Lanceomigine *N*-Oxide. A Structural Examination using ^{1}H Nuclear Magnetic Resonance and Transient Nuclear Overhauser Effect Measurements

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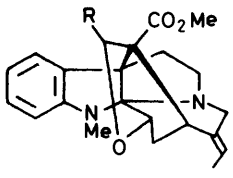
The structure of lanceomigine *N*-oxide (7) has been examined using a combination of ^{1}H–^{1}H spin–spin coupling and nuclear Overhauser enhancement data measured at 300 MHz. It is shown that all details of the structure are susceptible to elucidation using these methods including the recognition of the rare N-1–C-17-bond.

The alkaloids of various parts of the tree *Hunteria umbellata* (K. Schum) Hall F. whose structures have been established are corymine [1] (1a) and its acetate [1] (1b), isocoryminel [1–3] (2a), erinine [3,4] (2b), erinicine [3,4] (2b), eripine [5] (3), and a dimer umbellamine [6] (4). All these, except the eburnamenine ' half ' of umbellamine fall into that group [7] of indole alkaloids which have an unrearranged corynanthé carbon skeleton and additionally, a 7,16-bond.

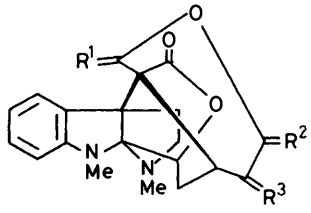
We have shown [3] how the use of a combination of ^{1}H–^{1}H spin–spin coupling and transient nuclear Overhauser enhancement data could be used to elucidate the fine stereochemical details of the structures of isocorymine, erinine, and erinicine with a precision which could not be matched other than by *X*-ray crystallographic studies of the solid state.

In examinations [8] of the minor alkaloids from *Hunteria umbellata* we isolated an amorphous base the structure of which it proved difficult to elucidate by conventional chemical degradative methods. For example although the polarity of the new alkaloid and the presence of a substantial $M - 16$ peak in its mass spectrum strongly suggested the presence of an *N*-oxide grouping, the compound was unaffected by treatment with triphenylphosphine.

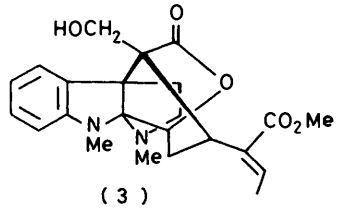
Lanceomigine (5) from *Alstonia lanceolata*,[9] *Hunteria congolana*,[10] and *Hunteria zeylanica*,[11] also an amorphous base, was reduced [12] with a combination of triethylsilane and trifluoroacetic acid to the crystalline deoxy-derivative (6) the structure of which was established [12] by *X*-ray crystallography. The location of the hydroxy-group at C-17 in the alkaloid itself was deduced from the presence of a sharp singlet signal for 17-H. That lanceomigine exists in a hexa-



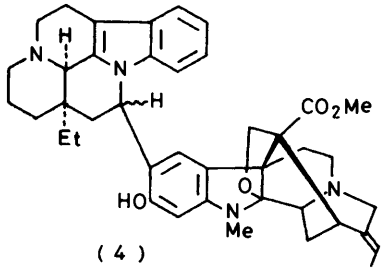
(1)
a; R = OH
b; R = OAc



(2)
a; R^1 = H,OH, R^2 = H_2, R^3 = CHMe
b; R^1 = H_2, R^2 = O, R^3 = CHMe
c; R^1 = H_2, R^2 = O, R^3 = H,Et



(3)



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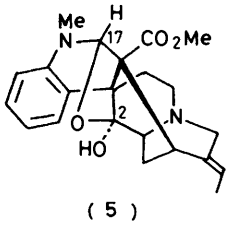
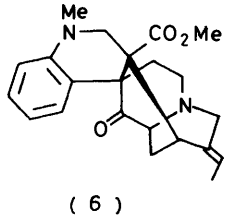
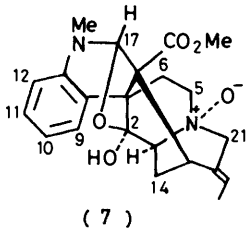


