chloroform) gave upon chromatography on column B a single band, nonbasic, R_f 0.83 (15 mg., 62%), with infrared spectrum superimposable upon that of starting material XVI.

20-N-Methyldihydrocyclobuxine (IIe).—Dihydrocyclobuxine (55 mg.) was dissolved in ethanol (4 ml.) containing 40% formaldehyde (0.05 ml.), and the solution added to reduced platinum oxide (50 mg.) in acetic acid (10 ml.), stirred under hydrogen. One mole equivalent of hydrogen was taken up rapidly (15 min.), the next very slowly (many hours). The hydrogenation was halted when the uptake was about 1.5 mole equiv., and the product (recovered with water, evaporation of the ethanol, ammonium hydroxide, and chloroform) purified on column C. The slower-running major band, R_f 0.75 (22 mg., 38%), crystallized from acetone–water as needles (12 mg.), m.p. 123–125°, $[\alpha]^{22}D$ +21° (*c* 0.29); infrared spectrum similar to that of N,N′-dimethyldihydrocyclobuxine, but stronger at 2.9 μ.

Anal. Calcd. for $C_{28}H_{45}ON_2$: C, 77.55; H, 11.52. Found: C, 78.05; H, 11.55.

The faster-running major band, R_f 0.95 (24 mg., 41%), had an infrared spectrum superimposable upon that of N,N′-dimethyldihydrocyclobuxine (IIb), and demonstrated similarly poor crystallization behavior.

O,N-Diacetyl-20-N-methyldihydrocyclobuxine (IIf).—20-N-Methyldihydrocyclobuxine (IIe, 8 mg.) was acetylated with acetic anhydride–pyridine, 42 hr. at room temperature; the product was purified on column C, and the band of R_f 0.65 (8 mg.) crystallized from acetone–isopropyl ether to give 3 mg., m.p. 202–205°; λ_{max} 3.60 (m), 5.82 (s), and 6.16 (s) μ.

Anal. Calcd. for $C_{30}H_{50}O_3N_2$: C, 74.03; H, 10.39. Found: C, 74.31; H, 10.34.

Hydrolysis of the O,N-diacetyl derivative IIf (20 mg.) in refluxing 80% ethanol (2.5 ml.) containing potassium hydroxide (1.0 g.) for 2 hr. gave a product, purified on column C (R_f 0.25, 7 mg.) showing λ_{max} 6.16 (s) μ; there was no trace of a band above R_f 0.5 corresponding to fully hydrolyzed 20-N-methyldihydrocyclobuxine.

N-Acetyldihydrocyclobuxine (IIg).—N,N′-Diacetyldihydrocyclobuxine *A*[18] (from 50 mg. of dihydrocyclobuxine) was hydrolyzed as above, and the product purified on column B to give a single sharp red band, R_f 0.48 (31 mg.), crystallized with difficulty from acetone to give 13 mg., m.p. 237–240°, λ_{max} 6.18 (s) μ.

Anal. Calcd. for $C_{27}H_{46}O_2N_2$: C, 75.30; H, 10.77. Found: C, 75.38; H, 10.53.

The monoacetyl derivative IIg (25 mg.) was methylated in ethanol (4 ml.) and acetic acid (12 ml.), using platinum oxide (50 mg.) and 40% formaldehyde (0.05 ml.). One mole equivalent of hydrogen was taken up, *rapidly*. The total product (recovered as for 20-N-methyldihydrocyclobuxine) was acetylated with acetic anhydride–pyridine, 10 hr. at room temperature, and the product purified on column C. The major band, R_f 0.59 (20 mg.), had an infrared spectrum superimposable upon that of O,N-diacetyl-20-N-methyldihydrocyclobuxine prepared as above; crystallization from acetone–isopropyl ether gave material, m.p. 198–201°; m.m.p. with IIf of m.p. 200–203°, 198–203°.

