Alkaloids from *Croton* species. Part III.* The Constitution of the Proaporphines Crotonosine, "Homolinearisine", Base A, and the Dihydroproaporphine Linearisine

By L. J. Haynes, K. L. Stuart, D. H. R. Barton, and G. W. Kirby

A full report on the proaporphine alkaloids isolated from *Croton linearis* Jacq. is presented. Crotonosine is shown to have the structure (I; R = R^1 = H), base A is *NO*-dimethylcrotonosine and is identical with pronuciferine, "homolinearisine" is L-(−)-*N*-methylcrotonosine, and linearisine is 8,9-(or 11,12)-dihydro-L-(−)-*N*-methylcrotonosine.

COMMUNICATIONS [1,2] on the structures of proaporphine alkaloids from *Croton linearis* Jacq. reported that crotonosine has the structure (I; R = R^1 = H), pronuciferine [3,4] (base A) is *NO*-dimethylcrotonosine, "homolinearisine" [5] is L(−)-*N*-methylcrotonosine (II) and linearisine is 8, 9- (or 11, 12)-dihydro-L(−)-*N*-methylcrotonosine. We now present a full account of the work leading to these structures.

The location of aromatic oxy-substituents was rigorously established by conversion of the alkaloids into compounds of known structures by unambiguous routes. Hofmann degradation of *NOO*-trimethylapocrotonosine methiodide, produced by methylation of the aporphine obtained by acid rearrangement of crotonosine, gave the phenanthrene (III) when the product was refluxed with methyl iodide. The melting point of this methiodide was in good agreement with that of the phenanthrene derivative obtained similarly from tuduranine [6] (IV; R = H, R^1 = R^2 = Me, R^3 = OH). The nuclear mag-

* Part II L. J. Haynes and K. L. Stuart, *J. Chem. Soc.*, 1963, 1789.

[1] L. J. Haynes, K. L. Stuart, D. H. R. Barton, and G. W. Kirby, *Proc. Chem. Soc.*, 1963, 280.

[2] L. J. Haynes, K. L. Stuart, D. H. R. Barton, and G. W. Kirby, *Proc. Chem. Soc.*, 1964, 261.

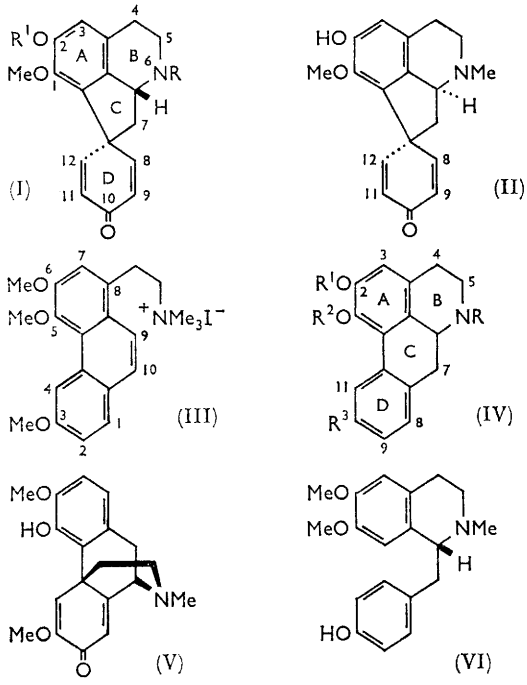
[3] K. Bernauer, *Helv. Chim. Acta*, 1963, **46**, 1783; 1964, **47**, 2119, 2122.

[4] M. P. Cava, K. Nomura, R. H. Schlessinger, K. T. Buck, B. Douglas, R. F. Raffauf, and J. A. Weisbach, *Chem. and Ind.*, 1964, 282.

[5] L. J. Haynes and K. L. Stuart, *J. Chem. Soc.*, 1963, 1784, 1789.

[6] K. Goto, *Annalen*, 1935, **521**, 175.

netic resonance (n.m.r.) spectrum was completely consistent with the structure (III). A direct comparison showed that *NOO*-trimethylapocrotonosine methiodide was identical (infrared spectrum, mixed m. p., thin-layer chromatography) with *N*-methyltuduranine methyl ether



methiodide prepared from authentic samples of tuduranine hydrochloride. It should be noted that acid rearrangement of proaporphines gives only one product, namely, the aporphine with the ring D oxygen substituent at C(10) and there is no evidence of the C(9) isomer's being formed.

