Tetrahedron. Vol. 29, pp. 4137 to 4148 Pergamon Press 1973. Printed in Great Britain

ALKALOIDS FROM THE LEAVES OF *STRYCHNOS ICAJA* BAILL.[a]

N. G. BISSET,* B. C. DAS and J. PARELLO†
Institut de Chimie des Substances Naturelles, 91190-Gif-sur-Yvette, France

(*Received in the U.K.* 14 *April* 1973; *accepted for publication* 13 *July* 1973)

Abstract—From the leaves of *Strychnos icaja* Baill. a further seven alkaloids have been isolated and their structures determined: 16-hydroxystrychnine (**1e**), 21,22-α-epoxy-4-hydroxy-3-methoxy-N-methyl-sec.-pseudostrychnine (**5c**), 21,22-α-epoxy-4-hydroxy-N-methyl-sec.-pseudostrychnine **5a**), 21,22-α-epoxy-2-methoxy-N-methyl-sec.-pseudostrychnine (**5b**), 21,22-α-epoxy-14-hydroxy-N-methyl-sec.-pseudostrychnine (**6a**), 21,22-α-epoxy-4,14-dihydroxy-N-methyl-sec.-pseudostrychnine (**6b**), and 14-hydroxy-N-methyl-sec.-pseudostrychnine (**7**). Jaminet's alkaloid B' is shown to be impure 21,22-α-epoxy-14-hydroxy-2,3-dimethoxy-N-methyl-sec.-pseudostrychnine (**6c**).

The young roots of *Strychnos icaja* Baill. have long been used in Central Africa as a source of arrow poison and ordeal poison.[1] Early investigations of the alkaloids were almost all interpreted in terms of strychnine (**1a**) and brucine (**1d**),[1] but Sandberg *et al.* have now shown that the main active principles present in the roots are strychnine and 4-hydroxystrychnine (**1b**).[2]

In 1951 Jaminet obtained three unidentified bases, A, B', and C, from the leaves of *S. icaja*.[3] Later, Bisset isolated icajine (N-methyl-sec.-pseudostrychnine) (**3a**) and vomicine (4-hydroxy-N-methyl-sec.-pseudostrychnine) (**3c**) from the leaves[4] and showed that Jaminet's alkaloid A is a mixture of these two bases.[5] The structure determination of a third alkaloid 21,22-α-epoxynovacine (21,22-α-epoxy-2,3-dimethoxy-N-methyl-sec.-pseudostrychnine) (**5d**) has been reported: the base is identical with Jaminet's alkaloid C.[6]

We now record the isolation, characterization, and structure determination of an additional seven alkaloids from the leaves of *S.icaja*. The alkaloids so far obtained from our material belong to five different series. Those of the first two series are previously known alkaloids and the sections dealing with them are devoted mainly to a discussion of the diagnostic features of their NMR and mass spectra. This lays the foundation for the structure determination, primarily by physico-chemical means, of the new alkaloids isolated, which is dealt with in subsequent sections.

Pseudo series. A small amount of 16-*methoxystrychnine* (**1f**) was isolated and its identity confirmed by comparison of the IR and mass spectra with those of an authentic sample and by the m.p. and mixed m.p.

16-Hydroxystrychnine (pseudostrychnine) (**1e**) on treatment with alcohols readily gives 16-alkoxystrychnines.[7] Since the product obtained had been crystallized in methanol, presumably the component present in the original alkaloid mixture was 16-hydroxystrychnine itself—a base which has been isolated from *S.nux-vomica* L.[8] and as 16-ethoxystrychnine (**1j**) from *S.ignatii* Berg.[7]

In the NMR spectrum of **1f** the H-1 signal is a multiplet centred at δ 7·78, whereas in the spectrum of **1a** this signal is part of a 3-hydrogen multiplet centred at δ 7·25. The considerable paramagnetic shift results from deshielding by the 16-OMe function which is in the α-configuration and near to H-1. Similar shifts have been observed in the spectra of **1e** and other *Strychnos* bases with a 16-OH group.[7,9,10] The signal for H-4 in **1f** is a multiplet centred at δ 8·13, which is about the same value as for **1a**, and its low-field position is clearly due to deshielding by the amide carbonyl at C-10; for **1e** there appears to be a slight upfield shift to δ 8·06.

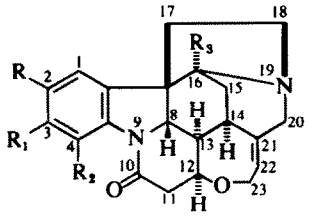
The important features in the mass spectrum of **1f** are: the mol ion peak at *m/e* 364, which is also the base peak; the loss of OMe to give a peak at *m/e* 333 ($M^+ - 31$), which is accompanied by a metastable peak (m^*_{obs} 304·7, m^*_{calc} 304·6); a small peak at *m/e* 185 (see below); and the "indole" peaks at *m/e* 144, 143, and 130.[11]

For **1e** the characteristic features of the mass

[a]To Prof. M. M. Janot and Dr. R. Goutarel—"l'équipe Janot–Goutarel"—on the occasion of their 70th and 65th birthdays, respectively.

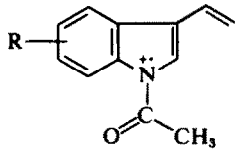
*From the Ph.D. thesis submitted to the University of London. Present address: Pharmacognosy Research Laboratories, Department of Pharmacy, Chelsea College, University of London, Manresa Road, London SW3 6LX.

†Present address: Équipe de Recherche C.N.R.S., Bâtiment Chimie Extension, Université des Sciences et Techniques du Languedoc, 34060-Montpellier, France.



1

1a: R = R₁ = R₂ = R₃ = H
1b: R = R₁ = H, R₂ = OH, R₃ = H
1c: R = OMe, :R₁ = R₂ = R₃ = H
1d: R = R₁ = OMe, R₂ = R₃ = H
1e: R = R₁ = R₂ = H, R₃ = OH
1f: R = R₁ = R₂ = H, R₃ = OMe
1g: R = OMe, R₁ = R₂ = H, R₃ = OH
1h: R = H, R₁ = OMe, R₂ = H, R₃ = OH
1i: R = R₁ = OMe, R₂ = H, R₃ = OH
1j: R = R₁ = R₂ = H, R₃ = OEt



2

R = H *m/e* 185
R = OMe *m/e* 215
R = 2×OMe *m/e* 245

spectrum are: the prominent mol ion peak at *m/e* 350; some loss of oxygen* and water as evidenced by small peaks at *m/e* 334 ($M^+ - 16$) and 332 ($M^+ - 18$); the base peak at *m/e* 185; and the "indole" peaks at *m/e* 144, 143, and 130. The spectra of **1g** and **1h** are similar, but with the peaks at 30 m.u. higher.[10] For **1i**, on the other hand, the mol ion peak at *m/e* 410 is also the base peak; there is considerable loss of oxygen ($M^+ - 16$) and water ($M^+ - 18$); the peak at *m/e* 245 is accompanied by a smaller *m/e* 244 peak and a larger *m/e* 243 peak; and the "indole" peaks are at *m/e* 204, 203, and 190.

The fact that the *m/e* 185 peak is found in the spectra of both **1e** and **1f** shows that the 16-oxygen function is not part of the entity which gives rise to this peak and the fact that *ar*-substituted 16-hydroxystrychnines have a similar peak at an appropriate number of m.u. higher indicates that the indole moiety is a part of the ion. Structure **2** may be postulated for it. The occurrence of this ion and its analogues is indicative of 16-hydroxy- or 16-alkoxystrychnines, i.e. alkaloids of the pseudo series (Table 1).

N-Methyl-sec.-pseudo series

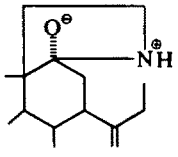
The isolation of N-*methyl-sec.-pseudostrychnine* (*icajine*)(**3a**) and (4-*hydroxy*-N-*methyl-sec.-pseudostrychnine* (*vomicine*) (**3c**) has already been reported.[4]† Some features of the NMR and mass spectra which have helped in elucidating the structures of the subsequent alkaloids are discussed.

In general the NMR spectrum of **3a** resembles that of **3c** (Table 2) except for the aromatic region which is similar to that in the spectrum of 16-hydroxy- or 16-methoxystrychnine (**1e**) or (**1f**) rather than strychnine (**1a**).

