Studies on the Constituents of *Corydalis* sp. VI.[1] Alkaloids from Chinese Corydalis and the Identity of *d*-Corydalmine with *d*-Corybulbine

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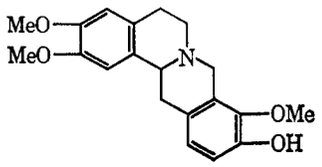
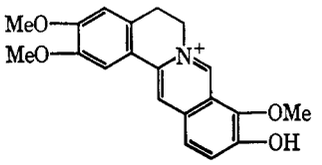
Several years ago, the first isolation of two new protoberberine bases named corydalmine (I), $C_{20}H_{23}O_4N$ (mp 238–239°, $[\alpha]_D$ 337.4°), and dehydrocorydalmine (II), $C_{20}H_{20}O_4N^+$ (iodide, $C_{20}H_{20}O_4NI \cdot 2H_2O$, mp 238–



I

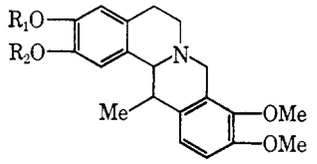


II

240° dec), from the commercially available *Corydalis* species[2] cultivated for use in Chinese medicine, and their structural assignments were reported by Imaseki and Taguchi.[3]

Recently, Telang and Bradsher[4] synthesized *dl*-10-hydroxy-2,3,9-trimethoxydibenzo[*a*,*g*]quinolizidine corresponding to the structure of I, and more recently Doskotch, *et al.*,[5] isolated dehydrocorydalmine iodide from *Stephania glabra*. It was noted by the above authors[4,5] and by Jeffs[6] that there were large differences in melting points between the synthetic product of *dl*-corydalmine,[4] mp 187.5–188.5°, and the natural *dl*-corydalmine,[3] mp 213–215°, and also between dehydrocorydalmine iodide from *Corydalis* species,[3] mp 238–240° dec, and that from *Stephania glabra*,[5] mp 195°. Up to now, those discrepancies in the melting points have remained unresolved because of the absence of a sample for direct comparison, and there is still some uncertainty which cannot be resolved satisfactorily by the accepted formulas I and II for these alkaloids.

In this paper, we wish to reveal that the alkaloid having mp 238–239°, and named *d*-corydalmine by Imaseki and Taguchi,[3] is identical with *d*-corybulbine[7]



III, $R_1 = H$; $R_2 = Me$
IV, $R_1 = Me$; $R_2 = H$

having the structure of III, and also to report further investigations on the tertiary alkaloids from the same plant.[2]

Reexamination of the tertiary alkaloid fraction from the plant resulted in the isolation of eleven crystalline alkaloids (Table I). From the tertiary nonphenolic

TABLE I
ALKALOIDS ISOLATED FROM COMMERCIALLY AVAILABLE CHINESE CORYDALIS[a]

Name	Yield,[b] %
Tertiary Phenolic Alkaloids	
l-Tetrahydrocolumbamine	0.0474
d-Corybulbine	0.0054
l-Scoulerine	0.0021
d-Tetrahydrojatrorrhizine	0.0015
Tertiary Nonphenolic Alkaloids	
d-Corydaline	0.1161
d-Glaucine	0.0822
dl-Tetrahydropalmatine	0.0530
Protopine	0.0282
l-Tetrahydrocoptisine	0.0182
α-Allocryptopine	0.0003
Noroxyhydrastinine	0.0001

[a] Reference 2. [b] Per cent yields calculated from the dried material.

alkaloid fraction, *d*-corydaline, *dl*-tetrahydropalmatine, *l*-tetrahydrocoptisine, and protopine were isolated by means of alumina column chromatography in the manner described by Imaseki and Taguchi.[3] Further elution with methanol from the alumina column gave a large amount of a crystalline aporphine-type alkaloid which was identified as *d*-glaucine[8,9] by direct comparison. On the other hand, the tertiary phenolic base fraction showed more than ten spots on the thin layer chromatogram, and eight alkaloids were isolated by means of silica gel column chromatography. Five of them were identified as *l*-tetrahydrocolumbamine,[3] *d*-corydaline, *l*-scoulerine,[10] noroxyhydrastinine,[11] and α-allocryptopine,[10] respectively, by direct comparison, *i.e.*, thin layer chromatography, infrared and nmr spectra, and mixture melting point determinations. One new base, mp 248–250°, having a characteristic infrared absorption at 1715 cm^{-1}, was isolated in so small an amount that it could not be further investigated. The other two tertiary phenolic bases were tentatively designated as base A and B.

