LYCOPODIUM ALKALOIDS

PART VIII. NEW ALKALOIDS FROM JAMAICAN LYCOPODIUM SPECIES¹

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ABSTRACT

Four more alkaloids have been isolated from Lycopodium fawcettii. Lycodine and des-Nmethyl- α -obscurine have been found in other Lycopodium species but base L, C₁₈H₃₁NO₄, and base R, C₁₆H₂₄N₂O₂, are new. Some transformations of base L are described including its conversion to the previously described base O. An unstable base from L. clavatum has been characterized as the picrate C₁₅H₂₅NO₃. C₆H₃N₃O₇. Further characterization of the two bases fawcettidine (base F) and fawcettimine (base A) is reported.

Many alkaloids have been reported (1 (a,b)) from *L. fawcettii*; most of these possess the lycopodine (2) type of skeleton, although neither lycopodine itself nor any of its simple relatives (those with one or no oxygen functions) have been isolated. Three bases have more recently been isolated from this Jamaican plant, all of which contain two nitrogen atoms. Two of these (from the weaker bases), lycodine (3) and des-N-methyl- α -obscurine (4), have been previously isolated and are of known structure but the other, obtained from countercurrent distribution of the stronger base fraction, is not previously described. The latter, base R, C₁₆H₂₄N₂O₂, forms only a monoperchlorate (B. HClO₄) and from infrared spectra its structure includes carbonyl and secondary amino groups. Acetylation of base R affords a neutral oily compound showing N-acetyl absorption in the infrared.

Another new strong base containing only one nitrogen atom has also been found. Base L, which analyzes best for $C_{18}H_{31}NO_4$, is shown from its infrared spectrum to contain an O-acetyl residue and two hydroxyl groups. Since the methiodide prepared from base L is normal (B. MeI) the nitrogen is presumably tertiary and this is confirmed by acetylation which gives the diacetate $C_{20}H_{33}NO_5$, identical with base O described elsewhere (1 (*b*)). Further acetylation of base O is ineffective so the other hydroxyl grouping was thought to be tertiary and this suspicion is strengthened by the chromium trioxide oxidation of base L to the hydroxy-keto-acetate, $C_{18}H_{29}NO_4$. The latter is readily hydrolyzed to the dihydroxy-ketone (desacetyldehydro-base L), $C_{16}H_{27}NO_3$, which shows infrared absorption attributable to a six-membered (or larger) ring ketone. Neither hydroxyl in desacetyl-dehydro-base L is suitably placed relative to the ketone for ready elimination.

Base L hydrolyzes to a trihydroxy compound, desacetyl-base L, $C_{16}H_{29}NO_3$, which consumes no periodic acid implying that it contains no cis α -glycol system. Acetylation of desacetyl-base L at room temperature gives rise to the diacetate, base O, but at low temperature (5°) a monoacetate is obtained which is not base L and must be the positional isomer, isobase L (cf. fawcettimine (5) and lycofoline (6)), since on further acetylation it also affords base O. These preliminary results suggest that base L is a tricyclic tertiary base possessing a tertiary hydroxyl, a secondary equatorial hydroxyl, and a secondary axial O-acetyl grouping.

¹Part VII. R. H. Burnell and D. R. Taylor. Tetrahedron, 18, 1467 (1962).

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Further characterization of fawcettimine (7), $C_{16}H_{25}NO_2$, shows that both oxygen atoms are present as carbonyl groups. While the base itself shows only one carbonyl peak in the infrared, the crystalline neutral N-acetyl derivative, the N-nitrosamide, and the fawcettimine methiodide all show two peaks attributable to carbonyls (the N-acetyl derivative also shows the amide peak at 1620 cm⁻¹).

The action of sodium borohydride on N-acetylfawcettimine results in the reduction of only one of the carbonyls; the product, dihydro-N-acetylfawcettimine, $C_{18}H_{29}NO_3$, must contain a hindered carbonyl, the N-acetyl group, and a secondary hydroxyl. Reduction of N-acetylfawcettimine with sodium and isobutanol gives rise to this same dihydro compound.