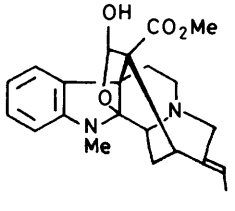
(5)



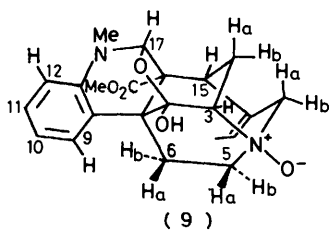
(6)



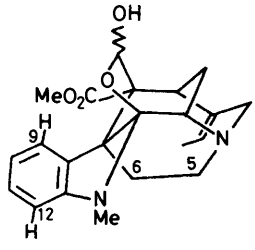
(7)



(8)



(9)



(10)

cyclic form with a C-17–O–C-2 hemiacetal link followed from the absence of a ^{13}C signal for C-2 in the carbonyl carbon region.

Lanceomigine is one of only two indole alkaloids so far reported, the other being lanceomigine *N*-oxide (7), which have an N-1–C-17 bond instead of the usual N-1–C-2-bond present originally in biogenetic precursor tryptophan–tryptamine. It was demonstrated [12] that although 17-hydroxy-ψ-akuammigine (8) which co-occurs with lanceomigine in *H. congolana* could be transformed smoothly into lanceomigine with mineral acid at room temperature, nevertheless the latter is *not* an artefact of the isolation procedure.

Lanceomigine *N*-oxide, isolated amorphous from *A. lanceolata*, was reduced [9] to the same ketone (6) obtained from lanceomigine and could be formed [9] from the latter by *N*-oxidation using *p*-nitroperbenzoic acid. A comparison of the published data [9] for lanceomigine *N*-oxide with those for the amorphous *H. umbellata* alkaloid, suggested their identity and this was confirmed by direct comparison with spectra and sample kindly provided by Professor Lévy.

Distinguishing between the usual picralima skeleton, *e.g.* (8) and that in lanceomigine (5) is by no means trivial without resort to *X*-ray crystallography and presents a problem which may well arise again. For example it is not inconceivable that alkaloids may be found in the ajmaline–quebrachidine series in which N-1 has switched from C-2 to C-17. One method which certainly does *not* distinguish the two types is u.v. spectroscopy for lanceomigine has absorption which in the indole alkaloid context would be described as typical of an indoline (2,3-dihydroindole) as displayed by a large number of indole alkaloids, whereas the chromophore in lanceomigine is a 1,2,3,4-tetrahydroquinoline: u.v. spectroscopically both are of course anilines.

We show below how it is possible to derive the structure of (5) in some stereochemical detail, including the fact that N-1 is attached to C-17 and not to C-2, by ^1H n.m.r. measurements.

Experimental

Spectra were recorded on a Varian Associates SC-300 pulsed Fourier transform spectrometer operating at 300 MHz. Selective inversion of single resonances was achieved by pulsing the homonuclear decoupler; π pulse widths of *ca.* 20 ms were employed. The transient n.O.e.s were evaluated by comparing the peak heights of spectra recorded at thermal equlibrium with those recorded during the relaxation of the inverted peak. To ensure an accurate representation of the peak profile, free induction decays were accumulated into 32 K data points and weighted with an exponential function of time constant −1 s to reduce noise.

2% w/v Solutions in [$^2H_6$]acetone were used. The samples were degassed and sealed *in vacuo*. The temperature of measurement was 23 ± 1 °C.

