The Alkaloids of *Stephania glabra*. A Direct Chemical Correlation of the Absolute Configuration of Some Benzyltetrahydroisoquinoline, Proaporphine, and Aporphine Alkaloids. A New Protoberberine Alkaloid

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Separation of the alkaloid bases of *Stephania glabra* yielded the proaporphines (+)-stepharine and (+)-pronuciferine, as well as the protoberberines (−)-tetrahydropalmatine, (−)-corydalmine, and (−)-stepholidine. Evidence is presented to support the structures assigned to the new alkaloids stepharine and stepholidine. The cleavage of proaporphines to benzyltetrahydroisoquinolines by sodium in ammonia is reported. The use of this cleavage reaction, considered together with the conversion of proaporphines into aporphines, makes possible a chemical correlation of the absolute configuration of several alkaloids of the benzyltetrahydroisoquinoilne, proaporphine, and aporphine groups.

In an earlier paper, we gave a preliminary account of the isolation of the dienone alkaloids stepharine (I) and pronuciferine (II) from *Stephania glabra*, as well as a simple chemical correlation of the absolute configuration of these compounds with that of related aporphines and benzyltetrahydroisoquinolines.[2] In this paper we will give additional details of this investigation, including the isolation and characterization of stepholidine (XII), a new protoberberine alkaloid.

Stephania glabra (Miers) is a large, climbing shrub belonging to the family Menispermaceae. The large tubers of this plant have been used in India for the treatment of a variety of disorders, including asthma, tuberculosis, dysentery, and fevers.[3]

The first report on the alkaloids of *S. glabra* tubers described the isolation of three alkaloids named gindarine, gindarinine, and gindaricine.[4] Subsequently, gindarine and gindarinine were shown to be identical with tetrahydropalmatine and palmatine, respectively; gindaricine was not further identified.[5,6] The work described in this paper concerns a reinvestigation of the tertiary bases of *S. glabra*;[7] a study of the quaternary alkaloids is reported elsewhere.[8,9]

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(2) M. P. Cava, K. Nomura, R. H. Schlessinger, K. T. Buck, B. Douglas, R. F. Raffauf, and J. A. Weisbach, *Chem. Ind.* (London), 282 (1964).

(3) R. N. Chopra, I. C. Chopra, K. L. Handa, and L. D. Kapur, "Chopra's Indigenous Drugs of India," 2nd ed, U. N. Chur and Sons, Ltd., Calcutta, 1958, p. 412.

(4) G. R. Chaudhry and S. Siddiqui, *J. Sci. Ind. Res.*, (India), **98**, 79 (1950).

(5) G. R. Chaudry, V. N. Sharma, and M. L. Dhar, *ibid.*, **118**, 337 (1952).

(6) The analytical values reported for gindaricine[4] suggest that it is quite possibly identical with stepharine; on the other hand, the melting point reported for gindaricine (193°) is more than 10° higher than our melting point of stepharine. It should be noted also that the rotation of gindarine (tetrahydropalmatine) is given as −261° in the discussion section of ref 4 but as +261° in the Experimental Section.

(7) Since the publication of our preliminary communication,[2] several reports have appeared concerning the alkaloids of *S. glabra*, mainly material of Caucasian origin: I. I. Shchelchokova, T. N. Il'inskaya, and A. D. Kuzovkov, *Khim. Prirodn. Soedin., Akad. Nauk SSSR, Inst. Khim. Privodn. Soedin.*, 271 (1965); K.-C. Fang, I. I. Fadeeva, and T. N. Il'inskaya, *ibid.*, 392 (1965); I. M. Rabinvich, P. N. Kibal'chick, I. I. Fadeeva, T. N. Il'inskaya, A. D. Kuzovkov, V. V. Bereshinskaya, E. A. Trutneva, and S. S. Nikitina, *Aptech. Delo*, **14**, 19 (1965). The total alkaloids reported in these papers are (−)-tetrahydropalmatine, (+)-stepharine, cycleanine, and four unidentified bases. Interestingly, (+)-stepharine was found only in Indian *S. glabra;* no product corresponding to stepholidine was encountered. In addition, (+)-stepharine has recently been isolated from *S. rotunda* [M. Tomita, M. Kozuka, and S. Uyeo, *Yakugaku Zasshi*, **86**, 460 (1966)] and *Pericampylus formosanus* [M. Tomita, M. Kozuka, and S.-T. Lu, *ibid.*, **87**, 315 (1967)].

(8) M. T. Wa, J. L. Beal, and R. W. Doskotch, *Lloydia*, in press.

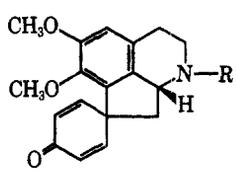
(9) R. W. Doskotch, M. Y. Malik, and J. L. Beal, *J. Org. Chem.*, **32**, 3253 (1967).

