

^{13}C NMR Spectra of Lichen Xanthenes. Temperature Dependent Collapse of Long-range Couplings to Hydrogen-bonded Hydroxyl Protons ^{1,2}

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^{13}C – ^1H Spin-spin coupling to the C-1 hydroxyl proton facilitated analysis of the single-resonance ^{13}C NMR spectrum of norlichexanthone *1*, the parent compound of all lichen xanthenes. The substituent effects of chlorination and *o*-methylation of *1* were studied and permitted interpretation of the spectra of seven lichen xanthenes. A temperature dependent collapse of the long-range couplings to the C-1 hydroxyl proton was found in the spectra of xanthenes with chloro substituents and is ascribed to an increase in the rate of exchange of the C-1 hydroxyl proton catalysed by the acidic protons in positions 3 and 6.

Only a few ^{13}C NMR spectra of naturally occurring xanthenes have been reported ³ and, since the oxygenation pattern for this group of

compounds varies considerably,⁴ each type has to be examined separately for successful interpretation of the spectra.

Norlichexanthone *1*, which has a 1,3,6-oxygenation pattern, is the parent compound of the lichen xanthenes ⁵ and for a few xanthenes of fungal origin ⁶ (e.g. griseoxanthenes B and C, compounds *8* and *4* in Table 1). ^{13}C NMR spectroscopy should be particularly useful for the structure determination of lichen xanthenes. Most lichen xanthenes are chlorinated (with up to four nuclear chlorine atoms) and chloro-substitution in simple aromatics is known to cause a down-field shift of approximately 6 ppm for the C-1 carbon, whereas the carbons in the *meta* positions are virtually unaffected.⁷

RESULTS AND DISCUSSION

The single resonance spectrum of *1* is presented in Fig. 1 (signals for CO, CH₃, and C-8 are excluded, since their assignment was trivial). Among the ring carbons, those bearing an oxygen substituent are expected to resonate at the lowest field (155–165 ppm) whereas the others are found in the region 90–120 ppm. Comparison between (a) and (b) in Fig. 1 shows that four carbons (C-1, C-2, C-3 and C-9a) are coupled to the C-1 hydroxyl, since the four multiplets change their pattern upon addition of D₂O. This phenomenon is attributable to the slow rate of exchange of the C-1 hydroxyl proton on the NMR time scale due to hydrogen bonding

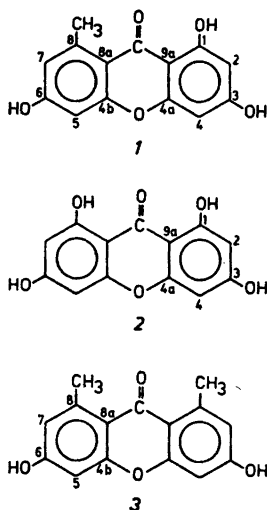


Table 1. ¹³C NMR shift values for norlichexanthone and related compounds.

