groups of the activation peptide have been amidated with glycine ethyl ester and suggest that calcium binding and carboxylate modification can be functionally equated in the activation process.

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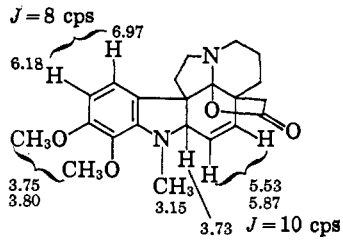
Haplophytine

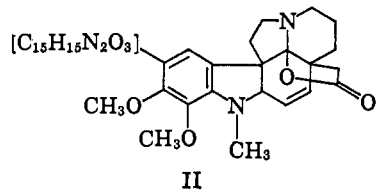
Sir:

Haplophytine[1] was first isolated by Snyder and his co-workers, who carried out an extensive investigation of its chemistry.[2] It is obtained as the principal alkaloid from *Haplophyton cimicidum* (Apocynaceae).[2-4]

High-resolution mass spectrometry[5] required the revision of its formula to $C_{37}H_{40}N_4O_7$ (calcd mol wt, 652.28990; found, *m/e* 652.29148).[6] Acid cleavage gave a crystalline compound (76%), $C_{22}H_{26}N_2O_4$ (calcd mol wt, 382; found: *m/e* 382), mp 201–203°, assigned structure I on the basis of its nmr spectrum (see I for δ values) and the following data. Its infrared spectrum ($CHCl_3$) shows a strong C=O band at 5.73 μ (*cf.* cimicine,[4] cimicidine,[4] and dichotamine[7]),



I



II

but no OH or NH bands. Its ultraviolet spectrum [λ_{max}^{EtOH} 222 mμ (ε 28,700), 256 (5900), and 304 (2400)] is in accord with an indoline chromophore. Its mass spectrum shows a strong peak at *m/e* M − 44, characteristic of lactonic alkaloids of the aspidoalbine skeletal type.[7,8] Hydrogenation of the cleavage product gave a zwitterionic tetrahydro derivative, mp 265° dec, formed by saturation of the ethylenic double bond accompanied by hydrogenolysis of the lactone (*cf.* dihydrocimicine and dihydrocimicidine[4]). Esterification with diazomethane gave a glass, $C_{23}H_{32}N_2O_4$, whose mass spectrum showed the molecular ion peak *m/e* 400 (base peak) and a single prominent fragment ion at *m/e* 168, a pattern very characteristic of nonlactonic alkaloids of the aspidospermine type.[9] Reduction of the cleavage product with sodium borohydride gave a zwitterionic product the spectra of whose methyl ester demonstrated that reductive cleavage of the lactone alone had occurred. The mass spectrum of this ester showed a molecular ion at *m/e* 398, confirming the extent of reduction; the absence of significant fragment ions at *m/e* 166 and 179 excluded a formulation in which the ethylenic double bond of I is placed at C-6–7.[9] The position assigned to the double bond is also in better accord with the chemical shift of the C-2 proton and the fine splitting of the olefinic proton signals in the nmr spectrum of I.

The many common features of the spectra of I and those of haplophytine [λ_{max}^{EtOH} 220 mμ (ε 48,500), 265 (14,300), and 305 (4500); $\lambda_{max}^{CHCl_3}$ 5.72 and 6.05 μ; δ^{CDCl_3} (ppm) 2.40 (~3 H), 3.00 (3 H), 3.17 (3 H), 3.65 (3 H), 3.72 (1 H), 5.55 (1 H, doublet, J = 10 cps), 5.85 (1 H, doublet, J = 10 cps), 6.27 (1 H, doublet of doublets, J = 7 and 2.5 cps), 6.9–7.2 (3 H, multiplet), 9.04 (1 H, absent after D_2O wash)] showed that the structure of haplophytine is related to that of I by replacement of a hydrogen atom in the latter by a moiety $C_{15}H_{15}N_2O_3$. That the linkage is at C-15 as in II is indicated by the absence of a doublet (J = 8 cps) in the nmr spectrum of haplophytine corresponding to the C-15 proton signal in the spectrum of I. Catalytic hydrogenation of haplophytine gave tetrahydrohaplophytine, $C_{37}H_{44}N_4O_7$, accompanied by spectral changes analogous to those occurring when I was converted to tetrahydro-I; acid cleavage of the methyl ester of O-methyltetrahydrohaplophytine, $C_{39}H_{48}N_4O_7$, gave tetrahydro-I.

