STUDIES ON THE ULEINE ALKALOIDS. II. SOME CHEMICAL TRANSFORMATIONS OF ULEINE[1,2]

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ABSTRACT.—The lithium aluminum hydride reduction and hydroboration of uleine (**1**) were further studied. Hydroboration of **1** afforded (16R)-17-hydroxy-16,17-dihydrouleine (**4**) in 23% yield in agreement with the earlier results of Djerassi *et al.* $LiAlH_4$ reduction afforded three products, the anticipated 16,17-dihydro derivative **2** having the 16*R*-configuration and, quite unexpectedly, the two 16-hydroxy-16,17-dihydrouleines **5** and **6**. The stereochemistry of the products was deduced from examination of their carbon-13 nmr spectra. Introduction of the hydroxyl group during $LiAlH_4$ reduction was thought to occur by way of the conjugated imine-enamine **7**.

In 1964 Joule and Djerassi reported (1) the formation of a 16,17-dihydrouleine derivative in high yield by the lithium aluminum hydride reduction of uleine (**1**). The product of this reaction (and aqueous work-up) was shown to be identical with the product from the catalytic hydrogenation of uleine by its tlc, mass spectrum and mixture melting point. Deuterium labeling studies ($LiAlD_4$, D_2O) indicated that both hydrogen atoms added across the double bond came from the metal hydride and none from the water used in the reaction work-up. The 16*R* stereochemistry of **2** was inferred by a probable attack, under catalytic conditions, from the least hindered (α) face forcing a 16β-methyl group.

In the present study, the lithium aluminum hydride reduction was conducted under slightly different conditions than those used by Joule and Djerassi (1) to afford three products, a 16,17-dihydro derivative, and two 16-hydroxy-16,17-dihydro derivatives in the approximate ratio 3:2:1. The introduction of a hydroxyl group following a hydride ion rearrangement is rather unusual and possibly stems from the stability of an intermediate postulated by Joule and Djerassi to account for the introduction of two hydride atoms from the reducing agent.

Analysis of the carbon-13 nmr spectrum of the three products on comparison with the data established for uleine (**1**) (2), permitted assignment of the C–16 stereochemistry unambiguously in all cases.

Djerassi *et al.* (3,4) had also reported a hydroboration reaction of uleine (**1**) to afford a single product considered to be 17-hydroxy-16,17-dihydrouleine (**3**), in which the 16*R*-configuration was tentatively assigned. We have repeated this reaction and have established through carbon-13 nmr analysis, both the structure and stereochemistry of the product.

EXPERIMENTAL[3]

HYDROBORATION OF ULEINE (**1**).—To uleine[4] (**1**, 100 mg) in dry ethyl ether (5 ml) was added

[1]For the previous paper in this series see R. P. Borris, D. C. Lankin and G. A. Cordell, *J. Nat. Prod.*, **46**, 200 (1983).

[2]Portions of this work were submitted by R. P. B. in partial fulfillment of the Ph.D. degree requirements of the Graduate College, University of Illinois at the Medical Center, August, 1981, and were first presented at the Joint Meeting of the American Society of Pharmacognosy and the Society for Economic Botany, Boston, Mass., July, 1981.

[3]Melting points were determined on a Kofler hot plate and are uncorrected. The uv spectra were obtained with a Beckman model DB-G spectrophotometer and the ir spectra with a Beckman model 18-A spectrophotometer with polystyrene calibration at 1601 cm^{-1}; absorption bands are recorded in wave numbers (cm^{-1}). Proton magnetic resonance spectra were recorded in $CDCl_3$ with a Varian T-60A instrument operating at 60 MHz with a Nicolet Modell TT-7 Fourier Transform attachment. Carbon magnetic resonance spectra were recorded in $CDCl_3$ with a JEOL FX-100 instrument operating at 25.05 MHz. Tetramethylsilane was used as an internal standard and chemical shifts are recorded in δ (ppm). Mass spectra were obtained with a Varian MAT 112S double focusing spectrometer operating at 70 eV.

[4]Obtained from *Aspidosperma subincanum* K. von Mart. as described previously (2).

dropwise with stirring (3 hr) a solution of boron trifluoride etherate in ethyl ether (1:19, 1.5 ml). After removal of ether (N_2 stream), the cream-colored precipitate of the uleine-boron trifluoride complex was dissolved in THF (5 ml), boron trifluoride etherate (0.2 ml) and sodium borohydride (250 mg) were added and the mixture stirred overnight at room temperature. Aqueous sodium hydroxide solution (30%, 1 ml) and hydrogen peroxide (30%, 2 ml) were added and the mixture refluxed for five hours and cooled to room temperature. After the addition of aqueous sodium hydroxide solution (10%, 20 ml), the mixture was extracted with chloroform-ethanol (9:1, 8 x 20 ml). The pooled organic phases were dried (Na_2SO_4), filtered and evaporated *in vacuo* to afford a brown residue.