The most prominent signal in the spectrum of **3c**‡ is the sharp 3-hydrogen singlet at δ 2·03 for the N-Me group; the high-field position of the signal is due to shielding by the 16-carbonyl group. There is an ill-resolved triplet at δ 6·02 for H-22 and irradiation of this signal shows that the signals for the two H-23 with which H-22 is coupled are in the δ 3·9–4·5 region. The broadening of the H-22 triplet appears to be caused by allylic coupling of H-22 with H-14 and probably also with the two H-20 (*cf* 14-hydroxy-N-methyl-sec.-pseudostrychnine, below). The H-8 doublet is centred on δ 4·34 and its relatively large coupling constant, $J_{8,13} = 11{\cdot}5$ Hz, is in keeping with the known *trans*-relationship of H-8 and H-13. Decoupling experiments locate the H-12 signal at δ *ca* 4·25 as a 6-line multiplet with $J_{12,13} = ca$ 3·5 Hz; this small coupling constant is in agreement with the known *cis*-relationship of H-12 and H-13. Details of the signals for the stereochemically important H-13 and H-14 are not clearly seen, even on decoupling, and the coupling constant cannot be determined. There is a sharp 1-hydrogen singlet at δ 11·66 which disappears with difficulty on deuteration and which is assigned to the H-bonded phenolic OH group.

The mass spectra of **3a**, **3c**, and **3e** are all rather similar, with in general little fragmentation. For **3c** and **3e** the mol ion peaks at *m/e* 380 and 424 are also the base peaks. In the spectrum of **3a**, on the other hand, while the mol ion peak is at *m/e* 364,

*Although **1e** and its analogues exist primarily in the carbinolamine form, from their chemical properties it is inferred that a (not detectable) proportion of the keto-amine form is also present.[16] Perhaps conditions in the mass spectrometer favour a slight shift towards the intermediate zwitterion.



†Ref 4, p. 5237, for "un pic important à *m/e* = 359 (M − 59)" read "un pic important à *m/e* = 321 (M − 59)". The UV data for vomicine should read "223 (log ε 4,23), 265 (3,70) et 299 mμ (3,57)".

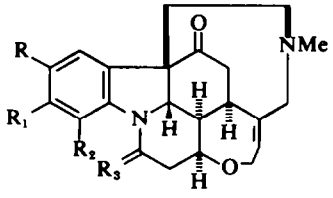
‡Some of the assignments given in Ref 12 are incorrect. The 60-MHz spectrum does not furnish enough detail to enable signals to be allocated to all 24 hydrogens.

Table 1. The principal diagnostic peaks in the mass spectra of *Strychnos icaja* alkaloids and related bases

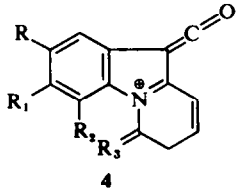
Alkaloid	"Indole" peaks	2	4	8,9	10	(M^+-71)	(M^+-59)	M^+
Pseudo series								
1e	130, 143, 144	185	—	—	—	—	—	350
1f	130, 143, 144	185	—	—	—	—	—	364
1g and **1h**	160, 173, 174	215	—	—	—	—	—	380
1i	190, 203, 204	245	—	—	—	—	—	410
N-*Methyl-sec.-pseudo series*								
3a	130, 143, 144	—	210	—	—	—	305	364
3a, no $\Delta^{21,22}$	130, 143, 144	—	210	—	—	295	307	366
3b	130, 143, 144	—	196	—	—	—	291	350
3c	146, 159, 160	—	226	—	—	—	321	380
3d	146, 159, 160	—	212	—	—	—	307	366
3e	190, 203, 204	—	(269), 270	—	—	—	365	424
21,22-α-*Epoxy-N-methyl-sec.-pseudo series*								
5a	146, 159, 160	—	226, (228)	—	—	325	337	396
5b	160, 173, 174	—	240	—	—	339	351	410
5c	176, 189, 190	—	256, (258)	—	—	355	367	426
5d	190, 203, 204	—	(269), 270	—	—	369	381	440
21,22-α-*Epoxy-14-hydroxy-N-methyl-sec.-pseudo series*								
6a	130, 143, 144	—	210	225	270	325	337	396
6b	146, 159, 160	—	226	241	286	341	353	412
6c	190, 203, 204	—	(269), 270	285	330	385	397	456
14-*Hydroxy-N-methyl-sec.-pseudo series*								
7	130, 143, 144	—	210	225, (226)	—	(309)	321	380

the base peak is at *m/e* 305 (= 364 − 59).[4] Accurate mass measurement shows that this ion has the composition $C_{19}H_{15}NO_3$ (Found: 305·1050; $C_{19}H_{15}NO_3$ requires: 305·1052) and it indicates removal of C_3H_9N, probably as ethylmethylamine, from the original molecule; this can be envisaged as loss of part of the nitrogen bridge along with two hydrogens. There is no corresponding metastable peak, although in the low-molecular-weight region there are peaks at *m/e* 58 and 57. A mechanism for the fragmentation, which is clearly not a one-stage process, has been proposed by Spiteller-Friedmann and Spiteller.[13] This loss of C_3H_9N or 59 m.u. is also found in the spectra of **3c** and **3e**[4] and it appears to be characteristic of alkaloids of the N-methyl-sec.-pseudo series (Table 1).

A peak in the spectrum of **3a** at *m/e* 210 corresponds to an ion of composition $C_{13}H_8NO_2$. It is postulated to have structure **4a**, i.e. it contains both the CO functions present in the original alkaloid (see 14-hydroxy-N-methyl-sec.-pseudostrychnine, below). **3c** and **3e** produce similar ions, at *m/e* 226 (**4c**) and 270 (**4f**), and the latter alkaloid also has one with *m/e* 269. Support for the suggested structure comes from the spectra of icajidine (**3b**) and vomicidine (**3d**). Although these compounds also show little fragmentation and the mol ion peaks are by far the most intense, there are small peaks at *m/e* 196 (**4b**) and 212 (**4d**), i.e. at 14 m.u. less than the corresponding peaks in the spectra of **3c** and **3e**, in agreement with the reduction of the amide carbonyl to methylene.



3



4

3a: R = R₁ = R₂ = H, R₃ = O
3b: R = R₁ = R₂ = H, R₃ = H₂
3c: R = R₁ = H, R₂ = OH, R₃ = O
3d: R = R₁ = H, R₂ = OH, R₃ = H₂
3e: R = R₁ = OMe, R₂ = H, R₃ = O

4a: R = R₁ = R₂ = H, R₃ = O *m/e* 210
4b: R = R₁ = R₂ = H, R₃ = H₂ *m/e* 196
4c: R = R₁ = H, R₂ = OH, R₃ = O *m/e* 226
4d: R = R₁ = H, R₂ = OH, R₃ = H₂ *m/e* 212
4e: R = OMe, R₁ = R₂ = H, R₃ = O *m/e* 240
4f: R = R₁ = OMe, R₂ = H, R₃ = O *m/e* 270

Table 2. NMR spectra of *Strychnos icaja* alkaloids*

Proton	3c	5d	5c	5a	6a	6b	6c	7
1	7·35 q *J* 7·5, 2	7·36 s	7·20 d *J* 9	7·36 q *J* 7, *ca* 1·5	7·83 m	7·33 dd *J* 8, 2	7·35 s	7·80 m
2	7·07 t *J* 7·5	OMe } 3·86, 3·88 s	6·67 d *J* 9	7·05 t *J* 7·5	} 7·15 m	6·98 t *J* 8	OMe } 3·88, 3·90 s	} 7·15 m
3	6·78 q *J* 7·5, 2	OMe	OMe 3·82 s	6·77 q *J ca* 8, *ca* 2		6·72 dd *J* 8, 2	OMe	
4	OH 11·66 s	7·83 s	OH 12·03 s	OH 11·66 s	8·16 m	OH 11·71 s	7·83 s	8·10 m
8	4·34 d *J* 11·5	4·49 d *J* 11·5	4·40 d *J* 11·5	4·47 d *J* 11·5	4·47 d *J* 12	4·30 d *J* 12	4·39 d *J* 12	4·38 d *J* 11·5
12	*ca* 4·25 m $J_{12,13}$ *ca* 3·5 *J ca* 14‡	4·14 m† $J_{12,13}$ *ca* 4 *J* 13‡	4·20 m† $J_{12,13}$ 5 *J ca* 13·5‡	4·16 m† $J_{12,13}$ *ca* 5 *J ca* 12·5‡	4·58 m† *J* 14‡	4·60 m† $J_{12,13}$ *ca* 5·5 *J ca* 14‡	(4·5–4·6)	4·87 q $J_{12,13}$ 4–5 *J* 11–12‡
13	(*ca* 1·70)	1·81 m $J_{13,8}$ 11·5 $J_{13,12}$ 4–5 $J_{13,14}$ *ca* 3**	1·83 m $J_{13,8}$ 11·5 $J_{13,12}$ *ca* 5 $J_{13,14}$ *ca* 3·5**	(*ca* 1·70)	1·98 q *J* 12, 5	1·98 q *J* 12, 6	1·97 q *J* 12, 5	1·90 q *J* 11·5, 4
14-OH	—	—	—	—	3·7 s	3·56 s	3·54 s	*ca* 3·0 s
19–NMe	2·03 s	2·00 s	1·96 s	2·00 s	1·99 s	1·98 s	2·04 s	2·03 s
20α, 20β	—	2·27, 3·02 d *J* 13·5–14	2·25. 3·00 d *J* 13	2·26, 3·04 d *J* 14	2·32, 3·07 d *J ca* 14	2·34, 3·06 d *J* 13	2·34, 3·10 d *J* 13·5	—
22	6·02 t *J* 13·5††	3·02 t *J* 9·5††	(*ca* 3·0)	3·05 m *J* 9·5–10††	3·20 t *J* 8††	3·18 t *J* 6·5††	3·20 t *J* 7††	6·00 t *J* 10††
23α	4·02 q *J* 15, 6	3·63 q *J* 14·5, 5	3·71 q *J* 15, 4·5	3·65 q *J* 14·5, 5	3·88 q *J* 14·5, *ca* 4	3·96 q *J* 15, 3	3·89 q *J* 15, 4	} (*ca* 4·35)
23β	4·26 q *J* 15, 7·5	4·52 q *J* 14·5, 4·5	4·59 q *J* 15, 4	4·55 q *J* 14·5, 4·5	4·55 q *J* 14·5, *ca* 3·5	4·60 q *J* 15, 3	—	

*Chemical shifts in δ; coupling constants in Hz.
†Appearance of a quartet.
‡$J_{12,11\alpha + 12,11\beta}$.
**By difference.
††$J_{22,23\alpha + 22,23\beta}$.