[CONTRIBUTION FROM THE DEPARTMENT OF PHARMACEUTICAL CHEMISTRY, UNIVERSITY OF WISCONSIN, MADISON, WISCONSIN, AND THE DEPARTMENT OF CHEMISTRY, STANFORD UNIVERSITY, STANFORD, CALIFORNIA]

Buxus Alkaloids. V.[1] The Constitution of Cyclobuxamine, a 4β-Monomethyl Cyclosteroid Alkaloid[2]

BY KEITH S. BROWN, JR.,[3] AND S. MORRIS KUPCHAN[4]

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The physical and chemical properties of cyclobuxamine (Ia, 3β-amino-20α-methylamino-4β,14α-dimethyl-9β,19-cyclo-5α-pregnan-16α-ol), an alkaloid isolated from the acetone-insoluble portion of the bases of *Buxus sempervirens* L., suggested that it was a 3-N-demethyldihydrocyclobuxine. However, trimethylcyclobuxamine (Ig) was found to be different from dimethyldihydrocyclobuxine. Molecular rotation comparisons indicated that, while dihydrocyclobuxine and its tri- and diacetyl derivatives IIIa–c possess a 4α-methyl group, the hydrogenation products Ih and Ii from tri- and diacetylcyclobuxine (IIb and IIc) have the 4β-methyl configuration. The triacetyl derivative Ih was synthesized from cyclobuxamine (Ia) *via* the 3-N-monoformyl derivative Im, confirming the structure and configuration of cyclobuxamine, which appears to be the first 4β-monomethyl steroid to be isolated from natural sources.

Cyclobuxamine (Ia), isolated by partition chromatography[1,5] from the acetone-insoluble portion of the strong bases from *Buxus sempervirens* L.,[2] is the most polar of the three major components of this fraction[6] and may be distinguished by its crystallization from acetone as the very stable isopropylideneimine Ib.[7] The imine Ib ($C_{27}H_{46}ON_2$) showed an infrared spectrum very close to that of dihydrocyclobuxine (IIIa)[1] with the addition of a strong sharp band at 6.01 μ for the C=N linkage. The n.m.r. spectrum[8] of Ib also showed its close similarity to IIIa, with similar peaks in the spectra of the two assignable to the 16β-proton (5.92 τ, octet, *J* 3, 7, and 9.5 c./sec.), an N-methyl (7.57 τ), two tertiary C-methyls (8.87 and 9.03 τ), two secondary C-methyls (8.92 and 9.32 τ), and a cyclopropyl methylene (9.58 and 9.83 τ, AB doublets of *J* 4 c./sec.); the spectrum of Ib also showed signals for the two methyl groups of the isopropylidene moiety (8.02 and 8.19 τ).

The imine could be hydrogenated with platinum in acetic acid, and the resulting isopropylamine Ic gave a triacetyl derivative Id showing typical infrared bands for an O-acetyl (5.77 μ) and two tertiary N-acetyl (6.15 μ) groups, and n.m.r. signals as observed before[1] for two N-acetyl and one N-methyl groups with restricted internal rotation. In the n.m.r. spectrum of Ic, the signals for the isopropylidene group of Ib are shifted upfield to appear as an isopropyl doublet (8.97 τ), while the signal for the 4β-methyl group is shifted downfield (to 9.00 τ).

(1) Parts III and IV: K. S. Brown, Jr., and S. M. Kupchan, *J. Am. Chem. Soc.*, **86**, 4414, 4424 (1964).

(2) This investigation was supported in part by research grants from the National Institutes of Health (H-2952 and CY-4500). We are deeply indebted to Ciba Pharmaceutical Co. for procurement and large-scale extraction of plant material, and especially to Dr. Emil Schlittler, Karl Heusler and Daniel Dickel for their kind interest and cooperation.

(3) National Science Foundation Cooperative Predoctoral Fellow in Chemistry (University of Wisconsin), 1960–1962; National Institutes of Health Postdoctoral Fellow (Stanford University), 1963.

(4) To whom inquiries concerning this paper should be directed.

(5) K. S. Brown, Jr., and S. M. Kupchan, *J. Chromatog.*, **9**, 71 (1962). Cyclobuxamine is band III, the alkaloid of R_f 0.59 in column 1 of Table II.

(6) The other two are cyclobuxine (IIa) (see ref. 1) and cyclovirobuxine (K. S. Brown, Jr., and S. M. Kupchan, *Tetrahedron Letters*, in press).

(7) For further examples of this behavior in 3- and 20-aminosteroids see S.A. Oletta, Belgian Patent 627,935 (May 5, 1963).

(8) We thank Mr. R. Matsuo and Mr. A. Krubsack for the n.m.r. determinations. All chemical shifts are reported in τ-values (p.p.m.).