Base A was obtained not only by the method previously reported for the isolation of *d*-corydalmine[3] but also by means of column chromatography described in the Experimantal Section. Base A, mp 220–222° dec, $[\alpha]_D$ 307°, analyzed for $C_{21}H_{25}O_4N$. Its infrared spectrum taken in a Nujol mull was superimposable on the reported spectrum of the "*d*-corydalmine" of Imaseki and Taguchi,[3] although we were unable to obtain an authentic sample for a direct comparison. The nmr spectrum of base A taken in DMSO-d_6 solution was

(1) For part V, see S. Naruto, S. Arakawa, and H. Kaneko, *Tetrahedron Lett.*, 1705 (1968).

(2) The original plant of Chinese corydalis is described as *Corydalis bulbosa* D. C. in the official Chinese pharmacognostical book "Chung Yao Chih," Vol. 1, Peking, 1959, p 263, but is quite different from *C. bulbosa* D. C. occuring in Europe. It rather seems to be morphologically a variety of *C. ambigua* Cham. et Schlecht. The pharmacognostical data of this plant will be reported elsewhere by our coworkers, Dr. S. Takahashi and Mr. K. Namba.

(3) I. Imaseki and H. Taguchi, *Yakugaku Zasshi*, **82**, 1214 (1962).

(4) S. A. Telang and C. K. Bradsher, *J. Org. Chem.*, **30**, 752 (1965).

(5) R. W. Doskotch, M. Y. Malik, and J. L. Beal, *ibid.*, **32**, 3253 (1967).

(6) P. W. Jeffs, "The Alkaloids, Chemistry and Physiology," Vol. 9, R. H. F. Manske, Ed., Academic Press Inc., New York, N. Y., 1967, pp 71–72.

(7) R. H. F. Manske, *Can. J. Res.*, **21B**, 13 (1943).

(8) J. Gadamer, *Arch. Pharm.*, **249**, 224 (1911).

(9) The isolation of *d*-glaucine served to identify one of the main spots which had not been identified hitherto on the two-dimensional thin layer chromatogram of the total alkaloid fraction of this plant. On the other hand, it was found that the peak of *d*-corydaline contained that of *d*-glaucine on the gas chromatogram of the total alkaloid, since the retention times of both alkaloids were almost equal on 2% QF-1 column: J. Iwasa, S. Naruto, and Y. Utsui, *Yakugaku Zasshi*, **86**, 396 (1966).

(10) R. H. F. Manske, *Can. J. Res.*, **14B**, 347 (1936).

(11) W. H. Perkin, Jr., and R. Robinson, *J. Chem. Soc.*, **97**, 305 (1910).