The spectrum of the partial synthetic 21,22-dihydro-N-methyl-sec.-pseudostrychnine (21,22-dihydroicajine) (**3a**, no 21,22-double bond) differs markedly in some respects from those of the compounds just discussed. Its mol ion peak at *m/e* 366 has only 13% relative abundance. The major fragmentation is the loss of part of the nitrogen bridge mentioned above—peak at *m/e* 307 ($M^+ - 59$) with 100% relative abundance, along with peaks at *m/e* 58 and 57. A second important fragmentation is loss of the entire nitrogen bridge, C_4H_9N: peak at *m/e* 295 ($M^+ - 71$) with 93% relative abundance, along with peaks at *m/e* 72, 71, and 70. This second fragmentation only takes place when the 21,22-double bond is absent, so that scission on either side of C-20 is possible. The occurrence of such a ($M^+ - 71$) peak may thus be taken as an indication for the probable absence of the 21,22-double bond in bases of the N-methyl-sec.-pseudo series (Table 1). A small *m/e* 210 peak is also present in the spectrum of 21,22-dihydroicajine and no doubt represents the same ion (**4a**) as is formed from icajine itself.

In the spectra of all the compounds discussed the "indole" peaks are observed as expected (Table 1).

21,22-α-Epoxy-N-methyl-sec.-pseudo series

21,22 - α - *Epoxy* - 2,3 - *dimethoxy* - N - *methyl* - *sec.-pseudostrychnine* (21,22-α-*epoxynovacine*) (**5d**). The elucidation of the structure by chemical methods, already reported,[6]* is supported by analysis of the NMR and mass spectra; some points are discussed in the following paragraphs.

Table 2 lists the NMR data for **5d**, part of which was obtained by decoupling experiments. As expected, the signal for H-13 is complex because of coupling with three adjacent hydrogens, H-8, H-12, and H-14. The coupling constants determined, $J_{8,13} = 11{\cdot}5$ Hz, $J_{13,12} = 4$–5 Hz, and $J_{13,14} = ca$ 3 Hz, agree with the configurations of the ring junctions established for strychnine, brucine, and their derivatives, i.e. 8,13-*trans*, 13,12-*cis*, and 13,14-*cis*.[16]

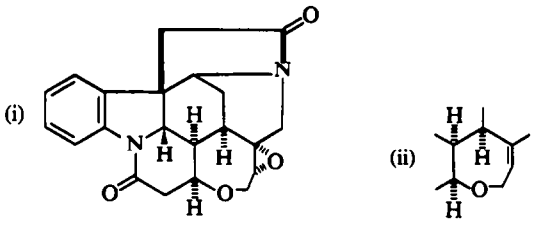
For the 7-membered ether ring the most stable conformation is the chair form with the oxygen up.[17,cf18] Dreiding models show that there is little change from this conformation in molecules with structures like **5** having a 21,22-α-epoxide ring instead of a 21,22-double bond.†

The coupling constants $J_{23,22} = ca$ 4·5 and 5 Hz do not enable the configuration of the epoxide ring to be determined or confirmed, since the values fit either the α- or the β-configuration depending on the conformation of the 7-membered ring. The chemical shifts for H-23α and H-23β in **5d** are 3·63 and 4·52 ppm, respectively, as compared with 4·02 and 4·26 ppm for **3c** (Table 2).‡ The considerable differences in the case of the epoxy base **5d** are consistent with an α-epoxide attached to the 7-membered ether ring in its most stable conformation, as the Newman projections show that the two hydrogens are then in very different positions with respect to the epoxide ring. H-20α and H-20β are in similar positions relative to the epoxide ring and here also there is a considerable difference between their chemical shifts (Table 2).

In the mass spectrum of **5d** the mol ion peak at *m/e* 440 is also the base peak. There is little fragmentation and besides the "indole" peaks at *m/e* 204, 203 and 190, there are only three other peaks of diagnostic value (Table 1). One is the peak at *m/e* 381 ($M^+ - 59$), indicating removal of part of the nitrogen bridge; this time, however, there is a definite metastable peak (m^*_{obs} 330, m^*_{calc} 329·9). The second peak is at *m/e* 369 ($M^+ - 71$), corresponding to loss of the whole nitrogen bridge; again, there is an appropriate metastable peak (m^*_{obs} *ca* 309·5, m^*_{calc} 309·5). Replacement of the double bond by the epoxide ring allows scission on either side of C-20, but not as readily as in the case of 21,22-dihydroicajine (**3a**, no 21,22-double bond) since the relative abundances of the ($M^+ - 59$) and ($M^+ - 71$) ions formed are quite low. The presence of the ($M^+ - 71$) peak can therefore be used as an argument for

*The IR spectrum of **5d** does not give any sure indication for the presence of the epoxide ring. There is additional absorption in the 850–900 cm^{-1} region as compared with the spectrum of **3e**, part of which may represent the asymmetric stretching vibration of the epoxide ring, but the assignment is uncertain.[14] Increased absorption in the 1250–1300 cm^{-1} region may also indicate a contribution by the epoxide ring, but the spectra of all the brucine-type alkaloids examined have a strong band in that region due to the antisymmetric stretching vibration of the aromatic OMe groups.

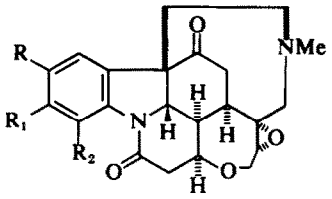
In the IR spectrum of 21,22-α-epoxy-18-oxostrychnine (i) Scheuer[15] assigned bands at 1299 and 1266 cm^{-1} ($CHCl_3$) to the epoxide function. We have observed another band, at 869 cm^{-1} (Nujol), not present in the spectrum of 18-oxostrychnine (ii), which may also derive from the epoxide function.



†When the oxygen is in the down position H-23β is then quite close to H-8 and non-bonded interactions are probable.

The presence of a 21,22-β-epoxide, whatever the conformation of the 7-membered ring, would give rise to considerable steric strain, because the oxygen is very near H-8 and could be expected to cause strong non-bonded interactions.

‡The correct comparison is with the data for **3e**. Unfortunately, these are not available, but they are not expected to differ greatly from those of **3c**.



5

5a: R = R₁ = H, R₂ = OH
5b: R = OMe, R₁ = R₂ = H
5c: R = H, R₁ = OMe, R₂ = OH
5d: R = R₁ = OMe, R₂ = H

the absence of the 21,22-double bond and as a further indication that the epoxide function is located at C-21 and C-22. The third important peak is at *m/e* 270 and it belongs to an ion of the type **4** which contains both the carbonyl functions present in the molecule; the peak is accompanied by a rather more intense one at *m/e* 269.

21,22 - α - *Epoxy* - 4 *hydroxy* - 3 - *methoxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**5c**). That this alkaloid, $C_{23}H_{26}N_2O_6$, is also an epoxy base is evident from the general similarity of its mass and NMR spectral properties (Tables 1 and 2) to those of 21,22-α-epoxynovacine just dealt with.

The pattern of aromatic substitution has been determined as follows: Absorption in the IR at 1248 cm⁻¹ and a sharp 3-hydrogen singlet in the NMR spectrum at δ 3·82 indicate the presence of an aromatic OMe group. Bands at 1672 and 1643 cm⁻¹ may be assigned to a 16-CO and a H-bonded amide carbonyl, respectively. A signal at δ 12·03, disappearing with difficulty on deuteration, points to the occurrence of a H-bonded phenolic OH group; presumably it is H-bonded to the amide carbonyl and so may be placed at C-4 (*cf* the IR and NMR spectra of vomicine (**3c**)).* Since the NMR spectrum shows two aromatic hydrogens as an AB quartet, the OMe group must be located at either C-1 or C-3. However, because the signals are centred at δ 6·67 and 7·20, they must be assigned to H-2 and H-1, respectively, the latter being deshielded by the 16-CO. The signals for these same hydrogens in henningsoline,[19] in which there is no 16-CO, are at δ 6·59 and 6·80 and the paramagnetic shifts of 0·08 and 0·40 ppm observed in the spectrum of the present base are only compatible with the suggested location of the two hydrogens concerned. The OMe function must therefore be attached to C-3. In the mass spectrum of **5c** the "indole" peaks are at *m/e* 190, 189, and 176 (Table 1) and accord with the presence of OMe and OH substituents in the aromatic ring of the indole nucleus.

