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OCCURRENCE OF *N*-CYANO ALKALOIDS IN ASIAN *STRYCHNOS* SPECIES

N G BISSET, A K CHOUDHURY* and MARGARET D WALKER

Pharmacognosy Research Laboratories, Department of Pharmacy, Chelsea College, University of London, Manresa Road, London SW3 6LX

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Abstract—The isolation of *N*-cyano-*sec*-pseudostrychnine and *N*-cyano-*sec*-pseudobrucine from the leaves of *Strychnos wallichiana* Steud ex DC is reported. *N*-Cyano-*sec*-pseudostrychnine and a *N*-cyano-*sec*-pseudocolubrine have been found among the alkaloids obtained from the stem bark of *S. ignatii* Berg.

INTRODUCTION

DURING an investigation of the alkaloids from the leaves of *Strychnos wallichiana* Steud ex DC[1] chromatography on silica gel of the total alkaloid mixture gave a series of fractions which were eluted after the pseudostrychnine/pseudobrucine fractions but before the icajine/novacine fractions. Repeated preparative TLC of the combined fractions enabled five alkaloids to be separated: pseudostrychnine (**2b**), icajine (**1a**), novacine (**1d**) and two new bases the elucidation of whose structures is the object of the present note.

RESULTS AND DISCUSSION

The first of the new alkaloids is considered to be *N*-cyano-*sec*-pseudostrychnine (**1b**) on the basis of the following evidence: accurate mass measurement of the molecular ion peak in the MS at *m/e* 375 indicates the formula $C_{22}H_{21}N_3O_3$. The UV spectrum is very similar to that of icajine (**1a**) and together with the intense IR absorption at 1655 cm^{-1} shows that the alkaloid is a N_a-acyldihydroindole[2]. That there is no substitution in the aromatic ring of the indole moiety follows from the strong IR band at 765 cm^{-1} (*o*-disubstituted benzene ring), the occurrence of signals for 4 hydrogens in the aromatic region (δ 6.9–8.3) of the NMR spectrum, and 'indole' peaks in the MS at *m/e* 130, 143, and 144. The moderately strong IR band at 1698 cm^{-1} shows the presence of a 16-carbonyl function, as in **1a**†. Additional evidence for this is the downfield shift of the H-1 NMR signal

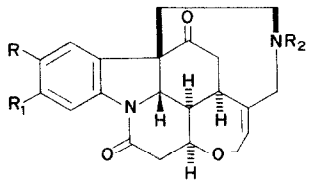
* Part of the work reported here is taken from the Ph.D. thesis submitted to the University of London (1972). Present address of A.K.C.: Natural Drugs Research Institute, B.C.S.I.R. Laboratories, Chittagong, Bangladesh.

† In the IR spectrum of **1a** only one (broad) carbonyl band is observed, at *ca* 1656 cm^{-1}; it is a combination of the C-10 (amide) carbonyl absorption at *ca* 1660 cm^{-1} and the C-16 carbonyl absorption which is lowered to *ca* 1646 cm^{-1} because of interaction with the nearby N_b trans-annular amide-type neutralization)[4]. In **1b** the mesomeric effect of the CN group will be to reduce the interaction and to allow the C-16 carbonyl to absorb at a higher frequency—hence the appearance of two carbonyl bands, at 1655 (C-10 C=O) and 1698 cm^{-1} (C-16 C=O).

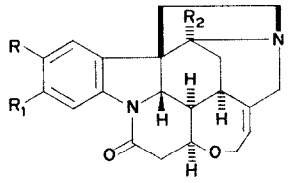
[1] BISSET, N. G. and CHOUDHURY, A. K. (1974) *Phytochemistry* **13**, 259.

[2] BRAND, J. C. D. and SCOTT, A. I. (1963) in *Technique of Organic Chemistry* (BENTLEY, K. W. ed.) Vol. 11, Part 1, p. 89, Interscience, New York.

from δ 7 25 as in the spectrum of **2a**, to δ 7 87, almost the same value as in the spectrum of **1a**, the effect is due to the deshielding by the 16-carbonyl group [3] The MS peak at *m/e* 210 is a further indication of such a function



(1)



(2)

(1a) R = R_1 = H, R_2 = Me
(1b) R = R_1 = H, R_2 = CN
(1c) R = OMe, R_1 = H, R_2 = CN or R = H, R_1 = OMe, R_2 = CN
(1d) R = R_1 = OMe, R_2 = Me
(1e) R = R_1 = OMe, R_2 = CN

(2a) R = R_1 = R_2 = H
(2b) R = R_1 = H, R_2 = OH
(2c) R = R_1 = OMe, R_2 = H
(2d) R = R_1 = OMe, R_2 = OH