Strong base cleavage of the imine Ib gave 1 equiv. of acetone (isolated as the 2,4-dinitrophenylhydrazone) and the parent alkaloid Ia, which could be crystallized from benzene, and which showed no band in the infrared for the imine, but in its place a band of medium intensity for the primary amino group (6.32 μ). Acetylation of either Ia or Ib with acetic anhydride–pyridine led to the triacetyl derivative Ie, with infrared bands for one O-acetyl (5.78 μ), one tertiary N-acetyl (6.14 μ), and one secondary N-acetyl (2.91, 6.02, and 6.60 μ) groups. Vigorous alkaline hydrolysis[1] of the triacetyl derivative Ie gave a secondary monoacetyl derivative (infrared bands at 6.00 and 6.60 μ), indicating (along with the shift in the n.m.r. signal of the 4β-methyl group upon hydrogenation of the imine Ib to Ic, above) that the primary amino group in the parent alkaloid Ia is at C-3.

N,N,N′-Trimethylcyclobuxamine (Ig) was prepared, but proved to be different in physical and chemical properties from dimethyldihydrocyclobuxine[1]; this led to the hypothesis that the two could differ in configuration at the 4-position. The 4α-configuration was assigned to dihydrocyclobuxine on the basis of the following arguments. Partial and complete acetylation of dihydrocyclobuxine (IIIa) led to the diacetyl derivative IIIc and the triacetyl derivative IIIb, which differed remarkably in melting point, rotation, and infrared and n.m.r. spectra from the corresponding isomers Ii and Ih produced by hydrogenation of diacetylcyclobuxine[1] (IIc)[1] and triacetylcyclobuxine (IIb),[1] respectively. In particular, the isomers from hydrogenation (Ii and Ih) had molecular rotations 125 and 139° more positive than those from acetylation of dihydrocyclobuxine (IIIc and IIIb). In four pairs of 4α- and 4β-methylcholestane derivatives reported previously (2-spirothian-3-ketone, 3-ketone, 3α-methyl-3β-hydroxy, 3β-methyl-3α-hydroxy),[9] the 4β-methyl isomers had an average molecular rotation 48° more positive than their 4α-methyl counterparts. Further support for the 4β-methyl configuration of Ih and Ii could be seen in the n.m.r. signals of the 19-cyclopropyl methylene, which were at considerably higher field in the spectra of Ih and Ii (average +0.25 and +0.16 p.p.m. for the lower and higher field doublets, respectively) than in those of IIIb and IIIc; this could be attributed to a blocking effect by the 4β-methyl group in the former pair on the long-range deshielding influence of the 3β-acetylamino group on the 19-methylene.[10] The assignment of 4α-methyl configuration to dihydrocyclobuxine is also supported by its direct conversion *via* Ruschig degradation and hydrogenation to a product of known configuration (IV) derived from cycloeucalenol[1,11]; strong basic treatment has been specifically shown to effect no epimerization of 4β-methylcholestan-3-one.[9,12] Examination of molecular models would appear to indicate that cyclobuxine (IIa) could readily undergo β-face hydrogenation (giving a 4α-methyl product), owing to the bowing of the molecule into an "L"-shape by the cyclopropane ring; substitution of bulky groups on the 3β-methylamino function, however, would make less likely this direction of catalyst approach[13] and the more normal α-face hydrogenation (to give the 4β-methyl product) would become predominant. Hydrogenation of N,N′-dimethylcyclobuxine (IId) seemed to give both possible products in a very difficultly separable mixture.

Cyclobuxamine (Ia) was converted to the O,N,N′-triformyl derivative Ij, which was rigorously purified by partition chromatography to remove all traces of cyclobuxine or its derivatives. Strong alkaline hydrolysis of this triformyl derivative was far less satisfactory than for the triacetyl derivative Ie; presumably, the rates of hydrolysis of the assisted[1] tertiary formamide at position 20 and the unassisted secondary formamide at 3 were comparable. By judicious choice of hydrolysis conditions, there was obtained in addition to the N,N′-diformyl derivative Ik (separated by extraction from acidic solution) a mixture of the desired 3-N-monoformamide Im and free cyclobuxamine (Ia), containing a small amount of the 20-N-monoformamide. This mixture was directly reduced with lithium aluminum hydride, and the total product acetylated with acetic anhydride–pyridine. The neutral fraction of the product (not including 20-dimethylamino material deriving from the 20-N-monoformamide) was separated by partition chromatography; the desired product (Ih) appeared at the expected R_f (0.75), while triacetylcyclobuxine (Ie) was retarded far behind it. The thus prepared triacetyl methylated cyclobuxamine (Ih) was completely identical with the same material obtained by hydrogenation of triacetylcyclobuxine, by infrared spectrum, melting point, and mixture melting point; a small sample was hydrolyzed to the diacetyl compound Ii, which while impure still melted at a temperature (270–275°, open capillary) consistent with its identity with Ii (m.p. 290–300°, Köfler block) rather than with IIIc (m.p. 224–231°).