Repeated preparative tlc of the residue on Silica gel 60 F_{254}[5] with ethyl acetate:isopropanol:28% ammonium hydroxide (22:15:1) as the developing solvent afforded a main component (R_f 0.51) which, when filtered through neutral alumina[6] (Brockman activity I), afforded (16*R*)-17-hydroxy-16,17-dihydrouleine (**4**) as a yellow-brown amorphous gum (22.4 mg, 23.5%), ms, *m/z* 284 (M^+, 48%), 253 (13), 228 (22), 227 (78), 223 (28), 222 (42), 210 (13), 209 (13), 198 (11), 197 (19), 196 (32), and 194 (16); ^{1}H-nmr, ($CDCl_3$, 60 MHz) δ 0.89 (5H, m, 18-H_3, 19-H_2), 2.35 (3H, s, 5-H_3), 3.96 (2H, AB system, 17-H_2), 4.20 (1H, br d, J=1.3 Hz, 21-H), 7.00–7.60 (4H, m, 4 x Ar-H), and 9.28 (1H, br s, N-H); ^{13}C-nmr, see figure 1.

LITHIUM ALUMINUM HYDRIDE REDUCTION OF ULEINE (**1**).—Uleine (**1**, 110 mg) in anhydrous ethyl ether (5 ml) was added dropwise to a suspension of $LiAlH_4$ (1.5 g) in anhydrous ethyl ether:THF (4:1, 50 ml) in an ice bath. The mixture was stirred at room temperature for 72 hrs., and excess reagent was decomposed by the addition of ethyl acetate (20 ml) and distilled water (10 ml). Dilution with distilled water (50 ml) followed by extraction with ethyl ether (5 x 50 ml), drying of the ethereal extract (Na_2SO_4) and removal of solvent *in vacuo* afforded a brown residue.

Preparative tlc of the residue on Silica gel-60 F_{254} with ethyl acetate:isopropanol:28% ammonium hydroxide (25:10:1) as the developing solvent permitted the delineation of uleine (R_f 0.55) and three major products (R_f 0.44, 0.30, and 0.23).

Further preparative tlc of the band at R_f 0.44 in the same solvent system, followed by filtration through neutral alumina (Brockman activity I) afforded (16*R*)-16,17-dihydrouleine (**2**) as a tan semi-solid (38 mg, 34%); ms, *m/z* 268 (M^+, 58%), 253 (10), 239 (17), 222 (20), 212 (32), 211 (100), 210 (18), 209 (18), 196 (20), 194 (20), 183 (33), 182 (68), 181 (21), 180 (26), 168 (21), and 167 (26); ^{1}H-nmr ($CDCl_3$, 60 MHz) δ 0.89 (5H, m, 18-H_3, 19-H_2), 1.36 (3H, d, J=7 Hz, 17-H_3), 2.28 (3H, s, 5-H_3), 3.06 (1H, br q, J=7 Hz, 16-H), 4.01 (1H, br d, J=2 Hz, 21-H), 7.00–7.60 (4H, m, 4 x Ar-H), and 7.98 (1H, br s, N-H); ^{13}C-nmr, see figure 1.

Further preparative tlc of the band at R_f 0.30 in the same solvent system and filtration through neutral alumina (Brockman activity I) afforded (16*R*)-16-hydroxy-16,17-dihydrouleine (**5**) as a yellow-brown oil (14.5 mg, 12%); ms, *m/z* 284 (M^+, 48%), 267 (12), 266 (38), 265 (11), 241 (13), 238 (11), 237 (33), 224 (15), 223 (70), 222 (18), 213 (19), 212 (12), 211 (20), 210 (34), 209 (84), 208 (33), 201 (26), 198 (13), 197 (12), 196 (14), 195 (22), 194 (48), 193 (16), 192 (10), 185 (21), 184 (17), 183 (22), 182 (21), 181 (22), 180 (38), 173 (34), 172 (100), 171 (18), and 170 (11); ^{1}H-nmr, ($CDCl_3$, 60 MHz) δ 0.96 (5H, m, 18-H_3, 19-H_2), 1.66 (3H, s, 17-H_3), 2.30 (3H, s, 5-H_3), 4.07 (1H, br d, J=1 Hz, 21-H), 7.10–7.70 (4H, m, 4 x Ar-H), and 8.38 (1H, br s, N-H); ^{13}C-nmr, see figure 1.

