ALKALOIDS OF SOME HIMALAYAN SPECIES OF *Meconopsis* GENUS*

J.Slavík and L.Slavíková

Institute of Medical Chemistry, Purkyně University, 662 43 Brno

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From *Meconopsis rudis* Prain amurensinine was isolated as the main alkaloid. Smaller amounts of 6-methoxy-2-methyl-1,2,3,4-tetrahydro-β-carboline, protopine and a new alkaloid, amurensinine methohydroxide, were also isolated, and the presence of small amounts of allocryptopine, amurensine, rhoeadine, isorhoeadine, papaverrubines A, E, D, and C, coptisine, magnoflorine and several additional unidentified alkaloids proved. The majority of these alkaloids was also detected in the species *M. horridula* Hook. f. et Thoms., *M. sinuata* Prain, *M. robusta* Hook. f. et Thoms., *M. paniculata* (D. Don) Prain, and *M. betonicifolia* Franch. Cryptopine was found in these species as another alkaloid.

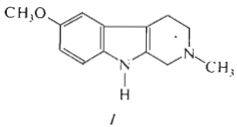
The genus *Meconopsis* from the *Papavereae* tribe comprizes about 40 species growing mainly in high mountain regions of the Himalayas and China, with the exception of a single West European species, *M. cambrica* (L.) Vig. Very little is known on the alkaloids of this genus. Only the alkaloids of the *M. cambrica*[1–5], *M. rudis* Prain[6], and newly *M. napaulensis* DC. (ref.[7]) species have been investigated in greater detail. The alkaloids of several additional species have also been studied by paper chromatography[1,8].

In this paper the results are given of the study of alkaloids of six Himalayan monocarpic *Meconopsis* species, among which *M. rudis* Prain, *M. horridula* Hook. f. et Thoms., and *M. sinuata* Prain are classified in the section *Aculeatae* Prain, and *M. robusta* Hook. f. et Thoms. and *M. paniculata* (D. Don) Prain in the section *Robustae* Prain. The species *M. betonicifolia* Franch. is sometimes classified in the independent *Cathcartia* genus under the name *C. betonicifolia* (Franch.) Prain (see[9]). These species usually display a very low alkaloid content (see also[1,7]). In view of the fact that these high-mountain plants can be cultivated under our climatic conditions only with difficulty, we were able to breed over a period of several years a relatively larger number of plants only of the species *M. rudis* and *M. betonicifolia*.

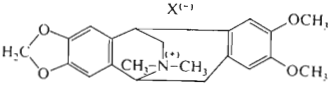
From the species *M. rudis* protopine has been already isolated earlier, and traces of sanguinarine and a further unidentified base were also detected in this species[1].

* Part LXIV in the series Alkaloids of the *Papaveraceae*; Part LXIII: This Journal *41*, 3343 (1976).

From the roots of this plant Gertig[6] isolated a small amount of protopine and magnoflorine and proved the presence of allocryptopine, sanguinarine, two additional non-quaternary and two quaternary alkaloids. From the plant material cultivated in our country we have now isolated alkaloids in 0·10% yield, calculated per whole dry plants. From the non-quaternary fraction of non-phenolic bases we isolated by direct crystallization an alkaloid of the composition $C_{13}H_{16}N_2O$ (see[7]), which was identified on the basis of its mass, IR and UV spectrum and other properties as 6-methoxy-2-methyl-1,2,3,4-tetrahydro-β-carboline[10,11] (*I*). The identity was confirmed by direct comparison with a synthetic specimen[11]. So far this alkaloid has been isolated from the Brazilian plants *Virola theiodora* WARB. (*Myristicaceae*) and *Adenanthera peregrina* (L.) SPEG. (*Viciaceae*)[10] and from the species *Phalaris arundinacea* L. (*Poaceae*)[11,12].