There is a moderately strong IR band at 2225 cm^{-1} which is interpreted as revealing the presence of a –C≡N group in the molecule, the position and intensity of the band indicate more particularly a cyanamide grouping (=N–C≡N) [5] Warming the alkaloid **1b** with Zn–HCl liberated HCN (sodium-picrate paper), this test is diagnostic for a CN group attached to a nitrogen atom [6] The MS of **1b** has a peak at *m/e* 305, corresponding to the loss of 70 m u The MS of **1a** also has a peak at *m/e* 305, which is due to the loss of 59 m u consequent on the scission of the nitrogen bridge [4] The increased loss in the case of **1b** from 59 to 70 m u is evidently due to the replacement of CH_3 by CN This fragmentation confirms that the new alkaloid has the same ring system as **1a** *N*-Cyano-*sec*-pseudostrychnine (**1b**) is thus indicated as the structure

TLC of the products formed on reducing **1b** with Zn–HCl show spots corresponding in R_f values with those of pseudostrychnine (**2b**) and strychnine (**2a**) Final proof of the structure comes from partial synthesis of **1b** by the reaction of **1a** with CNBr in CH_2Cl_2 [7] Identity of the product formed was established by comparison of the UV, IR, and MS and of the TLC properties in two systems

The spectral properties of the second new alkaloid (see Experimental) suggest that it is the 2,3-dimethoxy analogue of **1b** This has been confirmed not only by TLC of the Zn–HCl-reduction products, which show spots corresponding to pseudobrucine (**2d**) and brucine (**2c**) but also by comparison of the UV IR and MS of *N*-cyano-*sec*-pseudobrucine (**1e**), prepared from **1d** and CNBr in refluxing CH_2Cl_2 with those of the isolated compound

In studying the alkaloids from the stem bark of *S ignatii* Berg from Sabah (Borneo) a small fraction intermediate between pseudostrychnine and pseudobrucine was obtained on preparative TLC [8] The IR and MS of this fraction showed it to comprise mainly *N*-cyano-*sec*-pseudostrychnine (**1b**) with a little of a *N*-cyano-*sec*-pseudocolubrine (**1c**)

[3] BISSET N G DAS B C and PARELLO J (1974) *Tetrahedron* in press
[4] ANET F A L BAILEY A S and ROBINSON R (1953) *Chem Ind (Lond)* 944
[5] NAKANISHI K (1962) *Infrared Absorption Spectroscopy Practical* p 28 Table 4 Holden-Day San Francisco, Nankodo Tokyo
[6] FEIGL F (1960) *Spot Tests in Organic Analysis* 6th Edn, p 184 Elsevier Amsterdam
[7] ČERNY V and ŠORM F (1960) *Coll Czech Chem Commun* **25**, 2841, KASAI A ČERNY V and ŠORM F (1960) *Coll Czech Chem Commun* **25**, 2849 FODOR G (1971) *Chimia* **25**, 282
[8] BISSET N G and WALKER M D (1974) *Phytochemistry* **13**, in press

As far as is known, no other compounds containing the *N*-cyano or cyanamide function have yet been isolated from natural sources It might be thought that HCN, which is sometimes used to fumigate plant materials being sent from one country to another, could have reacted with the pseudostrychnine and pseudobrucine present,[1] but enquiries have produced no indication that the *S wallichiana* leaves and *S ignatii* stem bark were treated with HCN before arrival at the laboratory There is no evidence to suggest that the compounds were formed during work-up, so it seems that the *N*-cyano derivatives isolated must be looked upon as natural constituents Relevant to this is a report that the leaves, roots, and bark of *S nux-vomica* L have given positive tests for HCN [9] However, attempts to detect the presence of cyanogenetic compounds in the plant materials reported on here were unsuccessful, which, if such substances were indeed present, may have been due to their decomposition during drying, and it is possibly at this stage that the *N*-cyano alkaloid derivatives are formed However that may be, the negative findings are not conclusive and tests on fresh plant materials are required