Further preparative tlc of the band at R_f 0.23 in the same solvent system and filtration through neutral alumina (Brockman activity I) afforded (16*S*)-16-hydroxy-16,17-dihydrouleine (**6**) as a yellow brown oil (26.2 mg, 22%); ms, *m/z* 284 (M^+, 31%), 266 (11), 237 (15), 228 (12), 227 (28), 224 (13), 223 (31), 222 (19), 213 (16), 211 (17), 210 (35), 209 (74), 208 (19), 201 (15), 198 (14), 197 (13), 196 (20), 195 (28), 194 (28), 185 (19), 184 (20), 183 (25), 182 (17), 181 (15) and 180 (24); ^{1}H-nmr, ($CDCl_3$, 60 MHz) δ 0.90 (5H, m, 18-H_3, 19-H_2), 1.67 (3H, s, 17-H_3), 2.30 (3H, s, 5-H_3), 4.27 (1H, br s, 21-H), 7.10–7.60 (4H, m, 4 x Ar-H), and 9.87 (1H, br s, N-H); ^{13}C-nmr, see figure 1.

STRUCTURE ELUCIDATION OF THE REACTION PRODUCTS.—a) From hydroboration reaction. A single product was isolated in 23% yield having a molecular ion 18 amu more than uleine (**1**) with a significant loss of 31 amu, and a basic fragmentation pattern similar to that of uleine. In the ^{1}H-nmr spectrum, the exomethylene protons of uleine had been replaced by an AB system centered at δ 3.96. Slight downfield shifts of $-NCH_3$, H-21 and the indole NH were also observed. On this basis the gross structure of 17-hydroxy-16,17-dihydrouleine (**3**) was assigned without consideration of the C-16 stereochemistry at this point.

b) From $LiAlH_4$ reduction. The mass spectrum of the least polar product displayed a strong molecular ion, *m/z* 268, consistent with a molecular formula $C_{18}H_{24}N_2$ and a fragmentation pattern closely corresponding with the published data (1) for a dihydro uleine derivative. In agreement, the ^{1}H-nmr spectrum on comparison with uleine showed the addition of a three proton doublet at δ 1.36 and a one proton, broadened quartet at δ 3.06 with the concommitant loss of both exomethylene protons. The remainder of the spectrum was similar to that of uleine and together these observations confirmed the product to be a 16,17-dihydrouleine.

In the mass spectrum, the second isolate (R_f 0.30) displayed a molecular ion at *m/z* 284, consistent with the addition of the elements of water to uleine. Once again the exomethylene signals were absent in the ^{1}H-nmr spectrum, replaced by a three proton singlet at δ 1.66. This is consistent with a quaternary benzylic carbon bearing oxygen and a methyl group, indicating that the compound was an isomer of 16-hydroxy-16,17-dihydrouleine. The third isolate (R_f 0.23) displayed mass and ^{1}H-nmr properties similar to those of the second isolate except for

[5]E. Merck, Darmstadt, West Germany.
[6]M. Woelm, Eschwege, West Germany.

differences in the resonances of the ethyl side chain, the 21-H and the indole N-H, all of which were shifted downfield ($\Delta\delta$ +0.06, +0.20 and +1.49, respectively).

Preliminary evidence for the assignment of the configuration at C-16 was obtained by comparison of the tlc behavior and consideration of the ^{1}H-nmr spectra of the products. The less polar isomer, displaying less internal hydrogen bonding (upfield NH), was considered to be the isomer in which the hydroxyl group is more hindered and the angle between the indole nucleus and the C-16–O-bond is the largest (~70° from Dreiding models). This is the β-hydroxy isomer. Correspondingly, the more polar isomer was considered to have a more accessible hydroxy group and a smaller angle (~45°) between the indole nucleus and the C-16–O-bond, permitting more substantial internal H-bonding. This is the α-hydroxy isomer. These tentative stereochemical assignments were clarified by examination of the carbon-13 nmr spectra of the products.

A summary of the carbon-13 nmr data for the four reduction products under consideration is shown in figure 1 in comparison with the established data for uleine (2). Some tentative assignments are necessary because of the limited amount of available sample, but these do not affect the interpretation of the spectra for stereochemical purposes.