I



II

From the mother liquors after the mentioned alkaloid we isolated protopine, a small amount of a mixed fraction of protopine and allocryptopine, and we also detected two further alkaloids, preliminarily called MR 1 and MR 2, which probably also contain the β-carboline nucleus. However, neither of them could be obtained in a pure state. The alkaloid MR 1 was also found in *M. napaulensis*[7] and in all other Asian species of *Meconopsis* investigated. The largest alkaloidal fraction was composed of bases the hydrochlorides of which are extractable with chloroform[13,14]. Amurensinine, representing the main alkaloid of the *M. rudis* species, was isolated by chromatography of the bases of this fraction on alumina column. A mixed fraction of rhoeadine and isorhoeadine, with an admixture of papaverrubines A and E, was obtained in a very low yield. In the fraction of phenolic bases amurensine, papaverrubine D (porphyroxine) and papaverrubine C (epiporphyroxine)* were detected in addition to several unidentified alkaloids. The fraction of quaternary protoberberines contained a negligible amount of coptisine only. From the fraction of quaternary alkaloids which were obtained by extraction of their iodides with chloroform[16] and after their conversion to perchlorates amurensinine

* According to Hughes and coworkers[15] papaverrubine C might be an artifact formed during the isolation procedure from porphyroxine.

methoperchlorate (*II*, $X^{(-)} = ClO_4$) was isolated. It was identified by direct comparison with a sample prepared from amurensinine. This is the first finding of this alkaloid in nature. Further, the presence of a small amount of magnoflorine and of two additional quaternary alkaloids, provisionally indicated as MR 3 and MR 4, was also proved in the given fraction.

The alkaloids from the other five species of the *Meconopsis* genus could be studied by thin-layer and paper chromatography only, owing to the small amount of plant material available, or in the case of *M. betonicifolia* due to their unusually low alkaloid content. In *M. horridula* (0·13% of alkaloids), closely related to *M. rudis*, amurensinine was again the main alkaloid, while smaller amounts of protopine, cryptopine, 6-methoxy-2-methyl-1,2,3,4-tetrahydro-β-carboline, alkaloid MR 1, papaverrubine D and C, and traces of rhoeadine, isorhoeadine, papaverrubine E and coptisine were also detected. The main part of the quaternary fraction was composed of amurensinine methohydroxide, but it also contained a small amount of the MR 4 alkaloid. In the *M. sinuata* species (0·034% of alkaloids) protopine prevailed strongly, accompanied by a negligible amount of cryptopine, amurensinine, alkaloid MR 1, papaverrubine D, coptisine and the quaternary alkaloid MR 4. The species *M. robusta* (0·047% of alkaloids) contains protopine as the main alkaloid, and small amount of cryptopine, alkaloid MR 1, papaverrubine D, coptisine and magnoflorine and traces of 6-methoxy-2-methyl-1,2,3,4-tetrahydro-β-carboline, rhoeadine, papaverrubine E and C, corysamine and alkaloid MR 4. Also in *M. paniculata* (0·033% of alkaloids) protopine predominates, accompanied by smaller amounts of cryptopine, alkaloid MR 1, papaverrubine D, coptisine and alkaloid MR 4, and by traces of 6-methoxy-2-methyl-1,2,3,4-tetrahydro-β-carboline, rhoeadine, papaverrubine A, E and C, corysamine and magnoflorine. *M. betonicifolia* (less than 0·005% of alkaloids) contains protopine as the main alkaloid and negligible amounts of cryptopine, alkaloid MR 1, rhoeadine, papaverrubine D and C, coptisine, berberine, corysamine, as well as traces of allocryptopine, isorhoeadine, papaverrubine A and E and one quaternary alkaloid.

Some of the unidentified alkaloids of the *M. napaulensis* DC. species, mentioned in paper[7], were later identified as amurensinine (present in the overground part only), and the two alkaloids from the quaternary fraction from the root were identified as amurensinine methohydroxide and alkaloid MR 3. One of the two unidentified phenolic papaverrubines is evidently identical with papaverrubine C. We also demonstrated the presence of trace amounts of isorhoeadine and papaverrubine A (in the overground part and in the root) and alkaloid MR 4 (in the root only). It is remarkable that in contrast to earlier findings[1,6,8] not a trace of sanguinarine could be detected in any of the mentioned *Meconopsis* species.

