Am. Chem. Soc.* **71** (1949) 2249.
12. Battersby, A. R., Hall, E. S. and Southgate, R. *J. Chem. Soc.* **C 1969** 721.
13. Karrer, P. and Salomon, H. *Helv. Chim. Acta* **29** (1946) 1544.

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