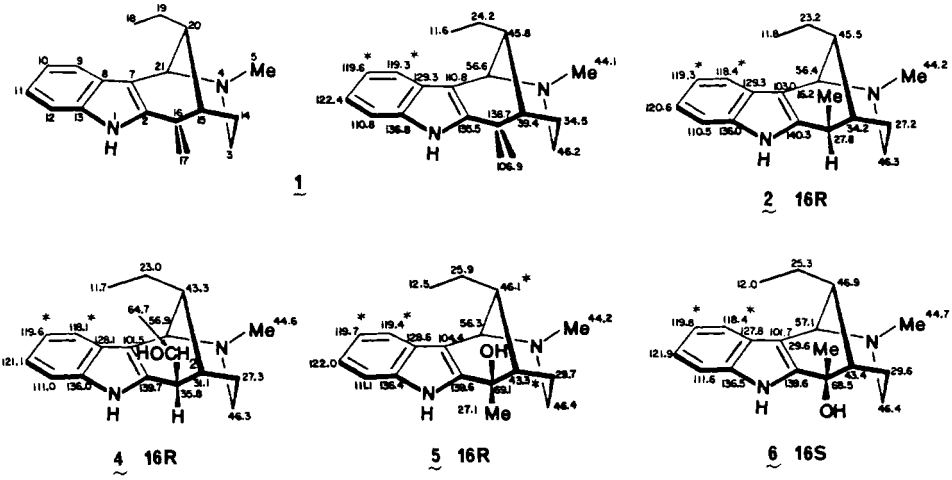


FIG. 1.

The carbon-13 nmr resonances of the two tertiary alcohol derivatives **5** and **6** could be assigned by inspection in comparison with uleine (**1**) (2) and then supported by consideration of data obtained for the other 16,17-dihydro derivatives, additivity parameter calculations and single-frequency off-resonance decoupling (SFORD) experiments (for **5**). Thus carbon-16 appeared downfield near δ 69, while C-17, the newly formed methyl group, appeared in the range 27.0–29.6 ppm in these compounds. Carbon-2 and carbon-15, the carbons β to the hydroxyl group, experience downfield shifts (4.4 and 3.9–7.5 ppm, respectively) relative to uleine. These data are consistent with saturation at the 16,17-bond and the presence of an oxygen atom at C-16. As expected, a substantial upfield shift of 5.4–9.3 ppm was observed for C-7 in the dihydro derivatives **2**, **4–6**.

The configuration at C-16 in these compounds was deduced as follows. In the least polar of the tertiary alcohols both C-18 and C-19 are approximately 0.5 ppm downfield of their frequencies in the more polar isomer, and C-7 is shifted downfield by 2.7 ppm. These chemical shift data suggest that the hydroxyl group is β-oriented in the less polar isomer.

All of the observed spectral data and tlc considerations are in accordance with an assignment of the 16*R* configuration to the less polar isomer and of the 16*S* configuration to the more polar isomer, which therefore have the structures **5** and **6** respectively.

Self-consistent tentative assignments could then be made for the 16,17-dihydro- and 17-hydroxy-16,17-dihydro-derivatives of uleine by inspection, SFORD experiments and additivity parameter calculation. In 16,17-dihydrouleine the signals of C-18, C-19 and C-20 are at higher field than in (16*S*)-16-hydroxy-16,17 dihydrouleine (**6**). These data are consistent with the presence of a β-oriented methyl group at C-16 involved with the ethyl side chain at C-20 in a steric interaction, i.e., C-16 has the *R*-configuration.

Additivity parameter calculations (5) could then be used to assist in the assignment of the remaining carbon resonances of this compound; the (16*S*)-tertiary alcohol **6**, having a β-CH_3 was the reference standard. Replacing the C-16 hydroxy group with a proton is expected to produce a dramatic (−50 ppm) shift in the resonance frequency of C-16 and significant (−10 ppm) shifts in C-15 and C-17.

A SFORD spectrum of **2** permitted assignment of the signal at δ 27.8 to C-16, consistent with the predicted upfield shift. Similarly, the signal at δ 27.2 was assigned to C-14, the change in chemical shift ($\Delta\delta$ −2.4) being dominated by the loss of the *peri*-oxygen deshielding effect. Assignment of the signal at δ 34.2 in **2** to C-15 is also consistent with the calculated shift value. Reversal of the C-15/C-16 assignments would lead to a shift in the C-15 resonance that is far too large (19 ppm) for the introduction of a hydroxy group at C-16 in **2**.