Thus constitution Ia (3β-amino-20α-methylamino-4β,14α-dimethyl-9β,19-cyclo-5α-pregnan-16α-ol) may be considered as established for cyclobuxamine. The occurrence of the abnormal steroid types represented by cyclobuxine (IIa, the first naturally occurring 4-methylene steroid as well as the first steroidal alkaloid discovered having substituents at 4 and 14 or a cyclopropane ring) and cyclobuxamine (Ia, evidently the first naturally occurring 4β-monomethyl steroid to be elucidated) in *B. sempervirens* raises interesting questions, which cannot as yet be answered, concerning possible modifications of the generally accepted pattern and sequence of steroid biogenesis.

Experimental[14]

Partition chromatography was performed on dry-packed columns of appropriate size (usually 1.8 cm. i.d., 20 ml. lower phase

(9) J. L. Beton, T. G. Halsall, E. R. H. Jones, and P. C. Phillips, *J. Chem. Soc.*, 753 (1957).

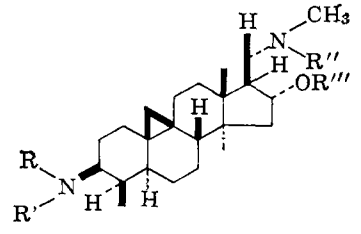
(10) The interactions here are numerous, and further complicated by the possibility of a flip of ring A to the twist conformation in which the effect of the 3β-acetylamino group on the 19-methylene would be greatly accentuated and unaffected by the conformation of the substituent at 4; however, it is almost certain that the observed shift *is* assignable to the interaction with the field of the 3β-acetylamino group, as direct interaction with the 4β-methyl group across the intervening space is highly unlikely.

(11) J. S. G. Cox, F. E. King, and T. J. King, *J. Chem. Soc.*, 514 (1959).

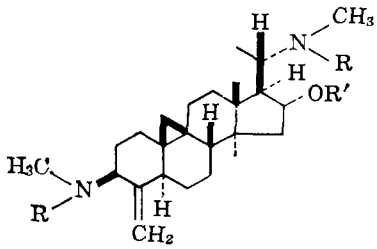
(12) However, note that the failure of this epimerization was attributed[9] to preferential enolization of the 3-ketone toward the 2-position, and the Ruschig degradation of dihydrocyclobuxine to the diketone IV proceeds through the 3-methylimine, which could enolize preferentially toward the 4-position.

(13) Note the restricted rotation of the 3-substituent observable in the n.m.r. spectrum of di- and triacetylcyclobuxine (IIc and IIb)[1]; models indicate that, in the more favored of the two difficultly interconvertible rotamers of this group, the β-face of ring A is protected by the acetyl group.

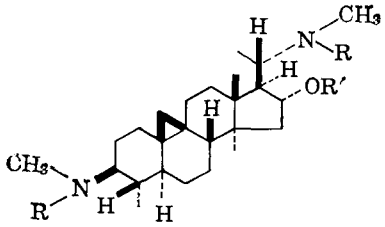
(14) Melting points, taken either on a Köfler block or in an open capillary, are corrected to the nearest degree. Infrared spectra and rotations



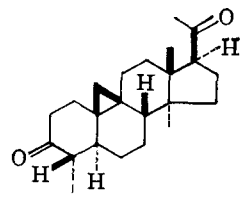
	R	R′	R″	R‴
Ia	H	H	H	H
b	$=C(CH_3)_2$		H	H
c	$CH(CH_3)_2$	H	H	H
d	$CH(CH_3)_2$	$COCH_3$	$COCH_3$	$COCH_3$
e	H	$COCH_3$	$COCH_3$	$COCH_3$
f	H	$COCH_3$	H	H
g	CH_3	CH_3	CH_3	H
h	CH_3	$COCH_3$	$COCH_3$	$COCH_3$
i	CH_3	$COCH_3$	$COCH_3$	H
j	H	CHO	CHO	CHO
k	H	CHO	CHO	H
m	H	CHO	H	H



	R	R′
IIa	H	H
b	$COCH_3$	$COCH_3$
c	$COCH_3$	H
d	CH_3	H



	R	R′
IIIa	H	H
b	$COCH_3$	$COCH_3$
c	$COCH_3$	H



IV

on 30 g. of Celite 545, retention volume 45 ml.), using phenol red as an indicator, and the system hexane–ethylene dichloride–methanol–water (100:50:20:3).[5]

Purified Cyclobuxamineisopropylideneimine (Ib).—Crude cyclobuxamineisopropylideneimine (3.5 g., m.p. about 240° dec.) was dissolved in absolute ethanol (75 ml.) and treated with 2% hydriodic acid (colorless) in 97% ethanol to litmus neutrality. The voluminous precipitate was cooled to 0° for several days and filtered, giving the dihydriodide (3.5 g.), m.p. 310–311° dec.