Fawcettidine, base F (1(a)), C₁₆H₂₃NO has been reduced to dihydrofawcettidine with sodium borohydride. The infrared spectra of these compounds (and that of acetyldihydrofawcettidine) show that the ketone in the parent base is in a six-membered (or larger) ring, the molecule contains a carbon-carbon double bond and that the secondary hydroxyl is probably axial. From the n.m.r. spectra only one olefinic proton is shown to be present as well as a secondary C-methyl group. The pK_a of fawcettidine is low (6.2) and that of the reduction product is still low (7.9) for a tertiary lycopodium base. The inference drawn is that both the double bond and the carbonyl are sufficiently close to the nitrogen as to greatly affect its basicity.

The unstable base referred to in an earlier report on the bases obtained from Jamaican *L. clavatum* (7) has now been obtained as the picrate, $C_{15}H_{25}NO_3$. $C_6H_3N_3O_7$. The free base, however, is extremely labile and rapidly develops a brown color in air.

EXPERIMENTAL

Melting points are uncorrected. Infrared spectral values are for Nujol mulls unless otherwise stated and pK_a determinations were done in 50% methanol.

Bases from L. fawcettii Lloyd and Underwood

The mixture of weak bases from *L. fawcettii* (1(*b*)) (2.2 g) was chromatographed over alumina in benzene affording first fawcettidine (base F: 150 mg), pK_a 6.2, $[\alpha]_D$ 161° (*c*, 0.6 EtOH).

Fawcettidine Methiodide

From methanol-acetone, m.p. 223-225°. Calc. for C₁₆H₂₃NO.CH₃I: C, 52.7; H, 6.8; I, 32.8; (N-Me)₁ 3.9%. Found: C, 52.6; H, 6.6; I, 32.8; N-Me, 4.1%.

Dihydrofawcettidine

A methanolic solution of NaBH₄ was added to fawcettidine (40 mg) in methanol and the mixture was left standing at room temperature for 6 hours. Dilution with water and chloroform extraction gave the dihydro base (40 mg) $[\alpha]_D$ 137° (c, 1.0 EtOH) which showed hydroxyl (3200 cm⁻¹) and double bond (1666 cm⁻¹) absorption in the infrared but no carbonyl peak. A sample was sublimed for analysis, m.p. 153–156°, pK_a 7.9. Calc. for C₁₆H₂₅NO requires: C, 77.7; H, 10.2%. Found: C, 77.6; H, 10.5%.

A cetyldihydrofawcettidine

Acetylation of dihydrofawcettidine in acetic anhydride – pyridine gave the O-acetate in the normal manner. The base was converted to the perchlorate, m.p. 220–221°. Calc. for $C_{18}H_{27}NO_2$. HClO₄: C, 55.5; H, 7.2; N, 3.6%. Found: C, 55.9; H, 7.5; N, 3.9%.

Ly codine

The next fraction (180 mg) eluted with benzene gave a crystalline hydrochloride which was recrystallized from methanol-acetone. Calc. for $C_{16}H_{22}N_2$. 2HCl. H_2O : C, 57.7; H, 7.9; N, 8.4; Cl, 21.3%. Found: C, 57.7; H, 8.1; N, 8.4; Cl, 21.3%.

The base, m.p. 117–118°, regenerated from the salt showed ultraviolet absorption at 268 m μ (log $\epsilon = 3.73$) and 276 m μ (log $\epsilon = 3.61$). On comparison (mixed melting point, infrared and ultraviolet spectra) with authentic lycodine (for which we thank Dr. W. A. Ayer, Alberta) the two were shown to be identical.

Des-N-methyl- α -obscurine

Closely associated with base I (1(b)) in the weak base mixture was a small amount of a crystalline base,

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m.p. 270–272° (sealed tube), showing ultraviolet absorption at 252 m μ (log ϵ = 3.75). Calc. for C₁₈H₂₄N₂O: C, 73.8; H, 9.3; N, 10.8%. Found: C, 74.1; H, 9.3; N, 10.7%.