EXPERIMENTAL

Generalities See Ref 10

Strychnos wallichiana *Separation of the* N-*cyano alkaloids* The *N*-cyano bases were isolated by further separation of the materials from groups 2 and 3 of the initial silica-gel chromatography of the *S wallichiana* leaf alkaloids [1] Preparative TLC (system CH_2Cl_2–MeOH (99 1), run 15 ×) of the *group* 2 bases (1 80 g) gave 6 bands (AA′, 277 mg of **1d**), *BB′*, 27 mg mixture, (CC′, 1000 mg of **1a**), (DD′, 60 mg not identified), *EE′*, 303 mg mixture, and (FF′, 15 mg not identified) The material from band *BB′* was separated by preparative TLC (system CH_2Cl_2, run 63 ×) into 4 zones (Bl, 5 mg **1d**), *B*2, 5 mg crystallizing in Me_2CO to give **1e**, (B3, 7 mg **1c**), and (B4 1 8 mg of **1a**) The material from band *EE′* was separated by preparative TLC (system CH_2Cl_2–MeOH (99 5 0 5), run 32 ×) into 2 zones, *E*1 120 mg crystallizing in Me_2CO to give **1b** and *E*2 135 mg mixture *E*2 was finally separated by preparative TLC (system EtOAc–iPrOH–conc NH_4OH (100 2 1), run 3 ×) into *E2a*, 70 mg crystallizing in Me_2CO to give a further amount of **1b**, and (E2b, 50 mg of **2b**) Additional **1b** was obtained by fractionation of the *group* 3 bases (9 01 g) Fraction 2 (320 mg) of the alumina chromatogram[1] was separated by preparative TLC (system CH_2Cl_2–MeOH (99 5 0 5), run 19 ×) into 2 bands (AA′ 265 mg of **1a**) and *BB′*, 31 mg mixture Repeated preparative TLC of the material from band *BB′* (system EtOAc–iPrOH–conc NH_4OH (100 2 1)) afforded 3 zones *B*1, 19 mg **1b**, (B2, 19 mg **2b**), and (B3, 7 mg **2b**)

N-*Cyano*-sec-*pseudostrychnine* (**1b**) The base crystallized in Me_2CO as needles, m p 235° (decomp) UV λ_{max}^{EtOH} 222 (log ε 4 62), 257 (4 76) and 283 (4 25) nm, λ_{min}^{EtOH} 233 (log ε 4 44) nm IR ν_{max}^{Nujol} 2225, 1698, 1655, 1590, 1415, 1280, 1170, 1155, 1110 and 765 cm^{-1} NMR δ 6 26 (1-H, broadened *t*, H-22), 7 26 (2-H, *m*, H-2 and H-3), 7 87 (1-H, *m*, H-1), 8 15 (1-H, *m*, H-4) (Found M^+ 375 1591 $C_{22}H_{21}N_3O_3$ requires 375 1583) MS *m/e* 375 (M^+, 100%) 347 (3), 333 (6), 332 (5), 305 (5), 272 (5), 237 (5), 234 (6), 225 (8), 222 (6), 210 (10), 209 (22), 208 (6), 196 (7), 194 (8), 183 (8), 182 (7), 180 (9), 168 (11), 167 (10), 156 (10), 154 (8), 144 (11), 143 (16) and 130 (22) The base **1b** was obtained through partial synthesis from icajine (**1a**) by treating it with CNBr in refluxing CH_2Cl_2 for 48 hr, yield *ca* 10% The TLC and spectral (UV, IR and MS) properties were identical with those of the compound isolated from the leaf alkaloid mixture

N-*Cyano*-sec-*pseudobrucine* (**1e**) The base crystallized in Me_2CO as needles UV λ_{max}^{EtOH} 215 (log ε 4 36), 266 (4 04) and 300 (3 88) nm, λ_{min}^{EtOH} 241 (log ε 3 80) and 288 (3 83) nm IR ν_{max}^{Nujol} 2225, 1695, 1635 and 1605 cm^{-1} MS *m/e* 435 (M^+, $C_{24}H_{25}N_3O_5$, 100%), 420 (8), 407 (16), 396 (8), 395 (30), 394 (30), 393 (53), 379 (7), 377 (8), 363 (7), 270 (8), 269 (9), 256 (7), 204 (10), 203 (14) and 190 (10) The base **1e** was obtained through partial synthesis from novacine (**1d**) by treating it with CNBr in refluxing CH_2Cl_2 for 6 days The TLC and spectral (UV, IR and MS) properties were identical with those of the compound isolated

Strychnos ignatii *Separation of the* N-*cyano alkaloid fraction* [8] IR ν_{max}^{Nujol} 2225, 1690, 1655, 1590, 1275, 1170, 1110 and 760 cm^{-1}, the more important MS peaks *m/e* 405 (M^+, $C_{23}H_{24}N_3O_4$), 375 (M^+, $C_{22}H_{21}N_3O_3$), 240, 239, 226, 210, 209, 196, 174, 173, 160, 144, 143 and 130

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[9] HERBERT, D A (1922) *Philipp Agric* **11**, 11

[10] BISSET, N G and CHOUDHURY, A K (1974) *Phytochemistry* **13**, 265

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