when compared with sterols isolated from the animal's seawater environment[13]. These crustacea, then, are not converting all the ingested campesterol and β-sitosterol to cholesterol, and a steady state concentration of these two compounds appears to exist in these organisms. More work, however, is clearly needed before this hypothesis is proven.

From our results, it appears that caution should be exercised when comparing sterol distributions among members of the same species sampled from different locations. The sterol content of the marine fauna and flora in the animal's surrounding oceanic environment should also be considered. The sterols present in the animal's seawater environment may be an indicator of this faunal and floral contribution to the overall sterol content of the animal[20].

Summary. In this study we have analyzed the sterol compositions of two continental shelf species of crustacea, the lobster (*Homarus americanus*) and the shrimp (*Pandalus borealis*). Cholesterol was found to be the most abundant sterol in these two species with smaller amounts of desmosterol, 24-methylcholesterol, 24-ethylcholesterol, 24-methylenecholesterol and 22-dehydrocholesterol.

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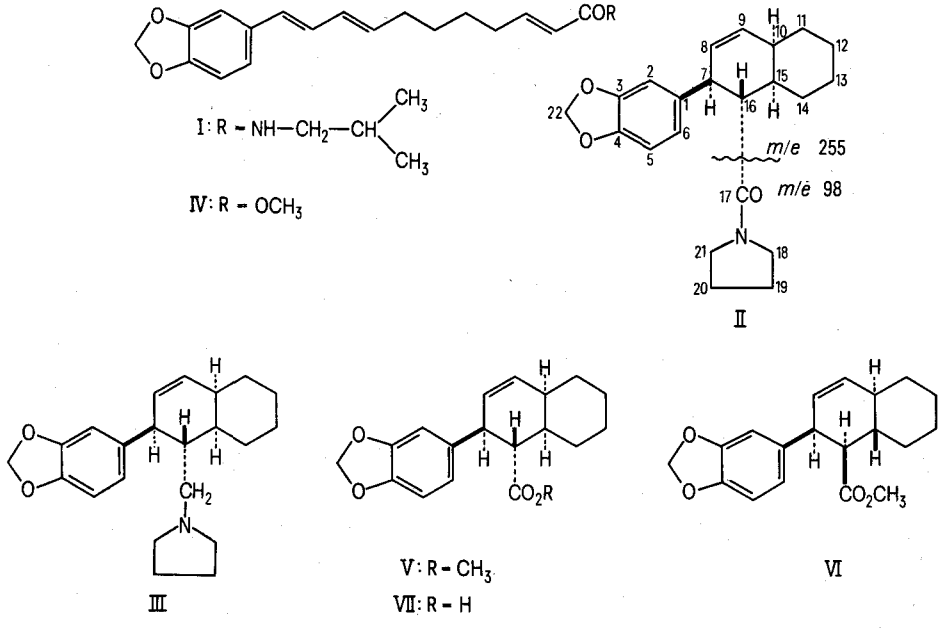
Piperaceae Alkaloids: Part II[1] Structure and Synthesis of Cyclostachine A, A Novel Alkaloid from *Piper trichostachyon* C. DC[2]

In a previous communication[2] we have assigned the structure (I) for piperstachine, isolated from the stem of *Piper trichostachyon* C. DC. We wish to report here the isolation and structure elucidation of a new alkaloid, cyclostachine A (II).

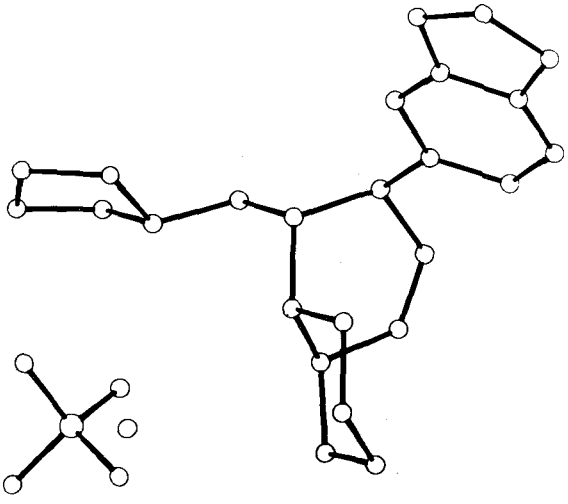
The alkaloid, isolated by cold percolation of the stems with hexane and chromatography over alumina had m.p. 136–138°. It analyzed for $C_{22}H_{27}NO_3$, and showed λ_{max}^{EtOH} 235, 287 nm (log ε 3.65, 3.62), ν_{max}^{KBr} 1630 cm^{-1} (tertiary amide). Its mass spectrum exhibited the molecular ion peak at *m/e* 353 and intense ions at *m/e* 255 and 98 due to the cleavage as shown in II and *m/e* 135 due to methylenedioxytropylium ion. The alkaloid is racemic as shown by CD and ORD determinations. In its ^{1}H-NMR-spectrum ($CDCl_3$, 100 MHz) cyclostachine A shows the following signals: δ (ppm from TMS) 6.7 (3H, m, $H_{2,5,6}$); 5.90 (2H, s, H_{22}); 5.92 (1H, d,d,d, $J_{9,8}$ = 10, $J_{9,10}$ = 5, $J_{9,7}$ = 2 Hz, H_9); 5.56 (1H, d(br), $J_{8,9}$ = 10 Hz, H_8); 3.68 (1H, d, q, $J_{7,16}$ = 10 Hz, H_7); 3.40 (2H, t, J = 6.5 Hz, H_{18} or H_{21}); 2.76 (1H, d,d, $J_{16,15}$ = 11, $J_{16,7}$ = 10 Hz, H_{16}); 3.0–3.3 (1H, m, H_{10}); 2.0–2.5 (3H, m, H_{15} and H_{21} or H_{18}); 1.0–2.0 (12H, m, $H_{11,12,13,14,19,20}$). The proton-noise decoupled ^{13}C-NMR-spectrum of the alkaloid ($CDCl_3$) shows 22 lines, and off-resonance partial decoupling experiments gave the multiplicity of each signal leading to the following assignments, δ (ppm from TMS): 173.6 (C_{17}), 147.7, 146.3 ($C_{3,4}$), 138.7 (C_1), 133.4, 128.5, 121.0, 108.4, 108.1 ($C_{2,5,6,8,9}$), 101.0 (C_{22}), 47.2, 46.8, 36.6, 35.6 ($C_{7,10,15,16}$) 46.4, 45.4 ($C_{18,21}$), 30.6, 28.8, 26.4, 26.0, 24.3 and 22.1 ($C_{11,12,13,14,19,20}$). The off-resonance ^{13}C-NMR-spectrum excluded the presence of quaternary sp^3-carbons in the alkaloid.