most informative, showing four aromatic protons at δ 6.85 (2 H), 6.72 (1 H), and 6.47 (1 H), three methoxyl groups at δ 3.85 (3 H) and 3.81 (6 H), one phenolic hydroxyl proton at δ 8.70, and a secondary methyl group at δ 0.83 (3 H) as a doublet (J = 6.8 cps). This secondary methyl signal could not be explained by the structure of I proposed for *d*-corydalmine.[3] Its methylation with diazomethane to *d*-corydaline served to characterize it as a 13-methylated protoberberine base having a 2,3,9,10-oxygenation pattern. The position of the phenolic hydroxyl group remained to be settled. Among the four possible isomers, two bases, *d*-corybulbine (III) and *d*-isocorybulbine (IV), were already known, the former of which was similar to base A in respect to physical constants. As a result, it was found that base A was identical with *d*-corybulbine by comparison of nmr and infrared spectra, thin layer chromatographic behavior, and mixture melting point. Accordingly, the name of *d*-corydalmine and its proposed structure I, first reported by Imaseki and Taguchi,[3] should be revised to those of *d*-corybulbine (III). Furthermore, it is suggested that the accompanying quaternary base, "dehydrocorydalmine," which on reduction afforded an optically inactive *dl*-corydalmine,[3] was identical with dehydrocorybulbine. Therefore the genuine "dehydrocorydalmine," represented by structure II, is considered to be that from *Stephania glabra*.[5] Recent investigation on the constituents of *S. glabra* has confirmed the presence of (−)-10-hydroxy-2,3,9-trimethoxydibenzo[*a*,*g*]quinolizidine (so-called *l*-corydalmine) as a natural product.[12]

Base B, mp 214–215°, $[\alpha]_D$ 302°, $C_{20}H_{23}O_4N$, gave *d*-tetrahydropalmatine by methylation with diazomethane. The infrared spectrum taken in the chloroform solution was superimposable on the infrared spectrum of *dl*-tetrahydrojatrorrhizine synthesized from berberine according to the method of Späth and Quietensky.[13] These results, in conjunction with the nmr spectrum of base B, indicated that it was *d*-tetrahydrojatrorrhizine.

In conclusion, *d*-glaucine, *l*-scoulerine, *d*-tetrahydrojatrorrhizine, noroxyhydrastinine, and α-allocryptopine were isolated for the first time from Chinese corydalis[2] cultivated in China. It is interesting to note from the chemotaxonomical viewpoint that three main alkaloids, *d*-corydaline, *d*-corybulbine, and dehydrocorydaline,[14] in this plant are identical with the major alkaloids of *Corydalis ambigua* Cham. *et* Schlecht. *var. amurensis* Maxim.[15,16] This fact suggests that the original plant of Chinese corydalis[2] is closely related to *C. ambigua* Cham. *et* Schlecht.

Experimental Section

Melting points are uncorrected. Ultraviolet spectra were measured in ethanol on a Hitachi Model EPS-2U recording spectrophotometer, and infrared spectra were taken in KBr disks unless otherwise specified, with a Hitachi Model EPI-S2 infrared spectrophotometer. Nmr spectra were determined in deuteriochloroform or DMSO-d_6 solution, with tetramethylsilane as an internal standard, with a Varian A-60 spectrometer. The optical rotations were determined with a Rex Model NEP-2 photoelectric polarimeter. Thin layer chromatography (tlc) was carried out with the use of silica gel G (Merck) and the solvent system used in tlc is abbreviated as follows: chloroform–methanol, 25:1 (solvent a). Alkaloid spots or bands were detected by spraying with Dragendorff's reagent. Microanalyses were performed by Mr. Y. Utsui and his associates.

Separation of the Crude Alkaloid.—The ether extract of the total tertiary alkaloids was obtained from the tubers of Chinese corydalis imported from China (5.0 kg) according to the procedure described previously.[14] This ether extract was concentrated to 0.05 volume and allowed to stand for 1 week. The crude crystals of *l*-tetrahydrocolumbamine were deposited. The etherial mother liquor was extracted with 5% aqueous sodium hydroxide solution. The ether layer was dried over anhydrous potassium carbonate and evaporated to dryness to give the nonphenolic alkaloid fraction (22 g). The 5% aqueous sodium hydroxide layer was saturated with ammonium chloride and extracted with ether. The ether layer was dried over anhydrous potassium carbonate and the solvent was removed to give the crude phenolic alkaloid fraction (4.5 g).