Decoupling experiments enabled details of the NMR signal for H-13 to be clearly seen and the coupling constants determined show that the base has the usual 8,13-*trans*, 13,12-*cis*, and 13,14-*cis* stereochemistry (Table 2). The arguments for the conformation of the 7-membered ether ring and for the presence of the 21,22-α-epoxide ring follow the same lines as before.

21,22 - α - *Epoxy* - 4 - *hydroxy* - N - *methyl* - *sec.* - *pseudostrychnine* (21,22-α-epoxyvomicine) (**5a**). Comparison of the spectral properties (Tables 1 and 2) of this alkaloid, $C_{22}H_{24}N_2O_5$, with those of vomicine (**3c**) and the epoxy bases already discussed shows that it must be 21, 22 - α - epoxyvomicine.

It may be noted that there is an IR band at 872 cm⁻¹, absent in the vomicine spectrum, which may perhaps be due to the asymmetric stretching vibration of the epoxide ring (*cf* Ref 14 and footnote p. 4141).

21,22 - α - *Epoxy* - 2 - *methoxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**5b**). Very little of this base, $C_{23}H_{26}N_2O_5$, was available for investigation. Peaks in the mass spectrum at *m/e* 351 ($M^+ - 59$) and 339 ($M^+ - 71$) suggest that the base is of the same type as the epoxy bases already considered (Table 1). The "indole" peaks at *m/e* 174, 173, and 160 point to the presence of a OMe substituent in the aromatic ring of the indole moiety; and the close resemblance of the UV spectrum to that of 9 - acetyl - 6 - methoxy - hexahydrocarbazole[21] which has the OMe group in the same position as in 2 - methoxystrychnine (β-colubrine) (**1c**) indicates that the function is located at the 2-position. The UV spectrum also shows that the base is a N_a-acyldihydroindole, while the occurrence of a peak in the mass spectrum at *m/e* 240, corresponding to the ion **4e**, indicates that in addition to the amide carbonyl a 16-carbonyl is present. The 21, 22 - α - epoxy function is assigned by analogy.

21, 22 - α - *Epoxy* - 14 - *hydroxy* - N - *methyl* - *sec.* - *pseudo series*

21, 22 - α - *Epoxy* - 14 - *hydroxy* - N - *methyl* - *sec.* - *pseudostrychnine* (21, 22 - α - *epoxy* - 14 - *hydroxyicajine*) (**6a**). The mass spectral and NMR evidence (Tables 1 and 2) indicates that this alkaloid, $C_{22}H_{24}N_2O_5$, probably has the same ring system as icajine (**3a**). However, there are two extra O atoms to be located. One is considered on the basis of indirect evidence to be present as a 21, 22 - α - epoxide. Thus, the NMR spectrum has no signal for an ethylenic hydrogen; the mass spectrum has a peak at *m/e* 325 ($M^+ - 71$), which points to absence of the 21,22-double bond; and the IR spectrum has

*Reduction of **5c** with $LiBH_4$ in THF, when only the amide carbonyl is reduced, gives a product whose NMR spectrum has a somewhat broadened signal for the phenolic OH at δ 5·32. In contrast with the original base, the reduction product gives a positive ferric-chloride test, thus showing that the phenolic OH must originally have been H-bonded to the amide CO.

bands at 863 and 1267 cm^{-1} (*cf* Ref 14 and foot-note p. 4141) which may be indicative of an epoxide ring. The alkaloid proved resistant to the action of refluxing AcOH and this, along with the fact that during the forcing conditions of acetylation (see below) the epoxide ring is not affected, testifies to the stability of the epoxide ring.*

The fifth oxygen is present as a OH group—IR band at 3480 cm^{-1} and broad NMR signal at δ *ca* 3·7 which disappears on deuteration. Attempted acetylation with Ac_2O/pyr. was unsuccessful, but under forcing conditions with Ac_2O/65% $HClO_4$ a monoacetate is obtained, which on dilute-acid hydrolysis régenerates the original base. The difficulty with which the acetate is formed and the fact that there is no downfield shift in the δ 4·0–5·0 region of the NMR spectrum suggest that the OH group is tertiary.†

In the NMR spectrum‡ the signal for H-13 at δ 1·98 is a pair of doublets, i.e. H-13 is coupled with only two hydrogens. Since the signals for H-8 and H-12 can be clearly recognized, it is evident that H-14 is missing and a reasonable supposition is that it has been replaced by the tertiary OH group. The H-12 signal is found at the low-field position of δ 4·58 because of its 1,3-diaxial relationship with the 14(α)-hydroxyl group. In the spectrum of 14-hydroxyicajine (**7**) H-12 is deshielded even more and the signal is at δ 4·87. The upfield shift of *ca* 0·3 ppm in the spectrum of the present alkaloid is attributed to long-range shielding by the epoxide ring which must have the α-configuration in order to be able to do this.

In the mass spectrum the mol ion peak at *m/e* 396 is also the base peak. The peaks at *m/e* 337 ($M^+ - 59$), 325 ($M^+ - 71$), and 210 (ion of type **4**) are also observed and the first two have metastable peaks close to the calculated values of m^*_{calc} 286·8 and 266·7. However, the presence of the 14-OH group brings about additional fragmentations (*cf* Scheme 1) and ions with *m/e* 270 (**10**), 226 (**9**), and 225 (**8**) are formed. Accurate mass measurement of the *m/e* 270 peak (Found: 270·0761; $C_{15}H_{12}NO_4$ requires; 270·0766) shows that the ion contains four of the five O atoms present in the original molecule. The presence of this type of ion appears to be diagnostic for the 21, 22 - α - epoxy - 14 - hydroxy group of bases (Table 1).

21, 22 - α - *Epoxy* - 4, 14 - *dihydroxy* - N - *methyl* - *sec.* - *pseudostrychnine* (21, 22 - α - *epoxy* - 14 - *hydroxyvomicine*) (**6b**). The spectral properties of this alkaloid, $C_{22}H_{24}N_2O_6$, establish that it is of the vomicine type (Tables 1 and 2).

Of the two extra O atoms, one is shown by the IR band at 3516 cm^{-1} to be in the form of a OH group and this is also evident from the broad 1-hydrogen singlet, disappearing on deuteration, at δ 3·56 in the NMR spectrum. The second oxygen is assumed to be present as a 21,22-α-epoxide—there is no NMR signal for an ethylenic hydrogen, while there are IR bands at 878 and 1298 cm^{-1} which may derive from an epoxide function; peaks in the mass spectrum at *m/e* 353 ($M^+ - 59$), 341 ($M^+ - 71$), and 286 (**10**, with OH at C-4) are also in accord with this formulation.

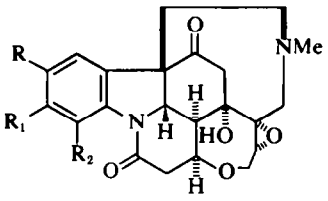
Analysis of the NMR spectrum supports the proposed structure. Irradiation of the H-8 signal which is at δ 4·30 enables the H-13 signal to be identified as a pair of doublets centred at δ 1·98. The H-12 signal is a pair of triplets centred at δ 4·60, but it is complicated by overlap of the signal for H-23β; decoupling H-12 collapses the signal for H-13 to a doublet and thus confirms the assignment. As H-13 is coupled with only two hydrogens, H-8 and H-12, there can be no H-14 and the OH group is placed there in its stead.

The coupling constants for H-8, H-13, and H-12 (Table 2) indicate the usual *trans*- and *cis*-configurations. H-12 is in a 1,3-diaxial relationship with the 14-OH group and this leads to a considerable low-field displacement (*ca* 0·45 ppm) of its signal as compared with that for the corresponding hydrogen in the spectrum of 21,22-α-epoxyvomicine (**5a**). Analogous downfield shifts have been observed in the spectra of steroid derivatives.[23] A similar deshielding effect is operative to a lesser degree on H-13, the signal for which relative to that in the spectrum of **5a** is downfield by *ca* 0·3 ppm. In the conformation of the 7-membered ether ring previously suggested, H-23α is in a diaxial relationship with the 14-OH group although somewhat further away than H-12. As compared with the two H-23 in **5a**, it would be expected that on introduction of the 14-OH group as in **6b** the axial H-23α will be deshielded while H-23β will not; this is borne out by the data given in Table 2.