The Proaporphine Bases.—One of the major alkaloids of *S. glabra* was a previously unreported,[6] nonphenolic base $C_{18}H_{19}NO_3$, mp 179–180°, which we have named stepharine (I). Stepharine is dextrorotatory, $[\alpha]D$ +143°, and contains two methoxyl groups as shown by direct analysis. Stepharine was shown to be a secondary amine containing an NH grouping, since it was readily converted by acetic anhydride into N-acetylstepharine (III), mp 235–236°. The infrared spectrum of III, compared to that of I, showed increased absorption in the 6- to 6.2-μ region, compatible only with an amide formulation for III. Similarly, the NH group of stepharine could be methylated by formaldehyde and formic acid to give N-methylstepharine (II): mp 128–129°; $[\alpha]D$ +87°. N-methylstepharine obtained in this way was identical with a second, hitherto unreported base present in the naturally occurring alkaloid mixture from *S. glabra*. The nature of the third oxygen atom of stepharine was revealed in the course of an attempted methylation of I by formaldehyde in the presence of hydrogen and palladium. The reaction product was not N-methylstepharine, but rather N-methyltetrahydrostepharine (X). The infrared spectrum of X (but not of II) showed the presence of a strong *unconjugated* carbonyl group (5.84 μ); on the other hand, the infrared spectra of I and II (but not the spectrum of X) showed strong absorption in the 6–6.2-μ region, which could now be assigned to a highly conjugated carbonyl group. At this stage, it seemed likely that we had encountered two examples of the unknown proaporphines,[10] a new alkaloidal class originally predicted by Barton and Cohen[11] as intermediates in the biosynthetic, oxidative transformation of benzylisoquinolines to aporphines. Our supposition was confirmed when stepharine underwent smoothly the dienone–phenol rearrangement on heating with aqueous hydrochloric acid to give the known aporphine alkaloid (−)-tuduranine (VII). It is of interest to note that none of the isomeric rearrangement product VIII could be detected. Finally, excellent additional support for the proaporphine structure was found in the sodium and liquid ammonia cleavage of N-acetylstepharine (III)

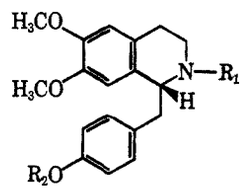
(10) The generic name proaporphine was independently proposed both by us[2] and by M. Shamma and W. A. Slusarchyk [*Chem. Rev.*, **64**, 59 (1964)]. The name appears to have received general acceptance by other workers in the field.

(11) D. H. R. Barton and T. Cohen, "Festschrift Arthur Stoll," Birkhauser A. G., Basel, 1957, p 117.

and N-methylstepharine (II) which gave (−)-N-acetylnorarmepavine (IV)[12] and (−)-armepavine (VI), respectively. At this time, Bernauer described the isolation and complete structure of pronuciferine (II), the first identified proaporphine alkaloid;[13] the same alkaloid, together with a closely related base, was reported simultaneously from a different plant source.[14] Since it was quickly determined by direct comparison that pronuciferine and N-methylstepharine (II) were identical, no further structural studies with the proaporphines I and II were carried out in our laboratories.[15]

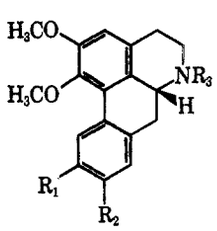


I, R = H
II, R = CH_3
III, R = $COCH_3$

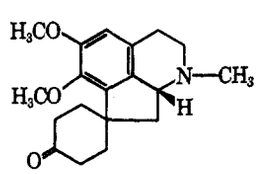


IV, R_1 = $COCH_3$; R_2 = H
V, R_1 = $COCH_3$; R_2 = CH_3
VI, R_1 = CH_3; R_2 = H

Other workers have determined the absolute configuration of armepavine.[16] We have found that the reductive cleavage product of pronuciferine (II) is (−)-armepavine, which has the D (or *R*) configuration at C-1, as indicated in structure VI. It follows, therefore, that both pronuciferine (II) and stepharine (I) must also have the D (or *R*) configuration at the corresponding asymmetric carbon atom. Furthermore, since we found that stepharine is rearranged by acid to (−)-tuduranine (VII),[17] and Bernauer reported that borohydride reduction of pronuciferine (N-methylstepharine) followed by acid treatment yields (−)-nuciferine (IX), it follows that both of the aporphine alkaloids (−)-tuduranine and (−)-nuciferine must belong stereochemically to the D (or *R*) series, as shown in structures VII and IX. The demonstration of the



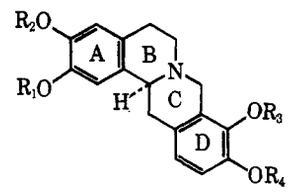
VII, R_1 = OH; R_2 = H; R_3 = H
VIII, R_1 = H; R_2 = OH; R_3 = H
IX, R_1 = R_2 = H; R_3 = CH_3

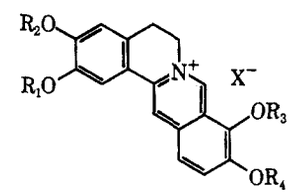


X

stereochemistry of nuciferine (IX) is particularly significant, since it has not been possible previously to prove chemically the absolute configuration of an aporphine alkaloid which lacks oxygen-containing functional groups on the benzyl-derived aromatic ring.[18,19]