On the basis of these results the conclusion can be drawn that the common biochemical character of all investigated Asian species of *Meconopsis* genus [1,7] is protopine which in many cases represents simultaneously the main alkaloid of the

plant. It is often accompanied by cryptopine or allocryptopine. The differences between the species mentioned consist mainly in a varying relative proportion of the individual alkaloids in the plant. The general occurrence of papaverrubines, or also small amounts of rhoeadine and isorhoeadine, indicates a close biochemical relatedness with the *Papaver* genus. The presence of amurensinine as the main alkaloid in the species *M. rudis* and *M. horridula* mainly indicates the relationship with the species of *Papaver* genus from the *Scapiflora* section. The occurrence of 6-methoxy-2-methyl-1,2,3,4-tetrahydro-β-carboline in several species of *Meconopsis* is also remarkable, even though little chemotaxonomic value can be attributed to this finding, owing to the insufficiently specific occurence of β-carboline alkaloids in the plants. However, up to now it is the first indole alkaloid found in the *Papaveraceae* family. It is worth mentioning that we have recently proved the presence of this alkaloid, together with the alkaloid MR 1, even in the roots of *Papaver rhoeas* L. Magnoflorine is not too widespread; it is mostly present in low concentrations only or is completely absent. It seems that the sole Western European polycarpic species *M. cambrica* (L.) VIG. (*Cambricae* section) has a rather isolated position, both by its geographic occurrence and the character of its alkaloids[1,2]. The main tertiary alkaloids, mecambrine and mecambridine, have not been detected in other *Meconopsis* species so far, the same as the minor alkaloids[1,7] roemerine, corytuberine and alkaloid $C_{19}H_{21}NO_4$. In addition to this we have also proved small amounts of papaverrubine D and C, protopine, coptisine and berberine, and from the root we isolated magnoflorine as the main alkaloid, similarly as the authors of paper[5].

EXPERIMENTAL

The melting points were determined both in capillaries and on a Kofler block and they were not corrected. The mass spectrum was measured on a AEI-MS 902 spectrometer, the IR spectra (in KBr) on a Unicam SP 1000 Infrared Spectrophotometer, and the UV spectra (in methanol) on a Unicam SP 1800 instrument. For thin layer chromatography both silica gel G (Merck) with gypsum (5 : 1), and Silufol UV 254 plates (Kavalier) were used. In the first case the following solvent systems were employed: cyclohexane–diethylamine 9 : 1 (S_1), cyclohexane–chloroform–diethylamine 7 : 2 : 1 (S_2), benzene–methanol 4 : 1 (S_3), benzene–acetone–methanol 7 : 2 : 1 (S_4), benzene–diethylamine 19 : 1 (S_5), ethanol–water–25% ammonia 15 : 9 : 1 (S_6), methanol–water–25% ammonia 15 : 3 : 1 (S_7) and 1-propanol–water–85% formic acid 12 : 7 : 1 (S_8). In the second case methanol–diethylamine 4 : 1 (S_9) was used for development. Paper chromatographies were carried out on Whatman paper No 1 (descending manner) with 1-butanol–acetic acid–water 10 : 1 : 3 (S_{10}) and ethanol–water 3 : 2 (S_{11}). Papaverrubines were detected as purple spots with concentrated hydrochloric acid fumes by 20 minutes' exposure. The detection of quaternary protoberberines was carried out in UV light, while the spots of other alkaloids were detected with potassium iodoplatinate and Dragendorff's reagent. The identity of known alkaloids was confirmed by comparison of their R_F values with those of parallely chromatographed authentic samples (in three to five solvent systems).

Extraction and Isolation of the Alkaloids

The plants were cultivated in the Experimental Botanical Garden, Medical Faculty, Brno, from the seeds obtained from various European botanical gardens. Only flowering specimens were collected (including roots), at the stage of flowers and unripe fruits at the end of June and the beginning of July. The plants were dried at room temperature and then ground.