The infrared spectrum, with a peak at 3250 (NH)cm⁻¹ and a complex band centered at 1675 cm⁻¹, was identical with that of des-N-methyl- α -obscurine. A mixed melting point with the latter showed no depression (we thank Dr. W. A. Ayer for the authentic sample).

Base R

The fraction (4.25 g) of the *L. fawcettiii* strong bases (1(b)) containing fawcettiine was distributed between chloroform and aqueous phosphate buffer, pH 7.4 for 20 transfers. The first five fractions contained weaker bases, including fawcettimine (base A: 140 mg perchlorate). Fractions 6–10 contained fawcettiine (1.24 g) and fractions 11–14 gave a new crystalline perchlorate (101 mg) which crystallized from acetone, m.p. 198–199°. Calc. for C₁₆H₂₄N₂O₂. HClO₄: C, 51.0; H, 6.7; O, 25.4; N, 7.4; Cl, 9.4%. Found: C, 51.2; H, 6.7; O, 25.5; N, 7.4; Cl, 9.2%.

The base crystallized from a concentrated acetone solution, m.p. 129–130°, p K_a 7.05. Calc. for C₁₆H₂₄N₂O₂: C, 69.5; H, 8.8; O, 11.6; (C—Me)₁, 5.4%. Found: C, 69.8; H, 8.8; O, 11.9; C—Me, 4.5%.

The infrared spectrum showed peaks at 3420, 3310, 3120, and 1720 cm⁻¹.

Base L

Fractions 19–20 of the distribution (above) gave a crystalline perchlorate, m.p. $290-294^{\circ}$ (decomp.). Calc. for C₁₈H₃₁NO₄: C, 50.8; H, 7.6; O, 30.1; N, 3.3; Cl, 8.3%. Found: C, 50.6; H, 7.7; O, 30.4; N, 3.3; Cl, 8.1%.

The base, pK_a 9.7, obtained from the perchlorate, was an oil showing infrared absorption at 3450 (OH), 1723, and 1250 cm⁻¹ (OAc).

The methiodide, m.p. 281–282° (from acetone). Calc. for C₁₈H₃₁NO₄. CH₃I: C, 48.8; H, 7.3; N, 3.3; O, 13.7%. Found: C, 48.6; H, 7.3; N, 3.3; O, 13.6%.

Acetylation of Base L (Base O)

Base L (108 mg) was acetylated in acetic anhydride (4 ml) and pyridine (4 ml) at room temperature for 20 hours. The mixture was then poured into iced water, basified with ammonia, and chloroform extracted giving a white solid (103 mg) which was sublimed twice for analysis, m.p. 179°. Admixture with base O (1(*b*)) (m.p. 180–181°) caused no depression in melting point and the two samples have identical infrared spectra. Calc. for $C_{20}H_{23}NO_5$: C, 65.4; H, 9.0; O, 21.8; (OAc)₂, 23.4%. Found: C, 65.5; H, 8.6; O, 21 9; OAc, 23.4%.

Desacetyl-base L

To base L perchlorate (92 mg) in methanol (5 ml) was added 3 N sodium hydroxide solution (1 ml). After 21 hours at room temperature the mixture was diluted with water and the chloroform was extracted, affording a crystalline solid (58 mg). Recrystallization was from acetone, m.p. 189–190°, pK_a 9.4. Calc. for $C_{18}H_{29}NO_3$: C, 67.8; H, 10.3; O, 16.9%. Found: C, 67.9; H, 10.4; O, 17.0%.

The base showed no carbonyl absorption in the infrared.- .-

Acetylation of Desacetyl-base L (Isobase \tilde{L})

Desacetyl-base L (106 mg) was acetylated in acetic anhydride and pyridine at 5° C. The white solid (100 mg) obtained by extraction was sublimed for analysis, m.p. 189–192°. Calc. for $C_{18}H_{81}NO_4$: C, 66.4; H, 9.6; O, 19.7; (O—Ac)₁, 13.2%. Found: C, 66.5; H, 9.4; O, 19.7; O—Ac, 12.9%.