The Protoberberine Bases.—As has been reported,[4-6] (−)-tetrahydropalmatine (XI) was present in a large quantity in *S. glabra* tubers. In addition, two phenolic bases were isolated, both of which yielded (−)-tetrahydropalmatine on treatment with diazomethane. The first phenolic base, mp 174–175°, had the composition $C_{20}H_{23}O_4N$, $[\alpha]_D$ −310°. On the basis of its composition and methylation to (−)-tetrahydropalmatine (XI), this alkaloid could be only one of the following four compounds: (−)-corydalmine (XV), (−)-tetrahydrocolumbamine (XVI), (−)-tetrahydrojatrorrhizine (XVII), or (−)-tetrahydropalmatrubine (XVIII). Direct comparison of its solution infrared spectrum with that of synthetic (±)-corydalmine[20] showed that it was (−)-corydalmine (XV). Only the enantiomeric (+)-corydalmine had been isolated previously from natural sources.[21,22]

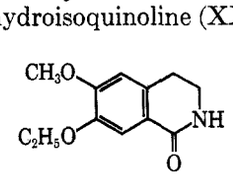




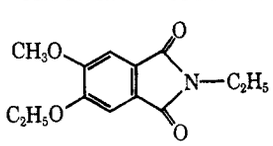
XI, R_1 = R_2 = R_3 = R_4 = CH_3
XII, R_1 = R_4 = H; R_2 = R_3 = CH_3
XIII, R_1 = R_3 = H; R_2 = R_4 = CH_3
XIV, R_1 = R_4 = C_2H_5; R_2 = R_3 = CH_3
XV, R_1 = R_2 = R_3 = CH_3; R_4 = H
XVI, R_1 = H; R_2 = R_3 = R_4 = CH_3
XVII, R_1 = R_3 = R_4 = CH_3; R_2 = H
XVIII, R_1 = R_2 = R_4 = CH_3; R_3 = H

XIX, R_1 = R_4 = H; R_2 = R_3 = CH_3; X = Cl

The second phenolic base was a new alkaloid which we have named stepholidine. Stepholidine crystallizes as a hydrate ($C_{19}H_{21}NO_4 \cdot H_2O$): mp 158–160°; $[\alpha]_D$ −311°. Its molecular weight (327) was confirmed by mass spectrometry. The composition of stepholidine and its methylation to give (−)-tetrahydropalmatine (XI) indicate that it has a structure corresponding to XI in which two of the methoxyls are replaced by hydroxyl groups. In order to determine the position of the hydroxyls, stepholidine was treated with excess diazoethane and the resulting O,O-diethylstepholidine (XIV) was oxidized with permanganate. The neutral fraction yielded 6-methoxy-7-ethoxy-1-keto-1,2,3,4-tetrahydroisoquinoline (XX). The isolation of XX served to fix the position of one methoxyl and one ethoxyl group in ring A of diethylstepholidine (XIV). The acidic fraction from the oxidation was converted into a



XX



XXI

(12) The synthesis of IV is included in the Experimental Section.

(13) K. Bernauer, *Helv. Chim. Acta*, **46**, 1783 (1963).

(14) L. J. Haynes, K. L. Stuart, D. H. R. Barton, and G. W. Kirby, *Proc. Chem. Soc.* (London), 280 (1963); see also footnote 5a in ref 13.

(15) Since 1963, seven proaporphines and eight reduced proaporphines have been isolated in the course of various phytochemical investigations. Some of this work has been summarized briefly in a recent review on the aporphine alkaloids: M. Shamma, in "The Alkaloids," Vol. IX, R. H. F. Manske, Ed., Academic Press Inc., New York, N. Y., 1967, p 1. A more complete review of the proaporphine alkaloids will appear shortly: K. L. Stuart and M. P. Cava, *Chem. Rev.*, in preparation.

(16) M. Tomita and J. Kunitomo, *Yakugaku Zasshi*, **82**, 734 (1962).

(17) An independent report of the acid-catalyzed rearrangement of I to VII appeared recently: M. Tomita and M. Kozuka, *ibid.*, **86**, 871 (1966).

(18) The structure of a new type of dienone alkaloid, androcymbine, was determined making use of our general dienone cleavage method: A. R. Battersby, R. B. Herbert, L. Pijewska, and F. Santavy, *Chem. Commun.*, 228 (1965). The cleavage of a different type of dienone base, (+)-O-methylsalutaridine, to (−)-laudanosine, has also been reported very recently by this method: D. H. R. Barton, D. S. Bhakuni, R. James, and G. W. Kirby, *J. Chem. Soc., Sect. C*, 128 (1967).

(19) The absolute configuration of a number of aporphines have been deduced on the basis of optical rotatory data: C. Djerassi, K. Mislow, and M. Shamma, *Experientia*, **18**, 53 (1962); M. Shamma, *ibid.*, **18**, 64 (1962).

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

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

(22) The melting point of our (−)-corydalmine is 174–175°, in reasonable accord with the recorded value[20] of 187.5–188.5° for synthetic (±)-corydalmine. On the other hand, natural (+)-corydalmine and (±)-corydalmine derived from it are said to melt at 238–239° and 213–215°, respectively.[21] We are unable to offer an explanation for these discrepancies.

mixture of ethylimides from which only 4-ethoxy-5-methoxy-N-ethylphthalimide (XXI), also derived from ring A of XIV, was isolated.

