ABSOLUTE CONFIGURATION
OF THE ALKALOID BREVICOLLINE*

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Successive oxidation of brevicolline (*I*) with potassium ferricyanide and chromium trioxide afforded (−)-hygrinic acid (*IV*), identical with the specimen obtained on methylation of L-(−)-proline. The alkaloid *I* was ascribed the (*S*)-(−)-configuration. The optical rotatory dispersion of brevicolline has been briefly discussed with respect to that of nicotine and anabasine.

The structure of the alkaloid brevicolline (*I*), isolated[1] from *Carex brevicollis* D. C., was determined by Lazurevskij and coworkers[2−4] on the basis of chemical (oxidative degradation) and spectral (NMR and mass spectra) evidence. In the present paper, we wish to report the assignment of its absolute configuration.

The attempted ozonolytic cleavage failed both with the base *I* and the corresponding bismethiodide in accordance with the reported results[5]. Another route which had proved useful in determinations of the absolute configuration of nicotine[6] and N-methylanabasine[7], has been therefore applied in the present case. The alkaloid *I* was converted to the methiodide *II*, the structure of which was unequivocally determined by NMR spectra (Table I). The melting point values of our methiodide *II* differs from that of a compound reported in the literature[8] as brevicolline methiodide. On the other hand, the melting point value of our bismethiodide and the reported bismethiodide[8] are identical. The potassium ferricyanide oxidation of the monomethiodide *II* afforded a non-crystalline product which was on chromatography more polar than the parent quaternary salt *II*. In spite of the close resemblance of main spectral characteristics of both compounds, the detailed differences may be used in structural determinations. Infrared spectra (Table II) indicate the absence of a proton at the indole nitrogen atom (absence of the otherwise[9] distinct stretching vibrations of the

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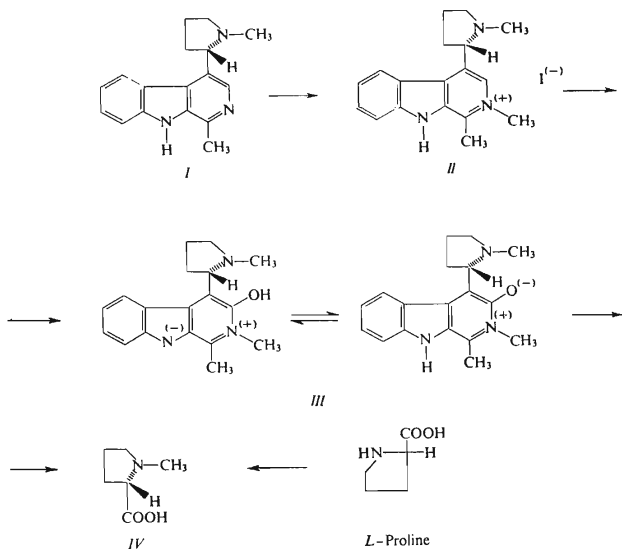
TABLE I

Nuclear Magnetic Resonance Spectra

The measurements were performed in trifluoroacetic acid on a 60 MHz apparatus (Jena, German Democratic Republic); hexamethyldisiloxane as internal standard; chemical shifts are given in δ (p.p.m.) after recalculation to tetramethylsilane.

Compound	⎧CH⎫ / N$^{(+)}$	N$^{(+)}$ / CH$_3$	⎧C⎫ / CH$_3$	⎧N—CH$_3$⎫
Brevicolline[a]	8·78 d	—	3·23 s	3·17 d
Brevicolline[b]	8·45 s	—	2·27 s	2·25 s
Methiodide II	9·15 s	4·50 s	3·19 s	3·08 d
Bismethiodide	8·93 s	4·57 s	3·34 s	3·24 s[c]
Oxidation product III[b]	—	4·34 s	3·35 s	2·43 s

[a] On a Varian 100 MHz apparatus; NH in the indole ring, 10·84 (broad singlet); N$^{(+)}$H of the protonised N$_{(2)}$ in the pyridine ring, 13·41; N$^{(+)}$H in the pyrrolidine ring, 8·76; [b] in deuterio-chloroform; [c] a double height peak in respect to peaks of other methyl groups.

TABLE II
Infrared Spectra (in cm^{-1})

Compound (technique)	δ(C—H)[a] out-of-plane	δ(C—H) in plane	ν(C—N) skeletal
Methiodide *II* (nujol)	740 m, 756 s, 787 w	1 048 m, 1 115 m, 1 206 m	1 346 s
Oxidation product *III* (chloroform)	—	1 050 m, 1 115 m, 1 160 m,	1 342 s
Oxidation product *III* (KBr)	738 s, 750 s, 780 w	1 040 m, 1 110 m, 1 150 m, 1 205 m	1 335 s

[a] Four adjacent hydrogen atoms.