Anal. Calcd. for $C_{27}H_{48}ON_2I_2 \cdot H_2O$: C, 47.25; H, 7.30; I, 36.76. Found: C, 47.25; H, 7.14; I, 38.95.

(approximated to the nearest degree) are in chloroform solution. N.m.r. spectra were measured on a Varian A-60 spectrometer. Microanalyses were performed by Mr. Joseph Alicino (Metuchen, N. J.) on samples dried at 80° and 0.1 mm. pressure (note that some materials were very hygroscopic, picking up water again before the analyses could be performed); we are grateful for his assistance.

The dihydriodide (3.5 g.) was transformed back to the free base with dilute ammonium hydroxide and chloroform; crystallization of the total thus obtained from acetone gave the imine Ib (2.0 g.), m.p. 244–246° dec. Pure material, obtained by basic hydrolysis (see below) and recrystallization from acetone, showed m.p. 248–250° dec., $[\alpha]^{25}D$ +67° (*c* 1.38); R_f in the partition system described above, 0.75; λ_{max} 6.01 (s) μ; n.m.r. 5.92 (1H, octet, *J* 3, 7, 9.5 c./sec.; $CH_2CHOHCH$), 7.57 (3H, unsplit; N-methyl), 8.02, 8.19 (6H, two sharp peaks; $=C(CH_3)_2$), 8.87, 9.03, (6H, two sharp peaks; 2 tertiary C-methyl), 8.92 (3H, doublet, *J* 6 c./sec.; secondary C-methyl at C-20), 9.32 (3H, doublet, *J* 6 c./sec.; secondary C-methyl at C-4), 9.58, and 9.83 τ (2H, AB doublets, *J* 4 c./sec.; cyclopropyl methylene).

Anal. Calcd. for $C_{27}H_{46}ON_2$: C, 78.20; H, 11.18; N, 6.76. Found: C, 77.72; H, 10.97; N, 7.07.

The diperchlorate was formed with 2% perchloric acid in 97% ethanol as above, and crystallized with the aid of ether; m.p. 314° and up with decomposition.

Anal. Calcd. for $C_{27}H_{48}O_9N_2Cl_2 \cdot H_2O$: C, 51.33; H, 7.93; Cl, 11.16. Found: C, 51.38; H, 7.67; Cl, 11.89.

3-N-Isopropylcyclobuxamine (Ic).—Cyclobuxamineisopropylideneimine (Ib, 200 mg., m.p. 244–246° dec.) was hydrogenated at atmospheric pressure with reduced platinum oxide (100 mg.) in acetic acid (30 ml.); hydrogen uptake stopped after 20 min. at about 94% of the theoretical volume. The solution was filtered and the product (200 mg.) recovered with dilute ammonium hydroxide and chloroform, and crystallized from methanol to give 139 mg. of Ic in two crops, m.p. 267–269° dec. (extremely difficult to handle, asbestos-like needles), no λ_{max} at 6.01 μ; n.m.r. 5.92 (1H, octet, *J* 3, 7, and 9.5 c./sec.; $CH_2CHOHCH$), 7.57 (3H, unsplit; N-methyl), 8.87, 9.03 (6H, two sharp peaks; 2 tertiary C-methyl), 8.90 (3H, doublet, *J* 6 c./sec.; secondary C-methyl at C-20), 9.00 (3H, doublet, *J* 6 c./sec.; secondary C-methyl at C-4), 8.97 (6H, doublet, *J* 8 c./sec.; isopropyl methyls) 9.65, and 9.90 τ (2H, AB doublets, *J* 4 c./sec.; cyclopropyl methylene).

Anal. Calcd. for $C_{27}H_{48}ON_2$: C, 77.83; H, 11.61; N, 6.73. Found: C, 77.27; H, 11.35; N, 7.14.