Compound	C-1	C-2	C-3	C-4	C-4a	C-4b	C-5	C-6	C-7	C-8	C-8a	C-9a	Me	CO	OMe	Temp. °C
1 (parent)	163.2	97.9	164.4	93.3	156.6	158.6	100.6	162.5	116.0	142.7	111.0	102.2	22.9	181.3	181.3	50
2	161.9	98.4	165.5	94.1	157.0	157.9	100.1	161.0	115.6	142.5	113.8	100.3	23.0	181.8	181.8	50
3	162.6	96.3	165.2	91.5	156.1	158.6	100.2	162.6	116.0	142.5	110.6	102.8	22.5	181.1	55.5	50 ^a
4 (3-OMe)	163.0	97.6	164.2	92.8	156.1	158.1	98.3	162.7	114.3	141.9	111.7	102.2	21.6	180.7	55.0	130 ^a
5 (6-OMe)	158.2	101.4	158.7	92.2	153.9	159.8	100.3	162.8	116.1	142.6	110.4	102.1	22.7	180.9	50	50
6 (2-Cl)	160.7	97.7	159.8	96.6	151.3	158.2	100.2	162.6	116.2	142.5	110.2	102.5	22.6	180.7	50	50
7 (4-Cl)	161.0	94.4	162.7	97.3	150.5	158.1	100.2	160.1	116.1	142.5	110.0	102.8	22.0	180.6	56.2	85 ^a
8 (4-Cl, 3-OMe)	162.9	97.7	164.7	93.1	156.3 ^c	156.1 ^c	101.1	158.4	119.3	139.2	111.2	102.1	17.9	180.8	50	50
9 (7-Cl)	156.3	102.7	155.4	98.0	149.2	158.0	100.2	162.9	116.5	142.6	109.7	102.1	22.6	180.1	50 ^a	50 ^a
10 (2,4-diCl)	158.0	101.6	159.9	93.1	153.4	156.0	100.1	158.4	119.4	139.0	110.5	101.8	17.8	180.2	50 ^a	50 ^a
11 (2,7-diCl)	160.4	97.8	160.0	96.7	150.9	155.6	100.8	158.3	119.5	139.0	110.5	101.2	17.6	180.1	50 ^a	50 ^a
12 (4,7-diCl)	160.5	98.2	160.6	97.2	151.3	153.5	105.0	158.7	115.7	140.0	111.1	102.2	22.5	180.6	50	50
13 (4,5-diCl)	166.1	103.1	155.9	98.5	149.1	153.2	105.0	158.8	115.7	140.1	110.6	101.9	22.4	180.1	50	50
14 (2,4,5-triCl)	155.8	103.0	155.9	98.4	148.6	153.0	105.0	158.7	115.8	140.0	110.5	101.8	21.3	180.0	60.3	130 ^{a,b}
15 (2,4,5-triCl,3-OMe)	155.9	103.4	156.2	98.5	148.6	151.2	106.9	154.8	120.5	136.9	110.6	101.7	17.7	179.6	50	50
16 (2,4,5,7-tetraCl)																

^a Spectra measured in micro probe. ^b Part of the sample decomposed. ^c Assignments may be reversed.

to the carbonyl group.^{8,9} The quartet for C-1 arises from coupling to the C-2 proton ($^2J = 3.0$ Hz, geminal coupling; all values given are ± 0.6 Hz) and to the C-1 hydroxyl proton ($^3J = 4.9$ Hz). The doublet of overlapping triplets for C-3 is attributed to equal couplings to the C-2 and C-4 protons ($^2J = 3.0$ Hz) and to the C-1 hydroxyl proton ($^4J = 1.8$ Hz). Carbon C-2 appears as a doublet ($^1J = 162.4$ Hz) of triplets. The broadening of the triplets is caused by unequal coupling to the C-4 proton ($^3J = 4.3$ Hz) and to the C-1 hydroxyl proton ($^3J = 6.1$ Hz). The quartet for C-9a, which collapses to a triplet upon addition of D_2O , is attributed to equal couplings to the C-2, C-4 protons and to the hydroxyl proton ($^3J = 4.9$ Hz).

The unresolved multiplets for C-7 ($^1J = 161.1$ Hz) and C-8a arise from vicinal coupling and coupling to the CH_3 -group. The doublet of doublets for C-4 ($^1J = 168.8$ Hz, $^3J = 3.7$ Hz) and C-5 ($^1J = 163.6$ Hz, $^3J = 3.7$ Hz) were assigned by comparison with model substances 2 and 3 (see Table 1; for comparison purposes the carbon numbering is referred to the equivalent carbon in 1). C-4a and C-4b could also be distinguished by comparison with 2 and 3 but, since the shift differences are small, a more reliable assignment was obtained by studying substituent effects (see below). It is noteworthy that coupling to the C-1 hydroxyl proton was not observed for C-4a, which couples to the C-4 proton only ($^2J = 3.7$ Hz); C-4b, on the other hand, appears as a distorted triplet caused by equal couplings to the protons in positions 5 and 7 ($^2J = ^4J = 2.4$ Hz).

