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2-Hydroxyvirensic Acid, a New Depsidone from the Lichen *Sulcaria sulcata**

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The new depsidone 2-hydroxyvirensic acid (4-formyl-2,3,8-trihydroxy-1,6,9-trimethyl-11-oxo-11*H*-dibenzo[*b*,*e*][1,4]dioxepin-7-carboxylic acid) (6) has been isolated from a chemical race of the lichen *Sulcaria sulcata* and the structure (6) deduced from a combination of spectroscopic data.

Keywords. Depsidone; lichen; mass spectrometry; natural product; n.m.r.

It appears likely that the majority of β-orcinol depsidones are derived from the common precursor, hypoprotocetraric acid (1), by stepwise oxidation reactions.[1–4] Secondary *O*-methylation, nuclear chlorination, decarboxylation or side-chain esterification can be observed at various oxidation levels, ultimately leading to the vast array of naturally occurring derivatives.[5] As a consequence a series of parallel biosynthetic schemes of sequentially related β-orcinol depsidones can be constructed, and Scheme 1 illustrates this relationship in the virensic acid chemosyndrome.

In convirensic acid (2) and virensic acid (3) the 4-methyl group of (1) has undergone sequential oxidation. Subsequent *O*-methylation and nuclear chlorination leads to methyl virensate (4)[6,7] and physciosporin (5),[6,7] further members of the virensic acid chemosyndrome.

In this paper we describe the isolation and structural elucidation of 2-hydroxyvirensic acid (6), a highly derived member of this chemosyndrome.

Thin-layer chromatographic and high-performance liquid chromatographic analysis of a chemical race of the lichen *Sulcaria sulcata* (Lev.) Bystrek ex Brodo & Hawksw. indicated the presence of the common, cortical depside atranorin, virensic acid (3) and a major metabolite of unknown constitution. Subsequent extraction of this species, and the separation of the atranorin and virensic acid, led to the isolation of this compound, which we have now identified as 2-hydroxyvirensic acid (6). High-resolution mass spectrometry established the molecular formula of 2-hydroxyvirensic acid (6) as $C_{18}H_{14}O_9$, i.e. having an extra oxygen atom when compared with virensic acid (3).

The structure of 2-hydroxyvirensic acid (6) followed from the ^1H and ^{13}C n.m.r. spectra. The ^1H n.m.r. spectrum showed three *C*-methyl resonances (δ 2.20, 2.38, 2.69), an aldehyde proton (δ 10.76) and an intramolecularly hydrogen-bonded hydroxy signal (δ 11.85).

More particularly, the marked similarities between the ^1H and ^{13}C n.m.r. spectra of virensic acid (3), methyl virensate (4), physciosporin (5) and 2-hydroxyvirensic acid (6) (Tables 1 and 2), as well as the expected differences, were completely consistent with the formulation (6). Further information regarding the structure of (6) was obtained from the mass spectral fragmentation pattern. Fragment ions observed at *m/z* 195 and 193 arising from the A-ring confirmed that 2-hydroxyvirensic acid (6) was substituted by an additional hydroxy group in this ring.[8] The mass spectral fragments (Scheme 2) at *m/z* 357, 356, 330 and 301, corresponding to the loss of OH, H_2O, CO_2 and CO_2+CHO respectively from the molecular ion, provided further confirmation for this formulation for 2-hydroxyvirensic acid.[8] 2-Hydroxyvirensic acid (6) represents the second β-orcinol depsidone known, where the A-ring has undergone a subsequent nuclear oxidation (hydroxylation) reaction.[9]

Experimental

General

The general experimental details have been described previously.[10]

Extraction of Sulcaria sulcata *(Lev.) Bystrek ex Brodo & Hawksw.*

The lichen *Sulcaria sulcata* was collected on dead *Salix*, 8 km south of Tongjug village, 360 km east of Lhasa, near bend of the river Tsangpo, north side of Gyala Peri, Nyainqêntanglha Shan, Xizang

