ALKALOIDS OF THE *Papaveraceae*. LII.*

THE CONSTITUTION OF ESCHOLININE AND THE IDENTITY OF ESHOLINE WITH MAGNOFLORINE

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For the quaternary alkaloid escholinine from *Eschscholtzia californica* CHAM. the constitution of (+)-romneine methohydroxide was deduced on the basis of spectral analyses and Hofmann degradation. It was found that the alkaloid esholine is identical with magnoflorine.

The quaternary alkaloid escholinine was isolated[1] from the root of *Eschscholtzia californica* CHAM. in the form of perchlorate; it is a minor component of the highly polar fraction of the alkaloids. From the UV spectrum and the optical rotation value the conclusion was drawn that it belongs to the group of benzyltetrahydroisoquinoline type alkaloids[1]. The results of a more detailed study of its structure is presented now.

The substance is of non-phenolic nature. In the IR spectrum of its perchlorate there are bands at 810 and 835 cm^{-1} (1,2,4-trisubstituted benzene ring), 865 and 875 cm^{-1} (1,2,4,5-tetrasubstituted benzene ring), 940 and 950 cm^{-1} (O_2CH_2), 1 090 cm^{-1} (ClO_4), 1 110 to 1 270 cm^{-1} (C—O—C bonds), and 1 490 to 1 590 cm^{-1} (aromatic vibrations). The PMR spectrum shows two singlets (3 H each) at 3·16 and 3·42 p.p.m. (2 N-methyl groups), two singlets at 3·75 and 3·85 p.p.m. (3 H each; 2 OCH_3), two singlets at 5·87 and 5·90 p.p.m. (O_2CH_2), and a complex multiplet at 6·50 to 6·80 p.p.m. (aromatic protons). During its mass spectrum measurement escholinine iodide was pyrolysed, while evaporating in the ionic source, in a manner analogous to other salts of quaternary alkaloids to a mixture of methine and a tertiary base. Methine appeared in the spectrum as a molecular peak of mass 355·1781, corresponding to the composition $C_{21}H_{25}NO_4$. The molecular peak of the tertiary base was not present in the spectrum although the majority of fragment ions was formed by its decomposition. Characteristic ions of the spectrum have mass 190 ("*a*", $C_{11}H_{12}NO_2$, base peak highly exceeding other peaks of the spectrum) and 151 ("*b*", $C_9H_{11}O_2$).

On the basis of this fragmentation pattern, recalling the behaviour of benzyltetrahydroisoquinoline alkaloids[2], and in agreement with other spectral results, we derived for escholinine iodide the structure *I* (X = I). The corresponding tertiary base has been isolated in the (−)-form under the name romneine from *Romneya coulteri var. trichocalyx* (EASTWOOD) JEPSON (*Papaveraceae*)[3]. The supposed structure of escho-

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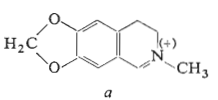
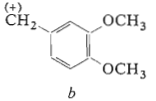
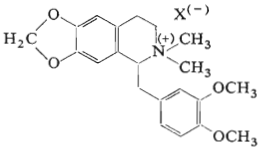
linine, corresponding to (+)-romneine methohydroxide, was then confirmed by direct comparison of the iodide and the perchlorate of escholinine and (±)-romneine methiodide (*I*, X = I) or methoperchlorate (*I*, X = ClO_4), which were prepared from a synthetic sample of (±)-romneine[3]. The UV and IR spectra, as well as the R_F values of the salts of escholinine and the methosalts of romneine were identical as were the mass spectra of escholinine iodide and romneine methiodide.



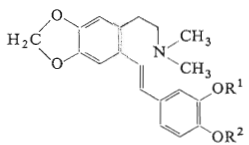
a



b



I



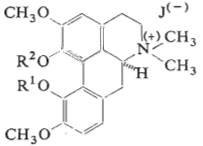
IIa, $R^1 = R^2 = CH_3$
IIb, $R^1 + R^2 = CH_2$

On Hofmann degradation of escholinine perchlorate a mixture of two optically inactive methines was obtained at the first step; they differed in their R_F values. The main reaction product gave a crystalline methine of a lower R_F value, probably the *trans* form of the structure *IIa*. Its UV spectrum is very similar to that of tetrahydroescholamine methine (*IIb*, see[1]) and it proves unambiguously the stilbene structure[4]. The second methine — probably the *cis* form — was oily. Both isomeric methines had identical mass spectra with the molecular peak at mass 355 (for mass spectrometric behaviour of escholinine methine see[5]).