On the basis of the above results, stepholidine could have only structure XII or structure XIII. Since XIII represents the known alkaloid scoulerine, a direct infrared comparison of scoulerine and stepholidine was made; it showed the two bases to be distinctly different. (−)-Stepholidine is thus securely represented by structure XII. Finally, stepholidine was dehydrogenated with iodine to the corresponding dehydro derivative, isolated as the yellow chloride (XIX), mp 270–273°. This compound was identical with stepharanine chloride, a minor constituent of the quaternary alkaloid mixture of *S. glabra*,[8] the structure of which was determined in quite an independent manner.[9]

Experimental Section[23]

Preliminary Fractionation of the Alkaloids.—Commercial (Indian) *Stephania glabra* root tubers (25 kg) were ground well and extracted exhaustively with ethanol, and the concentrated ethanol extract was digested with 0.2 *M* aqueous phosphoric acid. The pH of the aqueous acid extract was increased in three stages (pH 2, 7, and 10) by addition of alkali. At each stage the alkaloids were extracted with ether and any insoluble material was then removed by filtration. Each of the six fractions thus obtained (F_a–F_f) was put individually through the entire separation procedure in an attempted further separation. The final fractions obtained are described in Table I.

TABLE I

Stephania glabra FRACTIONS

Fraction	Wt, g	Description
F_a	43.5	Ether extractable at pH 2
F_b	12.0	Ether insoluble at pH 2
F_c	117.5	Ether extractable at pH 7
F_d	234.0	Ether insoluble at pH 7
F_e	14.0	Ether extractable at pH 10
F_f	100.0	Chloroform extractable at pH 10
Total	421.0	

Unfortunately, tlc patterns indicated that each of the above fractions was still a complex mixture with considerably carryover from one fraction to the next. Since all of the compounds isolated in this investigation were found in the major fractions (F_c and F_d), further details of isolation are recorded only for these fractions.

In addition to the weights recorded in Table I, crude crystalline (−)-tetrahydropalmatine (28.5 g) was isolated during the processing of fraction F_c, and crude crystalline stepharine (59.6 g) was isolated during the processing of fraction F_e.

Examination of Fraction F_c. A. Isolation of (−)-Tetrahydropalmatine (XI).—A solution of fraction F_c (94 g) in chloroform was extracted with dilute aqueous sodium hydroxide solution to remove phenolic constituents; the remaining solution of nonphenolic base in chloroform was extracted with 5% hydrochloric acid to remove stepharine (see below). The chloroform solution was evaporated to dryness *in vacuo* to give, after crystallization, 31 g of (−)-tetrahydropalmatine hydrochloride, mp 201–203°, identical with an authentic sample. The free base (XI) was crystallized from methanol: mp 140.5–142°; $[\alpha]^{25}D$ −299° (*c* 1.0, ethanol) (lit.[24] 142°, $[\alpha]D$ −292°).

B. Isolation of Stepharine (I).—The hydrochloric acid extract (section A, above) was made basic with ammonia and was extracted with chloroform. The chloroform layer was extracted in turn with McIlvaine buffer solutions of pH 5.2, 3.0, and 2.0, in that order. Each of the buffer extracts was worked up in the usual way with ammonia and chloroform.

The extract of pH 5.2 yielded 14.7 g of crude material which was recrystallized from acetone to give stepharine, colorless crystals: mp 179–180° dec; $[\alpha]^{27}D$ +144° (*c* 1.88, chloroform); λ_{max}^{KBr} 3.10 μ (NH), 3.28, 3.44, and 3.58 (CH), and 6.01 and 6.17 (cross-conjugated dienone). Stepharine is sensitive to air and light, even in the solid state, and gradually turns brown on standing. The analytical sample was recrystallized from 2-butanone.

Anal. Calcd for $C_{18}H_{19}NO_3$: C, 72.70; H, 6.44; N, 4.71; CH_3O, 27.49. Found: C, 72.84; H, 6.56; N, 4.69; CH_3O, 27.49.

C. Isolation of Pronuciferine (II).—The extract of pH 3.0 (section B, above) gave crude material, which was purified by chromatography on grade II neutral alumina (eluents, benzene and chloroform) and recrystallized from ether to give approximately 5 g of pure pronuciferine: mp 127–129° dec; $[\alpha]^{25}D$ +86.4° (*c* 1.94, chloroform) [lit.[25] mp 127–129°; $[\alpha]^{25}D$ +99.0 ± 1.0° (*c* 0.205 chloroform)]. This material was identical with an authentic specimen of pronuciferine.

The extract of pH 2.0 was worked up similarly but failed to give crystalline material.

Transformation Products of Stepharine (I) and Pronuciferine (II). A. Pronuciferine (II).—A mixture of stepharine (2.5 g), formic acid (5.5 ml), and formaldehyde (5.5 ml of a 35% aqueous solution) was heated on a steam bath for 4 hr. The reaction mixture was diluted with water and was washed with ether; it was then made basic with ammonia and was extracted with chloroform. The chloroform extract afforded a residue which was chromatographed with benzene on grade II neutral alumina to give 1.7 g (67%) of pronuciferine [mp 128–129°, $[\alpha]^{25}D$ +87° (*c* 1.71, chloroform)] identical with naturally occurring material (section C, above).