M. rudis: 497 g of dry plant material were extracted with methanol in a Soxhlet extractor, methanol was distilled off, the extract treated with 1% cold acetic acid, filtered and the undissolved matter was washed several times with 1% acetic acid. The filtrate was processed in the conventional manner (see[13,14,16]) to give alkaloid fractions *A*, *B*, *I* and *E*. Fraction *A* was further purified (the weight of crude bases was 0·84 g) and then separated using the method described in paper[18], to give fractions AC, AD_1 and AD_2. Fraction AC was further separated to non-phenolic bases AC_1 and phenolic bases AC_2. Fraction AC_1 (0·23 g) did not crystallize and it was chromatographed on a column of 45 g of neutral alumina according to Brockmann (Reanal), activity about 2. The column was packed in benzene and 20 ml fractions were collected. Their composition was controlled by thin layer chromatography in the system S_1, S_2, S_3 and S_4. Elution with benzene–ether 2 : 1 (fractions 12—20) gave 2·5 mg of a mixture containing predominantly isorhoeadine and papaverrubine A, in addition to a small amount of rhoeadine and papaverrubine E. Elution with benzene–ether 1 : 1 (fractions 21—34) gave amurensinine (total yield, including a small amount obtained from fraction AD_1, was 130 mg; 0·026%). Further fractions (35—56) obtained on elution with benzene–ether 1 : 2, pure ether, and chloroform–methanol 3 : 2 contained predominantly non-alkaloidal substances in addition to the trace residues of amurensinine and protopine. Fraction AC_2 (9·2 mg) remained amorphous, and according to thin-layer chromatography it consisted of several alkaloids among which one was identified as amurensine. Papaverrubine D and C and unidentified alkaloids of R_F values 0·03 and 0·15 (in S_1), 0·21 and 0·39 (in S_2) 0·28, 0·40 and 0·43 (in S_3), and 0·11, 0·35 and 0·39 (in S_4) were also detected in this fractions.

Fraction AD_1 (182 mg) yielded after crystallization from chloroform–methanol 6-methoxy-2-methyl-1,2,3,4-tetrahydro-β-carboline (68·2 mg; 0·014%). Crystallization of the remaining bases from methanol gave protopine (36·8 mg; 0·0072%) and 4·7 mg of a fraction with m.p. 170—200°C, which was a mixture of approximately equal parts of protopine and allocryptopine. The amorphous residue of the bases (64 mg) was separated on a column of alumina (6·5 g) of the same quality as above, applying the same procedure. Fractions of 5 ml each were collected. Fractions 6—8 (benzene–ether 1 : 1; 8·7 mg of bases) contained predominantly amurensinine, fractions 9—13 (benzene–ether 1 : 1 and ether; 29·5 mg of bases) contained mainly alkaloid MR 1 in addition to a smaller amount of amurensinine, protopine, allocryptopine and 6-methoxy-2-methyl-1,2,3,4-tetrahydro-β-carboline; fractions 14—19 (ether; 8·2 mg of bases) contained predominantly a mixture of alkaloids MR 1 and MR 2. Fractions 20—30 were non-alkaloidal. Combined fractions 9—13 were again separated on a column of alumina (6 g) under the same conditions as above and elution with benzene–ether 4 : 1, 2 : 1 and 1 : 1 gave almost pure alkaloid MR 1 (13·0 mg) which, however, was unstable and changed to a yellow, strongly polar substance, darkening further after several hours standing.

Fraction AD_2 (66 mg) was amorphous and according to TLC it contained a small amount of amurensine, papaverrubine D and C and further alkaloids of R_F 0·04, 0·08, 0·22 and 0·42 (in S_2), 0·27, 0·40 and 0·48 (in S_3) and 0·11, 0·31 and 0·35 (in S_4). In fraction *B* (2·8 mg) a small quantity of coptisine was found. Fraction *E* (180 mg) was practically non-alkaloidal. Fraction *I* (160 mg) also contained predominantly substances of non-alkaloid character. The quaternary alkaloids present in low concentration were separated by precipitation of the aqueous solution of fraction *I* with 20% sodium perchlorate solution, giving poorly soluble perchlorates. Crystal-

lization of crude perchlorates from methanol gave amurensinine methoperchlorate (2 mg), the remaining part (9·6 mg) remained amorphous. According to thin layer chromatography it contained, in addition to the remaining amurensinine methoperchlorate, alkaloid MR 3 and MR 4 and a small amount of magnoflorine.

