THE STRUCTURE OF LYCODINE[1]

W. A. Ayer and G. G. Iverach

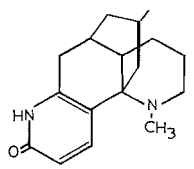
ABSTRACT

The molecular formula of the *Lycopodium* alkaloid lycodine has been revised to $C_{16}H_{22}N_2$. Transformation of β-obscurine (I) into N-methyl lycodine has shown that lycodine is the pyridine analogue of des-N-methyl-β-obscurine and is represented by structure II (R = H). The transformation of β-obscurine into α-obscurine is also described.

The alkaloid lycodine was first isolated from *Lycopodium annotinum* L. in 1958 by Anet and Eves (1), who assigned to it a molecular formula $C_{17}H_{24}N_2$. Lycodine was shown to be a tetracyclic diacidic base containing a secondary nitrogen atom, a 5,6,7,8-tetrahydroquinoline system unsubstituted in the heterocyclic ring, and a C-methyl group.

When structure I was advanced for β-obscurine (2) it was suggested that lycodine is formed by a similar biogenetic path,[2] with the exception that N-methylation has not occurred, and is represented by structure II (R = H). The correctness of this structure has now been confirmed by a series of experiments which are described in this communication.

The isolation of lycodine from *L. annotinum* L. involves a rather extended separation procedure (1). We have found that lycodine also occurs in *L. obscurum* L. and is readily separable by chromatography of the crude bases over alumina. Some lycodine was obtained directly by crystallization of the eluates. The remainder was separated from contaminating bases (mainly lycopodine and annotinine) by lithium aluminum hydride reduction, which transformed the contaminants into hydroxylic materials but did not affect the lycodine, followed by chromatography. The total yield of lycodine from dried plant material was approximately 0.003%.



I

II

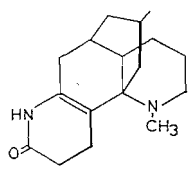
Structure II (R = H) requires that the formula of lycodine be revised to $C_{16}H_{22}N_2$. The analytical results quoted by Anet and Eves (1), especially those for the dipicrate of lycodine and the picrate of N-acetyl lycodine, do not clearly differentiate between the two possibilities. Our analytical results with lycodine and N-methyl lycodine, together with the preparation of N-methyl lycodine from β-obscurine ($C_{17}H_{24}ON_2$) described below, confirm the 16 carbon formulation. The proposed structure differs in two other respects from the partial structure advanced by Anet and Eves (1). Firstly, the more strongly basic nitrogen is attached to a carbon α to the pyridine ring. This attachment

[1]*Manuscript received June 13, 1960.*
Contribution from the Department of Chemistry, University of Alberta, Edmonton, Alberta. This work forms a portion of a thesis to be submitted by G. G. Iverach in partial fulfillment of the requirements for the degree of Master of Science.

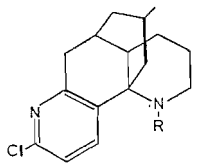
[2]*Recently, Professor H. Conroy (3) has outlined a biogenetic scheme which accounts for the formation, from acetate units and ammonia, of all the Lycopodium alkaloids of known structure.*

was regarded as improbable by the previous workers on the basis of a comparison of the pK_a's of the pyridine nitrogen atoms in lycodine and nicotine. We believe that the greater basicity (pK_a 3.97) of the pyridine nitrogen in lycodine as compared with that in nicotine (pK_a 3.35) is explained by the presence in lycodine of an electropositive substituent in the 2-position of the pyridine ring (4). Secondly, the C-methyl group of lycodine is not attached to a quaternary carbon atom. The assignment of the C-methyl group to such a grouping was made on the basis of the fact that the C-methyl peak in the nuclear magnetic resonance spectrum of lycodine is unsplit (1). The failure of the $\rangle CHCH_3$ peak to split is possibly attributable to the proximity of the aromatic ring, since in β-obscurine (I) the C-methyl peak is also unsplit, whereas in α-obscurine (III) the usual splitting occurs (2). In this connection, it is interesting to note that reduction of β-obscurine (I) with lithium and ammonia, a reagent which has been shown (5) to selectively reduce α-pyridones to the corresponding 3,4-dihydro-α-pyridones, yields α-obscurine (III).