The quaternary alkaloid esholine was isolated for the first time by Gertig[6] from the root of *E. californica* CHAM. in the form of perchlorate. This finding was later confirmed by us[1], but we also isolated esholine from the root of *E. douglasii* (ARN.) WALP. For the quaternary cation of escholine Gertig[6] gives the composition $C_{19}H_{22}.NO_3^+$ (from elementary analysis) and on the basis of its UV spectrum he concludes that it should possess the structure of the papaverine type. In view of the optical activity and other properties of esholine an aporphine skeleton seemed more probable[1]. The PMR spectrum of esholine iodide contains two characteristic singlets of N-methyl groups at 2·98 and 3·41 p.p.m., two singlets of methoxyl groups at 3·88 and 3·93 p.p.m., signals of three aromatic protons at 6·90 and 6·95 p.p.m. (2 H) and

further unresolved signals of aliphatic protons. The nuclear Overhauser effect observed between the signal of the low-field methoxyl and the aromatic proton signal at 6·90 p.p.m. (10·8%), and high-field methoxyl and the singlet at 6·95 p.p.m., (7·2%) indicates a mutual *ortho* orientation of methoxyls and aromatic protons. The two-proton singlet at 6·95 p.p.m. may be assigned to two aromatic *ortho* protons the chemical shifts of which are very close so that the usual AB type of the spectrum is changed to type A_2.

During the measurement of the mass spectrum of esholine iodide the decomposition of the quaternary salt to methine and tertiary base again took place. High resolution measurements indicated for the molecular peaks of both components the composition $C_{20}H_{23}NO_4$ (found 341·1623, calculated 341·1627) and $C_{19}H_{21}NO_4$ (found 327·1476, calculated 327·1470), respectively. In addition to peaks of iodine and methyl iodide the fragments of masses 312 (327 − 15), 310 (327 − 17), 283 (341 − 58) and 58 ($CH_2{=}N(CH_3)_2^+$) also deserve mention. The last two particles indicated the phenanthrene structure of the formed methine. Negligible fragmentation of ionised molecules and an appreciable amount of ions with two charges were in agreement with the aporphine structure.



IIIa, $R^1 = R^2 = H$
IIIb, $R^1 = CH_3$, $R^2 = H$
IIIc, $R^1 = H$, $R^2 = CH_3$
IIId, $R^1 = R^2 = CH_3$

From the presented findings structure *IIIa* followed which corresponds to magnoflorine iodide. Direct comparison of esholine iodide with authentic magnoflorine iodide (mass, IR and UV spectra, mixed melting point, optical rotation and R_F values) confirmed the identity of both substances. The constants of esholine perchlorate also agreed with the values given in literature for magnoflorine perchlorate[7,8]. The products of O-methylation of esholine were also found identical with authentic samples of the expected derivatives. From literature[9] it is known that O-methylation of magnoflorine takes place with considerable difficulty. On partial methylation of esholine with methyl iodide in alkaline medium we obtained O-monomethyl derivative identical with (+)-corydine methiodide (*IIIb*); the formation of the isomeric (+)-isocorydine methiodide (*IIIc*) was not observed. Under the reaction condi-

tions used earlier[9] we obtained O,O'-dimethylesholine iodide identical with (+)-O,O'--dimethylcorytuberine methiodide (*IIId*), which we prepared by methylation of (+)--isocorydine. The identity was proved on the basis of mass, IR and UV spectra, mixed melting point, optical rotation and R_F values.

The alkaloid magnoflorine which is widely distributed in many families belonging to the order of *Polycarpicae* was found in *Papaveraceae* only sporadically (*Papaver somniferum* L.[10], *Glaucium flavum* CR.[11,12], *G. fimbrilligerum* BOISS.[13] and *Meconopsis rudis* PRAIN[14]). According to our latest results (see Experimental) magnoflorine is the main alkaloid of the quaternary fraction of the root of *E. douglasii*, while in the root of *E. californica* it is a minor component. From the phytochemical point of view it is interesting that in the closely related species *E. glauca* GREENE it was not found even in traces[1].

EXPERIMENTAL

The melting points (uncorrected) were determined in capillaries and on a Kofler block. The PMR spectra were measured in hexadeuteriodimethyl sulfoxide or its mixture with deuteriochloroform on Varian HA-100 at 100 MHz. The mass spectra were recorded with a AEI-MS 902 spectrometer at energies of the ionising electrons of 70 eV. IR spectra were taken on Infrascan, Hilger and Watts, and the UV spectra on Unicam SP 500 apparatus. For thin-layer chromatography silica gel with gypsum (5 : 1) was taken and the chromatograms were run in ethanol–water–25% ammonia 15 : 9 : 1 (S_1), 1-propanol–water–formic acid 12 : 7 : 1 (S_2), and cyclohexane–chloroform–diethylamine 7 : 2 : 1 (S_3). The detection of spots was carried out with potassium iodoplatinate. Paper chromatography was carried out on paper Whatman No 1 (descending technique) in 1-butanol–acetic acid–water 10 : 1 : 3 (S_4) and the spots were detected by Dragendorff reagent.