Reduction of *N*-methylcrotonosine with sodium borohydride [7] and dehydration of the product gave 2-hydroxy-1-methoxyaporphine (IV; R = R^2 = Me, R^1 = R^3 = H) which by direct comparison was identical with a sample from *Nelumbo nucifera* Gaertn.[8]

The orientation of the methoxyl and hydroxyl groups in crotonosine was confirmed by n.m.r.-controlled deuterium experiments. By using alkaline deuterium oxide to exchange hydrogens *ortho* and *para* to the phenolic groups [9] it was shown that apoglaziovine (IV; R = R^1 = Me, R^2 = H, R^3 = OH) contains two such exchangeable hydrogens and apocrotonosine (IV; R = R^1 = H, R^2 = Me, R^3 = OH) three. *N*-Acetyltetrahydrocrotonosine under similar conditions showed the presence of one *ortho* phenolic hydrogen.

N.m.r. spectral characteristics of the proaporphines are clearly dissimilar from those of other dienone structures like salutaridine (V) which was synthesised [10] before its isolation from *Croton salutaris*,[11] and by comparing n.m.r. spectra of *NO*-diacetylcrotonosine and *NO*-tetrahydrocrotonosine it could be shown that *NO*-diacetylcrotonosine has an olefinic : aromatic proton ratio of 4 : 1 and not 2 : 2 as is the case for salutaridine.

The splitting pattern of the four olefinic protons in the proaporphines is as expected for the cyclohexadienone system seen in structures (I) and (II) (see Experimental section). It may be noted that in this and related structures containing an *N*-acetyl group, the methyl signal due to the acetyl group invariably appears at τ 7·78—7·82 when the n.m.r. spectra are determined in $CDCl_3$.

Pronuciferine, which has been synthesised by Bernauer,[12] is the *NO*-dimethyl derivative of crotonosine since both yield identical aporphine derivatives.[5] Pronuciferine and stepharine (I; R = H, R^1 = Me) were shown by chemical methods to have the D-configuration by their conversion into *D*-(−)-armepavine (VI).[4] From this correlation, the D-configuration of crotonosine is established and crotonosine therefore has structure (I; R = R^1 = H).

Further purification of "homolinearisine"[5] showed that it had the elementary composition $C_{18}H_{19}NO_3$ rather than $C_{19}H_{23}NO_3$ as previously reported, and by direct correlation with D-(+)-*N*-methylcrotonosine, $[\alpha]_D^{16}$ +122°, it was shown to be L-(−)-*N*-methylcrotonosine, $[\alpha]_D^{16}$ −116·5°, with the structure (II). All the spectral results are in full accord with this assignment.

Linearisine on hydrogenation gave a dihydro-compound, $[\alpha]_D^{20}$ −60·6°, which was similarly enantiomeric with D-(+)-*N*-methyltetrahydrocrotonosine, $[\alpha]_D^{20}$ + 59°: it shows the infrared and ultraviolet light absorption for a simple αβ-unsaturated ketone and its n.m.r. spectrum has an AB quartet at τ 3·16, 3·92 (J_{AB} = 10 c./sec.). It thus has the structure of either 8,9- or 11,12-dihydro L-(−)-*N*-methylcrotonosine: in Part V,[13] G. Snatzke shows by a study of the circular dichroism of this and other proaporphine alkaloids that linearisine is most probably the 8,9-dihydro-derivative of structure (II). His work also confirms the configurations assigned in this paper.

We are currently examining the alkaloidal content of other *Croton* species available in Jamaica and report the isolation of crotonosine from *C. discolor* Willd.

EXPERIMENTAL

All m. p.s. were determined on a Kofler hot-stage apparatus and are uncorrected. Nuclear magnetic resonance spectra were determined on a Varian 60 Mc./sec. instrument with tetramethylsilane as internal standard in deuterochloroform, except where otherwise stated; results are in Tables 1 and 2.