Addition of D_2O caused an up-field shift for the C-1 carbon (-0.55 ppm) but with C-2 and C-9a, in contradiction to reports for similar compounds,⁸ a down-field shift was observed ($+0.05$ and $+0.15$ ppm respectively). The latter values are small and the effects therefore difficult to interpret (e.g. C-6 is shifted -0.04 ppm).

Table 1 lists the noise- and off-resonance decoupled spectra of 14 xanthenes of type 1 in $DMSO-d_6$. Due to the low solubility of the substances and for comparison purposes most spectra were recorded at $50^\circ C$, but in some cases even higher temperatures had to be used. To facilitate analysis of the natural products, three monochloro- (6, 7 and 9) and two dichloro-xanthenes (11 and 12) were used as reference

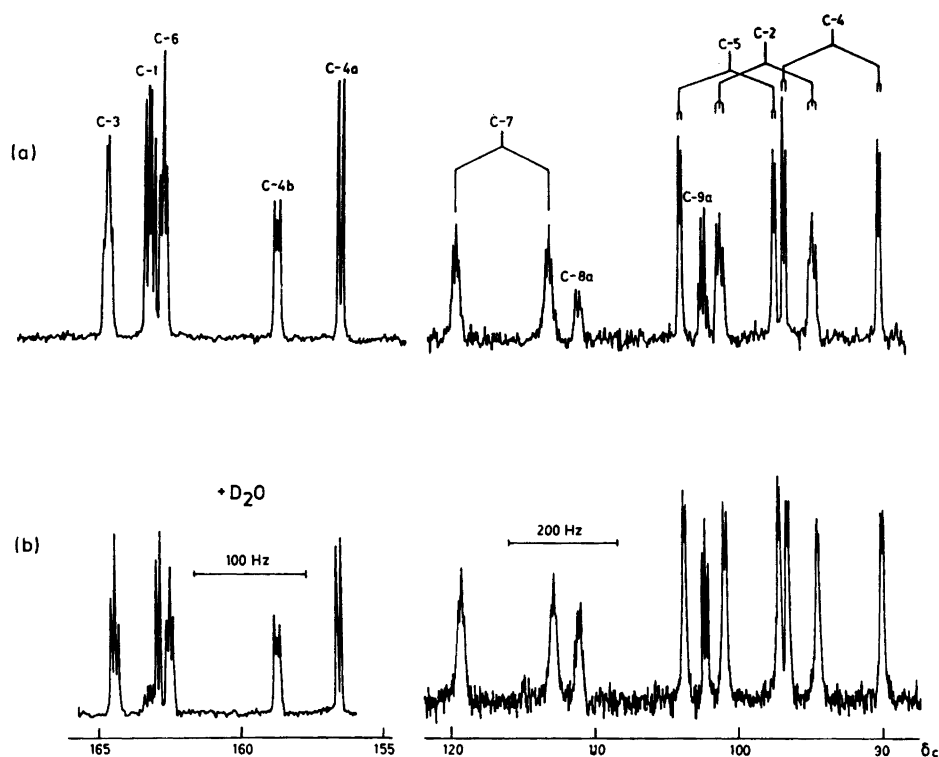


Fig. 1. 25 MHz proton-coupled ^{13}C NMR spectra of *1* in $\text{DMSO}-d_6$ at 25°C . (a) without and (b) with addition of 10 % D_2O .

substances. Since several lichen xanthenes are methyl ethers, the substituent effect of a methoxy group was examined separately (compounds *4* and *5*). Analysis of the shift values for the oxygen-substituted carbons was aided by the use of single-resonance spectra of *6*, *7* and *9*. In all cases the additivity principle was found to hold and therefore structures for other lichen xanthenes may easily be determined.