Isolation of the Alkaloids. A. Nonphenolic Alkaloids.—The crude alkaloid fraction (22 g) was placed on a column of basic alumina (350 g, activity I, Merck). *d*-Corydaline, mp 132–134° (3.87 g), *dl*-tetrahydropalmatine, mp 143–145° (0.37 g), *l*-tetrahydrocoptisine, mp 197–200° (0.35 g), and protopine, mp 205–207° (1.05 g), were isolated in the manner described by Imaseki and Taguchi[3] and were each identified with authentic samples. The mother liquors of *l*-tetrahydrocoptisine, *dl*-tetrahydropalmatine, and *d*-corydaline were combined and concentrated. The residue (9.56 g) was rechromatographed on silica gel column (100 g, silica gel, Merck). From benzene (6 l.) and benzene–ether (1:1) (1 l.) elutes, *d*-corydaline (0.97 g), *l*-tetrahydrocoptisine (0.57 g), and *dl*-tetrahydropalmatine (1.3 g) were isolated in a manner similar to the above procedure.[3] Ether (3 l.) and methanol (0.8 l.) elutes were combined and concentrated to give the residue (5.3 g), which was treated with a small amount of ether. The ether-soluble fraction was concentrated and crystallized from ether–petroleum ether to yield crystals of *d*-glaucine: mp 120–121.5° (2.958 g); $[\alpha]_D$ 125° (*c* 1.0, methanol); λ_{max}^{EtOH} 219 mμ (log ε 4.58), 282 (4.18), 302 (4.17).

Anal. Calcd for $C_{21}H_{25}O_4N$: C, 70.96; H, 7.09; N, 3.94. Found: C, 70.89; H, 6.92; N, 4.24.

The hydrobromide melted at 234–236° (from ethanol).

Anal. Calcd for $C_{21}H_{25}O_4N \cdot HBr$: C, 57.80; H, 6.01; N, 3.21; Br, 18.32. Found: C, 57.71; H, 6.08; N, 3.05; Br, 18.12. Identity was established by comparisons of the infrared spectrum of this hydrobromide with that of an authentic sample, and by mixture melting point determination.

The methanol elute from the first alumina column chromatography and the mother liquor of protopine were combined and concentrated. The residue (6.242 g) was applied to the top of the silica gel column (30 g). The chromatography was developed with chloroform to obtain further amounts of *d*-glaucin (1.150 g) and protopine (0.363 g).

B. Phenolic Alkaloids.—The crude phenolic alkaloid fraction (4.1 g) was combined with the similar material from another extraction (total, 19.6 g), and treated with chloroform (200 ml). The chloroform-insoluble fraction was recrystallized from chloroform–methanol to give *l*-tetrahydrocolumbamine, mp 222–224°, (4.3 g), which was identical with an authentic sample of *l*-tetrahydrocolumbamine. The mother liquor was combined with the chloroform-soluble fraction and concentrated. The brown residue (15.3 g) was dissolved in 4% methanol–chloroform applied to the top of a silica gel column (200 g, 4.8 × 23.4 cm, silica gel, Merck) and eluted in six fractions as shown in Table II.

TABLE II

COLUMN CHROMATOGRAPHY OF PHENOLIC ALKALOID (1)

Fraction	Eluent	Volume, ml	Yield, mg	R_f values on tlc, solvent a
1-1	4% MeOH–$CHCl_3$	100	911	0.93, 0.83
1-2	4% MeOH–$CHCl_3$	360	8890	0.66, 0.55, 0.43
1-3	4% MeOH–$CHCl_3$	360	1750	0.55, 0.43, 0.33 0.24
1-4	4% MeOH–$CHCl_3$	180	680	0.15, 0.11, 0.05
1-5	4% MeOH–$CHCl_3$	800	807	0.11, 0.05
1-6	6% MeOH–$CHCl_3$	800	310	0.02

Fraction 1-1 showed two spots (R_f 0.93 and 0.83) on tlc. These two components were separated by rechromatography on silica

(12) M. P. Cava, K. Nomura, S. K. Talapatra, M. J. Mitchell, R. H. Schlessinger, K. T. Buck, J. L. Beal, B. Douglas, R. F. Raffauf, and J. A. Weisbach, *J. Org. Chem.*, **33**, 2785 (1968).

(13) E. Späth and H. Quietensky, *Ber.*, **58**, 2267 (1925).