B. (−)-Armepavine (VI).—Small portions of sodium metal and a solution of pronuciferine in toluene were added alternately and with stirring to 500 ml of liquid ammonia at −50°. Addition of each successive portion of sodium was delayed until the color of dissolved sodium from the previous addition had been discharged. In this way, 0.9 g of sodium and 1.0 g of pronuciferine (in 100 ml of toluene) were added to the reaction mixture during 3 hr. The reaction product was worked up in the usual way to give 0.75 g of crude phenolic base, which was recrystallized several times from acetone to yield 0.13 g of pure (−)-armepavine (VI), mp 138–139°. Crystallization of a mixture of equal parts of (−)- and (+)-armepavine gave (±)-armepavine, mp 158–161°, which was identical with an authentic synthetic sample.

C. Tuduranine (VII).—A solution of stepharine (0.250 g) in 6 *N* hydrochloric acid was heated under nitrogen on a steam bath, for 15 hr. The reaction mixture was cooled and filtered to give tuduranine hydrochloride: mp 280° dec; $[\alpha]^{25}D$ −125° (*c* 1.45, aqueous methanol), which was obtained in good yield and was identical with an authentic sample.

Attempts to detect the presence of the other possible product of the reaction, VIII, which is an isomer of tuduranine, were unsuccessful.

D. N-Acetylstepharine (III).—A solution of stepharine (7.3 g) and acetic anhydride (4 ml) in 100 ml of ether was stirred at 0° for 1 hr. The resulting preciptate was collected by filtration and was recrystallized from methanol to give 6.3 g (76%) of N-acetylstepharine as glistening needles: mp 235–236° dec; $[\alpha]^{24}D$ −80° (*c* 1.57, chlorom); λ_{max}^{KBr} 3.42 (CH), 6.04 and 6.07–6.2 μ (cross-conjugated dienone and N-acetyl).

Anal. Calcd for $C_{20}H_{21}NO_4$: C, 70.78; H, 6.24; N, 4.13; CH_3O, 18.33. Found: C, 70.60; H, 6.34; N, 4.10; CH_3O, 17.92.

E. N-Methyltetrahydrostepharine (X).—A solution of 0.58 g of stepharine and 0.074 g of formaldehyde in methanol was stirred at room temperature for 1.5 hr. (The formaldehyde–methanol solution was prepared by diluting 1 g of a 37% aqueous formaldehyde solution to 10 g with methanol and taking a 2-g aliquot of the dilute solution.) The reaction mixture was then hydrogenated at room temperature under atmospheric pressure in the presence of 200 mg of 10% palladium-on-charcoal catalyst. The usual work-up gave a residue which was subjected to chromatography on neutral grade I alumina with benzene. N-Methyltetrahydrostepharine (X) was obtained as colorless needles: mp 114–115°; λ_{max}^{KBr} 3.43 and 3.6 (CH), 5.84 μ (unconjugated six-membered-ring ketone).

(23) Analyses were performed by Midwest Microlab, Inc., Indianapolis Ind. Melting points are uncorrected.

(24) H. G. Boit, "Ergebnisse der Alkaloid-Chemie bis 1960," Akademie-Verlag, Berlin, 1961, p 340.

(25) K. Bernauer, *Helv. Chim. Acta.*, **47**, 2119 (1964).

Anal. Calcd for $C_{19}H_{25}NO_3$: C, 72.35; H, 7.99; N, 4.44; CH_3O, 19.68; CH_3N, 4.76. Found: C, 72.35; H, 8.17; N, 4.91; CH_3O, 19.28; CH_3N, 3.50.

Transformation Products of N-Acetylstepharine (III). A. 1-(*p*-Hydroxybenzyl)-6,7-dimethoxy-2-acetyl-1,2,3,4-tetrahydroisoquinoline (IV).—N-Acetylstepharine (1.0 g) and sodium metal (0.92 g) were added alternately in small portions to 500 ml of liquid ammonia at −50° during 1.25 hr (see section B). The reaction mixture was treated with ammonium chloride and was worked up in the usual way to give 0.95 g of crude phenolic base. Recrystallization of the latter from methanol gave 0.55 g (55%) of 1-(*p*-hydroxybenzyl)-6,7-dimethoxy-2-acetyl-1,2,3,4-tetrahydroisoquinoline (IV), mp 234–235°, $[\alpha]^{25}D$ −90° (*c* 0.485, chloroform), which was identical with authentic material (see below).

B. 1-(*p*-Methoxybenzyl)-6,7-dimethoxy-2-acetyl-1,2,3,4-tetrahydroisoquinoline (V).—A solution of 0.19 g of IV and excess diazomethane–ether azeotrope in methanol was kept at 0° for 1 week. The reaction mixture was worked up in the usual way and the resulting crude nonphenolic base was chromatographed on grade I neutral alumina with benzene to give 1-(*p*-methoxybenzyl)-6,7-dimethoxy-2-acetyl-1,2,3,4-tetrahydroisoquinoline (V), mp 111–112° (from cyclohexane–ether).

Anal. Calcd for $C_{21}H_{25}NO_4$: C, 70.96; H, 7.09; N, 3.94; CH_3O, 26.19. Found: C, 71.16; H, 7.37; N, 3.96; CH_3O, 25.97.