Meconopsis betonicifolia: 1 600 g of dry plant material were worked up in principle in the same manner as in the case of *M. rudis*. Thus fractions *A*, *B* (3·1 mg), *I*(67·0 mg, traces of alkaloids only) and *E* (10·0 mg, non-alkaloidal) were obtained. Fraction *A* was separated to fractions AC_1 (74·8 mg, mainly non-alkaloidal), AC_2 (13·5 mg), AD_1 (8·3 mg) and AD_2 (14·1 mg, mainly non-alkaloidal). From fraction AC_2 2·3 mg of a non-alkaloidal substance were obtained by crystallization from ether (needles, m.p. 136—138°C), which was not further investigated. The other fractions did not crystallize and therefore they were investigated by thin-layer and paper chromatography only. The results are given in the introductory. part. The unidentified quaternary alkaloid present in fraction *I* had R_F in S_6 identical with that of amurensinine methiodide. In fraction AD_2 a small amount of a strongly polar alkaloid (R_F 0·08 in S_2) was detected.

Other species of Meconopsis (the date of harvest and the weight of the plant material obtained are given in brackets): *M. horridula* (29. 6. 1970; 5·0 g), *M. sinuata* (24. 6. 1975; 5·6 g), *M. robusta* (5. 7. 1968; 9·9 g) and *M. paniculata* (5. 7. 1968; 13·5 g) were extracted with methanol in the cold and the extract was worked up practically in the same manner as in the cases of the preceding two species. The obtained fractions *A*, *B*, *I* and *E* (practically non-alkaloidal) were investigated by thin-layer and paper chromatography. The results are given in the introductory part.

Characterization of the Alkaloids

Amurensinine: from ether, prisms, m.p. 138—139°C, undepressed with authentic specimen, $[\alpha]_D^{24}$ −117° ± 2° (*c* 0·23, methanol). The UV spectrum[19] and the R_F values in four solvent systems were identical with those of the reference sample. Amurensinine methiodide (*II*, $X^{(-)} = 1$): amurensinine (15 mg) was methylated with methyl iodide (0·2 ml) in methanol–ether solution. After five days the solvents were evaporated and the product crystallized from methanol on dilution with ether; m.p. 278—279°C (capillary) or 295—297°C (Kofler block), undepressed on admixture of an authentic[20] sample, $[\alpha]_D^{22}$ −106° ± 4° (*c* 0·13, methanol). UV spectrum: λ_{max} (log ε) 210 nm (4·79), 296 nm (3·92), shoulder at 238 nm (4·18), λ_{min} 265 nm (2·98). Methoperchlorate was prepared from methiodide by precipitation of an aqueous solution with a 20% solution of sodium perchlorate and crystallization of the precipitate from methanol; long needles of m.p. 287—289°C (Kofler block).

6-*Methoxy*-2-*methyl*-1,2,3,4-*tetrahydro*-β-*carboline*: long needles, m.p. 215—216°C from ether (Kofler block), undepressed in admixture with a synthetic specimen, prepared by reduction of 2-ethoxycarbonyl-6-methoxy-1,2,3,4-tetrahydro-β-carboline with lithium aluminum hydride according to[10] (literature[10] gives m.p. 215·5—216·5°C). Its mass spectrum contained the molecular peak at mass 216·1256, corresponding to the composition $C_{13}H_{16}N_2O$ (theory 216·1263), and it was identical with the spectrum described in literature[9]. IR spectrum (in KBr): bands at 790 and 830 cm^{-1} (1,2,4-trisubstituted aromatic ring), 1 488, 1 595 and 1 600 cm^{-1} (aromatic ring), 2755 and 2795 cm^{-1} (N—CH_3 and OCH_3 group) and 3 145 cm^{-1} (associated —NH--group). It was identical with the spectrum of a reference sample. The UV spectrum, λ_{max} (log ε) 225 nm (4·48), 279 nm (3·96), shoulder at 295 nm (3·87) and 308 nm (3·60), λ_{min} 251 nm (3·47) was also identical with the spectrum of the reference sample and in good agreement with the literature[9,10] data. The R_F values in four solvent systems confirmed the identity (after detection with potassium iodoplatinate, violet spot). With concentrated sulfuric acid the base became yellow, with Erdmann's reagent it turned pink and with concentrated nitric acid yellow.

Protopine: prisms, m.p. 206—207°C (from chloroform–ethanol), underpressed with an authentic specimen. The identity was confirmed by R_F values and colour reactions.