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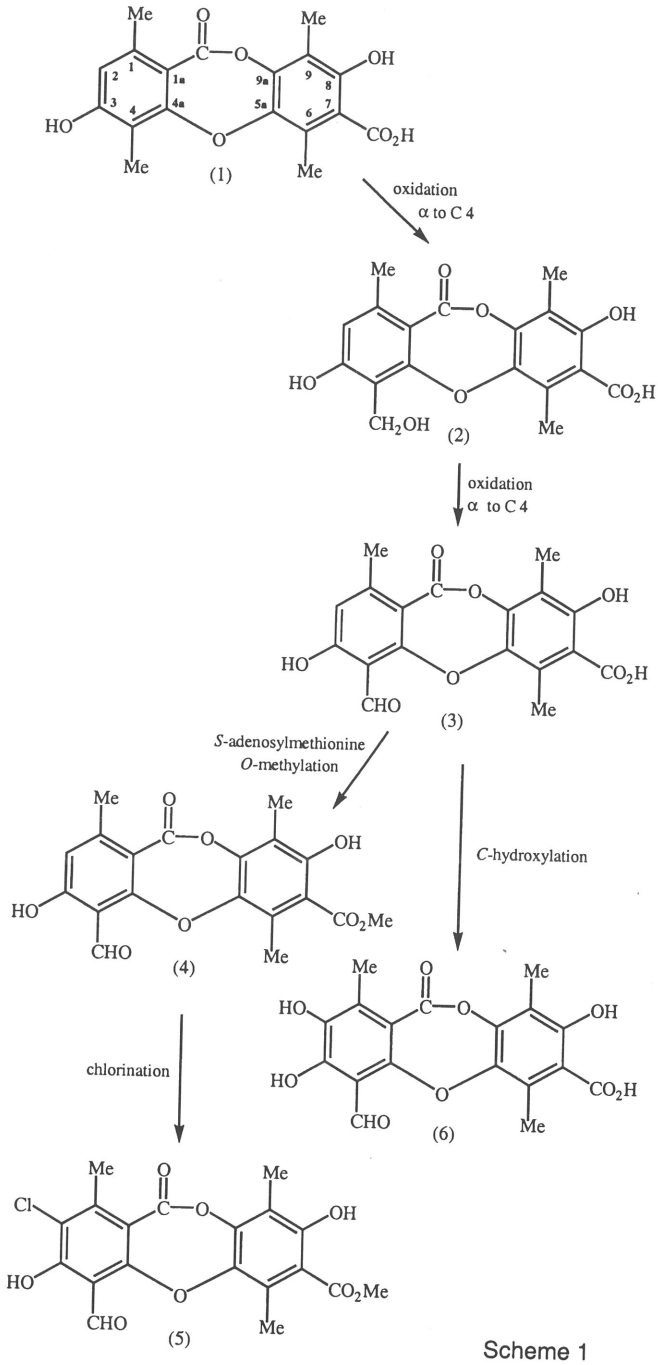


Scheme 1

Table 1. ^{1}H n.m.r. data (δ) for virensic acid (3), methyl virensate (4), physciosporin (5) and 2-hydroxyvirensic acid (6) in $(CD_3)_2CO$

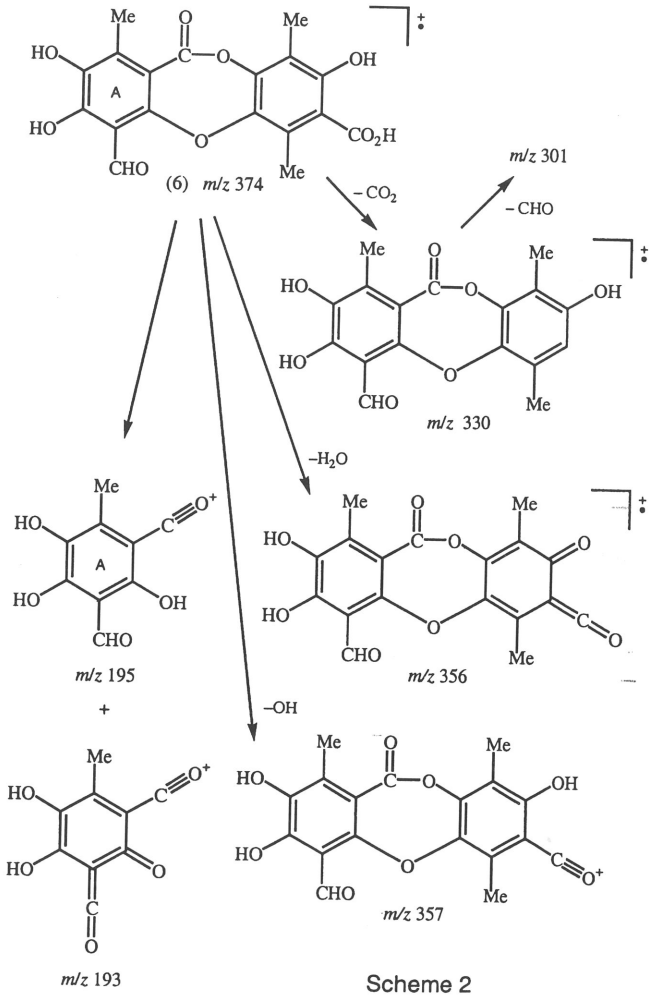
Protons	(3)	(4)	(5)	(6)
1-Me	2.52	2.51	2.54	2.38
6-Me	2.71	2.64	2.61	2.69
9-Me	2.21	2.27	2.20	2.20
4-CHO	10.79	10.80	10.79	10.76
H 2	6.82	6.84	—	—
7-CO_2Me	—	4.00	3.98	—
3-OH	12.27	12.26	12.97	11.85
8-OH	—	11.12	11.11	—

