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Opium Alkaloids V. Structure of Porphyroxine

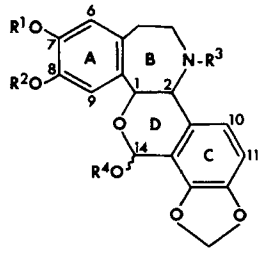
By EINAR BROCHMANN-HANSSEN and KENTARO HIRAI

Porphyroxine is a monophenolic alkaloid isolated from opium. It contains a methyl acetal group and has three asymmetric centers. The position of the phenolic hydroxyl group has been determined by NMR spectroscopy. Evidence has been presented for the configuration at position-14.

In 1837, MERCK (1) discovered a minor opium alkaloid which he named porphyroxine because it produced a red color when heated with dilute mineral acids. He suggested that this reaction might be used as a test for opium in forensic cases (2). The concentration of the alkaloid varies with the source of opium, and the color reaction is useful for determining the geographic origin of opium seized in illicit traffic (3, 4). Although there has been a great deal of interest in porphyroxine for more than 100 years and many investigators claim to have isolated it, it is only recently that it has been obtained in a form sufficiently pure for structure studies (5–7). In 1962 the empirical formula was determined by Pfeifer and Teige (6) as $C_{20}H_{21}NO_6$. In a series of investigations of the alkaloids of the genus *Papaver*, Pfeifer *et al.* (8, 9) described a total of six alkaloids which gave the characteristic red color with mineral acids. They coined the name "papaverrubines" for these compounds. All papaverrubines are secondary amines. The *N*-methyl derivatives, several of which have been found in the *Papaveraceae* family, do not give the red color reaction with acids. The gross structures of some of these alkaloids are illustrated in I–V.

Papaverrubine A (I), glaudine (III), isorhoeadine (IV), and glaucamine (V) have a *trans*-configuration at the junction of ring B and ring D, whereas papaverrubine E (I), oreodine (III), and rhoeadine (IV) have the *cis*-configuration (10–12).

Porphyroxine (II) has been established as a member of the B/D *trans*-series by methylation to glaudine (13). The only structural features of porphyroxine which have not yet been reported are the exact position of the hydroxyl group in ring A and the stereochemistry at position-14. During studies



I, $R^1 + R^2 = CH_2$, $R^3 = H$, $R^4 = CH_3$; papaverrubine A and E
II, $R^1 = H$ or CH_3, $R^2 = CH_3$ or H, $R^3 = H$, $R^4 = CH_3$; papaverrubine D (porphyroxine)
III, $R^1 = R^2 = R^3 = R^4 = CH_3$; glaudine and oreodine
IV, $R^1 + R^2 = CH_2$, $R^3 = R^4 = CH_3$; isorhoeadine, rhoeadine
V, $R^1 = R^2 = R^3 = CH_3$, $R^4 = H$; glaucamine

of the minor alkaloids of opium, this alkaloid was isolated. The isolation and subsequent studies of the structure of porphyroxine are described.

EXPERIMENTAL

Isolation—Four pounds of powdered opium of Indian origin were extracted, and a preliminary separation of alkaloid groups was carried out as described in a previous communication (14). The phenolic alkaloid fraction was subjected to preparative thin-layer chromatography on silica gel with chloroform–methanol (9:1). Bands were obtained of reticuline, isoboldine, and scoulerine. One band with a high R_f value was a mixture of several alkaloids which were separated on an alumina column (Merck, neutral, activity IV) with chloroform. The first fractions contained an alkaloid which, when crystallized from methanol, melted at 190° (micro m.p. K.). [Lit. m.p. 192° (6).] It gave a single peak by gas chromatography on silicone rubber SE-30 (3.8%, 4 ft., 200°) and single spots by thin-layer chromatography on silica gel in two different solvent systems (15). The spots produced a red color when

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TABLE I—CHEMICAL SHIFTS (τ) AND COUPLING CONSTANTS (c.p.s.) FOR THE PROTON RESONANCES OF PORPHYROXINE, GLAUDINE, EPIGLAUDINE, AND *N*-METHYLPORPHYROXINE IN $CDCl_3$ AT 60 Mc./sec.