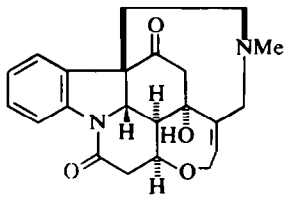
*Scheuer[15] was unable to cleave the epoxide ring in 21, 22-α-epoxy-18-oxostrychnine [(i), foot-note p. 4141] on heating the compound with H_2SO_4 for several hours. In the authors' hands, refluxing AcOH was also unsuccessful. As shown elsewhere,[6] this reagent does, however, cleave the epoxide ring in 21,22-α-epoxynovacine (**5d**), but with *cis*- instead of the more usual *trans*-(diaxial)-opening. Perhaps the presence of a third protonated oxygen, at C-14, in the epoxy-hydroxy bases renders approach of the OAc^- ion to the α-side of C-22 even more difficult than it already is in the case of the epoxy bases.

†Before this was realized, an attempt was made to oxidize the base with Ac_2O/DMSO—a method of oxidizing secondary alcohols which has proved of value in investigating indole alkaloids.[22] After a week, an almost quantitative yield of the beautifully crystalline methylthiomethyl ether was obtained. The formation of such ethers as a side reaction during the oxidation is well known and in some cases, as here, the ether may be the major product.[22]

‡The detailed analysis of the NMR spectrum of the epoxy-hydroxy type base was carried out on 21, 22 - α - epoxy - 14 - hydroxyvomicine (**6b**), but the similarity of the spectrum of the present base with that of **6b** (Table 2) makes it clear that the two alkaloids are of the same type.



6



7

6a: R = R₁ = R₂ = H
6b: R = R₁ = H, R₂ = OH
6c: R = R₁ = OMe, R₂ = H

Such a deshielding effect is unlikely when the oxygen in the 7-membered ring is down, since neither H-23 is then in an axial relationship with the 14-OH group.

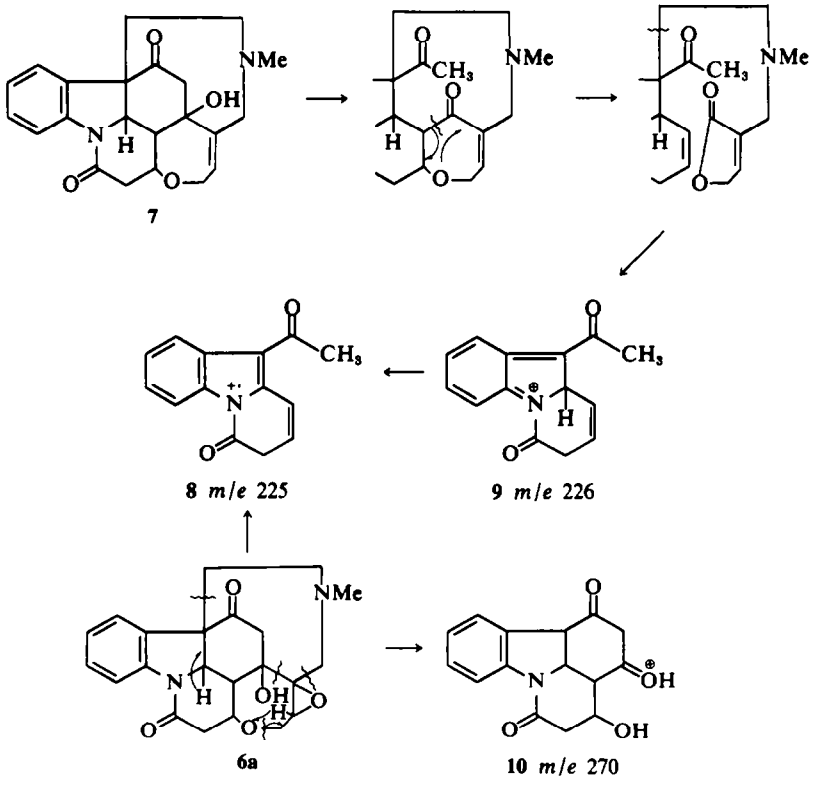
21, 22 - α - *Epoxy* - 14 - *hydroxy* - 2, 3 - *dimethoxy* - N - *methyl* - *sec.* - *pseudostrychnine* (21, 22 - α - *epoxy* - 14 - *hydroxynovacine*) (= Jaminet's alkaloid B') (**6c**). TLC of Jaminet's material[3] showed that it was not a pure substance. However, preparative TLC readily enabled the main component to be separated and obtained in crystalline form. The compound was not isolated from the plant material examined by the authors.

Again, the spectral evidence (Tables 1 and 2) shows that the purified main component, $C_{24}H_{28}N_2O_7$, belongs to the epoxy-hydroxy series and by analogy with the other members of this group it may be formulated as 21,22-α-epoxy-14-hydroxynovacine (**6c**).

14-Hydroxy-N-methyl-sec.-pseudo series

14 - *Hydroxy* - N - *methyl* - *sec.* - *pseudostrychnine* (14 - *hydroxyicajine*) (**7**). The base was easily recognized on TLC, since after spraying with Dragendorff's reagent the central area of the spot turned red and then on standing blue to bluish grey.

The spectral properties of the alkaloid, $C_{22}H_{24}N_2O_4$, indicate that it is of the icajine type



SCHEME 1

(Tables 1 and 2). While there is a signal in the NMR spectrum at δ 6·00 for an ethylenic hydrogen, showing that the 21,22-double bond is present, an IR band at 3460 cm^{-1} and a broad signal at δ *ca* 3·0, disappearing on deuteration, point to the occurrence of a OH group as well. As in the NMR spectrum there is no signal for H-14, the OH group is located there instead. Noteworthy is the very considerable deshielding of H-12 (by *ca* 0·6 ppm) consequent on the introduction of the 14-OH group, as compared with the equivalent hydrogen in icajine (**3a**). The replacement of H by OH at the 14-position removes the allylic coupling with H-22, whose signal is therefore a more clearly resolved triplet than is found in the spectrum of **3a**, for example.

7 would form a suitable material for establishing a direct correlation with the known alkaloid **3a**. However, in spite of the fact that the 14-OH group is allylic to the 21,22-double bond, attempts to remove it by treatment with Zn/AcOH, $LiBH_4$ in THF, or Pd-C/60% AcOH and thus obtained **3a** were unsuccessful. On TLC, the various products which had been formed were found in general to be more polar than **3a**.

The mass spectrum of **7** allows it to be distinguished from members of the other groups of bases present in *S.icaja*. Thus, although there is a *m/e* 321 ($M^+ - 59$) peak with concomitant peaks at *m/e* 58 and 57, there is only a very small peak at *m/e* 309 ($M^+ - 71$) and the usual peaks at *m/e* 72–70 are negligible. Rather intense peaks are at *m/e* 226 (Found: 226·0860; $C_{14}H_{12}NO_2$ requires: 226·0868) (**9**) and 225 (Found: 225·0793; $C_{14}H_{11}NO_2$ requires: 225·0790) (**8**), as in the spectra of the other 14-OH bases, but the peak at *m/e* 270 (**10**) is missing. The absence of this last peak serves to separate the combination 14-OH/21,22-double bond from the combination 14-OH/21,22-epoxide. The peak at *m/e* 210 (Found: 210·0552; $C_{13}H_8NO_2$ requires: 210·0555) (**4a**) is also present, as expected for an alkaloid with both C-10 and C-16 carbonyl functions.

It is noteworthy that the greater part of the alkaloids isolated comprises N-methyl-sec.-pseudo derivatives. Analyses of the alkaloid mixtures from other samples of *S.icaja* leaves have given similar results.[24] However, as Sandberg and Kristianson have shown,[25] the various types of N-methyl-sec.-pseudo alkaloid are very much less active pharmacodynamically than are strychnine and 4-hydroxystrychnine, the main active principles of the root bark. The alkaloids of the leaves of *S.nux-vomica* L. and *S.wallichiana* Steud. ex DC., two Asian species, have also been found to consist largely of N - methyl - sec. - pseudo compounds.[26,27] Small amounts of 14-hydroxyicajine (**7**) and its 2,3-dimethoxy analogue, 14-hydroxynovacine, have recently been isolated from the leaves of *S.wallichiana* collected in Bangla Desh.[27]

EXPERIMENTAL

M.p's are uncorrected. Optical rotations were measured in $CHCl_3$ in a 0·25-cm cell using a Roussel-Jouan Quick electronic polarimeter. NMR spectra were determined at 60 and/or 100 MHz in $CDCl_3$ soln. with TMS (δ = 0·00) as internal standard. Mass spectra were determined with an AEI MS9 high-resolution spectrometer; peak intensities are given as % of the largest peak above 100 m.u., which is taken as the base peak. TLC was carried out using neutral SiO_2 plates run in CH_2Cl_2/MeOH systems, the MeOH varying from 2 to 10%; the plates were sprayed with Dragendorff reagent (Munier-Macheboeuf modification[28]).