Synthesis of 1-(*p*-Hydroxylbenzyl)-6,7-dimethoxy-2-acetyl-1,2,3,4-tetrahydroisoquinoline (IV). A. 1-(*p*-Benzyloxybenzyl)-6,7-dimethoxy-2-acetyl-1,2,3,4-tetrahydroisoquinoline.—A solution of 1-(*p*-benzyloxybenzyl)-6,7-dimethoxy-3,4-dihydroisoquinoline hydrochloride[26] (5.0 g) in methanol (50 ml) was treated with sodium borohydride (1.2 g), added in small portions. After 30 min the reaction mixture was poured into water and extracted with ether and the ether extracts were dried over magnesium sulfate and evaporated to an oil [1-(*p*-benzyloxybenzyl)-6,7-dimethyl-1,2,3,4-tetrahydroisoquinoline].

The oil was dissolved in pyridine (10 ml), acetic anhydride (10 ml) was added, and the resulting reaction mixture was warmed on the steam bath for 15 min. After the usual work-up with water, ether, aqueous base, and aqueous acid solutions, 1-(*p*-benzyloxybenzyl)-6,7-dimethoxy-2-acetyl-1,2,3,4-tetrahydroisoquinoline was obtained as colorless microcrystals, mp 119–122° (from aqueous methanol), 4.0 g (79%). The analytical sample, mp 136–138°, was obtained by two additional recrystallizations.

Anal. Calcd for $C_{27}H_{29}NO_4$: C, 75.15; H, 6.77; N, 3.25. Found: C, 74.84; H, 6.50; N, 3.06.

B. 1-(*p*-Hydroxybenzyl-6,7-dimethoxy-2-acetyl-1,2,3,4-tetrahydroisoquinoline.—A solution of 1-(*p*-benzyloxybenzyl)-6,7-dimethoxy-2-acetyl-1,2,3,4-tetrahydroisoquinoline (3.0 g) in 170 ml of methanol was hydrogenated for 16 hr in the presence of 0.78 g of 5% palladium on charcoal under 40-psi initial hydrogen pressure. A colorless oil was obtained which crystallized from ethanol as colorless plates (1.7 g, 72%), mp 215–216°.

Anal. Calcd for $C_{20}H_{23}NO_4$: C, 59.59; H, 5.28; N, 7.72. Found: C, 59.60; H, 5.35; N, 7.66.

Examination of Fraction F_d. A. Isolation of (−)-Corydalmine (XV).—Fraction F_d (210 g) was extracted with chloroform in a Soxhlet apparatus. The chloroform-insoluble material (120 g) was discarded. The chloroform-soluble material (90 g) was separated into a phenolic fraction and a nonphenolic fraction and the latter was worked up with hydrochloric acid to give an additional 80 g of tetrahydropalmatine hydrochloride and small amounts of stepharine and N-methylstepharine.

The phenolic fraction (an aqueous alkaline solution) was adjusted to pH 7 with ammonium chloride and was extracted with chloroform to give a mixture of crude phenolic bases (19.7 g) which was reextracted with 1:2 acetone–chloroform. The material soluble in acetone–chloroform (11.2 g) was chromatographed on grade III neutral alumina with the same solvent mixture to give 0.5 g of (−)-corydalmine (XV): mp 173–174° dec from methanol; $[\alpha]^{30}D$ −310° (*c* 1.385, ethanol); λ_{max}^{EtOH} (neutral) 214 μ (log ε 4.43), 276 (3.78); λ_{max}^{EtOH} (acid) 212 mμ (log ε 4.40), 231 (4.20), 274 (3.82); λ_{max}^{EtOH} (base) 217 mμ (log ε 4.47), 278–284 (3.85). The infrared spectrum of (−)-corydalmine ($\lambda_{max}^{CHCl_3}$ 2.79, 3.31, 3.39, 3.52, 6.21 μ) was identical with that of an authentic sample of synthetic (±)-corydalmine. Treatment of (−)-corydalmine with diazomethane gave (−)-tetrahydropalmatine: mp 141°; $[\alpha]^{28}D$ −271° (*c* 1.63, ethanol).

B. Isolation of Stepholidine (XI).—The material not soluble in 1:2 acetone–chloroform (8.5 g) (see section A) yielded a substance crystallizing directly from acetone in fine needles, mp 126–138° (*in vacuo*), which was designated stepholidine: $[\alpha]^{18}D$ −311° (*c* 0.523, ethanol); λ_{max}^{EtOH} (neutral) 214 mμ (log ε 4.40), 287 (3.79); λ_{max}^{EtOH} (base) 219 mμ (log ε 4.46), 243–248 (4.09), 297–298 (3.82) (no ultraviolet absorption shift in acidic medium); λ_{max}^{KBr} 2.80 (OH), 3.08 (OH), 3.44 and 3.60 (CH), 6.18, and 6.28 μ.

The infrared spectrum of stepholidine differs in many respects from that of 1-scoulerine, mp 193–195° (lit.[27] mp 195° or 204°).

The analytical sample was prepared by thin layer chromatography on silica gel with 1:3 methanol–chloroform (to remove traces of corydalmine), mp 158–160° dec.

Anal. Calcd for $C_{19}H_{21}NO_4 \cdot H_2O$: C, 66.07; H, 6.71; N, 4.06. Found: C, 66.08; H, 6.81; N, 4.06. Calcd for $C_{19}H_{21}NO_4$: mol wt, 327.4. Found: mol wt, 327 (mass spectrum).