Table 2. ^{13}C n.m.r. data (δ) for virensic acid (3), methyl virensate (4), physciosporin (5) and 2-hydroxyvirensic acid (6) in $(CD_3)_2CO$

The assignment of chemical shifts bearing lower-case superscripts may be interchanged with shifts bearing the same superscript

Carbon	(3)	(4)[A]	(5)	(6)
1-Me	22.0	22.0	19.8	23.3
6-Me	15.6	15.5	15.5	15.6
9-Me	9.2	9.3	9.3	9.2
4-CHO	194.0	194.5	194.5	194.8
7-CO	173.4	172.1	171.6	173.5
7-OMe	—	53.1	53.1	—
C 1	154.5	154.5	150.6	153.9
C 1a	114.0	113.8	115.2	114.0
C 2	117.9	118.2	121.2	160.3
C 3	166.2	166.2	163.5	161.9[b]
C 4	114.0	111.9	112.0	112.8
C 4a	163.3[a]	161.8	161.8	167.4[b]
C 5a	143.1	143.3	143.3	142.5
C 6	133.5	130.2	130.3	130.8
C 7	110.2	111.5	111.7	111.5
C 8	157.5	158.5	158.5	157.8
C 9	117.0	117.2	117.3	117.0
C 9a	146.2	147.8	147.3	148.0
C 11	163.7[a]	161.8	161.8	163.4

[A] Assignments confirmed by ^{1}H–^{13}C HMQC, ^{1}H–^{13}C HMBC and DEPT experiments.



Scheme 2

Province, Tibet, China, 29° 56′ N, 94° 54′ E, 3350 m altitude, *W. Obermayer 6848*, 18 August 1994 (GZU).

The dried lichen thallus (0.7 g) was extracted in a Soxhlet extractor with anhydrous ether for 48 h. The ethereal solution was concentrated to 100 ml and filtered to remove the precipitate of atranorin. The solution was then evaporated to dryness and the residue (32 mg) was crystallized from acetone/light petroleum to yield *2-hydroxyvirensic acid* (6) (2.5 mg, 0.4%) as colourless microcrystals, m.p. >350° (dec.) (Found: mol. wt 374.0632. $C_{18}H_{14}O_9$ requires mol. wt 374.0637). The homogeneity of this compound was confirmed by high-performance liquid chromatography and ^{1}H n.m.r. spectroscopy. Mass spectrum: *m/z* 375 (17%), 374 (M, 86), 357 (18), 356 (70), 340 (12), 330 (35), 328 (42), 313 (20), 312 (30), 301 (25), 288 (17), 274 (15), 273 (16), 272 (20), 261 (11), 258 (21), 257 (11), 245 (13), 196 (43), 195 (63), 180 (46), 179 (31), 178 (19), 177 (11), 166 (20), 165 (35), 164 (73), 163 (14), 152 (23), 151 (17), 150 (24), 149 (11), 145 (13), 143 (11), 138 (14), 137 (17), 136 (83), 135 (21), 133(13), 131 (12), 129 (14), 125 (14), 124 (11), 123 (18), 122 (18), 121 (22), 119 (13), 117 (11), 115 (19), 111 (16), 110 (15), 109 (29), 108 (22), 107 (36), 106 (19), 105 (35), 103 (25), 67 (100). Standard t.l.c. R_F values:[11,12] R_F (A) 0.24; R_F (B′) 0.45; R_F (C) 0.27. Standard h.p.l.c.:[13,14] RI 16; RT 22.94 min.

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