Compd.	N—CH_3	14-OCH_3	Ar-OCH_3	14-H OCH-OCH_3	—OCH_2O—[a]	6-H	9-H	10- and 11-H	2-H[a]	1-H[a]
Porphyroxine	...	6.33	6.12	4.17	4.08[d], 3.93[d] J = 1.2	3.37	2.60	3.31, 3.17, 2.98, 2.84 AB quartet J = 8.5	6.13[d] J = 9	5.31[d] J = 9
Glaudine (10)	7.76	6.31	6.12 (6H)	4.23	4.08[d], 3.96[d] J = 1.5	3.33	2.63	3.28, 3.14, 3.01, 2.87 AB quartet J = 8.5	5.92[d] J = 9	4.81[d] J = 9
Epiglaudine	7.70	6.44	6.10 (6H)	4.25	4.02[d], 3.95[d] J = 1.7	3.32	2.68	3.28, 3.14, 3.02, 2.88 AB quartet J = 8.5	5.98[d] J = 9	4.43[d] J = 9
N-Methyl-porphyroxine	7.77	6.32	6.12	4.23	4.05[d], 3.93[d] J = 1.5	3.33	2.63	3.27, 3.13, 3.00, 2.86 AB quartet J = 8.5	5.93[d] J = 9	4.83[d] J = 9

[a] d, Doublet.

sprayed with dilute hydrochloric acid and heated. The IR and UV spectra were identical with those reported for porphyroxine (6). The NMR spectrum[1] in $CDCl_3$ (internal TMS standard) was consistent with the structure assigned to porphyroxine (II).

RESULTS AND DISCUSSION

The chemical shifts and coupling constants are listed in Table I. The C-1 and C-2 protons resonated as an AX quartet, $J_{1,2}$ 9 c.p.s., indicative of a B/D *trans*-configuration and in good agreement with the values reported for glaudine (10) (*cf.* Table I), glaucamine (11), and isorhoeadine (12). A smaller coupling constant ($J_{1,2}$ 2–2.5 c.p.s.) is observed for the B/D *cis*-series (10, 12).

Recently, the position of phenolic hydroxyl groups of alkaloids has been determined by deuteration of the *ortho* and/or *para* aromatic protons, followed by NMR spectroscopy (16–19). Attempts to apply this technique to porphyroxine were unsuccessful because of the instability of the alkaloid. Instead, a comparison of the chemical shifts of the aromatic protons in the phenolate ion and the undissociated phenol was used (20–22). In hexadeutero DMSO, the signals of the C-6 and C-9 protons appeared at 3.27 and 2.83 τ, respectively. Small amounts of sodium deuteroxide were added successively until no further changes took place in the chemical shifts. The results are listed in Table II. Porphyroxine

TABLE II—CHANGES IN CHEMICAL SHIFTS OF C-6 AND C-9 PROTONS OF PORPHYROXINE ON ADDITION OF NaOD IN $(CD_3)_2SO$

τ	1	2	3	4	$\Delta\tau$
C-6 proton	3.27	3.33	3.43	3.57	0.30
C-9 proton	2.83	2.99	3.03	3.25	0.42
	phenol ——→ phenolate				

could be recovered from the solution, proving that no structural changes occurred in the alkaloid as a result of the alkali treatment. The C-6 proton was shifted upfield 0.30 p.p.m. by conversion to the phenolate, and the C-9 proton was shifted 0.42 p.p.m. in the same direction. The following ranges have been reported (20) for anion shifts in alkyl and alkoxyl substituted phenols: *ortho*, 0.42–0.59 p.p.m.; *meta*, 0.19–0.39 p.p.m.; and *para*, 0.71–0.79 p.p.m. Therefore, it may be concluded that the C-6 proton in porphyroxine is *meta* to the phenolic hydroxyl group, and C-9 proton is *ortho*. Thus, the hydroxyl group in ring A was assigned to position-8.

Most of the naturally occurring papaverrubine alkaloids and their *N*-methyl analogs have a thermodynamically unstable configuration at position-14 and are readily epimerized by heating with methanol containing catalytic amounts of acid (23, 24). Porphyroxine has the same configuration at position-14 as glaudine, which can be epimerized to epiglaudine (13, 23).

The C-1 proton resonance of porphyroxine appears at a much higher τ-value than that of glaudine (Table I) due to the *N*-methyl group. *N*-Methylporphyroxine, prepared by reacting porphyroxine with formaldehyde and sodium borohydride (18, 25), gave essentially the same NMR spectrum as glaudine, except for the *Ar*-OMe group in position-8 (Table I).

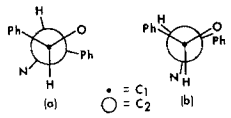


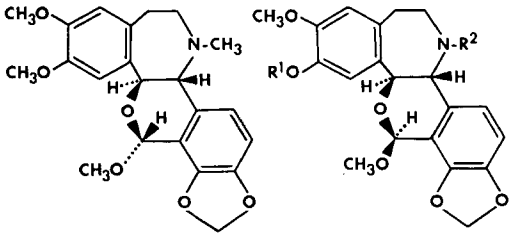
Fig. 1—Possible conformations and configurations of ring D.