Transformation Products of Stepholidine (XII). A. 2,10-Dihydroxy-3,9-dimethoxy-5,6-dihydrobenz[*a*]acridinium Chloride (XIX).—A solution of stepholidine (0.33 g, 1 mmol) and iodine (0.76 g, 3 mmol) in 50 ml of ethanol was refluxed on a steam bath for 1 hr. On treatment with a dilute aqueous solution of sodium bisulfite, the reaction mixture deposited 0.2 g of a yellow precipitate of the dihydrobenz[*a*]acridinium iodide. The precipitate was dissolved in the minimum volume of 1:1 acetone–water and the resulting solution was passed through a column of IRA-410 ion exchange resin (chloride form). The product was recrystallized from 0.1% hydrochloric acid to give 2,10-dihydroxy-3,9-dimethoxy-5,6-dihydrobenz[*a*]acridinium chloride (XIX) as yellow needles, mp 270–273° dec, identical in all respects with a sample of natural stepharanine chloride.[8]

B. (−)-Tetrahydropalmatine (XI).—A solution of (−)-stepholidine (XII) and excess diazomethane–ether azeotrope in methanol was kept at 0° for several days to give (−)-tetrahydropalmatine (XI): mp 141°, $[\alpha]^{25}D$ −258° (*c* 1.6, ethanol).

C. (±)-Stepholidine.—An aqueous solution of 0.08 g of XIX (section A) was treated with sodium borohydride (0.05 g) at room tempeature. The resulting precipitate was recrystallized from methanol to give colorless needles of (±)-stepholidine, mp 126–130° dec, the infrared spectrum of which was superimposable on that of (−)-stepholidine (XII) (chloroform solution spectra).

D. O,O-Diethylstepholidine (XIV).—A solution of (−)-stepholidine (XII) (0.950 g) and excess diazoethane ether azeotrope in methanol (25 ml) was kept at 0° for several days to give O,O-diethylstepholidine (XIV). The crude product was oxidized directly without further purification or characterization (see below).

Degradation Products from O,O-Diethylstepholidine (XIV). A. 6-Methoxy-7-ethoxy-1-keto-1,2,3,4-tetrahydroisoquinoline (XX).—A solution of 0.58 g of O,O-diethylstepholidine (XIV) in 1.5% sulfuric acid was adjusted to pH 7 with an aqueous sodium carbonate solution and was treated dropwise at room temperature with a solution of 1.04 g of potassium permanganate in 45 ml of water. The reaction mixture was heated at 60–70° for 10 min. It was cooled, made basic with potassium hydroxide, and extracted with chloroform. The chloroform extract was worked up in the usual way to give approximately 0.1 g of O,O-diethylstepholidine and a brown oil which was chromatographed with benzene on neutral grade IV alumina to give 0.02 g of 6-methoxy-7-ethoxy-1-keto-1,2,3,4-tetrahydroisoquinoline (XX), mp 189–190°, identical in all respects with authentic material.

B. 4-Ethoxy-5-methoxy-N-ethylphthalimide (XXI).—The basic aqueous layer from the degradation of O,O-diethylstepholidine (see section A, above) was worked up in the usual way to give an acidic residue (0.08 g), which was then treated with acetyl chloride (0.3 ml) in benzene (3 ml). After removal of excess acetyl chloride and solvent, the presumed diacid anhydride was converted into the corresponding half amide by treatment with ethylamine in benzene–ether. The crude half amide was heated with acetic anhydride (1 ml) and sodium acetate (0.1 g) on a steam bath for 0.5 hr to give the crude imide XXI, obtained in the form of a brown gummy residue. The residue was chromatographed with 1:1 benzene–petroleum ether (bp 30–60°) on grade III neutral alumina to give two 3-ml eluate fractions containing only noncrystalline material, followed by two 3-ml fractions containing 4 mg of 4-ethoxy-5-methoxy-N-ethylphthalimide (XXI), mp 190–194°, identical with authentic material.

(26) M. Tomita and H. Yamaguchi, *Pharm. Bull.* (Tokyo), **1**, 10 (1953).

(27) See ref 24, p 334.

Registry No.—I, 2810-21-1; 1-(*p*-hydroxybenzyl)-6,7-dimethoxy-2-acetyl-1,2,3,4-tetrahydroisoquinoline, 16562-16-6; III, 4880-87-9; IV, 16562-06-4; V, 16562-07-5; VI; 524-20-9; VII, 517-97-5; X, 16562-08-6; XV, 16562-09-7; XIX, 13509-87-0; XX, 16562-11-1; (−) XXI, 16562-12-2; (−) XII, 16562-13-3; (±) XII, 16562-14-4; 1-(*p*-benzyloxybenzyl)-6,7-dimethoxy-2-acetyl-1,2,3,4-tetrahydroisoquinoline, 14347-98-9.