N-Methyl-OO-dimethylapocrotonosine Methine Methiodide

[7] B. Gilbert, M. E. A. Gilbert, M. M. De Oliveira, O. Ribeiro, E. Wenkert, B. Wickberg, U. Hollstein, and H. Rapoport, *J. Amer. Chem. Soc.*, 1964, **86**, 694.
[8] M. Tomita, Y. Watanabe, and H. Furukawa, *J. Pharm. Soc. Japan*, 1961, **81**, 942.
[9] G. W. Kirby and L. Ogunkoya, *J. Chem. Soc.*, 1965, 6914, and references there cited.
[10] D. H. R. Barton, G. W. Kirby, W. Steglich, and G. M. Thomas, *Proc. Chem. Soc.*, 1963, 203.
[11] D. H. R. Barton, G. W. Kirby, W. Steglich, G. M. Thomas, A. R. Battersby, T. A. Dobson, and H. Ramuz, *J. Chem. Soc.*, 1965, 2423.
[12] K. Bernauer, *Experientia*, 1964, **20**, 380.
[13] G. Snatzke, and G. Wollenberg, *J. Chem. Soc.* (*C*), 1966, 1681.

(*III*).—*NOO*-Trimethylapocrotonosine methiodide[5] (0·048 g.) was refluxed in 5% methanolic potassium hydroxide (10 ml.) for 12 hr. The methanol was removed *in vacuo* and the base extracted into chloroform. A wax was obtained by removal of the chloroform and this was refluxed with methyl iodide (1 hr.) to give the substituted phenanthrene (0·025 g.) (III). Purification was by passage through

J = 3 c./sec.] and bands at τ 5·99, 6·08, 6·16 (3 OMe) (Found: C, 54·9; H, 5·9. $C_{22}H_{28}INO_3$ requires C 54·9; H, 5·9%).

N-*Acetyl*-O-*ethylcrotonosine* (I; R = Ac, R^1 = Et).—*NO*-Diacetylcrotonosine[5] (0·63 g.) was dissolved in 5% sodium hydroxide (20 ml.) and methanol (15 ml.). The yellow solution was allowed to stand for 2 hr. The

TABLE 1

N.m.r. results for aporphines (IV)

Structure	Alkaloid	R	R^1	R^2	R^3
(IVa)	*NO*-Diacetylasimilobine	Ac	Me	Ac	H
(IVb)	*NO*-Diacetyllanolobine	Ac	$-CH_2-$		H (C9) (OAc)
(IVc)	*NO*-Diacetylmichelalbine	Ac	$-CH_2-$		H (C7) (OAc)
(IVd)	*N*-Acetyllaurotetanine	Ac	Me	Me	OMe (C9) (OH)
(IVe)	*N*-Acetylnornuciferine	Ac	Me	Me	H
(IVf)	*N*-Acetyl-*OO*-dimethylapocrotonosine	Ac	Me	Me	OMe
(IVg)	*NO*-Diacetyl-*O*-methylapocrotonosine	Ac	Ac	Me	OMe
(IVh)	Apocrotonosine	H,HCl	H	Me	OH

Location of proton (values in τ)

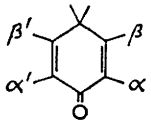
Structure	R	1	2	3	6a	7	8	9	10	11
(IVa)	7·80	7·64	6·42 *							
(IVb)	7·80	3·96	4·08 [a]					7·71 *		
(IVc)	7·79					8·13 *				
(IVd)	7·78	6·27	6·03	3·27			3·18		6·03	1·72 †
(IVe)	7·80	6·29	6·08	3·25			2·65 [b]	2·65 [b]	2·65 [b]	1·53 †
(IVf)	7·82	6·28	6·14 or 6·09	3·28	~5·20		2·76 [c]	3·08—3·23 [d]	6·09 or 6·14	1·84
(IVg)	7·81	6·39	7·63	3·10	~5·0		2·78 [f]	3·05—3·25 [g]	6·17	1·95 [h]
(IVh) [i]				3·40			2·95	3·35—3·50 [k]		2·60 [l]