Isolation of Alkaloids

For the isolation of quaternary alkaloids from the root of *E. californica* and *E. douglasii* the crude fraction *J* (see[1]) was used, obtained from the plant material described in[15]. Individual alkaloids were separated in the form of crystalline perchlorates or iodides (the yields are calculated as perchlorates) in a manner similar to that described in[1]. From the root of *E. californica* (1934 g) escholidine was isolated (1·53 g; 0·079%), further escholinine (0·36 g; 0·019%), (−)-α-canadine methohydroxide (0·25 g; 0·013%), esholine (0·18 g; 0·009%), californidine (0·09 g; 0·005%), perchlorate of the tertiary base ED (see[16]) (0·02 g; 0·001%) and small amounts of crystalline perchlorates of several further non-identified alkaloids. From the root of *E. douglasii* (3265 g) esholine (3·32 g; 0·10%) was obtained, further escholidine (2·81 g; 0·086%), (−)-α-canadine methohydroxide (0·38 g; 0·012%), californidine (0·10 g; 0·003%), perchlorate ED (0·17 g; 0·005%) and other unidentified alkaloids.

Escholinine: iodide crystallised from methanol melted at 197—198°C (in capillary and on Kofler block). The mass and the IR spectrum (in KBr) were identical with those of authentic (±)-romneine methiodide. Perchlorate from methanol gave prisms of m.p. 205—207°C (Kofler block) or 209—210°C (capillary), $[\alpha]_D^{25}$ +74° ± 3° (*c* 0·31, methanol). IR spectrum (in KBr) was identical with the spectrum of (±)-romneine methoperchlorate, as also was the UV spectrum (in methanol), λ_{max} (log ε) 234 nm (4·04), 286 nm (3·83), λ_{min} 225 nm (4·00), and 258 nm (3·23). The

R_F values (0·29 in S_1 and 0·66 in S_2) of escholinine salts were identical with the values of (±)-romneine methosalts. The same is true of colour reactions with concentrated sulfuric acid of both preparations (slow appearance of red-violet colour).

Methosalts of (±)-romneine: from (±)-romneine hydrobromide (m.p. 193–194°C; see[3]) the base was prepared, dissolved in 2 ml of methanol and 0·3 ml of methyl iodide added. After 20 h standing solvents were evaporated to give crude methiodide (quantitative yield), which was crystallised twice from methanol–ether, m.p. 174–177°C (Kofler block). Methoperchlorate was prepared from the methiodide by dissolution in water, precipitation with 20% sodium perchlorate solution and crystallisation from methanol; m.p. 175–177°C (Kofler block) or 185–187°C (capillary); mixed melting point with escholinine perchlorate was 200 — 205°C (capillary).

Escholinine methine: escholinine perchlorate (62·0 mg) was boiled with 10 ml of 20% potassium hydroxide in methanol for 6 h under reflux. The reaction mixture was diluted with water, acidified with dilute sulfuric acid, methanol was evaporated, and the solution alkalised with ammonia and extracted with ether. After evaporation of ether 51·8 mg of an amorphous product were obtained, which on a thin layer (in S_3) gave a main spot of R_F 0·45 and a weaker spot of R_F 0·54. On crystallisation from ether–hexane a product was obtained (37·0 mg) of m.p. 64–66°C (capillary), or 66–68°C (Kofler block), $[\alpha]_D^{23}$ 0° ± 3° (*c* 0·24, methanol). Molecular weight 355 (mass spectrometry). UV spectrum (methanol): λ_{max}(log ε) 220 nm (4·31), 298 nm shoulder (4·18), 330 nm (4·24), λ_{min} 261 nm (3·84). R_F value in S_3: 0·45. On evaporation of the mother liquor after the crystalline product an oily material was obtained which contained predominantly the second methine of R_F 0·54. Its mass spectrum (M^+ 355) was identical with that of the crystalline methine.

Esholine iodide: from methanol needles of m.p. 270–272°C (Kofler block) or 249–250°C (capillary), mixed melting point with authentic magnoflorine iodide was undepressed, $[\alpha]_D^{22}$ +193° ± 2° (*c* 0·20, methanol); literature gives various values for $[\alpha]_D$, in the range from +198° (*cf.*[17]) to +220° (*cf.*[9]). UV spectrum (methanol), λ_{max}(log ε), 225 nm (4·81), 270 nm (4·13), 311 nm (4·00), λ_{min} 258 nm (4·06), 292 nm (3·85) as well as the IR spectrum (nujol) were identical with the spectra of magnoflorine iodide. The R_F values (0·65 in S_1 and 0·45 in S_4) were also identical with the values of authentic magnoflorine iodide. Under UV light violet-blue fluorescence, after detection with potassium iodoplatinate a brown spot.