(14) J. Iwasa, S. Naruto, and N. Ikeda, *Yakugaku Zasshi*, **86**, 437 (1966).

(15) H. Taguchi and I. Imaseki, *ibid.*, **83**, 578 (1963).

(16) H. Taguchi and I. Imaseki, *ibid.*, **84**, 773 (1964).

gel column (20 g, 2.3 × 13 cm) with methanol–chloroform (1:100) as a developing solvent. The component of R_f 0.93 was obtained as colorless needles, mp 248–250°, after recrystallization from methanol. This material, showing an infrared absorption at 1715 cm^{-1}, was obtained in such a small amount (3 mg) that it could not be further investigated. The other component (R_f 0.83) was *d*-corydaline, mp 132–134°, identical (tlc behavior, infrared spectrum, and mixture melting point determination) with an authentic sample. Fraction 1-2 showed three spots (R_f 0.66, 0.55, and 0.43) on tlc. Repeated crystallization of this fraction from chloroform–methanol yielded a material showing R_f 0.55, which proved to be identical with *l*-tetrahydrocolumbamine by direct comparison. The mixture consisted of *l*-tetrahydrocolumbamine, and a material showing R_f 0.66 was deposited from the mother liquor. These two components were separated by means of silica gel column chromatography with methanol–chloroform (3:100) as a developing solvent. The component (R_f 0.66) was obtained from the first elute in pure form, which was designated as base A described below. The last elute gave *l*-tetrahydrocolumbamine. The mother liquors of these two bases were combined and concentrated to yield the residue (2.0 g) which was rechromatographed on silica gel column (60 g, 2.8 × 20 cm) and eluted in four fractions as shown in Table III.

TABLE III

COLUMN CHROMATOGRAPHY OF PHENOLIC ALKALOID (2)

Fraction	Eluent	Volume, ml	Yield, mg	R_f values on tlc, solvent a
2-1	4% MeOH–$CHCl_3$	50	150	0.83, 0.66
2-2	4% MeOH–$CHCl_3$	100	804	0.66, 0.55, 0.43
2-3	4% MeOH–$CHCl_3$	40	494	0.43, 0.33
2-4	4% MeOH–$CHCl_3$	40	285	0.33, 0.24

Additional amounts of *l*-tetrahydrocolumbamine (200 mg) and base A (500 mg) were obtained by recrystallization of fraction 2-2 from chloroform–methanol. The mother liquors were combined with the fraction 2-3 and recrystallized from the same solvent to give base B (R_f 0.43) (341 mg). Fraction 2-4 was converted to the hydrochloride and crystallized from ethanol–ether to yield pale yellow crystals of *l*-scoulerine hydrochloride (74 mg). Identity was established by comparison of the infrared spectrum of its base (mp 194–196°, R_f 0.24) with that of an authentic sample, and by mixture melting point determination.

Fraction 1-3 was recrystallized from chloroform–methanol to give *l*-tetrahydrocolumbamine (351 mg). This mother liquor was concentrated to give the residue (1.4 g), which was rechromatographed on silica gel column (70 g, 3.2 × 20 cm) and eluted in three fractions as shown in Table IV.

TABLE IV

COLUMN CHROMATOGRAPHY OF PHENOLIC ALKALOID (3)

Fraction	Eluent	Volume, ml	Yield, mg	R_f values on tlc, solvent a
3-1	4% MeOH–$CHCl_3$	20	198	0.55, 0.43
3-2	4% MeOH–$CHCl_3$	60	1250	0.33, 0.24
3-3	4% MeOH–$CHCl_3$	60	68	0.33, 0.24

Fraction 3-1 was treated as in the case of fraction 2-2 to give *l*-tetrahydrocolumbamine (51 mg) and base B (108 mg). Fraction 3-2 was converted to the hydrochloride and treated as in the case of fraction 2-4 to give *l*-scoulerine hydrochloride (451 mg). The mother liquor of the hydrochloride was reconverted to the base. This basic fraction (714 mg) showed the presence of a component of R_f 0.33 which was isolated by means of silica gel column chromatography (24 g, 2.1 × 16.5 cm) using chloroform–methanol (25:1) as a developing solvent. The compound, isolated as pale yellow needles (R_f 0.33), mp 189–190°, was identical with an authentic sample of noroxyhydrastinine.