[a] Doublet at each. [b] Complex pattern.† [c] *ortho* coupled with C(9); J = 8 c./sec. [d] *ortho* coupled with C(8); J = 8 c./sec. and *meta* coupled with C(11); J = 3 c./sec. [e] J = 3 c./sec. [f] J = 8·5 c./sec. [g] *ortho* coupled with C(8); J = 8·5 c./sec.; *meta* coupled with C(11); J = 2·5 c./sec. [h] J = 2·5 c./sec. [i] Run in D_2O with Bu^tOH as internal standard (τ = 8·77). [j] J = 8 c./sec. [k] Partly masked by C(3) proton; *meta* splitting J = 2·5 c./sec. [l] J = 2·5 c./sec.

* M. Tomita and M. Kozuka, *Yukugaku Zasshi*, 1965, **85**, 77. † W. H. Baarchers, R. R. Arndt, K. Pachler, J. A. Weisbach, and B. Douglas, *J. Chem. Soc.*, 1964, 4778.

TABLE 2

N.m.r. results on proaporphines and related compounds (τ values)



Alkaloid	*N*-Substituent	C(1) (OMe)	C(2)	C(3)	C(6a)	
Crotonosine [a]				3·43		2·96 [b]; 3·80 [c]
NO-Diacetylcrotonosine	7·82 (Ac)	6·42	7·71 (Ac)	3·07	4·8 [d]	2·95 [e]; 3·65 [f]
L-(−)-*N*-Methylcrotonosine [g]		6·48?		3·37		2·85 [h]; 3·76 [i]
Linearisine	7·58 (Me)	6·30		3·58		[3·16; 3·92] [j]
NO-Diacetyltetrahydrocrotonosine	7·80 (Ac)	6·18	7·55 (Ac)	3·25	5·1	

[a] Run in dimethyl sulphoxide (DMSO). [b] ββ′ Protons giving 8 lines; $J_{\beta\beta'}$ = 2·5, $J_{\alpha\beta}$ = $J_{\alpha'\beta'}$ = 10 c./sec. [c] αα′ Protons giving 8 lines; $J_{\alpha\alpha'}$ = 1·5 c./sec. [d] Deshielded by *N*-acetyl group and coupled with the C(7) methylene protons which by decoupling were found to absorb at τ 7·68 and 7·73. [e] $J_{\beta\beta'}$ = 2·5 c./sec. [f] $J_{\alpha\alpha'}$ = 1·6 c./sec. [g] Run in DMSO. [h] ββ′ Protons; J = 2·5 c./sec. [i] αα′ Protons, $J_{\alpha\beta}$ = $J_{\alpha'\beta'}$ = 10 c./sec. [j] Dihydro-ring D giving an AB quartet, J_{AB} = 10 c./sec.

alumina in methanol and finally by recrystallisation from methanol, m. p. 286° [lit., 279° [6]; 276—277° (decomp.) [14]]; $\nu_{max.}$ 1600 and 1500 (aromatic) cm.$^{-1}$.

The n.m.r spectrum in dimethyl sulphoxide with tetramethylsilane as internal standard showed the following bands: τ 0·9 [C(4) proton, J = 2·5 c./sec.]; τ 2·12 [C(1) proton, J = 9 c./sec.]; τ 2·30 [C(9) and [(C10) protons]; τ 2·40 [C(7) proton]; τ 2·77 [C(2) proton, J = 9 c./sec.;

[14] A. Chatterjee, P. L. Majumder, R. Mukherjee, S. K. Saha, and S. K. Talapatra, *Tetrahedron Letters*, 1965, 1539.

methanol was removed and the phenolic base was extracted into chloroform. After removal of the chloroform, addition of methanol–acetone gave *N*-acetylcrotonosine, m. p. 205°; $\nu_{max.}$ 3150 (OH), 1652, 1625 (cross-conjugated dienone) cm.$^{-1}$ (Nujol mull). *N*-Acetylcrotonosine was refluxed with ethyl iodide in ethanol and with sodium carbonate (2 hr.). The ethanol was removed *in vacuo*, water added, and made acidic with dilute hydrochloric acid, and then extracted with chloroform (3 × 10 ml.). Crystals obtained (0·40 g.) were recrystallised from ethyl acetate–benzene, m. p.