The O,N,N′-triacetyl derivative Id was prepared with acetic anhydride–pyridine, 30 hr. at room temperature; crystallized from acetone, m.p. 280–281° dec.; λ_{max} 5.77 (s) and 6.15 (vs) μ; n.m.r. 5.03 (1H, complex; $CH_2CH(OAc)CH$), 7.27, 7.32 (3H, split; N-methyl with restricted rotation), 7.87, 7.90, 7.98, 8.03 (6H, 2 split peaks; two N-acetyls with restricted rotation), 8.05 (3H, unsplit; O-acetyl), 8.50–9.50 (18H, exceedingly complex; 6 C-methyl), 9.58, and 9.83 τ (2H, AB doublets, *J* 4 c./sec.; cyclopropyl methylene).

Anal. Calcd. for $C_{33}H_{54}O_4N_2 \cdot 1.5H_2O$: C, 69.55; H, 10.08; N, 4.92. Found: C, 69.27; H, 9.44; N, 5.27.

Cyclobuxamine (Ia).—Cyclobuxamineisopropylideneimine (Ib, 200 mg.) was hydrolyzed 1 hr. with potassium hydroxide (1.0 g.) in refluxing 80% ethanol (10 ml.). A glass tube led from the closed reaction vessel into 2,4-dinitrophenylhydrazine–sulfuric acid reagent (5 ml.). A yellow crystalline solid slowly formed in the receiving solution; it was recovered by filtration (110 mg. = 97%, m.p. 124–127°, m.m.p. with authentic acetone 2,4 dinitrophenylhydrazone 126–129°). The alkaloidal product, recovered with dilute hydrochloric acid, evaporation of the ethanol, ammonium hydroxide, and chloroform, was purified by partition chromatography (see above); the single band (184 mg., infrared, no 6.01 μ) crystallized from acetone to give back the starting imine Ib (159 mg., 248–250°, λ_{max} 6.01 (s) μ); or (160 mg.) from benzene to give pure cyclobuxamine (Ia, 110 mg.), m.p. 209–211° dec., $[\alpha]^{24}D$ +30° (*c* 1.08); λ_{max} 6.32 (m), no 6.01 μ.

Anal. Calcd. for $C_{24}H_{42}ON_2$: C, 76.95; H, 11.30; N, 7.48. Found: C, 77.02; H, 11.86; N, 8.33.

The O,N,N′ triacetyl derivative Ie was produced from either cyclobuxamine (Ia) or the imine Ib with acetic anhydride–pyridine (room temperature, 45 hr. or 100°, 75 min.): crystallized from acetone, m.p. 261–263° dec., R_f on the above partition system 0.34; λ_{max} 2.91 (m), 5.78 (s), 6.02 (s), 6.14 (vs), and 6.60 (s) μ.

Anal. Calcd. for $C_{30}H_{48}O_4N_2 \cdot 1.5\ H_2O$: C, 70.71; H, 9.69; N, 5.50. Found: C, 70.81; H, 9.67; N, 5.69.

3-N-Monoacetylcyclobuxamine (If).—O,N,N′-Triacetylcyclobuxamine (Ie, 60 mg.) was hydrolyzed 2 hr. with potassium hydroxide (1 g.) in refluxing 80% ethanol (2.5 ml.); the basic

product (60 mg.) was amorphous, with a sharp infrared spectrum (λ_{max} 2.90 (m), 6.00 (s), and 6.60 (s) μ).

N,N,N′-Trimethylcyclobuxamine (Ig).—Cyclobuxamine (Ia, 123 mg.) was methylated 8 hr. at room temperature and 7 hr. at reflux with 40% formaldehyde (0.4 ml.) and 88% formic acid (0.6 ml.). The product (146 mg.), recovered with dilute hydrochloric acid, ether, ammonium hydroxide, and chloroform, was crystallized from isopropyl alcohol to give Ig (88 mg.), m.p. 205–219°. Pure material showed m.p. 217–218°, $[\alpha]^{24}D$ +33° (*c* 0.50), infrared spectrum very different from that of N,N′-dimethyldihydrocyclobuxine.[1]

N,N′-Diacetyldihydrocyclobuxine (*A*) (**IIIc**).[1]—Dihydrocyclobuxine (IIIa, 50 mg., m.p. 206–209°) was stirred 75 min. at room temperature with acetyl chloride (46 mg.) and potassium carbonate (1 g.) in benzene (10 ml.); the product (60 mg.), recovered by evaporation of the benzene and partition of the residue between water and chloroform, was purified by partition chromatography. The band of R_f 0.49 (46 mg.) crystallized from ethanol to give 34 mg. (IIIc), m.p. 224–231°, $[\alpha]^{24}D$ −41° (*c* 3.33); λ_{max} 2.94 (m), and 6.20 (vs) μ; n.m.r. 6.00 (1H, complex; $CH_2CHOHCH$), 7.18 (3H, unsplit; N-methyl), 7.15, 7.20 (3H, split peak; N-methyl with restricted rotation), 8.00 (3H, unsplit; N-acetyl), 7.92, 7.98 (3H, split peak; N-acetyl with restricted rotation), 8.93, 9.00 (6H, 2 sharp peaks; 2 tertiary C-methyl), 8.98 (3H, doublet, *J* 6 c./sec.; secondary C-methyl at C-20), 9.24 (3H, doublet, *J* 7 c./sec.; secondary C-methyl at C-4), 9.38, and 9.75 τ (2H, broad; cyclopropyl methylene).