Esholine perchlorate: from methanol prisms of m.p. 278–279°C (Kofler block) or 261–262°C (capillary), $[\alpha]_D^{23}$ +214° ± 2° (*c* 0·20, methanol); literature gives for esholine perchlorate[6] m.p. 256–257°C, for magnoflorine perchlorate m.p. 257–258°C[7] or 261°C[8] and $[\alpha]_D^{22}$ +213° (methanol)[8] or +216° (methanol)[7]. UF and IR spectra[1] are in good agreement with the data for esholine perchlorate[6].

O-Methylation of Esholine

1) Esholine iodide (201·5 mg) was refluxed in 20 ml methanol with 0·4 g potassium hydroxide and 5 ml methyl iodide for 9 hours on a water bath. The solution was diluted with water, acidified with dilute sulfuric acid, methanol and methyl iodide were evaporated, potassium iodide added and the mixture extracted several times with chloroform. The product obtained after evaporation of chloroform gave on crystallisation from methanol 43·4 mg of unreacted esholine iodide and from the mother liquors 116·6 mg of a product from which corydine methiodide (75 mg) was obtained after several crystallisations, m.p. 190–210°C (capillary), or 210–215°C (Kofler block), undepressed on admixture of a sample prepared from (+)-corydine, $[\alpha]_D^{24}$ −148° ±

± 4° (*c* 0·31, methanol). UV spectrum and R_F values (0·31 in S_1 and 0·66 in S_4) were identical with the values of the reference sample.

2. Esholine iodide (201·4 mg) was refluxed in 20 ml methanol with 0·4 g potassium hydroxide and 10 ml methyl iodide on a water bath. After 7 h boiling another 0·4 g of potassium hydroxide and 4 ml methyl iodide were added and the mixture boiled, and the procedure was repeated three times more. The reaction mixture was diluted with water, potassium hydroxide added, and extracted with chloroform to separate the non-phenolic product. From the concentrated chloroform solution 184·4 mg of substance (86·4%) crystallised out which when recrystallised from methanol had m.p. 264—265°C (Kofler block) or 251—252°C (capillary), undepressed on admixture with O,O'-dimethylcorytuberine methiodide, $[\alpha]_D^{24}$ +155° ± 3° (*c* 0·30, methanol). The identity was proved by mass, UV and IR spectrum and R_F values (0·35 in S_1 and 0·73 in S_4).

(+)-*Corydine methiodide*: 100 mg of (+)-corydine were dissolved in 2 ml of methanol and 5 ml of ether and 0·5 ml of methyl iodide were added to it. The next day the separated needles (162·2 mg, solvate) were collected under suction and crystallised from methanol; m.p. 190—210°C (capillary). or 210—214°C (Kofler block), $[\alpha]_D^{22}$ +153° ± 2° (*c* 0·20, methanol). UV spectrum (methanol): λ_{max}(log ε) 225 nm (4·84), 270 nm (4·25), 302 nm (3·98); λ_{min} 250 nm (3·87), 288 nm (3·83). R_F values: 0·31 (S_1) and 0·66 (S_4).

(+)-*Isocorydine methiodide*: 100 mg of (+)-isocorydine were dissolved in 2 ml of boiling methanol, 5 ml of ether and 0·5 ml of methyl iodide were added, and the mixture allowed to stand overnight. The separated product (140 mg) was filtered off under suction and crystallised from methanol; needles of m.p. 226—228°C (Kofler block), or 231—232°C (capillary), $[\alpha]_D^{23}$ +139° ± ± 2° (*c* 0·21, methanol). UV spectrum (methanol): λ_{max} (log ε) 220 nm (4·78), 270 nm (4·20), 304 nm (3·83), λ_{min} 250 nm (3·79), 293 nm (3·77). R_F values 0·29 (S_1) and 0·61 (S_4).

(+)-O,O'-*Dimethylcorytuberine methiodide*: a mixture of 205 mg of (+)-isocorydine, 5 ml of methanol, 0·1 g of potassium hydroxide and 1 ml of methyl iodide was refluxed for 2·5 h on a water bath. The reaction mixture was diluted with water, potassium hydroxide was added until the solution was alkaline and the non-phenolic material was separated by extraction with several portions of chloroform. After evaporation of chloroform and crystallisation of the residue from methanol the yield was 131·7 mg of product (needles) melting at 263—265°C (Kofler block) or 251—252°C (capillary), $[\alpha]_D^{24}$ +156° ± 3° (*c* 0·30, methanol). UV spectrum (methanol): λ_{max}(log ε) 222 nm (4·80), 271 nm (4·19), 307 nm shoulder (3·71), λ_{min} 250 nm (3·78). R_F values 0·35 (S_1) and 0·73 (S_4).

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