170—174°, $\nu_{max.}$ 1650 (cross-conjugated dienone) cm.$^{-1}$ (Found: C, 71·2; H, 6·5; N, 3·8; O, 18·2. $C_{21}H_{23}NO_4$ requires C, 71·4; H, 6·6; N, 4·0; O, 18·1%).

N-*Methylcrotonosine* (I; R = Me; R^1 = H).—Crotonosine (0·107 g.) was refluxed with formaldehyde (3 ml.) and 80% formic acid (3 ml.) [15] for 3 hr. The formaldehyde was removed *in vacuo* rather than by repeated treatment with hydrochloric acid followed by evaporation to dryness. (This was done to safeguard against possible acid rearrangement.) Water was added, the mixture basified, and extracted with chloroform. *N*-Methylcrotonosine (0·065 g.), crystallised from acetone–methanol, had m. p. 216—218° (decomp.), $[\alpha]_D^{16}$ +122° (*c* 0·45, MeOH), $\nu_{max.}$ 1653, 1622 (dienone), 1600, and 1500 (aromatic) cm.$^{-1}$. On chromatographic comparison on buffered Whatman No. 4 paper (pH 6·2) with development in butanol at 24° it had R_f = 0·82, while in the same system pronuciferine (base A) had R_f = 0·87, and glaziovine [7] 0·77—0·78 (Found: C, 72·5; H, 6·5. $C_{18}H_{19}NO_3$ requires C, 72·7; H, 6·4%).

D-(+)-N-*Methyltetrahydrocrotonosine*.—*N*-Methylcrotonosine (0·05 g.) was hydrogenated in ethanol using 5% palladium on carbon, and showed an uptake of 2 mol. of hydrogen. The product (0·04 g.) was recrystallised from ethanol. Decomposition started at ~210° and crystals melted at 225—228° (decomp.), $[\alpha]_D^{20}$ +59° (*c* 0·51, MeOH) (Found: C, 71·8; H, 7·8. $C_{18}H_{23}NO_3$ requires C, 71·7; H, 7·7%). Its thin-layer chromatograms and infrared spectrum were identical to those of dihydrolinearisine, $[\alpha]_D^{20}$ − 60·6 (*c* 0·33, MeOH); decomposition started ~210° and it melted at 225—227°; $\nu_{max.}$ 1700 (>C=O) cm.$^{-1}$ in Nujol.

2-*Hydroxy*-1-*methoxyaporphine* (IV; R = R^2 = Me, R^1 = R^3 = H).—*N*-Methylcrotonosine (0·073 g.) was stirred in 5% aqueous methanol (10 ml.) with sodium borohydride (91 mg.) for 50 min.[7] The solution was acidified with dilute hydrochloric acid to effect dehydration, then basified and extracted into chloroform. The chloroform was evaporated, and a few drops of ether–acetone added produced crystals (0·045 g.), m. p. 194—196°, $[\alpha]_D^{20}$ −253° (*c* 0·45, $CHCl_3$) lit.,[8] $[\alpha]_D^{22}$ −255° ($CHCl_3$)[8], which gave an identical infrared spectrum and showed no depression of m. p. when mixed with an authentic sample from *Nelumbo nucifera* Gaertn.

L-(−)-N-*Methylcrotonosine* ("*Homolinearisine*") (II).—Thin-layer chromatographic separation of "homolinearisine" on silica in methanol–ammonium hydroxide (97 : 3) showed that "homolinearisine" was contaminated by linearisine (t.l.c. plates treated with iodine vapour showed a characteristic red spot for linearisine). Pure "homolinearisine" was obtained by countercurrent distribution of the mixture (0·66 g.) using 0·2N-acetate buffer (pH 5·59) as the moving phase and chloroform as the stationary phase. Thirty transfers gave L-(−)-*N*-methylcrotonosine (0·45 g.) in tubes 1—10 and linearisine in tubes 20—30. This base can, however, be obtained directly from the crude alkaloid mixture as earlier outlined [5] if thin-layer chromatography is used to determine the separation of bases. L-(−)-*N*-Methylcrotonosine was recrystallised from ethanol, m. p. 218—220° (decomp.); $\nu_{max.}$ 1664, 1625 (dienone) cm.$^{-1}$, $[\alpha]_D^{16}$ −116·5° (*c* 0·85, MeOH) and is the enantiomer of D-(+)-*N*-methylcrotonosine (Found: C, 72·6; H, 6·5; N, 4·9; OCH_3, 10·8, *N*-CH_3, 4·9. $C_{18}H_{19}NO_3$ requires C, 72·7; H, 6·4; N, 4·7; OCH_3, 10·4; *N*-CH_3, 5·0%).