The intensities of the ultraviolet maxima of haplophytine indicated that the $C_{15}H_{15}N_2O_3$ moiety also possesses an indoline system. This can be expanded to a 7-hydroxy-1-acylindoline system on the basis of the relationship between the infrared and nmr spectra of haplophytine and the spectra of its O-substituted derivatives, as earlier deduced by Snyder.[2,10] The nmr signal at 6.27 ppm, which can be assigned to an aromatic proton coupled with *ortho* and *meta* protons, requires that the 7-hydroxy-1-acylindoline system be unsubstituted at the C-4, -5, and -6 positions. The signal at 2.40 ppm indicated the presence of an aliphatic N–CH_3 in the $C_{15}H_{15}N_2O_3$ moiety, while the infrared spectrum showed that the third oxygen atom can only be present in an ether linkage.

(1) Much of the earlier work on haplophytine has been summarized by J. E. Saxton, *Alkaloids*, **8**, 673 (1965).

(2) E. F. Rogers, H. R. Snyder, and R. F. Fischer, *J. Am. Chem. Soc.*, **74**, 1987 (1952); H. R. Snyder, R. F. Fischer, J. F. Walker, H. E. Els, and G. A. Nussberger, *ibid.*, **76**, 2819, 4601 (1954); H. R. Snyder, H. F. Strohmayer, and R. A. Mooney, *ibid.*, **80**, 3708 (1958).

(3) M. P. Cava, S. K. Talapatra, K. Nomura, J. A. Weisbach, B. Douglas, and E. C. Shoop, *Chem. Ind.* (London), 1242 (1963).

(4) M. P. Cava, S. K. Talapatra, P. Yates, M. Rosenberger, A. G. Szabo, B. Douglas, R. F. Raffauf, E. C. Shoop, and J. A. Weisbach, *ibid.*, 1875 (1963).

(5) We thank Mr. L. Weiler, Harvard University, for the mass spectral data.

(6) Satisfactory elemental analytical data have been obtained in this or earlier investigations[2] for all compounds whose molecular formulas are given.

(7) K. S. Brown, Jr., H. Budzikiewicz, and C. Djerassi, *Tetrahedron Letters*, 1731 (1963).

(8) K. S. Brown, Jr., W. E. Sanchez L., A. de A. Figueiredo, and J. M. Ferreira Filho, *J. Am. Chem. Soc.*, **88**, 4984 (1966).

(9) H. Budzikiewicz, C. Djerassi, and D. H. Williams, "Structure Elucidation of Natural Products by Mass Spectrometry," Vol. 1, Holden-Day, Inc., San Francisco, Calif., 1964, Chapter 7.

(10) The presence of this second aromatic ring is considered to account for the unusual shielding of the protons of one of the methoxyl groups in haplophytine; in I, where this second ring is absent, the chemical shifts of the protons of both methoxyl groups are normal.

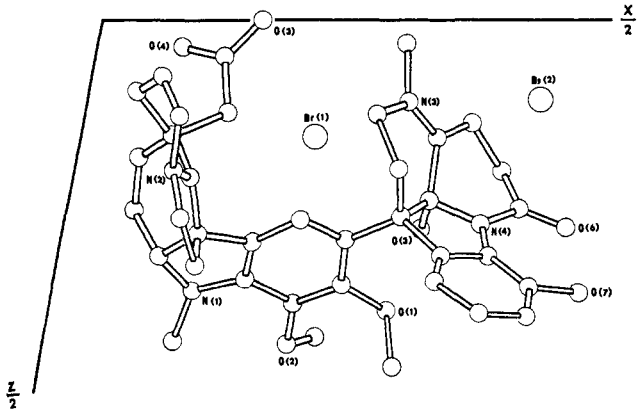
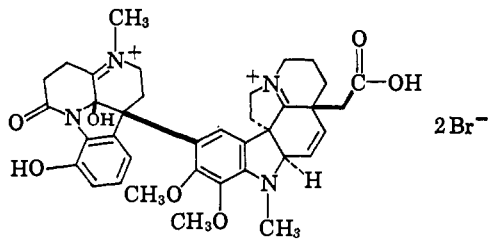


Figure 1.