Amurensinine methohydroxide: perchlorate from methanol, needles, m.p. 287—288°C (Kofler block), undepressed in admixture with a standard prepared from amurensinine. The R_F values were identical with those of a reference sample. The same was true of colour reactions: with concentrated sulfuric acid no coloration was observed, with Erdmann's reagent it turned first blue-green for a short time, then changed to yellow-green and ochre; with Fröhde's reagent it gave a green and with concentrated nitric acid a brown-red, later brown, coloration.

R_F Values

In systems S_1, S_2, S_3 and S_4, respectively: alkaloid MR 1 0·11, 0·20, 0·08, 0·09; alkaloid MR 2 0·14, 0·29, —, —; allocryptopine 0·25, 0·63, 0·17, 0·13; amurensine 0·06, 0·24, 0·35, 0·22; amurensinine 0·28, 0·61, 0·42, 0·30; cryptopine 0·21, 0·68, 0·28, 0·16; isorhoeadine 0·61, 0·84, 0·97, 0·96; 6-methoxy-2-methyl-1,2,3,4-tetrahydro-β-carboline 0·05, 0·10, 0·36, 0·19; papaverrubine A 0·37, 0·67, 0·93, 0·93; papaverrubine C 0·13, 0·41, 0·72, 0·79; papaverrubine D 0·07, 0·29, 0·68, 0·61; papaverrubine E 0·37, 0·67, 0·58, 0·47; protopine 0·40, 0·73, 0·35, 0·23; rhoeadine 0·58, 0·81, 0·76, 0·77.

In system S_5: papaverrubine A 0·83; papaverrubine C 0·41; papaverrubine D 0·30; papaverrubine E 0·58.

In systems S_6, S_7 and S_8, respectively: alkaloid MR 3 0·55, 0·43, 0·70; alkaloid MR 4 0·56, 0·19, 0·68; amurensinine methiodide 0·27, 0·11, 0·58; magnoflorine 0·64, 0·54, 0·48 (violet fluorescence).

In systems S_9, S_{10} and S_{11}, respectively: berberine 0·17, 0·63, 0·20; coptisine 0·50, 0·49, 0·07; corysamine 0·09, 0·73, 0·66.

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REFERENCES

1. Slavík J.: This Journal *25*, 1663 (1960).
2. Slavík J., Slavíková L.: This Journal *28*, 1720 (1963).
3. Slavík J.: This Journal *30*, 914 (1965).
4. Slavík J.: This Journal *31*, 4184 (1966).
5. Hemingway S. R., Phillipson J. D.: J. Pharm. Pharmacol. *27*, 84 P (1975) (Suppl.).
6. Gertig H.: Ann. Pharm. (Poznań) *1969*, 111.
7. Slavík J., Slavíková L.: This Journal *41*, 3343 (1976).
8. Hakim S. A. E., Mijović V., Walker J.: Nature (London) *189*, 198 (1961).
9. Fedde F. in the book: *Das Pflanzenreich — Regni vegetabilis conspectus* (A. Engler, Ed.), Part IV, Band 104. Leipzig 1909.

10. Agurell S., Holmstedt B., Lindgren J. E.: Acta Chem. Scand. *23*, 903 (1969).
11. Shannon P. V. R., Leyshon W. M.: J. Chem. Soc. (C) *1971*, 2837.
12. Viiayanagar H. M., Audette R. C. S., Bolan J., Clark J. W.: Lloydia *38*, 442 (1975).
13. Slavíková L., Slavík J.: This Journal *24*, 559 (1959); Chem. Listy *51*, 1923 (1957).
14. Slavík J., Slavíková L.: This Journal *26*, 1839 (1961).
15. Hughes D. W., Kühn L., Pfeifer S.: J. Chem. Soc. (C) *1967*, 444.
16. Slavíková L., Slavík J.: This Journal *31*, 3362 (1966).
17. Slavík J., Novák V., Slavíková L.: Unpublished results.
18. Slavík J., Slavíková L.: This Journal *41*, 290 (1976).
19. Novák V., Slavík J.: This Journal *39*, 883 (1974).
20. Slavík J., Slavíková L., Dolejš L.: This Journal *33*, 4066 (1968).

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