Of particular analytical value is the large shift difference observed for carbons C-5 and C-7, since protons in these positions are not easily distinguished by ^1H NMR.² Thus it is clearly seen that arthothelin *14*¹⁰ and thuringione *15*¹¹ both have the 2,4,5-trichloro pattern (formerly given a 2,4,7 pattern) and that the proper structure of the dichloro-xanthone of *Lecanora straminea*¹² is 4,5-dichloronorlichexanthone *13* (formerly called 2,7-dichloronorlichexanthone). These findings have recently been confirmed by total syntheses of lichen xanthenes.^{2,13}

As shown in Table 1, chloro-substitution causes a down-field shift of 3.3–4.4 ppm, typically 3.3–3.5 ppm, for the carbon to which the chlorine atom is attached, whereas the *meta* carbons are unaffected. The *ortho*-effect of the halogen is large (3.5–5.8 ppm up-field) and in this case it is partly explained as a steric effect observed when a substituent is flanked by two *ortho* substituents.¹⁴ This steric effect is particularly pronounced for the carbon of the methyl group, which is shifted up-field 5.0–5.3 ppm. The *para*-effect of a chloro substituent is smaller (2.5–3.3 ppm up-field), but still significant, and in this case could be explained by mesomeric effects.

The *ortho*-effect of a methoxy substituent in compounds *4* and *5* is an up-field shift of 1.6–2.3 ppm. A chloro substituent in the *ortho* position, however, opposes this effect as is seen for carbons C-2 and C-4 in *15* and C-4 in *8*. In the case of *15* a marked shift to lower field (4.8 ppm relative to that of *4*) is observed for

the methoxy carbon. This is opposite to that found for the C-8 methyl and, although the influence of two *ortho*-substituents must be considered, this effect is not understood at present.¹⁵

The carbonyl absorptions show only small variations in shift-values and are therefore of little analytical value. A small up-field shift, however, was observed for the more heavily substituted xanthenes.

A temperature dependence of the rate of exchange of the hydrogen-bonded hydroxyl proton was observed for the xanthenes with chloro substituents. At 50 °C coupling was seen for all compounds examined, but disappeared, for example at 75 °C for 13 and at 130 °C for 7. No attempts were made to elucidate this dependence, but in general lower temperatures were required for the more highly chlorinated xanthenes. A similar effect of temperature on ¹³C-¹H couplings was observed for methyl salicylate⁹ but, in that case, the dependence was attributed to rotation of the carbonyl group.¹⁶ With the rigid xanthenes an increased rate of exchange of the hydroxyl proton is responsible for the loss of coupling, and this exchange could also be observed as a broadening of the low-field proton signal in the ¹H NMR spectra. With chloro substitution, the acidity of the hydroxyl protons in positions 3 and 6 increases and this could catalytically enhance the rate of exchange of the hydroxyl proton. The exchange-rate was found to be dependent on concentration, but addition of 14 to a dilute solution of 7 caused a significant broadening of the hydroxyl proton signal in the ¹H NMR spectrum of 7. The influence of trace amounts of water in the solvent (commercial DMSO-*d*₆ was used) cannot be excluded, but this is not a dominant factor since 1, with no chloro substituents, did not show this temperature dependence below 150 °C. The dependence on temperature must be considered when examining similar compounds of low solubility at lower temperatures and offers an alternative to the method of analysing coupling to hydrogen-bonded protons by addition of D₂O.