A single-crystal X-ray diffraction study of haplophytine dihydrobromide revealed the structure shown in III (see Figure 1). The following crystal data were



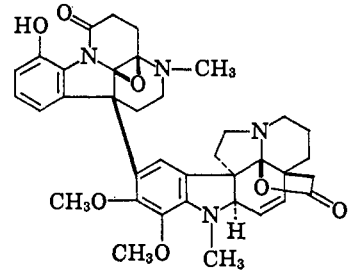
III

obtained for the dihydrobromide with Cu Kα radiation: monoclinic, space group C2, with a = 25.535, b = 7.490, c = 18.861 A, β = 101° 19′, V = 3537.2 A^3, Z = 4, D_x = 1.530, D_m = 1.528 g/cm^3.

Three-dimensional diffraction intensity data were recorded at room temperature on a Picker four-circle automatic diffractometer for 2001 independent reflections. The positions of the bromine atoms in the unit cell were deduced from a three-dimensional Patterson synthesis. After fixing the origin of the unit cell by setting the y coordinate of one of the bromine atoms at zero, the positions of these heavy atoms were used to calculate the phases of the observed structure amplitudes. Successive three-dimensional Fourier syntheses and structure factor calculations progressively disclosed the electron-density distribution of the molecule. The R factor $(\Sigma||F_o| - |F_c||)/|F_o|$, is 8.4% with anisotropic temperature factors for the bromine atoms and isotropic for the light atoms excluding hydrogen. The bond distances and angles are within the range of accepted values.

There are two intramolecular hydrogen bonds in the molecule, as indicated by oxygen–oxygen distances of 2.6 A. One of these is in the 7-hydroxy-1-acylindole system, and the other is between the tertiary alcoholic hydroxyl group and the oxygen atom of the most proximal methoxyl group. The effect of the latter bond is to fix the orientation of the two large moieties with respect to one another in the crystal lattice. The absolute configuration of the molecule was determined by the anomalous dispersion method and is as shown in III and in the accompanying perspective diagram, in which the positive y direction extends out of the page toward the viewer (*i.e.*, conventional right-handed coordinate system). Further refinements of the structure will be reported in detail in due course.

Consideration of structure III in relation to the chemical and spectroscopic properties of haplophytine and the observation that the dihydrobromide is reconverted to the latter at pH 8 lead to the assignment of structure IV to haplophytine. This structure also permits the assignment of structures to the transformation products of haplophytine described earlier.[2]



IV

Acknowledgment. Acknowledgment is gratefully made by P. Y. to the National Research Council of Canada for a grant which supported this work in part.

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Trimethylenemethane, $C(CH_2)_3$

Sir:

Theoretical treatments[1,2] of trimethylenemethane (II) predict a triplet ground state and a high delocalization energy relative to the classical structure of one double bond and two localized electrons. Trimethylenemethane, stable in low-temperature matrixes, has recently been prepared by the photolysis of 4-methylene-1-pyrazoline[3a] or 3-methylenecyclobutanone,[3b] and the esr spectrum confirmed a triplet ground state. Trimethylenemethane and its derivatives have been postulated to explain the formation of "rearranged" methylenecyclopropanes in the pyrolyses[4] or photolyses[5] of 4-alkylidene-1-pyrazolines and in the thermal isomerization of methylenecyclopropanes.[6]

We have examined the gas-phase reaction of 2-iodomethyl-3-iodopropene (I)[7] with alkali metal vapor

(1) A. Streitwieser, Jr., "Molecular Orbital Theory for Organic Chemists," John Wiley and Sons, Inc., New York, N. Y., 1961, p 43.
(2) D. P. Chong and J. W. Linnett, *J. Chem. Soc.*, 1798 (1965).
(3) (a) P. Dowd, *J. Am. Chem. Soc.*, **88**, 2587 (1966); (b) P. Dowd and K. Sachdev, *ibid.*, **89**, 715 (1967).
(4) R. J. Crawford and D. M. Cameron, *ibid.*, **88**, 2589 (1966).
(5) (a) A. C. Day and M. C. Whiting, *J. Chem. Soc., Sect. C*, 464 (1966); (b) S. D. Andrews and A. C. Day, *Chem. Commun.*, 667 (1966).
(6) (a) J. K. Crandall and D. R. Paulson, *J. Am. Chem. Soc.*, **88**, 4302 (1966); (b) J. P. Chesick, *ibid.*, **85**, 2720 (1963); (c) E. F. Ullman, *ibid.*, **82**, 505 (1960).
(7) 2-Iodomethyl-3-iodopropene (mp 32–33°, bp 83–85° (10 mm)) was prepared by the reaction of KI in acetone on 2-chloromethyl-3-chloropropene (see ref 8).