[1] For Part I see B. S. JOSHI, N. VISWANATHAN, D. H. GAWAD and W. VON PHILIPSBORN, Helv. chim. Acta, in preparation.

[2] Contribution No. 402 from Ciba-Geigy Research Centre; ^{13}C-NMR-spectroscopy Part 8, for Part 7 see Ref.[1].



Acid hydrolysis of (II) gave pyrrolidine and catalytic reduction afforded the dihydro derivative m.p. 163°. The presence of a disubstituted double bond was confirmed by osmylation to the diol m.p. 167° which on cleavage with periodate gave the dialdehyde m.p. 119°. Cyclostachine A on reduction with $LiAlH_4$ formed the amine (III) isolated as its sulphate, m.p. 110°, the structure of which has been determined by X-ray crystal analysis. $[(C_{22}H_{30}NO_2)^+ (HSO_4)^- H_2O$; monoclinic, a = 15.874, b = 6.584, c = 21.729 Å; β = 98.33°, Z = 4; space group $P2_1/a$ (which confirms the racemic nature of these



compounds); refined to R 0.046 for 2958 reflexions]. The figure depicting molecule III as found in the crystal shows that atoms C (7–10, 15) in the unsaturated ring are coplanar and C(1), C(14) and C(17) are all axially substituted. This is in contrast to NMR results which show that II in solution has these three substituent atoms equatorial – a rather crowded conformation.

The difference is presumably due to the greater volume required by the non-planar amine ring compared with the amide form, and the need to proximate the sulphate ion to the nitrogen atom for the ionic and hydrogen bonded linkage.

The structure of cyclostachine A has been independently confirmed by synthesis. Intramolecular Diels-Alder reaction of the ester (IV)[3] yielded a mixture of (V) m.p. 137° and (VI) m.p. 88°. Hydrolysis of (V) gave the acid (VII), m.p. 141–142°, the chloride of which was condensed with pyrrolidine to yield (II) identical in all respects with the natural sample[4].

Summary. A novel alkaloid designated cyclostachine A has been proved to have the structure (II) on the basis of spectral and degradative data. The structure has been confirmed by synthesis, and by X-ray analysis of the derived amine sulphate (III).

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[3] B. S. Joshi, N. Viswanathan, V. Balakrishnan and W. von Philipsborn, Helv. chim. Acta, in preparation.

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[8] Dedicated to Professor T. R. Govindachari on the occasion of his 60th birthday.

The Identification of Dansyl Sarcosine and its Occurrence in Molluscs

A convenient, sensitive procedure for the analysis of tissue amines and amino acids, involving the two-dimensional chromatography of their dansyl derivatives, has been developed and optimised by Neuhoff et al.[1,2]. This technique has been used to screen for the existence and distribution of putative neurotransmitters in nervous tissue[3–6]. To facilitate interpretation of the resulting chromatographs, several reference maps have been published[1,2,7], based on the migration of standards. However, there are several unknowns among the frequently observed chromatographic spots. In trying to identify the transmitter at the squid giant synapse (G. A. Cottrell, unpublished observations), we encountered an intriguing unknown that appeared to be localized to the post-synaptic axon but absent from the pre-synaptic axon. Purification and characterization of the compound suggest it is the N-dansylated derivative of sarcosine (N-methyl glycine). This report gives the evidence for the identification and some preliminary results of the distribution in molluscan nervous systems.

Materials and methods. Specimens of *Eledone cirrhosa* were obtained locally and maintained in aquaria. *Helix pomatia* were obtained from Gerrard and Haig (Surrey, England), and *Loligo* from Naples, Italy.

Dansylation and subsequent chromatography were executed as described elsewhere[2,7].

Fluorimetry. Comparisons of the unknown and standards were performed in 0.17 ml quartz microcuvettes on an Aminco-Bowman spectrophotofluorometer. Since cephalopod brain was a rich source of the unknown, homogenates of octopus (*Eledone cirrhosa*) brain were dansylated and chromatographed. Pooled unknown spots were extracted with absolute ethanol[2] and compared to standard dansylsarcosine in terms of their excitation spectra (emission set at 522 nm) and emission spectra (excitation set at

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