EXPERIMENTAL

The ¹³C NMR spectra were recorded on a JEOL-FX 100 spectrometer, operating at

25.05 MHz. The deuterium signal of the DMSO-*d*₆ solvent was used as the lock-signal. Solutions were 0.3–0.6 M and recorded in 5 mm and 1.7 mm outer diameter tubes. δ Values, in ppm down-field from TMS, were converted using [$\delta(\text{TMS}) = \delta(\text{DMSO-}d_6) + 39.5$ ppm]. The syntheses of compounds 1, 4–9, 11 and 12 have been performed at this Institute^{2,17} and compounds 10, 13–16 were of lichen origin.^{10–12}

1,3,6,8-Tetrahydroxyxanthone 2, was prepared from 2,4,6-tribenzyloxybenzoic acid¹⁸ and 1,3,5-tribenzyloxybenzene² by improved methods. Total yield 88 %, m.p. 347–349 °C (lit.¹⁸ 350 °C.).

3,6-Dihydroxy-1,8-dimethylxanthone 3, was obtained by heating 100 mg of 2,2',4,4'-tetrahydroxy-6,6'-dimethylbenzophenone¹⁹ with 5 ml of water in a sealed tube at 205–215 °C for 2 h. TLC on silica gel (Merck precoated plates, 0.5 mm) with dichloromethane–acetone (4:1) as eluent yielded 21 mg (22 %) of 3 (the rest consisted mainly of orcinol). The product was recrystallized from benzene, m.p. 303–305 °C (dec.). Anal. C₁₅H₁₂O₄: C, H. MS (IP 70 eV): 256 (M). IR (KBr): 1592 (s), 1604 (s), 1620 (s) cm⁻¹. ¹H NMR [60 MHz, (CD₃)₂CO]: δ 2.77 (6 H, s) 6.65 (4 H, s), 9.32 (2 H, s).

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REFERENCES

1. Part 35 of the series Chemical Studies on Lichens.
2. Part 34: Sundholm, E. G. *Tetrahedron. In press.*
3. Castela, Jr., J. F., Gottlieb, O. R., DeLima, R. A. and Mesquita, A. L. *Phytochemistry* 16 (1977) 735, and references therein.
4. Carpenter, I., Locksley, H. D. and Scheinmann, F. *Phytochemistry* 8 (1969) 2013.
5. Culberson, C. F. *Chemical and Botanical Guide to Lichen Products*, Univ. of N. Carolina Press, Chapel Hill 1969.
6. a. McMaster, W. J., Scott, A. I. and Trippett, S. *J. Chem. Soc.* (1960) 4628; b. Rhodes, A., Boothroyd, B., McGonagle, M. P. and Somerfield, G. A. *Biochem. J.* 81 (1961) 28.
7. Levy, G. C. and Nelson, G. L. *Carbon-13 Nuclear Magnetic Resonance for Organic Chemists*, Wiley, New York 1972.
8. Wehrli, F. W. *Chem. Commun.* (1975) 663.
9. Chang, Ching-er *J. Org. Chem.* 41 (1976) 1881.
10. Santesson, J. *Ark. Kemi* 30 (1969) 449.
11. Huneck, S. and Santesson, J. *Z. Naturforsch. Teil B* 24 (1969) 756.
12. Santesson, J. *Ark. Kemi* 30 (1969) 455.

13. Independent of this work, Huneck has come to the same conclusions regarding the structures of 14 and 15. *Personal communication*.
14. Stothers, J. B. *Carbon-13 NMR Spectroscopy*, Academic, New York 1972.
15. Dhimi, K. S. and Stothers, J. B. *Can. J. Chem.* 44 (1966) 2855.
16. For a general discussion, see Grunwald, E. and Ralph, E. K., in *Dynamic Nuclear Magnetic Resonance Spectroscopy*, Academic, New York 1975, Chapter 15.
17. Sundholm, G. *Acta Chem. Scand. B* 28 (1974) 1102.
18. Grover, P. K., Shah, G. D. and Shah, R. C. *Chem. Ind. London* (1955) 62.
19. Hendrickson, J. B., Ramsay, M. V. and Ross Kelly, T. *J. Am. Chem. Soc.* 94 (1972) 6834.

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