Information about the Structure of Phthiocerol from Mass Spectral and X-Ray Crystallographic Data

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Phthiocerol, the optically active wax alcohol found in the waxes of several types of tubercle bacilli [1,2] has the empirical composition $C_{34}H_{67}(OH)_2OCH_3$. The carbon skeleton of phthiocerol corresponds to 4-methyltritriacontane [3-5]. Recent attempts to determine the structure of phthiocerol by degradative methods have led Demarteau-Ginsburg and Lederer [6], and Hall and Polgar [7] to formulate phthiocerol as 4-methyl-5-methoxy-8,10-dihydroxytritriacontane and 4-methyl-4-methoxy-9,10-dihydroxytritriacontane,respectively.

The above formulations appeared to be at variance with the results of monolayer experiments [8] which suggested that one of the polar groups must be situated near one end of the chain. We have therefore tried to obtain further evidence on the structure of phthiocerol from a study of its mass spectrum and X-ray diffraction pattern [9,10].

A striking feature of mass spectrum is an extremely strong peak at mass number 73. The corresponding fragment must contain one oxygen atom, which in this case means that it must contain the intact methoxyl group. For this to be possible, the methoxyl group must occupy position 2 or 3 in the intact phthiocerol molecule. The strongest peak in the mass spectrum of methyl-*n*-heneicosyl ether is due to the fragment $CH_3—O—CH_2—$; by analogy position 3 appears more probable than position 2 for the methoxyl group in phthiocerol. There is evidence that elimination of methoxyl or hydroxyl groups in the fragmentation process in the mass spectrometer introduces a double bond in α,β-position to the carbon atom initially carrying the oxygen atom. A similar reaction mechanism would explain that Demarteau-Ginsburg and Lederer [6] were able to isolate methyl-*n*-propyl ketone after ozonization of the ene-diol obtained after removal of the methoxyl group.

The mass spectrum of phthiocerol shows further points of interest and may allow a direct determination of the position of the two hydroxyl groups when mass spectral data have been obtained for simple long chain glycols.

The X-ray diffraction pattern of phthiocerol [4] shows a series of orders of a long spacing of 46 Å with a characteristic intensity distribution. Using reasonable assumptions regarding the arrangements of the molecules in the lattice we have calculated the intensities of the 00*l* reflexions for different molecular models with the methoxyl group in position 2, 3, 4 and 5, respectively. Good agreement with experiment was found only when the methoxyl group occupied position 2 or 3.

Details of this work will be published later.

1. Stodola, F. H. and Anderson, R. J. *J. Biol. Chem.* **114** (1936) 467.
2. Cason, J. and Anderson, R. J. *J. Biol. Chem.* **119** (1937) 549.
3. Ställberg-Stenhagen, S., Stenhagen, E., Sheppard, N., Sutherland, G. B. B. M. and Walsh, A. *Nature* **160** (1947) 580.
4. Ställberg-Stenhagen, S. and Stenhagen, E. *J. Biol. Chem.* **173** (1948) 383.
5. Ställberg-Stenhagen, S. and Stenhagen, E. *J. Biol. Chem.* **183** (1950) 223.
6. Demarteau-Ginsburg, H. and Lederer, E. *Compt. rend.* **1955** 815.
7. Hall, J. A. and Polgar, N. *Chemistry & Industry* **1954** 1293.
8. Ställberg-Stenhagen, S. and Stenhagen, E. *J. Biol. Chem.* **143** (1942) 171.
9. The specimen of phthiocerol used was obtained from Professor R. J. Anderson of Yale university several years ago. The infrared spectrum showed the presence of a small amount of ketonic material, *cf.* Ref.[7] and Demarteau-Ginsburg, H., Ginsburg, A. and Lederer, E. *Biochim. et Biophysica Acta* **12** (1953) 587.
10. The mass spectrometer was built by one of us (R.R.). It has a heated intake system, *cf.* O'Neal, M. J. Jr. and Wier, T. P. Jr. *Anal. Chem.* **23** (1951) 830.