Fraction 1-4 was converted to the hydrochloride and allowed to stand for 4 days. Pale yellow crystals of protopine hydrochloride separated, which, after recrystallization from ethanol, were converted to the base. This base was identified with an authentic sample of protopine by comparison of the infrared spectrum and by mixture melting point determination. The mother liquor of fraction 1-4 and fraction 1-5 contained more than two components (R_f 0.15 and 0.11), none of which could be isolated in pure form.

Crystallization of fraction 1-6 from ethanol yielded a small amount (120 mg) of the material showing R_f 0.02, mp 163–164°, which proved to be identical with α-allocryptopine. Identity was established by comparison of the infrared spectrum with that of an authentic sample, by tlc behavior, and by mixture melting point determination. The remainder of the fraction could not be separated into pure components.

Base A (*d*-**Corybulbine** = *d*-**Corydalmine**) (**III**).—Base A, mp 220–222° dec, $[\alpha]_D$ 307° (*c* 0.38, chloroform), was identical with *d*-corybulbine [nmr (described in the text) and infrared spectra, tlc behavior, and mixture melting point determination].

Anal. Calcd for $C_{21}H_{25}O_4N$: C, 70.96; H, 7.09; N, 3.94. Found: C, 71.30; H, 6.96; N, 3.84.

A solution of 5 mg of base A in 10 ml of methanol was added to a solution of an excess of diazomethane in ether, and the mixture was allowed to stand for 3 hr. The solution was concentrated under reduced pressure and the residue was recrystallized from ethanol to give pale yellow prisms of mp 134–136°. This compound was identified with *d*-corydaline by comparison of its infrared spectrum and by mixture melting point determination.

Base B (*d*-**Tetrahydrojatrorrhizine**).—Base B had mp 214–215°, $[\alpha]_D$ 302.2° (*c* 0.65, chloroform).

Anal. Calcd for $C_{20}H_{23}O_4N$: C, 70.36; H, 6.79; N, 4.10. Found: C, 70.57; H, 7.09; N, 4.04. Infrared (in chloroform solution) and nmr spectra and tlc behavior of this compound were identical with those of synthetic *dl*-tetrahydrojatrorrhizine.[13] Base B was methylated as in the case of base A by diazomethane to give pale yellow prisms, mp 143–145°. Tlc behavior and infrared spectrum of this methylated compound were identical with those of *dl*-tetrahydropalmatine.

Registry No.—*l*-Tetrahydrocolumbamine, 20504-94-3; *d*-corybulbine, 518774; *l*-scoulerine, 6451-73-6; *d*-tetrahydrojatrorrhizine, 6018-39-9; *d*-corydaline, 3907-48-0; *d*-glaucine, 475-81-0; *dl*-tetrahydropalmatine, 2934-97-6; protopine, 130-86-9; *l*-tetrahydrocoptisine, 20504-98-7; α-allocryptopine, 485-91-6.

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A Novel Method of Converting Aldehydes into Nitriles under Mild Conditions. The Reaction of Dialkyl Hydrogen Phosphonates with Oximes

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March 10, 1969

Present methods of converting aldehydes into nitriles generally require rather vigorous conditions.[2] We wish to report a very mild method of effecting this conversion in two simple steps.

(1) Plastics Department, E. I. du Pont de Nemours and Co., Experimental Station Laboratory, Wilmington, Del. 19898.

(2) See, for example, W. Theilheimer ["Synthetic Methods of Organic Chemistry," Vol. 1–22, S. Karger, A G., Basel, Switzerland, 1946–1968] for representative techniques. For a mild method, see J. H. Pomeroy and C. A. Craig, *J. Amer. Chem. Soc.*, **81**, 6340 (1959).