Separation of the alkaloids

The crude bases (11·5 g)[4] were purified by a preliminary chromatography over Al_2O_3 (activity I); 250-ml fractions were collected. After check TLC the fractions were combined as follows:

Group 1/1			
C_6H_6	Frns	1–3	0·3 g
C_6H_6/CH_2Cl_2 9:1		4–6	
C_6H_6/CH_2Cl_2 5:5		7–8	
CH_2Cl_2		9–10	
CH_2Cl_2/MeOH 99:1		11–12	
Group 1/2			
CH_2Cl_2/MeOH 99:1		13–17	9·4
Group 1/3			
CH_2Cl_2/MeOH 99:1	Frns	18	0·8
CH_2Cl_2/MeOH 98:2		19–22	
CH_2Cl_2/MeOH 95:5		23–25	
CH_2Cl_2/MeOH 50:50		26	
Group 1/4			
CH_2Cl_2/MeOH 50:50		27–28	0·2
MeOH		29–30	

Group 1/2. The alkaloids were rechromatographed on Al_2O_3 (activity III; 6% H_2O) and 150-ml fractions collected which after check TLC were combined as follows:

Group 2/1			
C_6H_6	Frns	1–7	—
C_6H_6/Et_2O 90:10		8–9	
Group 2/2			
C_6H_6/Et_2O 90:10		10–11	1·7 g
C_6H_6/Et_2O 75:25		12–16	
Group 2/3			
C_6H_6/Et_2O 50:50		17–26	0·3
Et_2O		27–30	
Group 2/4			
Et_2O/EtOH 99:1		31–34	—
Et_2O/EtOH 97·5:2·5		35	
Group 2/5			
Et_2O/EtOH 97·5:2·5	Frns	36–37	1·0
Et_2O/EtOH 95:5		38–41	
Et_2O/EtOH 90:10		42–44	
Group 2/6			
Et_2O/EtOH 90:10		45–62	5·0

Group 2/7			
Et_2O/EtOH	90:10	63–67	0·2
Group 2/8			
Et_2O/EtOH	80:20	68–71	0·2
Et_2O/EtOH	50:50	72–73	

Group 2/2. TLC showed the presence of 2 major components. Rechromatography on SiO_2 and elution with CH_2Cl_2/MeOH mixtures gave amounts of the pure components *vomicine* (**3c**) and *icajine* (**3a**), which crystallized in MeOH, as well as mixed fractions.[4]

Group 2/3. TLC indicated the presence of 3 major components. Preparative TLC (system CH_2Cl_2/MeOH 98:2; run 3×) gave 3 zones: the residue from the least polar one furnished 16-*methoxystrychnine* (**1f**) (25 mg) after crystallization from MeOH, while the residue from the middle zone (86 mg) was further purified by a 2nd preparative TLC to remove small front and rear zones and finally afforded 21, 22 - *α* - *epoxy* - 4 - *hydroxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**5a**) (79 mg).

Group 2/5. Repeated preparative TLC (system CH_2Cl_2/MeOH 97:3; run 3×) and repeated SiO_2 column chromatography (elution with CH_2Cl_2/MeOH 99:1) finally yielded 21, 22 - *α* - *epoxy* - 2 - *methoxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**5b**) (10 mg) and 21, 22 - *α* - *epoxy* - 2, 3 - *dimethoxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**5d**) (*ca* 640 mg).

Group 2/6. Fractionation by repeated SiO_2 column chromatography (elution with CH_2Cl_2 containing up to 2% MeOH), followed by repeated preparative TLC (system CH_2Cl_2/MeOH 96:4; run 3×) gave 2 sets of fractions. The material from the less polar ones was again subjected to preparative TLC (system CH_2Cl_2/MeOH 96:4) and separated into 3 zones: the least polar zone gave a residue (*ca* 1220 mg) part of which was finally purified by SiO_2 column chromatography (elution with CH_2Cl_2) to furnish 21, 22 - *α* - *epoxy* - 4 - *hydroxy* - 3 - *methoxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**5c**) (*ca* 240 mg), while the residue from the middle zone was converted to the hydrochloride salt of 21, 22 - *α* - *epoxy* - 4, 14 - *dihydroxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**6b**) (*ca* 1100 mg). The more polar fractions gave 21, 22 - *α* - *epoxy* - 14 - *hydroxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**6a**) (*ca* 900 mg).

Group 1/3. Preparative TLC (system CH_2Cl_2/MeOH 95:5) afforded a main zone which was further purified by preparative TLC (system CH_2Cl_2/MeOH 90:10; run 2×) to give as the main product 14 - *hydroxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**7**) (180 mg).

Characterization and identification of the individual alkaloids

16-*Methoxystrychnine* (**1f**)

The product crystallized from MeOH as feathery needles, m.p. 195–200° [lit.[29] 196–198° (dec)]; $[\alpha]_D$ − 66° (*c* = 0·67, $CHCl_3$) [lit.[29] $[\alpha]_D$ − 64° ($CHCl_3$)]; ν_{max}^{Nujol} 760, 1396, 1592, and 1667 cm^{-1}; *δ* 3·32 (3-H, s; OMe), 5·92 (1-H, m; H-22), and 6·85–8·25 (4-H, m; H-1, H-2, H-3, and H-4); MS: *m/e* 364 (M$^+$, $C_{22}H_{24}N_2O_3$; 100%), 333 (50), 322 (5), 193 (12), 192 (15), 191 (15), 185 (10), 164 (18), 144 (24), 143 (18), and 130 (18).

The m.p. of the naturally-occurring product was undepressed on admixture with authentic 16-methoxystrychnine (see below). Comparison of the spectral properties confirmed the identification.

21, 22 - *α* - *Epoxy* - 4 - *hydroxy* - 3 - *methoxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**5c**)

The base was not obtained crystalline, but TLC and GLC confirmed that the material was homogeneous: $[\alpha]_D$ + 78° (*c* = 1·05, $CHCl_3$); λ_{max}^{EtOH} 206, 237, and 271 nm (log *ε* 4·11, 4·21, and 3·81); $\nu_{max}^{CHCl_3}$ 1590, 1608, 1643, and 1672 cm^{-1}; [found: M$^+$ 426·1777, $C_{23}H_{26}N_2O_6$ requires M$^+$ 426·1791]; MS: *m/e* 426 (M$^+$, 100%), 398 (3), 367 (14), 355 (12), 258 (8), 256 (5), 230 (8), 229 (6), 190 (8), 189 (8), and 176 (5); for the NMR spectrum, see Table 2.

21, 22 - *α* - *Epoxy* - 4 - *hydroxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**5a**)

This base crystallized from MeOH as small needles, m.p. 252–255° (dec); $[\alpha]_D$ + 112° (*c* = 1·00, $CHCl_3$); λ_{max}^{EtOH} 229, 267, and 300 nm (log *ε* 4·44, 4·04, and 3·75); $\nu_{max}^{CHCl_3}$ 1484, 1582, 1605, 1642, and 1670 cm^{-1}, [found: M$^+$ 396·1689, $C_{22}H_{24}N_2O_5$ requires M$^+$ 396·1685]; MS: *m/e* 396 (M$^+$, 100%), 368 (3), 337 (16), 325 (25), 254 (12), 228 (15), 226 (12), 200 (19), 160 (17), 159 (16), and 146 (9); for the NMR spectrum, see Table 2.

21, 22 - *α* - *Epoxy* - 2 - *methoxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**5b**)

The base crystallized from MeOH as flat prisms, m.p. 265–267° (dec); λ_{max}^{EtOH} 208, 261, and 298·5 nm (log *ε* 4·37, 4·23, and 3·73); [found: M$^+$ 410·1844, $C_{23}H_{26}N_2O_5$ requires M$^+$ 410·1842]; MS: *m/e* 410 (M$^+$, 100%), 351 (4), 339 (14), 268 (10), 240 (13), 2414 (12), 174 (9), 173 (7), and 160 (5).

21, 22 - *α* - *Epoxy* - 14 - *hydroxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**6a**)

This alkaloid crystallized from MeOH as rods, m.p. 266–268° (dec); $[\alpha]_D$ + 3° (*c* = 1·00, $CHCl_3$); λ_{max}^{EtOH} 254, 282·2 (infl.), and 290 nm (log *ε* 4·28, 3·81, and 3·67); ν_{max}^{KBr} 766, 772 (infl.), 1493, 1602, 1668, and 3480 cm^{-1}; [found: M$^+$ 396·1684, $C_{22}H_{24}N_2O_5$ requires M$^+$ 396·1685]; MS: *m/e* 396 (M$^+$, 100%), 368 (1), 367 (1), 341 (6), 337 (2), 326 (5), 325 (22), 270 (15), 226 (3), 225 (9), 210 (11), 168 (5), 167 (4), 154 (12), 144 (15), 143 (10), and 130 (8); for the NMR spectrum, see Table 2.