Results

The spectrum of (7) was well resolved and essentially first-order. The majority of chemical shift assignments resulted from the application of standard chemical shift structural correlations and spin-decoupling experiments. At this stage, the remaining problems were the assignment of the a and b protons within the C-5, C-6, C-14, and C-21 methylene proton pairs and, in the aromatic region the distinction of 9- and 12-H (both doublets) and 10- and 11-H (both triplets). These tasks were accomplished with the aid of measurements of ^1H spin–lattice relaxation times (T_1) and transient nuclear Overhauser enchancements (n.O.e.s). The T_1 values are given in the Table, together with the assignments deduced from the

Spin–lattice relaxation times, chemical shifts, and coupling constants for lanceomigine *N*-oxide. 20 mg ml^{-1} Solution in [$^2H_6$]acetone at 23 °C

Proton	T_1/s	δ	*J* (coupling proton)/Hz
3	0.53	4.66	2.5(14a), 2.5(14b), 1(15)
5a	0.28	4.52	10(5b), 4(6a), 15(6b), 1.5(21a)
5b	0.27	3.41	10(5a), 1(6a), 4.5(6b)
6a	~0.2	2.68	4(5a), 1(5b), 16(6b)
6b	0.30	3.03	15(5a), 4.5(5b), 16(6a)
9	0.44	7.44	1(11), 6(10)
10	1.38	6.85	6(9), 6(11), 1(12)
11	1.42	7.21	1(9), 6(11), 6(12)
12	1.19	6.67	1(10), 6(11)
14a	~0.20	2.66	2.5(3), 14(14b), 1.5(15)
14b	0.23	2.41	2.5(3), 14(14a), 3(15)
15	0.66	3.75	1(3), 1.5(14a), 3(14b), <1(19)
17	0.63	5.07	
18	0.56	1.61	7(19), 2(21a)
19	0.83	5.66	<1(15), 7(18), 1.5(21a)
21a	0.25	4.35	1.5(5a), 2(18), 1.5(19), 15(21b)
21b	0.23	3.96	<1(19), 15(21a)
OCH_3	0.92	3.65	
NCH_3	0.73	3.07	

T_1 and n.O.e. data. The arguments used to obtain these assignments are as follows.

In order to draw reliable structural conclusions from n.O.e. data, it is necessary to determine the extent to which spin–lattice relaxation is controlled by intramolecular dipole–dipole interactions. That this mechanism is in fact dominant is shown by the relaxation times shown in the Table. The T_1 values cover a wide range, reflecting the variety of proton environments. All the methylene protons have similar short T_1 values of *ca.* 0.24 s arising from the geminal interaction at *ca.* 178 pm. The longest values arise from the aromatic 10-, 11-, and 12-H reflecting the larger *ortho*-proton separation of *ca.* 250 pm. Assuming intramolecular nearest-neighbour interactions only, the ratio of T_1 values for a methylene proton and an aromatic proton with two *ortho*-neighbours should be approximately $2 \times (178/250)^6 = 0.26$. After making an allowance for non-geminal interactions, we estimate the T_1 for an isolated geminal group to be *ca.* 0.3 s, giving an experimental CH_2/ArH T_1 ratio of 0.3/1.4 = 0.21, in reasonable agreement with the theoretical value.

Spatial relationships between protons were investigated using the transient n.O.e. method, details of which are given in ref. 13. Briefly, this method entails the measurement of the transient enchancements resulting from intensity transfer during relaxation of a selectively inverted peak. The maximum enchancement is denoted by the symbol η*. Approximate internuclear distances were obtained from a Dreiding molecular model.

The coupling constant, T_1, and n.O.e. data of particular structural importance were these.

(a) The T_1 values of the two aromatic doublets (9- and 12-H) were shorter than those of the aromatic triplets (10- and 11-H), indicating the proximity of both 9- and 12-H to non-aromatic protons since if the only interactions had been with the other aromatic protons, the T_1 values of 9- and 12-H would have been about twice as great as those for 10- and 11-H. A significant transient n.O.e. (η* *ca.* 0.07) of the doublet at δ 6.67 was observed on inversion of the 1-CH_3 peak, giving the assignment of 12-H. The assignment of 11-H resulted from decoupling 12-H and the assignments of 9- and 10-H followed by default.