Hydrogenation of N,N′-Diacetylcyclobuxine. N,N′-Diacetyldihydrocyclobuxine (B) (**Ii**).[1]—N,N′-Diacetylcyclobuxine (IIc, 35 mg., m.p. 283–285° dec.) was hydrogenated at atmospheric pressure with reduced platinum oxide (20 mg.) in 20% ethanolic acetic acid (12 ml.); uptake was 1 mole equiv. in 2 hr. The product (31 mg.) was recovered with water, evaporation of the ethanol, treatment with ammonium hydroxide and chloroform, and crystallization from ethanol to give 15 mg. Ii, m.p. 290–300° dec., $[\alpha]^{21}D$ −14° (*c* 0.62); infrared spectrum quite different from that of IIIc in the 6.5–12.0 μ region; n.m.r. 5.93 (1H, complex; $CH_2CHOHCH$), 7.16 (3H, unsplit; N-methyl), 7.25, 7.30 (3H, split peak; N-methyl with restricted rotation), 7.97 (3H, unsplit; N-acetyl), 7.92, 8.00 (3H, split peak; N-acetyl with restricted rotation), 8.82, 8.93 (6H, two sharp peaks; 2 tertiary C-methyl), 8.95 (3H, doublet, *J* 6 c./sec.; secondary C-methyl at C-20), 9.21 (3H, doublet, *J* 7 c./sec.; secondary C-methyl at C-4), 9.67, and 9.90 τ (2H, broad; cyclopropyl methylene).

O,N,N′-Triacetyldihydrocyclobuxine (*A*) (**IIIb**).[1]—Dihydrocyclobuxine (IIIa, 50 mg., m.p. 205–208°) was acetylated with acetic anhydride (1.0 ml.) in pyridine (0.5 ml.), 40 hr. at room temperature; the product (61 mg.), recovered with dilute ammonium hydroxide and chloroform, was purified by partition chromatography. The band of R_f 0.74 (57 mg.) was crystallized from acetone–isopropyl ether to give 31 mg. of IIIb, m.p. 230–234°; pure material showed m.p. 233–235°, $[\alpha]^{23}D$ −62° (*c* 3.47); λ_{max} 5.80 (s) and 6.17 (vs) μ; n.m.r. 5.00 (1H, complex; $CH_2CH(OAc)CH$), 7.05 (3H, unsplit; N-methyl), 7.18, 7.22 (3H, split peak; N-methyl with restricted rotation), 7.98 (3H, unsplit; N-acetyl), 7.87 (3H, broadened; N-acetyl with restricted rotation), 8.01 (3H, unsplit; O-acetyl), 8.87, 8.88 (6H, two sharp peaks; 2 tertiary C-methyl), 8.92 (3H, doublet, *J* 6 c./sec.; secondary C-methyl at C-20), 9.17 (3H, doublet, *J* 7 c./sec.; secondary C-methyl at C-4), 9.37, and 9.68 τ (2H, AB doublets, *J* 4 c./sec.; cyclopropyl methylene).

Anal. Calcd. for $C_{31}H_{50}O_4N_2 \cdot 0.5H_2O$: C, 70.98; H, 9.81; N, 5.35. Found: C, 70.64; H, 9.42; N, 5.58.