O-*Acetate.* To the base (74 mg) dissolved in redistilled Ac_2O (2 ml) was added 65% $HClO_4$ (0·25 ml). The soln. was left in the dark overnight, then concd. *in vacuo*, and, after addition of H_2O and NH_4OH, worked up to give a mixture which was separated by prep. TLC using CH_2Cl_2/MeOH 95:5. Elution of the main zone afforded the *acetate* (37 mg) as feathery needles from Me_2CO, m.p. 191–194° (dec); ν_{max}^{Nujol} 1243, 1585, 1660, and 1730 cm^{-1}; *δ* 2·06 (3-H, s; NMe), 2·08 (3-H, s; OAc), 4·2–5·5 (3-H, m), and 6·9–8·2 (4–H, m; H-1, H-2, H-3, and H-4); MS: *m/e* 438 (M$^+$, $C_{24}H_{26}N_2O_6$; 100%), 395 (3), 383 (6), 379 (5), 378 (3), 368 (3), 367 (14), 319 (21), 307 (43), 273 (30), 210 (16), 168 (15), 167 (17), 154 (14), 144 (51), 143 (32), 130 (29), 60 (130), and 43 (415).

Hydrolysis of the acetate (15 mg) with 2N HCl (2 ml) by heating on the water bath for 45 min, followed by the usual work-up, separation of the resulting mixture by prep. TLC using CH_2Cl_2/MeOH 97:3, and elution of the more polar zone gave rods (10 mg) from MeOH, identified as the original alkaloid by TLC and its IR and mass spectra.

O-*Methylthiomethyl ether.* The alkaloid (75 mg) added to a mixture of DMSO (3 ml), previously dried over mol sieve type 4A (Union Carbide International Co), and

freshly distilled Ac_2O (2 ml) did not completely dissolve. The whole was left in the dark for 7 days and shaken occasionally. At the end of this period all the alkaloid had gone into solution. The reaction mixture was then concd. *in vacuo* and the residue gave an almost quantitative yield of the *ether* as needles from MeOH, m.p. 272–273° (dec); $[\alpha]_D - 77°$ (c = 1·01, $CHCl_3$); ν_{max}^{Nujol} 1265, 1585, and 1650 cm^{-1}; found: S 7·02, 7·03%, $C_{24}H_{28}N_2O_5S$ requires S 7·01%; δ 2·05 (3-H, s; NMe), 2·22 (3-H, s; SMe), 3·70 (1-H, dd; J = 13·5 Hz, J' = 6·5 Hz), 4·35–5·00 (5-H, m), and 6–95–8·35 (4-H, m; H-1, H-2, H-3, and H-4); MS: m/e 456 (M^+, $C_{24}H_{28}N_2O_5S$; 100%), 441 (3), 395 (5), 380 (4), 379 (6), 351 (15), 337 (18), 309 (31), 307 (15), 273 (14), 210 (14), 168 (15), 167 (13), 154 (13), 144 (59), 143 (45), 130 (29), 77 (18), and 61 (142).

21, 22 - α - *Epoxy* - 4, 14 - *dihydroxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**6b**)

The base crystallized from Me_2CO as rods, m.p. 239–242°; $[\alpha]_D + 86°$ (c = 1·01, $CHCl_3$); λ_{max}^{EtOH} 228, 266, and 298 nm (log ϵ 4·28, 3·90, and 3·60); $\nu_{max}^{CHCl_3}$ 1483, 1582, 1606, 1643, 1672, and 3516 cm^{-1}; [found: M^+ 412·1632, $C_{23}H_{24}N_2O_6$ requires M^+ 412·1634]; MS: m/e 412 (M^+, 100%), 396 (3), 382 (6), 368 (9), 357 (6), 353 (5), 341 (9), 286 (7), 226 (9), 160 (22), 159 (18), and 146 (11); for the NMR spectrum, see Table 2.

Hydrochloride. Fine needles from MeOH, decomposing from 180° onwards without melting.

21, 22 - α - *Epoxy* - 14 - *hydroxy* - 2, 3 - *dimethoxy* - N - *methyl* - *sec.* - *pseudostrychnine* (**6c**)

Jaminet's alkaloid B′ (30 mg) was purified by prep. TLC using CH_2Cl_2/MeOH 95:5 (run 2×). Elution of the main zone gave the alkaloid (24 mg) which crystallized from Me_2CO as rods, m.p. 251–253° (dec); $[\alpha]_D + 3°$ (c = 0·96, $CHCl_3$); λ_{max}^{EtOH} 219, 264, and 301 nm (log ϵ 4·39, 4·11, and 3·91); ν_{max}^{Nujol} 1264, 1273, 1288, 1405, 1494, 1660, and 3535 cm^{-1}; [found: M^+ 456·1886, $C_{24}H_{28}N_2O_7$ requires: M^+ 456·1896]; MS: m/e 456 (M^+, 100%), 442 (9), 441 (7), 428 (1), 426 (5), 397 (< 1), 385 (3), 330 (2), 285 (3), 270 (7), 269 (9), 204 (6), 203 (6), and 190 (4); for the NMR spectrum, see Table 2.

14-*Hydroxy*-N-*methyl*-*sec.*-*pseudostrychnine* (**7**)

The base was obtained from MeOH as needles, m.p. 262–265° (decomp.); $[\alpha]_D - 60°$ (c = 1·10, $CHCl_3$); λ_{max}^{EtOH} 254, 279 (infl.), and 291 nm (log ϵ 4·25, 3·79, and 3·61); ν_{max}^{KBr} 765, 1430, 1493, 1600, 1666, and 3460 cm^{-1}; [Found: M^+ 380·1725, $C_{22}H_{24}N_2O_4$ requires M^+ 380·1736]; MS: m/e 380 (M^+, 100%), 352 (14), 323 (10), 321 (8), 309 (8), 305 (7), 226 (22), 225 (54), 210 (31), 196 (22), 184 (23), 183 (34), 168 (25), 167 (21), 154 (20), 144 (28), 143 (29), and 130 (42); for the NMR spectrum, see Table 2.

Reference compounds

Authentic *vomicine* (**3c**) had MS: m/e 380 (M^+, 100%), 352 (6), 337 (4), 321 (92), 306 (14), 304 (8), 267 (7), 251 (18), 226 (7), 199 (12), 184 (11), 183 (7), 170 (8), 160 (13), 159 (18), and 146 (18); for the NMR spectrum, see Table 2.

16-*Hydroxystrychnine* (**1e**) was prepared from strychnine (**1a**) by the procedure of Bailey and Robinson[29] and was obtained from $CHCl_3$ as a white powder, m.p. 248–251° (dec) [lit.[30] m.p. 262–268° (dec)]; ν_{max}^{Nujol} 762, 1590, 1663, and 3405 cm^{-1}; MS: m/e 350 (M^+), 210, 209, 198, 197, 196, 186, 185, 172, 144, 143, and 130

O-*Methylation.* **1e** was boiled with MeOH and after purification of the material by prep. TLC yielded 16-*methoxystrychnine* (**1f**) which crystallized from MeOH as needles, m.p. 197–200° [lit.[29] m.p. 196–198° (dec), $[\alpha]_D - 64°$ ($CHCl_3$)]; ν_{max}^{Nujol} 761, 1396, 1593, and 1666 cm^{-1}; MS: m/e 364 (M^+, $C_{22}H_{24}N_2O_3$; 100%), 333 (60), 322 (5), 193 (20), 192 (26), 191 (24), 185 (14), 164 (31), 144 (53), 143 (29), and 130 (33).

N-*Methyl*-*sec.*-*pseudostrychnine* (*Icajine*) (**3a**), prepared from **1e** by N-methylation according to the procedure of Leuchs and Boit,[31] was obtained from MeOH as needles, m.p. 269–270° (dec) [lit.[32] prisms (MeOH), m.p. 273°]; $[\alpha]_D - 11°$ (c = 1·08, $CHCl_3$); λ_{max}^{EtOH} 253 and 291 nm (log ϵ 4·23 and 3·52); $\nu_{max}^{CHCl_3}$ 1598 and 1665 cm^{-1}; δ 2·06 (3-H, s; NMe), 3·65–4·6 (4-H, m; H-8, H-12, H-23α, and H-23β; d centred at 4·41, J = 11·5 Hz, belongs to H-8), *ca* 6·0 (1-H, ill-defined t; H-22), 6·9–7·4 (2-H, m; H-2 and H-3), *ca* 7·6–8·0 (1-H, m; H-1), *ca* 8·0–8·3 (1-H, m; H-4): MS: m/e 364 (M^+, $C_{22}H_{24}N_2O_3$; 84%), 336 (14), 305 (100), 290 (26), 288 (14), 251 (15), 210 (15), 168 (21), 167 (20), 154 (17), 144 (16), 143 (21), 130 (30), 126 (22), 58 (19), and 57 (25).