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Steroids. LXXIX. Synthesis and Reactions of Oxiranes Obtained from 3- and 17-Keto Steroids[1]

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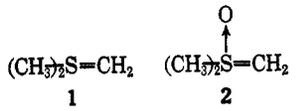
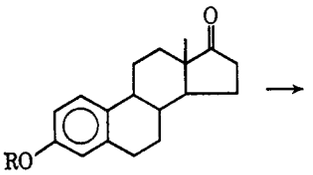
The synthesis of oxiranes from 3- and 17-keto steroids is described. Dimethylsulfonium methylide (1) reacted with estrone in a highly stereoselective manner by α-side addition of CH_2. α-Side addition also predominated in the reaction of 1 with 3-keto-5α steroids (although an earlier claim of stereospecificity has to be modified to stereoselectivity), whereas dimethyloxosulfonium methylide (2) gave products resulting from β-side attack on the 3-keto group. Stereochemistry of the products was demonstrated by chemical conversions. The oxiranes reacted with amines to yield amino alcohols. Spiro-17β-oxiranylestra-1,3,5(10)-trien-3-ol (4a) reacted with sodium cyanide at steam-bath temperature to yield 17α-cyanomethyl-3,17β-estradiol (5e), but under more strenuous conditions the product of this reaction was estrone.

The report by Corey and Chaykovsky[2] that dimethylsulfonium methylide (1) and dimethyloxosulfonium methylide (2) react with ketones and aldehydes to yield oxiranes opened a convenient route to a number of new steroid derivatives by reaction with readily available steroidal ketones. We report here on the preparation and properties of some new steroid oxiranes formed in this manner.[3]

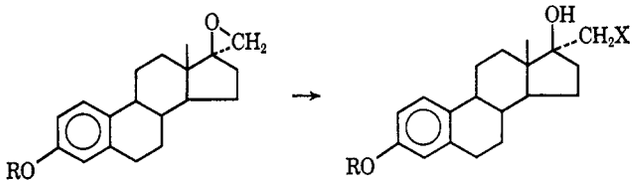


1 2

Estrone (**3a**) reacted smoothly with the sulfonium ylide 1 to yield a single oxirane (**4a**), identified as such by an infrared band at 3040 cm^{-1} and an AB quartet in the nmr spectrum.[4] The stereochemistry at C-17 was demonstrated by reduction with lithium aluminum hydride to the known 17α-methyl-3,17β-estradiol (**5a**).[5] The melting point of the crude diol was not depressed by admixture with authentic **5a** prepared from estrone and methyllithium.



3a, R = H
b, R = CH_3



4a, R = H
b, R = CH_3

5a, R = X = H
b, R = CH_3; X = H
c, R = H; X = $-NC_5H_{10}$
d, R = H; X = $-NHCH_2C_6H_5$
e, R = H; X = −CN

The nmr peak for the 18-CH_3 of oxirane **4a** was at 56 cps while that of the diol **5a** was at 54 cps. The observation that the 18-CH_3 is only slightly less shielded in the oxirane **4a** than in the alcohol **5a** is consistent with the results of Bertin and Nedelec in the androstane series. They reported the 18-CH_3 to shift upfield by δ 0.03 (*ca.* 2 cps) in going from 17β-oxirane to 17α-methyl-17β-ol. On the other hand, there was an upfield shift of δ 0.12 (7 cps) on going from a 17α-oxirane to a 17β-methyl-17α-ol.[3b]

While the conversion in high yield into essentially pure 17α-methyl-3,17β-estradiol was good evidence for stereospecific formation of the 17β-oxirane, the melting range of **4a** was wide enough to suggest a possibility of the presence of the C-17 epimer. When the reaction was repeated using the 3-methyl ether of estrone (**3b**) as substrate, oxirane **4b** (18-methyl resonance at 55.5 cps) and alcohol **5b** (18-methyl resonance at 54 cps) were obtained in high yield by the same sequence

(1) (a) For part LXXVIII, see D. Rosenthal, C. F. Lefler, and M. E. Wall, *Tetrahedron*, **23**, 3583 (1967). (b) This research was carried out under Contract SA-43-ph-4351 of the Cancer Chemotherapy National Service Center, National Cancer Institute, National Institutes of Health. (c) Abstracted in part from work done by R. C. Corley in partial fulfillment of the requirements for the Ph.D. degree at North Carolina State University at Raleigh. (d) Portions of this work were presented at the 149th National Meeting of the American Chemical Society, Detroit, Mich., April 1965, Abstracts 92P. See also C. E. Cook, R. C. Corley, and M. E. Wall, *Tetrahedron Lett.*, 861 (1965).

(2) (a) E. J. Corey and M. Chaykovsky, *J. Amer. Chem. Soc.*, **84**, 3782 (1962); (b) E. J. Corey and M. Chaykovsky, *ibid.*, **87**, 1353 (1965).

(3) For results of other workers in this area, see, *inter alia*, (a) G. Drefahl, K. Ponsold, and H. Schick, *Ber.*, **97**, 3529 (1964); (b) D. Bertin and L. Nedelec, *Bull. Soc. Chim. Fr.*, 2140 (1964); (c) H. G. Lehmann, O. Engelfried, and R. Wiechert, *J. Med. Chem.*, **8**, 383 (1965).

(4) Unless otherwise noted, all nmr spectra were obtained at 60 Mc in deuteriochloroform with tetramethylsilane as internal standard.

(5) (a) E. Haack, G. Stoek, and H. Voigt, *Naturwissenschaften*, **41**, 429 (1954); (b) L. F. Fieser, *Experientia*, **6**, 312 (1950).