The signal at δ 16.2 in **2** was assigned to C–17 from the SFORD experiment and is consistent with the loss of the inductive deshielding of the tertiary alcohol. The resonances of the remaining carbons are all similar and are consistent with the assignments made for the corresponding carbons in uleine (**2**) and the two tertiary alcohol derivatives. The 16,17-dihydro derivative can therefore be assigned the complete structure and stereochemistry shown in **2**.

The remaining compound to be discussed is the hydroboration product, 17-hydroxy-16,17-dihydrouleine. Similar arguments are again used to establish the stereochemistry at C–16 in this compound. Resonances for C–18 and C–19 are both shifted to higher field than in any other derivative, reflecting a steric interaction of a bulky hydroxy methyl group at the 16β-position. By use of additivity parameter calculations (5) and (16*R*)-16,17-dihydrouleine (**2**) as the reference, assignment of the carbon resonances for this compound could be achieved. These calculations predict a very large (+50 ppm) shift for C–17, a smaller (+10 ppm) shift for C–16 and small upfield shifts (−3 ppm) for both C–2 and C–15. Little or no change is predicted for the resonance frequencies of C–14 and C–20.

The signal at δ 64.7 is readily assigned to C–17 as it is the only oxygenated carbon, and the observed downfield shift of 48.5 ppm agrees with the calculated value. The resonance at δ 35.8 is assigned to C–16. Alternatives considered were δ 31.1 or δ 27.3, but these would result in downfield shifts from the assignment of C–16 in the dihydro derivative (δ 27.8) of +3.3 or −0.5 ppm, too small to be consistent with the predicted shift.

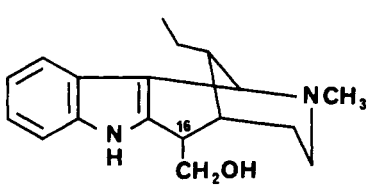
This attribution of C–16 is in agreement with the assignment of the signal at δ 31.1 to C–15. If the resonance at δ 27.3 had been assigned to this carbon, the observed change in chemical shift (6.9 ppm) would also have been too large and inconsistent with calculations. This would also have produced a significant (+3.8 ppm) shift in the resonance of C–14, inconsistent with the predicted absence of change. Assigning δ 31.1 to C–15 and δ 27.3 to C–14 is more completely in agreement with expected values.

Carbon-20, although predicted by an inductive effect to remain unchanged, in fact shifts downfield by 2.2 ppm. This is typical however for the γ-*gauche* interaction arising from the steric compression of an ethyl side chain and a bulky hydroxymethyl group in a β-orientation at C–16. On this basis the hydroboration product of uleine is assigned the structure (16*R*)-17-hydroxy-16,17-dihydrouleine (**4**).

Although the magnitude of the chemical shift differences are small between the various derivatives, taken together they permit a self-consistent assignment of the stereochemistry in each compound.

DISCUSSION

Uleine (**1**) is one of the rare indole alkaloids derived from tryptophan in which the amino ethyl bridge has been fragmented (6). In order to provide reference compounds which might be produced during the microbial transformation of uleine (7), two chemical reactions of uleine were repeated (1,3,4). Lithium aluminum hydride reduction of uleine (**1**) afforded (16*R*)-16,17-dihydrouleine (**2**) as the principal product. This is in agreement with the earlier work by Djerassi *et al.* (1) who had also shown that both hydrogen atoms introduced at C–16 and C–17 arose from the reducing agent and postulated an intermediate imine-enamine **7** to explain this result.



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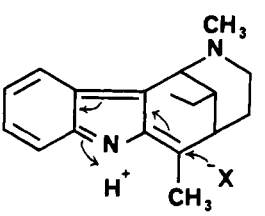

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Lithium aluminum hydride reduction also afforded the two 16-hydroxy-16,17-dihydrouleines **5** and **6**. This rather surprising result, which overall comprises the addition of water to the 16,17-double bond, may also be interpreted in terms of a relatively stable form of **7**, which subsequently undergoes nucleophilic attack.

Hydroboration of uleine (**1**) had previously (3,4) produced 17-hydroxy-16,17-dihydrouleine (**3**), in which the 16*R*-configuration was suggested based on stereomechanistic considerations. In our hands, this reaction also yielded a single reaction product whose structure and stereochemistry were deduced to be **4** by comparison of the carbon-13 nmr spectral data of the product with those of **1**, **2**, **5** and **6**. In the assignment of C–16 stereochemistry in these derivatives, observa-

tion of slight shielding and deshielding effects afforded by the steric interaction of groups at C–16 with the ethyl side chain and C–14 was critical.

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