ALKALOIDS FROM *Papaver litwinowii* FEDDE ex BORNM.*

Jiří SLAVÍK and Leonora SLAVÍKOVÁ

Department of Medical Chemistry and Biochemistry, J. E. Purkyně University, 662 43 Brno

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From *Papaver litwinowii* FEDDE ex BORNM. aporheine ((+)-roemerine) was isolated as the dominant alkaloid in addition to minor alkaloids aporheine methohydroxide, corytuberine, hordenine, (—)-mecambrine, cryptopine, protopine and coptisine. The presence of small amounts of allocryptopine, corydine, isocorydine, scoulerine, corysamine and papaverrubine A, C, D and traces of E(?) was also proved. Corytuberine was now isolated from *P. dubium* L. as well.

Papaver litwinowii FEDDE ex BORNM. is an annual plant from the section *Orthorhoeades* FEDDE (*Rhoeadium* SPACH) occurring in Central Asia, Iran and Afghanistan, and it is related to the species *P. dubium* L. (*cf.*[1,2]). Little has been known about the alkaloids of this plant so far. Preininger and coworkers[3] isolated from it alkaloids of m.p. 239–241°C, 272–273°C and a Dragendorff negative substance of m.p. 348–352°C, which were not identified.

From the plants cultivated from the seeds from the Botanical Garden in Tashkent, USSR, we now isolated the sum of alkaloids in a 0·15% yield, from which we separated aporheine ((+)-roemerine, *Ia*) as the main alkaloid. It represented 76% of the sum of the "non-quaternary" fraction of the base. From non-phenolic bases of the same fraction we further isolated (−)-mecambrine (*II*), cryptopine and a small amount of protopine. From the phenolic fraction we separated a base of m.p. 118°C as the main component, which we identified as hordenine (*III*) on the basis of its m.p., UV, IR and mass spectra. This nonspecific base was thus detected in *Papaveraceae* for the first time. Using TLC the presence of negligible amounts of allocryptopine, corydine, isocorydine, scoulerine and papaverrubines C and D, in addition to trace amounts of papaverrubines A and, maybe, of E as well, was proved. Neither the presence of rhoeadine nor that of oxysanguinarine could be proved. Coptisine, accompanied by a small amount of corysamine, represented the fraction of the quaternary protoberberines.

From the fraction of the alkaloids extractable in the form of iodides with chloroform[4] aporheine methiodide was isolated as the main component. The isolation

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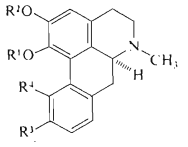
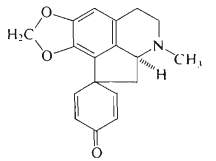
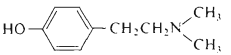
of this alkaloid from *P. litwinowii* has already been published earlier[5]. From the mother liquors we isolated corytuberine (*Ib*) in the form of its hydriodide (0·0055%). This alkaloid was isolated recently from *Papaver dubium* L. in 0·0024% yield.



Ia, $R^1 + R^2 = CH_2$, $R^3 = R^4 = H$
Ib, $R^1 = H$, $R^2 = CH_3$, $R^3 = OCH_3$, $R^4 = OH$



II



III

From these findings it follows that the species *P. litwinowii* is closely related to *P. dubium* (*cf.*[6,7]) not only botanically but biochemically as well, in that they have a common main alkaloid, aporheine, and some minor components, as for example (−)-mecambrine, corytuberine, protopine and aporheine methohydroxide; they differ, however, in the presence of cryptopine and hordenine in *P. litwinowii*, which were not detected in *P. dubium*. According to its m.p., UV and IR spectra the alkaloid of m.p. 239−241°C, isolated by Preininger and coworkers[3] is evidently identical with aporheine hydrochloride.

EXPERIMENTAL

The melting points were determined on a Mettler FP 51 apparatus and they were not corrected. The UV spectra were measured on a spectrophotometer Unicam SP 1800. For thin-layer chromatography (TLC) both Silica gel G Merck and Silufol plates UV 254 (Kavalier) were used. For the first material the following solvent systems were used: cyclohexane–diethylamine 9 : 1 (S_1), cyclohexane–chloroform–diethylamine 7 : 2 : 1 (S_2), 5 : 4 : 1 (S_3) and 1 : 8 : 1 (S_4), methanol–25% ammonia 200 : 1 (S_5), benzene–methanol 4 : 1 (S_6), benzene–acetone–methanol 7 : 2 : 1 (S_7), benzene–diethylamine 19 : 1 (S_8), chloroform–diethylamine–ethanol 8 : 1 : 1 (S_9) and ethanol–water–25% ammonia 15 : 9 : 1 (S_{10}). For the second material the systems methanol–diethylamine 4 : 1 (S_{11}) and 1 : 1 (S_{12}) were employed. The spots of fluorescing alkaloids were detected in UV light, the spots of papaverrubines by exposure to concentrated hydrochloric acid fumes for 25 min (formation of purple spots), while the spots of other alkaloids were detected with potassium iodoplatinate.