N—H bond at 3 460 cm^{-1}). The additional band at 3 060 cm^{-1} may be ascribed to stretching vibrations of the OH group in a strong hydrogen bonding. It was not possible to identify the bending vibrations of the O—H bond by the presence of a distinct band because of the known[10] association with skeletal vibrations; the bending vibrations, however, are assumed to cause the considerably increased background in the whole 1 300 — 1 400 cm^{-1} region. The hydroxylic function is probably localized at position 3 of the carboline system. This assumption is not at variance with NMR spectra of the oxidation product *III* (see Table I) which lack the peak at the position corresponding to the proton at $C_{(3)}$ as well as the peak due to the indole nitrogen proton. The shift of signals due to the methyl group at the carbon atom to higher δ (p.p.m.) values (3·35 in deuteriochloroform) in comparison with that of alkaloid *I* (2·77 in deuteriochloroform) might be explained by the presence of the quaternary nitrogen atom. The dipolar lactim structure *III* is also in accordance with the close resemblance of the ultraviolet spectra (Table III) of alstonine[11] (*V*) and the oxidation
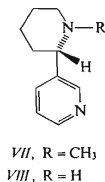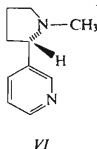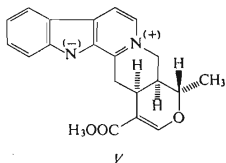

V

VI

VII, R = CH₃
VIII, R = H

TABLE II
(*Continued*)

ν (ring)	ν(OH) bonded	ν(N—H)	
1 501 m, 1 532 m, 1 560 w 1 575 w, 1 620 sh m, 1 635 s 1 680 w	—	3 150 s	3 395 m 3 460 m
1 505 m, 1 533 m, 1 565 sh w, 1 580 sh w, 1 600 sh w 1 620 sh w, 1 633 s, 1 660 sh m	3 060 m	3 150 sh w,	—
1 450 m, 1 495 m, 1 527 m, 1 555 w, 1 570 w, 1 618 m, 1 628 s, 1 650 w	3 070 s	3 150 sh w	—

product *III*. The alternative lactam structure is highly improbable in respect of the character of infrared spectra in the $1\,600 - 1\,700\ \text{cm}^{-1}$ region of compounds $I - III$ (*cf.*[12]).

Compound *III* was oxidized with chromium trioxide in aqueous sulfuric acid and the reaction mixture processed with the use of ion exchange resins to afford an amino acid, identical on paper chromatography as well as silica gel thin-layer chromatography with $(-)$-hygrinic acid (*IV*), obtained by methylation of L-$(-)$-proline[13,14]. The structural identity of both compounds is indicated also by gas-chromatographic analysis of the corresponding methyl esters[15] and by mass spectroscopy. As shown by optical rotatory dispersion curves of the free acid both in methanol and 1M-HCl, the isolated acid — though not quite optically pure — belongs to the L-series of amino

TABLE III

Ultraviolet Spectra in Methanol

Compound	λ_{max}, nm (log ε)				
Brevicolline (*I*)	243 (4·49)	288 (4·49)	—	336 (3·72)	351 (3·75)
Monomethiodide *II*	253 (4·49)	—	305 (4·20)	336 (3·15)[a]	374 (3·77)
Oxidation product *III*	253 (4·36)	—	305 (4·07)	338 (3·20)[a]	375 (3·67)
Alstonine[b] (*V*)	252 (4·54)	—	309 (4·36)	336 (3·39)	369 (3·60)

[a] After resolution of bands; [b] ref.[11].

acids and the alkaloid *I* is of the (*S*)-configuration. The same configuration was found with (−)-nicotine[6] (*VI*) and (−)-N-methylanabasine[7] (*VII*).

The identical configuration of all three alkaloids, (−)-*I*, (−)-*VI*, and (−)-*VII* should reflect on their chiroptical properties. The total chirality of the molecule of the alkaloids mentioned depends on the relative position of the single occurring chiral atom in the pyrrolidine or piperidine ring, 1: in respect to the planar heteroaromatic ring. The chiral atom is attached to the latter ring by a single bond and there is no obvious reason why the fundamental conformational features should differ. The nature of optical rotatory dispersion curves at longer wavelengths should be to the highest degree influenced by transitions of the heteroaromatic electrons into the first antibonding orbital; their polarisation should be the same in all cases because of the structural similarity (pyridine ring substituted at position 3 is always involved). Consequently, it might be expected that the sign of the Cotton effect at the longest wavelengths would reflect in this case the absolute configuration at the chiral center which causes chiral perturbation of the corresponding electronic transitions. In accordance with this expectation, the first Cotton effect of (*S*)-(−)-nicotine[16] (*VI*), and (*S*)-(−)-anabasine[16] (*VIII*) on the one hand and of (*S*)-(−)-brevicolline (*I*) on the other is always negative.