Epiglaudine was prepared from glaudine[2] as described by Pfeifer *et al.* (23) and its NMR spectrum determined in $CDCl_3$ (Table I). There was no change in the coupling constants for the C-1 and C-2 proton signals, indicating that the epimerization did not involve a change in the conformation of ring D, the two possible conformations of which are shown in Fig. 1. Conformation (a) is close to the staggered form and is the more stable, while conformation (b) approaches the eclipsed form. The large coupling constants (9 c.p.s.) of the C-1 and C-2 protons (Table I) tend to indicate an approximately diaxial orientation corresponding to Fig. 1 (a) (26) and illustrated in structures VI and VII. The conversion of glau-

[1] Varian A-60A NMR spectrometer.

[2] The authors are indebted to Professor S. Pfeifer for a sample of glaudine.

dine to epiglaudine caused the 14-OMe signal to be shifted upfield 0.13 p.p.m. while a strong downfield shift of 0.38 p.p.m. was observed for the C-1 proton. This downfield shift may be explained on the basis of a 1,3-diaxial orientation of the C-1 proton with respect to the O—CH_3 group. Thus, it may be attributed to anisotropy effects of the C—OMe single bond, to effects due to C—OMe electric dipoles (27) or, perhaps equally well, to van der Waals interactions deshielding the C-1 proton (28). Therefore, one may conclude that the C-1 proton and the OMe group of epiglaudine are on the same side of the acetal oxygen bridge, with the OMe group in the axial position, as illustrated in VI. It follows that



VI VII

glaudine and porphyroxine may be represented by structure VII or its mirror image (glaudine: $R^1 = R^2 = CH_3$; porphyroxine: $R^1 = R^2 = H$).

There was no significant difference in the ORD and CD spectra[3] of glaudine, epiglaudine, and *N*-methylporphyroxine {glaudine ORD: $[\phi]_{299}23325$ (pk), $[\phi]_{282}0$ (tr.); CD: $[\theta]_{309}0$, $[\theta]_{293}15640$, $[\theta]_{269}0$, $[\theta]_{251}-5536$, $[\theta]_{244}0$}. These compounds have three asymmetric centers adjacent to benzene chromophores, and the weak effect of the configurational change at C-14 appears to be covered by strong active absorption due to the C-1 and C-2 centers. Only porphyroxine showed a slightly different spectrum in the short wavelength region {ORD: $[\phi]_{333}6010$, $[\phi]_{298}15025$ (pk), $[\phi]_{281}2394$ (tr.), $[\phi]_{242}19240$ (pk), $[\phi]_{237}18028$ (tr.), $[\phi]_{249}60092$; CD: $[\theta]_{313}0$, $[\theta]_{292}10314$, $[\theta]_{267}0$, $[\theta]_{255}0$, $[\theta]_{238}8727$}.

SUMMARY AND CONCLUSIONS

Porphyroxine has been isolated from the phenolic alkaloid fraction of opium. It belongs to the "papaverrubine" group of alkaloids which contain a 7-membered heterocyclic nitrogen ring (ring B) fused to a cyclic lactol ether (ring D). The nitrogen is a secondary amine, and the B/D ring junction of porphyroxine (II) has a *trans*-configuration. Based on NMR spectroscopy at different pH values, the phenolic hydroxyl group has been assigned to position-8. The steric configuration at position-14 of porphyroxine is the same as that of glaudine (III) and is thermodynamically unstable. Epimerization of glaudine results in a strong downfield shift of the C-1 proton signal, while the coupling constants for the C-1 and C-2 protons are unchanged. This indicates that the conversion of glaudine to epiglaudine has not changed the conformation of ring D, but only the stereochemistry at the asymmetric center at position-14. From model considerations and the large coupling constants for the C-1 and C-2 protons, it may be concluded that these protons have an approximately diaxial orientation as illustrated in Fig. 1 (a) and in structures VI and VII. The downfield shift of the C-1 proton signal on epimerization may be explained by anisotropy effects of the C—OMe single bond or by a deshielding caused by van der Waals forces, thereby indicating a 1,3-diaxial orientation of the C-1 proton and the O—Me group in epiglaudine (VI). Consequently, glaudine and porphyroxine must have the configuration at position-14 illustrated in structure VII.

The ORD and CD spectra were not useful for elucidating the configuration of ring D because the effect of the configuration change at position-14 was masked by the stronger absorption due to the asymmetric centers at C-1 and C-2.

[3] Jasco ORD/CD-5 instrument.

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