Extraction and Isolation of Alkaloids

The plants were cultivated in the Experimental Garden of the Medical Faculty in Brno from the seeds obtained from the botanical garden in Tashkent, USSR, and harvested including roots at the stage of flowering and unripe fruits on the 6th July, 1978. A herbarium specimen is deposited at the Department of Medical Chemistry and Biochemistry in Brno. The plants were dried at room temperature and the dry, ground material (3·57 kg) was extracted with methanol in a Soxhlet extractor. Methanol was distilled off, the residue dissolved in cold 1% acetic acid and filtered. Using a procedure aimed to isolate oxysanguinarine we isolated from the basic fraction a substance with m.p. 290—292°C (methanol) which was not identical, however, with oxysanguinarine.

The acid filtrate was extracted with ether first after alkalization with sodium carbonate (fraction *A*) and then after alkalization with sodium hydroxide above pH 13 (fraction *B*). The aqueous layer was neutralized with 20% sulfuric acid to pH about 7, a saturated solution of potassium iodide was added and the mixture extracted several times with chloroform or a mixture of chloroform with 20% of ethanol (fraction *I*). After alkalization with ammonia the aqueous layer was reextracted with chloroform (fraction *E*).

The crude bases of fraction *A* (4·10 g) were dissolved in 5% acetic acid and a saturated solution of potassium chloride was added. The aporheine hydrochloride which crystallized out (total yield of the aporheine base was 2·41 g) was filtered off under suction and the filtrate was partitioned by extraction with chloroform to a fraction of hydrochlorides extractable with chloroform (*AC*) and unextractable with it (*AD*). These were then separated in the usual manner[8,9] to non-phenolic (AC_1 and AD_1) and phenolic (AC_2 and AD_2) fractions. From the fraction AC_1 (0·40 g) a further smaller amount of aporheine hydrochloride was isolated in the above manner and the rest of the bases from the mother liquors was obtained after alkalization with ammonia by extraction with ether. The ethereal solution was gradually concentrated to crystallization, affording thus 77 mg of (—)-mecambrine (total yield 83 mg) and 15 mg of cryptopine. In the remaining amorphous bases (115 mg after purification) the presence of a negligible amount of papaverrubine A, an unidentified papaverrubine (not identical with papaverrubine B) and traces of papaverrubine E (?) as well as a further five unidentified alkaloids were detected by TLC in addition to the residues of aporheine and mecambrine. From the crude fraction AD_1 (0·14 g) 31 mg of cryptopine (total yield 46 mg), 6·5 mg of mecambrine and 1·0 mg of protopine were separated on crystallization from ether. After purification 29 mg of remaining amorphous bases were obtained which according to TLC contained, in addition to the residues of the three mentioned alkaloids, small amounts of allocryptopine and three additional unidentified alkaloids. In fraction AC_2 (24 mg), which remained amorphous, TLC showed that corydine was the main component, accompanied with smaller amounts of scoulerine, hordenine, papaverrubine D and C, trace amounts of isocorydine and five additional unidentified alkaloids. From the ethereal solution of fraction AD_2 (0·31 g) 171 mg of hordenine crystallized out. In the amorphous residue (43 mg after purification) scoulerine and three unidentified alkaloids could be detected in addition to the residues of hordenine and traces of papaverrubine D.

From the purified fraction *B* 12·2 mg (0·0003%) of a golden-yellow base were obtained in the usual manner[8,9], which was identical with coptisine. The presence of negligible amounts of corysamine was also proved. When crystallized fractionally from methanol–ether, fraction *I* afforded totally 0·35 g of aporheine methiodide and from the mother liquors 195 mg of corytuberine hydriodide were obtained using the same solvent mixture. An amorphous residue (0·27 g) was thus obtained in which no alkaloid could be detected by TLC, except the two mentioned ones. In this residue non-alkaloidal substances predominated. The amorphous fraction *E* (0·20 g) also contained mainly non-alkaloidal substances in addition to negligible amounts of corytuberine and traces of two unidentified alkaloids.