EXPERIMENTAL

Melting points (uncorrected) were taken on a heated microscope stage (Kofler block). Infrared spectra (Zeiss Jena UR 10 apparatus), ultraviolet spectra (Zeiss Jena apparatus, sodium chloride prism), mass spectra (MCh 1303 apparatus, Soviet Union), proton magnetic resonance (60 MHz apparatus, Jena), and optical rotatory dispersion (Jasco UV/ORD-5 apparatus) were recorded on the apparatus given in parentheses unless stated otherwise. Descending chromatography was performed on paper Whatman No 4 in the solvent systems S_1, 1 : 1 chloroform–benzene saturated with 10 vol. % of a 15% ethanolic solution of formamide containing 5% of ammonium formate, and S_2, 100 : 20 : 39 1-butanol–hydrochloric acid–water (spots were detected with the Dragendorff reagent). Thin-layer chromatography was performed on silica gel (Kieselgel G, Merck) in the solvent systems S_3, 85 : 7·5 : 7·5 2-butanol–25% aqueous ammonia–water, S_4, 2-butanol–90% formic acid–water 75 : 13·5 : 11·5 and S_5, 1-butanol–pyridine–water–acetic acid 30 : 20 : 12 : 6 (detection with iodine).

Brevicolline Methiodide (*II*)

Methyl iodide (0·4 ml) was added at room temperature to a solution of brevicolline (*I*; 400 mg; m.p. 224—226°C; R_F 0·75 in S_1) in acetone (30 ml) and the resulting mixture kept at room temperature for 15 hours to deposit crystals which were collected with suction, washed with acetone, and dried at 0·1 Torr. Yield, 460 mg of the methiodide *II*, m.p. 274—276°C (decomposition); R_F value 0·5 in the solvent system S_1. The attempted recrystallisation from methanol or ethanol is accompanied by decomposition. For NMR (Table I), infrared (Table II), and ultraviolet (Table III) spectra see the tables stated. For $C_{18}H_{22}IN_3$ (407·3) calculated: 53·08% C, 5·44% H, 31·16% I, 10·32% N; found: 52·73% C, 5·57% H, 33·00% I, 10·09% N. The literature[8] reports m.p. 210 to 212°C for the monomethiodide.

Potassium Ferricyanide Oxidation of the Methiodide *II*

A suspension of the methiodide *II* (447·0 mg) in water (9 ml) was treated at room temperature under stirring with a solution of potassium ferricyanide (1·3 g) in water (6 ml). The resulting

mixture was stirred for 3 days, saturated with solid potassium carbonate, allowed to stand at room temperature for 24 hours, diluted with water (80 ml), and filtered. The insoluble material on the filter was washed with chloroform and the filtrate was extracted repeatedly with chloroform. The washings and extracts were combined, dried over magnesium sulfate, and evaporated under diminished pressure. The glassy residue of compound *III* (254·7 mg; R_F 0·45 in S_1) was used for spectral measurements (see Tables I—III) and for the subsequent chromium trioxide oxidation.

Picrate. Saturated ethereal solution of picric acid was added to a solution of compound *III* (5 mg) in ethanol (0·5 ml), the picrate collected with suction, and washed with ether. M.p. 153 to 156°C (decomposition). For the C_2H_5OH solvate of the dipicrate, $C_{32}H_{33}N_9O_{16}$ (799·6), calculated: 48·06% C, 4·16% H, 15·76% N; found: 48·71% C, 4·06% H, 15·30% N.

