

Studies on the Chemistry of Lichens

21.* The Isolation and Synthesis of Methyl 3,5-Dichlorolecanorate, a New Depside from *Ramalina* sp.

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A new depside has been isolated from the *Ramalina ceruchis* group and identified as methyl 3,5-dichlorolecanorate, the synthesis of which is also described.

In connection with an investigation concerning the chemical constituents of the *Ramalina ceruchis* group, chromatographic evidence indicated the presence of a previously unknown depside in the chloroform extracts of *R. flaccescens* Nyl. and of one of the two chemical strains of *R. ceruchis* (Ach.) DNot.¹

The depside was isolated from *R. ceruchis* in a poor yield by preparative thin layer chromatography. It has been obtained in a far better yield from *R. flaccescens*, the purification here achieved by crystallisation.

The depside melted at 177–177.5° (decomp.). It gave a red colour reaction with ferric chloride, but showed no distinct acidic properties. The analytical values agreed with an empirical composition of $C_{17}H_{14}Cl_2O_7$.

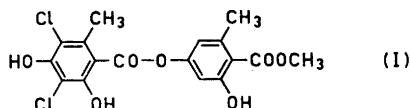
The ultraviolet spectrum in ethanol exhibited two well defined maxima at 259 μ and 318 μ thus suggesting an orcinol depside structure.² The infrared spectrum showed carbonyl bands at 1685 and 1715 cm^{-1} . The NMR spectrum indicated the presence of two aromatically bound methyl groups as well as of one methyl ester or methyl ether group. No other aliphatic protons were detected.

On methanolysis, the depside yielded two phenolic components, separated by preparative thin layer chromatography. One of the methanolysis products was found to be free from chlorine and was identified as methyl orsellinate. The identity of the second component was established as methyl 3,5-dichloro-

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orsellinate through comparison with an authentic specimen, synthesised as described by Fujikawa *et al.*³

On brief treatment with boiling water the depside afforded two hydrolysis products as established by thin layer chromatography. One of these was isolated by preparative thin layer chromatography and was identified as methyl orsellinate. These results leave methyl 3,5-dichlorolecanorate (I) as the most reasonable structure of the depside.



Consistently, the naturally occurring depside was proved to be identical with a synthetic specimen of methyl 3,5-dichlorolecanorate obtained in a fair yield by chlorination of methyl lecanorate with the calculated amount of sulphuryl chloride.

Among the known chlorine-containing lichen compounds the two depsidones, gangaleoidin⁴ and vicanicin⁵ both have their chlorine atoms in the "acid" component of the molecule. Thus it might be suggested that the chlorination in Nature occurs after the formation of the depside linkage.

EXPERIMENTAL

All melting points are uncorrected. The thin layer chromatography was carried out according to Stahl,⁶ using silica gel G as adsorbent and chloroform-acetone (1:1, v/v) as solvent. UV light (365 m μ) and bis-diazotized benzidine were used to reveal the spots.

The origin of the lichen material has been reported previously.¹

Isolation of methyl 3,5-dichlorolecanorate. Air-dried and powdery *R. flaccescens* (7.0 g) was extracted in darkness with chloroform (3 \times 70 ml) at 50° for 30 min. The combined extracts were concentrated in vacuum and the separated crystals were collected (0.27 g = 3.8 %). After crystallisation in darkness from dry acetone, a compound was obtained as needles, m.p. 175–176° (decomp.) (0.16 g = 2.2 %). It was identical (mixed melting point, IR spectra) with the depside isolated as follows from *R. ceruchis*. The chloroform extract from *R. ceruchis* was applied on the plate as a thin band. After development, the zone containing the band with $R_F = 0.45-0.55$ was removed and extracted with acetone. Evaporation of the extract yielded crystals (m.p. 169–172°) (decomp.) (0.002 g = 0.1 %). An analytical sample of the depside prepared by crystallisation from dry benzene in darkness melted at 177–177.5° (decomp.). (Found: C 50.4; H 3.52; Cl 17.5. C₁₇H₁₄Cl₂O₇, requires: C 50.9; H 3.52; Cl 17.7). This compound showed no depression on admixture with methyl 3,5-dichlorolecanorate synthesised as described below.

The depside was insoluble in sodium hydrogen carbonate. An ethanolic solution of the compound gave a red colour with ferric chloride. With Gibb's reagent (2,6-dichloroquinone-chloroimide⁷) a blue colour was obtained and with bis-diazotized benzidine a red-brown colouration.

Ultraviolet spectrum in ethanol (95 %) ($c = 5 \times 10^{-5}$ M): $\lambda_{\max} = 259$ m μ (log $\epsilon = 4.2$), $\lambda_{\max} = 318$ m μ (log $\epsilon = 4.1$), $\lambda_{\min} = 244$ m μ (log $\epsilon = 4.1$), $\lambda_{\min} = 286$ m μ (log $\epsilon = 3.9$).

The infrared spectrum (KBr-phase) had the following main bands in cm⁻¹: 3400, 1715, 1685, 1620, 1600, and 1575 (Perkin-Elmer 237).

NMR spectrum in CDCl₃-pyridine (9:1 v/v): signals at 2.53, 2.70, and 3.91 ppm, integrated 1:1:1 (internal reference TMS).

Methanolysis of the isolated depside. The depside (7 mg) was dissolved in methanolic potassium hydroxide (0.5 ml 0.001 M) and kept at 50° in a nitrogen atmosphere for 30 min. By means of thin layer chromatography three compounds with R_F values = 0.38, 0.51, and 0.62 were isolated from the reaction mixture. The compound with $R_F = 0.62$ was identified as methyl orsellinate through comparison with an authentic sample (IR, mixed m.p. 137 – 139°). The compound with $R_F = 0.38$ was identified as methyl 3,5-dichlororsellinate through comparison with a synthetically prepared sample³ (IR, mixed m.p. 110 – 113°). The compound with $R_F = 0.51$ corresponded to unchanged starting material.

Hydrolysis of the depside. A suspension of the depside (5 mg) in water (1 ml) was heated to 100° for 30 min. The cooled reaction mixture was extracted with chloroform and the extract chromatographed. Spots with $R_F = 0.06$, 0.50, and 0.61 were obtained. The compound with $R_F = 0.61$ was isolated through preparative thin layer chromatography and identified as methyl orsellinate (IR, mixed m.p. 136 – 138°). The compound with $R_F = 0.50$ was the starting material.

Synthesis of methyl 3,5-dichlorolecanorate. Methyl lecanorate (2.15 g, 6.5 mmole) was dissolved in dry ether (80 ml) the solution cooled to 0°, and sulphuryl chloride (1.85 g, 13.7 mmole) in dry ether (10 ml) added dropwise under stirring. After 30 min, the solution was slowly heated to 25°. After about 10 min a white precipitate appeared which was filtered off, washed with small portions of sodium hydrogen carbonate solution (1 %), water and then dried. Crystallisation from dry benzene in darkness yielded methyl 3,5-dichlorolecanorate (0.85 g), m.p. 175 – 176° (decomp.). The remaining ethereal solution after washing with sodium hydrogen carbonate solution (1 %) (2 × 50 ml) and water (3 × 50 ml), evaporation, and repeated crystallisations from benzene in darkness yielded another fraction of methyl 3,5-dichlorolecanorate (0.70 g), m.p. 174 – 175.5°. (Total yield 1.55 g = 60 % of the theoretical amount).

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