Characterization of the Alkaloids Isolated

The alkaloids isolated from *P. litwinowii* were identified by melting points and mixed melting points, optical rotation values, IR and UV spectra and mass spectra, as well as R_F values, which were compared with those of authentic samples. The yields of individual alkaloids in % per dry plant material are given in brackets.

Aporheine (0·068%): From ether–hexane needles in clusters, m.p. 101—102°C, $[\alpha]_D$ +80° ±2° (*c* 0·50, methanol). UV spectrum: λ_{max} (log ε) 210 nm (4·56), 234 nm (4·29), 272 nm (4·35), 317 nm (3·65), λ_{min} 229 nm (4·29), 254 nm (4·10), 303 nm (3·58). Hydrochloride m.p. 247—248°C (water).

Aporheine methiodide (0·010%): From methanol–ether (1 : 1) needles of m.p. 234—235°C, UV spectrum and other properties see ref.[5].

Corytuberine hydriodide (0·0055%): From methanol leaflets, m.p. 216—217°C, UV spectrum: λ_{max} (log ε) 224 nm (4·71), 273 nm (4·12), 312 nm (3·91), λ_{min} 260 nm (4·04), 293 nm (3·77).

Hordenine (0·0048%): From ether prisms, m.p. 117·5—118·5°C. UV spectrum: λ_{max} (log ε) 227 nm (3·92), 280 nm (3·25), λ_{min} 251 nm (2·78), and IR spectrum agreed with the literature data[10]. In the mass spectrum a molecular peak was found at mass 165·1153 (for $C_{10}H_{15}NO$ calculated 165·1154) and further fragments at 121 (M—$(CH_3)_2N$), 120 (M—$(CH_3)_2NH$), 107 (M—$CH_2N(CH_3)_2$), 91, 77 and 58 ($CH_2{=}N(CH_3)_2$) mass units. After labelling with [O—^{2}H]-ethanol in ion source all peaks, except the fragment of *m/z* 58, were shifted by one mass unit to higher values.

(—)-*Mecambrine* (0·0023%): From ether prisms, m.p. 179—180°C, $[\alpha]_D^{22}$ —96° ± 3° (*c* 0·20, methanol), UV spectrum: λ_{max} (log ε) 210 nm (4·52), 220 nm (4·52), 232 nm (4·51), 294 nm (3·57), λ_{min} 215 nm (4·51), 227 nm (4·50), 270 nm (3·45).

Cryptopine (0·0013%): From chloroform–methanol prisms, m.p. 223—224°C, undepressed on admixture of an authentic specimen.

Protopine (0·00003%): From methanol prisms, m.p. 208—209°C, undepressed with an authentic sample.

Isolation of Corytuberine from *P. dubium* L.

After the separation of aporheine methiodide (0·0034%, see ref.[5]) from the fraction *I* obtained from a small sample (384 g) of *P. dubium* the remaining mother liquor was purified and submitted to crystallization from methanol–ether, affording 9·3 mg (0·002%) of corytuberine hydriodide, m.p. 212—212°C, undepressed on admixture of an authentic preparation. The R_F values of both samples were also identical.

R_F Values

In systems S_1, S_2, S_3 and S_5, respectively: Allocryptopine 0·30, 0·65, —, 0·34; aporheine 0·70, 0·86, —, 0·69; corydine 0·19, 0·53, 0·76, 0·66; cryptopine 0·23, 0·65, —, 0·48; hordenine 0·20, 0·32, 0·47, 0·44; isocorydine 0·26, 0·63, 0·89, 0·64; mecambrine 0·19, 0·59, —, 0·78; protopine 0·47, 0·78, —, 0·52; scoulerine 0·04, 0·21, 0·44, 0·76. In systems S_1, S_6, S_7 and S_8, resp.: Papaverrubine A 0·32, 0·86, 0·91, 0·83; papaverrubine C 0·10, 0·54, 0·75, 0·53; papaverrubine D 0·05, 0·42, 0·68, 0·43; papaverrubine E 0·28, 0·39, 0·50, 0·69. In systems S_4, S_9 and S_{10}, resp.: Aporheine methiodide 0·03, 0·04, 0·24; corytuberine 0·16, 0·60, 0·87. In systems S_{11} and S_{12}: coptisine 0·50, 0·80; corysamine 0·18, 0·50.

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