(b) Selective inversion of 9-H gave a large n.O.e. (η* *ca.* 0.1) for the 6-H at δ 2.68. This n.O.e., and the short T_1 of H-9

(0.44 s) compared with 10-H (1.38 s), suggest that 9- and a 6-H are closer than 9- and 10-H. The value of 190 pm measured for the 9-H–6a-H distance using a Dreiding model, is entirely consistent with these measurements. Only in a model of the lanceomigine structural type [see (9) for a conformational representation of (7)] can 9-H and one 6-H approach this closely. For example in the ψ-akuammigine structural type a model [see (10) for conformational representation of (8)] indicates that the nearest methylene proton to 9-H is *ca.* 290 pm away. Assuming that the relaxation of 10-H arises entirely from interactions with its two *ortho*-neighbours, the relaxation time of 9-H from interaction with 10-H alone should be *ca.* 2.8 s. If the difference between this value and the observed value of 0.44 s arises from the interaction with 6b-H only, then the contribution to 9-H relaxation from 6b-H amounts to 0.52 s. This relaxation time requires an internuclear distance of 190 pm, in excellent agreement with the distance measured from the model.

The assignment of 6b-H followed by default and the assignments of 5a- and 5b-H were made on the basis of spin–spin coupling data. In the Dreiding model, one of the 5-H–6-H vicinal interactions is *trans* (6b-H–5a-H) and the remaining three are *gauche*. This structure is consistent with the four coupling constants observed (1, 4, 4.5, and 15 Hz). The *trans* coupling gives the assignment of 5a-H to the resonance at δ 4.52.

(c) A small n.O.e. (η* *ca.* 0.06) of the 14-H peak at δ 2.66 was observed on inversion of the 21-H peak at δ 4.35, and *vice versa*. This is consistent with the 1,4-diaxial interaction of 14b- and 21a-H (*ca.* 200 pm) in the boat-shaped C-3–C-14–C-15–C-20–C-21–N-4 ring (see structural diagram), and allows the assignment of the C-14 and -21 proton pairs.

(d) Small enhancements of 17-H (η* *ca.* 0.04) were observed on inversion of either 15- or 14a-H. These are consistent with 1,3- and 1,4-diaxial relationships found in the Dreiding model which gives *R*(17-H–15-H) 280 and *R*(17-H–14a-H) 240 pm.

(e) An enhancement of 15-H (η* *ca.* 0.07) on inverting the 18-Me signal confirms the *E*-configuration of the C-19–C-20 olefinic bond, a feature found almost universally in the indole alkaloids.

(f) An unusually large four-bond coupling of 1.5 Hz was observed between 5a- and 21a-H. This is consistent with the *trans–trans*-conformation of the 6a-H–C-6–N-4–C-21–21a-H fragment in the model.[14] A coupling of 1 Hz is observed between 3- and 15-H which are similarly disposed.

(g) The small vicinal couplings between 3-, 14a-, 14b-, and 15-H (1.5, 2.5, 2.5, and 3 Hz) are consistent with the *gauche*-arrangements found in the model.

(h) There are two further, structurally significant features of the T_1 data in the Table. Firstly, the T_1 values for (7) are a factor of five or six *smaller* than those of the similarly sized alkaloids apparicine[13] and isocorymine.[3] The difference indicates that (7) is tumbling much more slowly, presumably due to stronger intermolecular forces arising from the very polar *N*-oxide group. Secondly, the T_1 value of the OCH_3 group is only slightly greater than that of the $N{-}CH_3$ group. This indicates that rotations about the C-16–CO and CO–O bonds are strongly hindered, otherwise the superposition of these motions onto rotation about the $O{-}CH_3$ bond would give a much longer T_1 for the OCH_3 group compared to the $N{-}CH_3$ group. The Dreiding model shows considerable ester group restriction by the 1-CH_3 and 18-CH_3 groups. The T_1 values for the $N{-}CH_3$ and 18-CH_3 protons are about twice that estimated for an isolated methylene group rigidly fixed in the molecular frame, showing that internal methyl rotation is rapid compared to molecular tumbling.

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