The infrared spectrum differed from that of base L and seeding concentrated solutions of the latter with this product caused no crystallization.

Oxidation of Base L (Dehydro-base L)

To a cooled solution of base L (250 mg) in pyridine (5 ml) was added a cold mixture of chromium trioxide (1 g) in pyridine (5 ml). After 90 minutes at 5° the mixture was poured into ice water and made basic with ammonia. Chloroform extraction and evaporation gave a pale brown solid, affording crystals from acetone. A sample was sublimed for analysis, m.p. 196–197°. Calc. for $C_{18}H_{29}NO_4$: C, 66.9; H, 9.0%. Found: C, 66.9; H, 9.1%.

Desacetyldehydro-base L

Dehydro-base L (187 mg) was hydrolyzed in methanol with sodium hydroxide (75 mg) at room temperature for 5 hours. Extraction gave a white solid (124 mg) which was recrystallized from acetone and sublimed for analysis, m.p. 199–200°. Calc. for $C_{16}H_{27}NO_3$: C, 68.6; H, 9.6; O, 16.8%. Found: C, 68.3; H, 9.7; O, 17.0%.

L. clavatum-weak base

The weak base residues from *L. clavatum* (7) were chromatographed over alumina in benzene. The first eluate proved to be extremely labile, colored rapidly when exposed, but afforded a relatively stable picrate, m.p. 125–127°. Calc. for $C_{15}H_{25}NO_3$. $C_6H_3N_3O_7$: C, 50.8; H, 5.7; N, 11.3; O, 32.2%. Found: C, 51.1; H, 5.7; N, 11.1; O, 32.2%.

Further elution of the column gave the previously described weak bases (7) and then a substantial amount of fawcettimine (base A).

Acetylation of Fawcettimine (N-Acetylfawcettimine)

To fawcettimine (950 mg) in pyridine (11 ml) was added acetic anhydride (11 ml). After 20 hours at room temperature the volatile materials were removed on a rotary evaporator and the residue taken up in chloroform. After washing thoroughly with dilute hydrochloric acid, dilute ammonium hydroxide, and water, the neutral product (856 mg), obtained on evaporation of the solvent, was recrystallized from acetone-ether, m.p. 144-145°. Calc. for C18H27NO3: C, 70.8; H, 8.9; N, 4.6; O, 15.7%. Found: C, 70.7; H, 9.1; N, 4.7; 0, 15.7%.

Infrared peaks were at 1735 (C=O), 1710 (C=O), and 1620 (N-COCH₃) cm⁻¹.

Sodium–Isobutanol Reduction of N-Acetylfawcettimine

N-Acetyl fawcettimine (400 mg) was dissolved in toluene (40 ml) containing isobutanol (1 ml) and the solution added to a fine dispersion of sodium (600 mg) in toluene (50 ml) under purified nitrogen. The mixture was stirred for 24 hours, the undissolved sodium decomposed with ethanol, and then the solution was poured into water, shaken, and separated. The aqueous portion was extracted with chloroform. The organic layers were combined, washed with dilute hydrochloric acid and then water, dried, and evaporated to give an oil (359 mg). A crystalline product (90 mg), which was obtained by being dissolved in a small volume of acetone and then cooled, was sublimed for analysis, m.p. 205-206°. Calc. for C18H29NO3: C, 70.3; H, 9.5; O, 15.6; N, 4.6%. Found: C, 70.6; H, 9.5; O, 15.4; N, 4.5%.

The infrared spectrum (Nujol) showed peaks at 3370 (OH), 1735 (C=O), and 1620 (N-COCH_a) cm⁻¹.

The material from the mother liquors above slowly crystallized when they were left standing and gave an infrared spectrum identical with that of the sublimed dihydro-N-acetylfawcettimine.

This same reduction product was obtained when N-acetylfawcettimine was treated with sodium borohydride in methanol, but crystallization was only possible after chromatography over alumina.

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