Hydrogenation of O,N,N′-Triacetylcyclobuxine. O,N,N′-Triacetyldihydrocyclobuxine (*B*) (**Ih**).[1]—O,N,N′-Triacetylcyclobuxine (IIb, 100 mg., m.p. 246–252° dec.) was hydrogenated with reduced platinum oxide (25 mg.) in 10% ethanolic acetic acid (20 ml.); the uptake was 1 mole equiv. in 1 hr. The product (105 mg.), recovered as for the diacetate above, was purified by partition chromatography; the band of R_f 0.70 (103 mg.) was crystallized from ethyl acetate to give 48 mg. of Ih, m.p. 225–227°, $[\alpha]^{23}D$ −35° (*c* 2.19); infrared spectrum quite different from that of IIIb in the 6.5–12.0 μ region; n.m.r. 5.00 (1H, complex; $CH_2CH(OAc)CH$), 7.18 (3H, unsplit; N-methyl), 7.17, 7.25 (3H, split peak; N-methyl with restricted rotation), 8.00 (3H, unsplit; N-acetyl), 7.89, 7.95 (3H, split peak; N-acetyl with restricted rotation), 8.02 (3H, unsplit; O-acetyl), 8.87, 8.88 (6H, two sharp peaks; 2 tertiary C-methyl), 8.92 (3H, doublet, *J* 6 c./sec.; secondary C-methyl at C-20), 9.20 (3H, doublet, *J* 7 c./sec.; secondary C-methyl at C-4), 9.58, and 9.85, τ (2H, AB doublets, *J* 4 c./sec.; cyclopropyl methylene).

Anal. Calcd. for $C_{31}H_{50}O_4N_2 \cdot 0.5\ H_2O$: C, 70.98; H, 9.81; N, 5.35. Found: C, 70.45; H, 9.97; N, 5.42.

O,N,N′-Triformylcyclobuxamine (Ij).—Cyclobuxamine (Ia, 250 mg., purified through the dihydriodide of the imine) was treated 67 hr. at room temperature with formylating mixture (20 ml.) prepared by heating together at 70° for 1 hr. a 1:1 mixture of formic acid (98%) and acetic anhydride. At the end of this period, the formylating solution was removed under reduced pressure at 60°, and the product recovered with water, dilute ammonium hydroxide, and methylene chloride (backwashing the organic extracts with dilute hydrochloric acid to remove residual amine). The neutral product (250 mg.) crystallized from ethyl acetate to give the triformyl derivative Ij (141 mg.), m.p. 260–264°. Material rigorously purified by partition chromatography (R_f 0.33, widely separated from O,N,N′-triformylcyclobuxine[1] at R_f 0.56) crystallized from ethyl acetate in needles, m.p. 275–276° dec.; λ_{max} 5.78 (s), 5.98 (s), 6.01 (vs), and 6.63 (s) μ.

Anal. Calcd. for $C_{2:}H_{42}O_4N_2$: C, 70.71; H, 9.23; N, 6.11. Found: C, 70.55; H, 9.47; N, 6.02.

O,N,N′-Triacetyldihydrocyclobuxine (*B*) (**Ih**).—O,N,N′-Triformylcyclobuxamine (Ij, 130 mg.) was hydrolyzed 2 hr. with sodium hydroxide (400 mg.) in refluxing 90% methanol (10 ml.). The product was divided into neutral and basic fractions by partition between dilute hydrochloric acid and methylene chloride; the neutral fraction (100 mg.) showed bands in the infrared spectrum at 5.98 (s), 6.01 (vs), and 6.63 (s) μ, indicative of the N,N′-diformamide Ik. A sample was crystallized from methanol to give **N,N′-diformylcyclobuxamine** (**Ik**), m.p. 330–331° dec.

Anal. Calcd. for $C_{26}H_{42}O_3N_2$: C, 72.52; H, 9.83. Found: C, 72.64; H, 9.79.

The basic fraction from the above hydrolysis (25 mg.) was reduced 1 hr. with lithium aluminum hydride (50 mg.) in refluxing ether (10 ml.). The product (25 mg.), recovered with saturated sodium sulfate solution, refluxing 30 min., and filtration, showed only a low residual band at 6.0 μ in the infrared spectrum. The total was acetylated with acetic anhydride (2 ml.) and pyridine (2 ml.) for 18 hr. at room temperature and 1 hr. at 65°. The neutral product, recovered with dilute hydrochloric acid and methylene chloride (backwashing the organic extracts with dilute ammonium hydroxide), was purified by partition chromatography; the fraction of R_f 0.75 (11 mg.) crystallized from ether to give 3 mg., m.p. 220–226°, m.m.p. with authentic O,N,N′-triacetyldihydrocyclobuxine (B) (Ih) of m.p. 223–226°/223–226°; the infrared spectra of the two were completely superimposable.

The mother liquor from the above crystallization (5 mg.) was hydrolyzed 15 min. in refluxing 5% ethanolic potassium hydroxide (2 ml.); the product (4 mg., λ_{max} 2.94 (m), 6.17 (vs), no 5.80 μ) gave upon trituration with ether small crystals, m.p. 270–275° dec. (open capillary).