Chromium Trioxide Oxidation of Compound *III*

A solution of compound *III* (164·4 mg) in water (2·9 ml) was treated under stirring with a solution containing chromium trioxide (820 mg), water (4·9 ml), and concentrated sulfuric acid (1·15 ml), and the whole mixture was refluxed for 5 hours. On cooling, the excess chromium trioxide was destroyed with sulfur dioxide and the mixture was precipitated with saturated aqueous barium hydroxide. The voluminous precipitate was filtered off and washed with water. The aqueous filtrates were evaporated to dryness under diminished pressure to afford 98·7 mg of an oil. The residue (75·0 mg) was dissolved in 1M-HCl (3 ml), the solution applied to a column of Zerolite 225 SRC 13 ion exchange resin, and the column washed with water (150 ml) until the eluate was free of any residue on evaporation (total yield, 39 mg of a Dragendorff-negative substance). The subsequent elution of the column with 1M-NH$_4$OH (100 ml) and evaporation of the eluate under diminished pressure afforded 35 mg of an oil. This oil was dissolved in a little water and the solution applied to a column of Zerolite G SRA 96 (OH$^-$) ion exchange resin. The column was washed with water (150 ml) and then 1M-CH$_3$COOH (150 ml). The acetic acid eluate was treated with a few drops of 1M-HCl and evaporated to dryness under diminished pressure. As shown by paper chromatography, the residue contained a single Dragendorff-positive substance the R_F value of which was identical with that of authentic (−)-hygrinic acid (R_F 0·45 in S_2). The semisolid residue was extracted with a little ethanol. The insoluble portion was identified as ammonium chloride. Concentration of the ethanolic extract afforded another crop of ammonium chloride which was filtered off. Evaporation of this filtrate afforded 23 mg of an oil which failed to crystallize. A sample of this oil was used for measurement of the optical rotatory dispersion curve (*vide infra*). Thin-layer chromatography, R_F values: 0·10 in S_3, 0·04 in S_4, and 0·22 in the solvent system S_5, all in accordance with R_F values of authentic (−)-hygrinic acid[12].

The residue (15 mg) was dissolved in methanol (0·5 ml), the solution saturated with gaseous hydrogen chloride, and allowed to stand at room temperature overnight. The solution was evaporated under diminished pressure, the residue dissolved in chloroform, and made alkaline with a solution of ammonia in chloroform. The ammonium chloride was filtered off, the filtrate evaporated under diminished pressure, and the residue (6 mg) distilled in a microdistillation apparatus at 15 Torr/100°C (bath temperature) to afford 4 mg of a liquid. The distillate was homogeneous and identical with authentic methyl L-(−)-hygrinate on gas chromatography (Perkin Elmer Model F-11 apparatus, flame-ionisation detection, 4 meter column, inner diameter 2·2 mm, packed with Chromosorb W (80—100 mesh) containing 1·5% of FS 1265, hexamethyldisiloxane as internal standard, nitrogen as carrier gas) as well as on mass spectrometry: molecular peak, 143; base peak, m/e 84 (M — COOCH$_3$); m/e 82 (M — COOCH$_3$ — 2 H); and m/e 42. Both samples exhibited a negative optical rotatory dispersion curve in the 400—225 nm region (a trough at 255 nm). Due to the lack of material the quantitative evaluation of the curve of the degradation product *IV* was not performed.

Methyl L-(—)-*hygrinate* for comparison was prepared by esterification of the acid *IV* with methanol and hydrogen chloride; b.p. 80°C/18 Torr (*cf.* ref.[15] for optically inactive compound). For $C_7H_{13}NO_2$ (143·2) calculated: 58·72% C, 9·15% H, 9·78% N; found: 58·94% C, 9·18% H, 9·93% N.

Spectropolarimetric Measurements

(—)-*Nicotine*, *c* 0·11, methanol, λ (nm), [Φ]: 300, —1100°; sh 286, —2000°; min 275, —3550°; sh 267, —900°; 264, 0°; sh 261, +2000°; max 255, +3250°; 244, 0°; end value 230, —3500°.

(—)-*Brevicolline* (*c* 0·07, methanol) [Φ]: 400, —1620°; 380, —2750°; min 360, —4160°; 350, —2900°; max 330, —760°; 300, —1730°; min 291, —4160°; 275, —1100°; 262, 0°.

L-(—)-*Hygrinic acid*, (authentic specimen), *c* 0·15, methanol: $[\alpha]_{325}$, —300°; min $[\alpha]_{240}$ —600°; max $[\alpha]_{229}$ —570°; $[\alpha]_{220}$ —760°; (*c* 0·1, 1M-HCl): $[\alpha]_{300}$, —300°; min $[\alpha]_{260}$, —380°; $[\alpha]_{245}$, —350°; max $[\alpha]_{228}$, +20°; $[\alpha]_{215}$, —950°; min $[\alpha]_{207}$, —2700°.

L-(—)-*Hygrinic acid* (oxidative degradation product of brevicolline), *c* 0·37, methanol: $[\alpha]_{325}$, —215°; min $[\alpha]_{240}$, —400°; max $[\alpha]_{230}$, —390°; $[\alpha]_{220}$, —560°; (*c* 0·1,1M-HCl): min $[\alpha]_{260}$, —180°; $[\alpha]_{245}$, —145°; max $[\alpha]_{228}$, +10°; $[\alpha]_{215}$, —500°; min $[\alpha]_{208}$, —1600°.

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