It seemed at this point that structure II could be confirmed by direct transformation of β-obscurine (I) into lycodine (II, R = H) or, since some difficulty was anticipated in the removal of the N-methyl group of β-obscurine, into N-methyl lycodine (II, R = CH_3).



III



IV

Treatment of lycodine with formaldehyde and formic acid gave, in good yield, N-methyl lycodine (II, R = CH_3). The new base had no NH stretching band in the infrared and the ultraviolet spectrum was almost identical with that of lycodine. The nuclear magnetic resonance spectrum was similar to that of lycodine (1), with the addition of a strong (3-proton) signal at τ = 7.38 due to the N-methyl group (6). The C-methyl peak (at τ = 9.21) was again unsplit.

The transformation of an α-pyridone into the corresponding 2-chloropyridine is usually accomplished by the treatment with phosphorus oxychloride, phosphorus pentachloride, or a mixture of these two reagents (7). β-Obscurine was recovered unchanged after prolonged refluxing with phosphorus oxychloride and phosphorus oxychloride – phosphorus pentachloride. However, treatment with phosphorus pentachloride at 200° or, preferably, with phenylphosphonic dichloride (8) yielded an oily product whose spectral properties (ultraviolet maximum at 275 mμ, peak in the infrared at 1565 cm^{-1}) were consistent with those expected for the desired chloropyridine IV. The crude chloro compound was hydrogenated over platinum in acetic acid. Chromatography of the products gave N-methyl lycodine (41% yield from β-obscurine) and, unexpectedly, lycodine (23% yield). Presumably, loss of the N-methyl group occurred during the treatment with phenylphosphonic dichloride and the oily product was a mixture of the chloropyridines IV (R = CH_3) and IV (R = H). The appearance of a weak NH band at 3330 cm^{-1} in the infrared spectrum of the crude product is consistent with this view.

This transformation establishes that lycodine is the pyridine analogue of des-N-methyl-β-obscurine and, on the basis of the proposed (2) structure for β-obscurine, is represented by structure II (R = H).

EXPERIMENTAL

Ultraviolet spectra were measured in ethanol and, unless otherwise stated, infrared spectra in chloroform. Microanalyses are by Pascher Mikroanalytisches Laboratorium, Bonn, West Germany, and Micro-Tech Laboratory, Skokie, Illinois.

Isolation of Lycodine

Finely ground *L. obscurum* L. (10 kg) was extracted by the procedure of Manske and Marion (9). The crude alkaloids (9.3 g) yielded some lycopodine (2.0 g) on crystallization from ether. The mother liquors were dissolved in chloroform and placed on a column of basic alumina (150 g). Thorough elution with chloroform gave 3.75 g of basic material which was rechromatographed over alumina (100 g). Fraction A (2.1 g), eluted with benzene (900 ml), was mainly lycopodine. Fraction B (0.63 g), eluted with ether (850 ml), was a mixture of lycopodine and lycodine (as shown by infrared and ultraviolet measurements). Fraction C (0.17 g), eluted with 1:1 ether – methylene chloride (500 ml), contained annotinine, lycodine, and unidentified carbonyl-containing compounds. Crystallization of fraction B from pentane gave lycodine (172 mg). The residues from the crystallization were combined with fraction C, dissolved in ether, and reduced with lithium aluminum hydride in the usual manner. The product was chromatographed over alumina. Elution with benzene–ether gave crystalline lycodine (130 mg). The combined lycodine was recrystallized several times from pentane to give colorless blocks, m.p. 118–119°. Calc. for $C_{16}H_{22}N_2$: C, 79.29; H, 9.15; N, 11.56%. Found: C, 79.49, 79.33; H, 9.13, 9.09; N, 11.61, 11.47%; no N-methyl.

N-*Methyl Lycodine*

Lycodine (52 mg) was dissolved in 98% formic acid (0.25 ml) and formalin (0.25 ml) added. The solution was refluxed for 2 hours, then maintained at 50° for 12 hours. The reaction mixture was diluted with water (50 ml), basified with concentrated ammonium hydroxide, and extracted several times with chloroform. Evaporation of the dried chloroform extracts gave a pale yellow oil (0.05 g) which solidified on standing. Three recrystallizations from acetone gave N-methyl lycodine as colorless blocks (23 mg), m.p. 91–92°. Calc. for $C_{17}H_{24}N_2$: C, 79.63; H, 9.44; N, 10.93; one N—CH_3, 5.86%. Found: C, 79.61, 79.59; H, 9.36, 9.27; N, 11.25, 10.75; N—CH_3, 5.19%. Ultraviolet spectrum: λ_{max} 268 mμ (log ϵ = 3.61), shoulder at 275 mμ (log ϵ = 3.49). Infrared spectrum (Nujol): γ_{max} 3040, 1574, 1472, 812, 737 cm^{-1} (pyridine ring).