$LiBH_4$ reduction. To **3a** (100 mg) in THF (20 ml) was added $LiBH_4$ (500 mg) and the whole was refluxed for 4 hr. After addition of dil. HCl to destroy the remaining borohydride and work-up as usual, the resulting product was purified by prep. TLC using CH_2Cl_2/MeOH 98:2 to give N-*methyl*-*sec.*-*pseudostrychnidine* (*icajidine*) (**3b**) (23 mg), plates from MeOH/$CHCl_3$, m.p. 279–281°(dec) [lit.[20] felted needles, m.p. 283–285° (dec)]; λ_{max}^{EtOH} 214, 258, and 318·5 nm (log ϵ 4·31, 4·00, and 3·48); $\nu_{max}^{CHCl_3}$ 1602 and 1655 cm^{-1}; δ 2·05 (3-H, s; NMe), 5·99 (1-H, m; H-22), 6·25–7·65 (4-H, m; H-1, H-2, H-3, and H-4); MS: m/e 350 (M^+, $C_{22}H_{26}N_2O_2$; 100%), 291 (30), 276 (6), 262 (7), 248 (7), 196 (21), 182 (7), 180 (5), 144 (7), 143 (9), and 130 (5).

21, 22 - *Dihydro* - N - *methyl* - *sec.* - *pseudostrychnine* (21, 22 - *Dihydroicajine*). **3a**, (208 mg) dissolved in 10% aq. AcOH (55 ml) was hydrogensted over an excess of Adam's catalyst for 2½ hr.[33] Filtration from the catalyst, followed by the usual work-up and crystallization from MeOH gave the *dihydro-compound* as rods, m.p. 292–296° (dec) [lit.[33] m.p. 296–297°]; $[\alpha]_D + 170°$ (c = 1·05, $CHCl_3$); λ_{max}^{EtOH} 253 and 291 nm (log ϵ 4·36 and 3·76); $\nu_{max}^{CHCl_3}$ 1598 and 1668 cm^{-1}; δ 2·09 (3-H, s; NMe), 4·53 (1-H, d, J = 11 Hz; H-8), 6·8–7·55 (3-H, m; H-1, H-2, and H-3), and 7·9–8·15 (1-H, m; H-4); MS; m/e 366 (M^+, $C_{22}H_{26}N_2O_3$; 13%), 307 (= $M^+ - 59$; 100), 295 (= $M^+ - 71$; 93), 210 (7), 144 (15), 143 (18), and 130 (9).

Novacine (**3e**) was obtained by N-methylation according to the procedure of Boit[31] of pseudobrucine, prepared from brucine by the method of Bailey and Robinson.[29] After purification by prep. TLC using CH_2Cl_2/MeOH 95:5, the *compound* crystallized from Me_2CO as prisms, m.p. 225–228° [lit.[34] m.p. 231–232°]; λ_{max}^{EtOH} 215·5, 267, and 301 nm (log ϵ 4·56, 4·30, and 4·11); ν_{max}^{KBr} 1510, 1615, and 1662 cm^{-1}; δ 2·04 (3-H, s; NMe), 3·87, 3·90 (2 × 3-H, 2 × s; 2- and 3-OMe), 6·02 (1-H, q; H-22), 7·36 (1-H, s; H-1), and 7·86 (1-H, s; H-4); MS: m/e 424 (M^+, $C_{24}H_{28}N_2O_5$; 100%), 365 (11), 270 (5), 269 (4), 204 (3), 203 (2), and 190 (3).

Acknowledgements—We thank Prof. M. M. Janot and Dr. R. Goutarel for their close interest in this work. We are indebted to Prof. J. Le Men, Reims, for providing the sample of *S.icaja* leaves investigated. Prof. A. Denoël, Liège, kindly made available samples of Jaminet's alkaloids and of known *Strychnos* alkaloids.

We thank Mme L. Allais for recording the 60-MHz NMR spectra and Mme F. Picot-Havard for the UV spectra.

The manuscript has benefited from the comments of Dr. P. J. Hylands, Department of Pharmacy, Chelsea College, London.

REFERENCES

[1]N. G. Bisset and A. J. M. Leeuwenberg, *Lloydia* **31**, 208 (1968)

[2]F. Sandberg, K. Roos, K. J. Ryrberg and K. Kristianson, *Tetrahedron Letters* 6217 (1968); *Acta pharm. suec.* **6**, 103 (1969)

[3]F. Jaminet, *Lejeunia* **15**, 9 (1951); *J. Pharm. Belg.* [n.s.], **8**, 449 (1953). A. Denoël, F. Jaminet, G. Detilleux, M. van Sumsen and L. Merveille, *Contribution à l'étude chimique des Strychnos du Congo Belge*, pp. 105–127 Ministère des Colonies, Direction de l'Agriculture, Bruxelles (1953)

[4]N. G. Bisset, *C.r.Acad. Sci., Paris* **261**, 5237 (1965)

[5]N. G. Bisset and P. Fouché, *J. Chromatog.* **37**, 172 (1968). N. G. Bisset, M. Dejestret and P. E. Fouché, *Ann. pharm. franç.* **27**, 147 (1969)

[6]N. G. Bisset, *Tetrahedron Letters* 3107 (1968)

[7]N. G. Bisset, C. G. Casinovi, C. Galeffi and G. B. Marini-Bettòlo, *Ric. sci.* **35** (II-B), 273 (1965)

[8]F. delle Monache, A. Gelabert de Brovetto, E. Corio and G. B. Marini-Bettòlo, *J. Chromatog.* **32**, 178 (1968). G. B. Marini-Bettòlo, F. delle Monache, A. Gelabert de Brovetto and E. Corio, *J. Assoc. off. anal. Chem.* **51**, 185 (1968)

[9]M. Koch, M. Plat, B. C. Das and J. Le Men, *Tetrahedron Letters* 2353 (1966)

[10]G. B. Marini-Bettòlo, E. Miranda delle Monache, C. Galeffi, M. A. Ciasca Rendina and E. Villar del Fresno, *Ann. Chim., Rome* **60**, 444 (1970)

[11]H. Budzikiewicz, C. Djerassi, and D. H. Williams, *Structure Elucidation of Natural Products by Mass Spectrometry. Vol.* 1: *Alkaloids*, pp. 42–45 Holden-Day, San Francisco, London, Amsterdam (1964)

[12]A. Chatterjee and S. C. Basa, *J. Ind. Chem. Soc.* **44**, 663 (1967)

[13]M. Spiteller-Friedmann and G. Spiteller, *Liebigs Ann.* **711**, 205 (1968)

[14]L. J. Bellamy, *The Infra-red Spectra of Complex Molecules*, pp. 118–119 (2nd Edition) Methuen, London, (1958). K. Nakanishi, *Infrared Absorption Spectroscopy–Practical*, p. 36 Holden-Day, San Francisco, and Nankodo, Tokyo, (1962)

[15]P. J. Scheuer, *J. Am. Chem. Soc.* **82**, 193 (1960)

[16]G. F. Smith, *The Alkaloids. Chemistry and Physiology* (Edited by R. H. Manske) Academic Press, New York and London, vol. **8**, chap. 17 (*Strychnos* Alkaloids), pp. 598, 636 (1965)

[17]J. A. Deyrup, H. Schmid and P. Karrer, *Helv. chim. Acta* **45**, 2266 (1962)

[18]G. L. Buchanan and J. M. McCrae, *Tetrahedron* **23**, 279 (1967)

[19]K. Biemann, J. S. Grossert, J. Occolowitz, and F. L. Warren, *J. Chem. Soc.* 2818 (1965)

[20]H. G. Boit and L. Paul, *Chem. Ber.* **87**, 1859 (1954)

[21]J. B. Chalmers, H. J. Openshaw, and G. F. Smith, *J. Chem. Soc.* 115 (1957)

[22]J. D. Albright and L. Goldman, *J. Am. Chem. Soc.* **87**, 4214 (1965). F. W. Sweat and W. W. Epstein, *J. org. Chem.* **32**, 835 (1967). W. W. Epstein and F. W. Sweat, *Chem. Rev.* **67**, 247 (1967)

[23]K. Tori and T. Komeno, *Tetrahedron* **21**, 309 (1965). R. C. Sheppard and S. Turner, *Chem. Commun.* 77 (1968)

[24]N. G. Bisset and J. D. Phillipson, *Lloydia* **34**, 1, 31 (1971)

[25]F. Sandberg and K. Kristianson, *Acta pharm. suec.* **7**, 329 (1970)

[26]N. G. Bisset and J. D. Phillipson, *J. Pharm. Pharmacol.* **23**, Suppl., 244S (1971)

[27]A. K. Choudhury and N. G. Bisset, unpublished work

[28]R. Munier and M. Macheboeuf, *Bull. Soc. Chim. biol.* **31**, 1144 (1949)

[29]A. S. Bailey and R. Robinson, *J. Chem. Soc.* 703 (1948)

[30]K. Warnat, *Helv. chim. Acta* **14**, 997 (1931)

[31]H. Leuchs and H. G. Boit, *Ber. Deut. Chem. Ges.* **73**, 885 (1940)

[32]B. K. Blount and R. Robinson, *J. Chem. Soc.* 2305 (1932)

[33]B. K. Blount and R. Robinson, *Ibid.* 595 (1934)

[34]W. F. Martin, H. R. Bentley, J. A. Henry, and F. S. Spring, *Ibid.* 3603 (1952)