(With Dr. D. S. Bhakuni) *Apocrotonosine* (N-*nor*-2, 10-*dihydroxy*-3-*methoxyaporphine*).—Crotonosine (50 mg.) in 6N-hydrochloric acid (1 ml.) was heated at 100°. The product began to separate as needles within *ca.* 5 min. and after 1 hr. the mixture was cooled in ice and centrifuged. The crystals were washed with a little 6N-hydrochloric acid and dried *in vacuo* over potassium hydroxide. This material (50 mg.) was sufficiently pure for further experiments (see below). A specimen was dissolved in water, treated with charcoal, then caused to crystallise out by addition of concentrated hydrochloric acid. *Apocrotonosine hydrochloride hemihydrate* formed needles, m. p. 218—220°, $[\alpha]_D$ −148° (*c* 0·42, water) (Found: C, 62·5; H, 5·95. $C_{17}H_{18}ClNO_3$, $\frac{1}{2}H_2O$ requires C, 62·2; H, 5·8%).

Deuteration of Apocrotonosine and Apoglaziovine.—Apocrotonosine hydrochloride hemihydrate (40 mg.) and potassium t-butoxide (50 mg.) in deuterium oxide (*ca.* 1 ml.) were sealed under nitrogen in an n.m.r. tube and heated at 100°. After 1 hr. the sharp n.m.r. singlet corresponding to the proton at C(3) had disappeared, there being little change in the remainder of the spectrum. More prolonged heating caused the signals corresponding to protons at C(9) and C(11) to disappear also. Apoglaziovine [7] was deuterated under the same conditions for 8 hr. Only two protons [positions C(9) and C(11)] were replaced by deuterium.

Deuteration of Diacetyltetrahydrocrotonosine and Tetrahydroglaziovine.—Diacetyltetrahydrocrotonosine (70 mg.) and potassium t-butoxide (140 mg.) in deuterium oxide (0·5 ml.) were heated at 100° under nitrogen for 7 hr. Water (2 ml.) and excess of solid carbon dioxide were added and the precipitated product extracted with chloroform (3 × 4 ml.). The chloroform was evaporated and the phenolic residue acetylated in the usual way. The resulting deuterated diacetyltetrahydrocrotonosine (60 mg.), m. p. 106—108°, $\nu_{max.}$ 1760, 1715, and 1630 cm.$^{-1}$ (in chloroform) showed no aryl proton band in the n.m.r. spectrum. When tetrahydroglaziovine was deuterated under the same conditions no change of the aryl proton occurred.

Isolation of Crotonosine from C. discolor *Willd.*—1 kg. of dried and powdered *C. discolor* was extracted with 4 l. of 2% tartaric acid and worked up in the usual way.[5] 3·25 g. (0·325% yield) of the crude alkaloid mixture was obtained. The mixture was seeded with crotonosine to give 0·59 g. of crotonosine which was identical in all respects to this base from *C. linearis* Jacq. (thin-layer chromatography, infrared spectrum, optical rotation).

We thank Dr. B. Gilbert and Professors M. P. Cava, K. Goto, and M. Tomita for alkaloid samples, the Scientific Research Council of Jamaica and the Tropical Products Institute, London, for financial assistance, and Dr. D. S. Bhakuni for the experiments indicated.

Chemistry Department,
University of the West Indies,
Kingston 7, Jamaica.
Imperial College,
London S.W.7.

[6/263 *Received, March* 1*st*, 1966]

[15] S. M. Kupchan, B. Dasgupta, E. Fujita, and M. L. King, *Tetrahedron*, 1963, **19**, 227.