N-*Methyl Lycodine and Lycodine from β-Obscurine*

A solution of β-obscurine (168 mg, isolated from *L. annotinum* L. by established procedures (10, 11)) in phenylphosphonic dichloride (50 ml) was maintained at 200–210° for 45 minutes. The reaction solution was decomposed with ice and water and made basic by the addition of concentrated ammonium hydroxide. Chloroform extraction yielded the crude chloro compounds IV (R = CH_3) and IV (R = H) as a pale yellow oil (0.16 g), ultraviolet maximum 275 mμ, infrared peaks at 3330 cm^{-1} (NH) and 1565 cm^{-1} (pyridine ring). The oil was dissolved in glacial acetic acid and shaken with hydrogen (50 p.s.i.) in the presence of Adam's catalyst (100 mg) for 3 hours. The catalyst was filtered off and the filtrate concentrated, diluted with water, made basic with ammonium hydroxide, and extracted thoroughly with chloroform. Removal of the chloroform left a viscous yellow oil (0.12 g) which was chromatographed over basic alumina (3 g). Elution with benzene and crystallization from acetone gave N-methyl lycodine (65 mg), m.p. 91–92°. This did not depress the melting point of the N-methyl compound prepared from lycodine and their infrared spectra were identical.

Elution of the column with ether yielded a fraction (35 mg) which crystallized from acetone as colorless blocks, m.p. 115–116°. The infrared spectrum was identical with that of lycodine and a mixture melting point was undepressed.

α-Obscurine from β-Obscurine

A solution of β-obscurine (31 mg) in dry tetrahydrofuran (100 ml) was added during 1/2 hour to a vigorously stirred solution of lithium (0.1 g) in ammonia (40 ml). The solution was stirred for a further $2\frac{1}{2}$ hours, then the reaction mixture was evaporated to dryness. The residue was dissolved in aqueous HCl, washed with chloroform, basified, and extracted with chloroform. Evaporation of the chloroform left a solid residue which was chromatographed over alumina (1 g). Elution with chloroform and crystallization from methanol gave α-obscurine (20 mg), m.p. 285–287° (uncorrected). The mixture melting point with authentic α-obscurine was undepressed, and the infrared spectra of the two were identical.

Further elution of the column with chloroform–methanol (4:1) gave a fraction (7 mg) whose spectral properties were identical with those of β-obscurine.

ACKNOWLEDGMENTS

We wish to thank the National Research Council for financial support, Professor Z. Valenta for the nuclear magnetic resonance spectrum, and Mr. R. N. Swindlehurst for many of the ultraviolet and infrared spectra.

REFERENCES

1. F. A. L. Anet and C. R. Eves. Can. J. Chem. **36**, 902 (1958).
2. W. A. Ayer and G. G. Iverach. Tetrahedron Letters. In press.
3. H. Conroy. Tetrahedron Letters. In press.
4. E. A. Braude and F. C. Nachod. Determination of organic structures by physical methods. Academic Press, New York. 1955. p. 597.
5. J. A. Berson and J. S. Walia. J. Org. Chem. **24**, 756 (1959).
6. L. M. Jackman. Applications of nuclear magnetic resonance spectroscopy in organic chemistry. Pergamon Press, New York. 1959. p. 56.
7. R. C. Elderfield. Heterocyclic compounds. Vol. 1. John Wiley and Sons, New York. 1950. p. 513.
8. Z. Valenta, H. Yoshimura, E. F. Rogers, M. Ternbah, and K. Wiesner. Tetrahedron Letters. In press.
9. R. H. F. Manske and L. Marion. Can. J. Research, B, **20**, 87 (1942).
10. R. H. F. Manske and L. Marion. Can. J. Research, B, **21**, 92 (1943).
11. B. P. Moore and L. Marion. Can. J. Chem. **31**, 952 (1953).