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THE ^{13}C-NMR SPECTRA OF 1,2,10-TRIOXYGENATED APORPHINES

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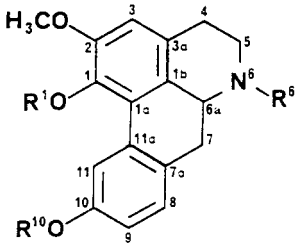
Since the appearance of the first studies on the ^{13}C-nmr spectra of aporphines (1,2), several tabulations of carbon chemical shifts have been published for a large number of aporphinoids (3-5). Some useful relationships between structure and chemical shifts are stated explicitly in these papers, and others can be deduced from the tables and from a more recent listing of the properties and occurrence of these compounds (6). Perusal of these tables reveals that out of more than 60 aporphinoid ^{13}C-nmr spectra reported only 9 represent the four possible ring-D monooxygenation patterns.

As a considerable amount of zenkerine [**1**] had been isolated a decade ago in our laboratory and partially characterized (7), we decided to attempt its purification from the remnants of the *Isolona zenkeri* (Annonaceae) fractions from which it was originally obtained with the hope of being able to determine at least the sign of its optical rotation and its ^{13}C-nmr spectrum. The successful purification of zenkerine allowed this modest goal to be reached, and also the ^{13}C analysis of the previously unknown *O*-methylzenkerine [**2**], of pulchine (*N*-methylzenkerine) [**3**] isolated from *Ocotea pulchella* (Lauraceae) (8), and of 1,2,10-trimethoxyaporphine [**4**] isolated from *Thalictrum foliolosum* (Ranunculaceae) (9).



	R^1	R^6	R^{10}
1	H	H	CH_3
2	CH_3	H	CH_3
3	H	CH_3	CH_3
4	CH_3	CH_3	CH_3
5	H	CH_3	H
6	CH_3	CH_3	H

Although the free base of zenkerine is rather unstable and could only be obtained as a highly colored glass, it was possible to determine its optical rotation at 589 nm. It proved to be levorotatory, and its absolute configuration is, therefore, 6a(*R*) (10), establishing it as a biogenetic derivative of (*R*)-coclaurine presumably via crotsparine and sparsiflorine. Therefore, the *N*-methylcrotsparine which co-occurs with zenkerine in *I. zenkeri* (7) most probably belongs to the same stereochemical and biogenetic series.

The ^{13}C-nmr chemical shifts of compounds **1**-**4** are listed in Table 1. Resonances were assigned by correlation with published values (3,5) and by ^{1}H off-resonance decoupling experiments. On comparing the aromatic ring carbon resonance assignments given in the literature for 1-hydroxy-2-methoxyaporphines it became apparent that references (3) and (5) disagree with regard to the chemical shifts of C-1b and C-3a. Severini Ricca and Casagrande (3) based their attempted identification of the C-1b signal on the upfield shift expected for the carbon resonances upon protonation of the nitrogen atom. Unfortunately, these authors did not carry out any direct comparisons of the ^{13}C-nmr spectra of base-salt pairs, but relied on the spectra of four salts, two of which correspond to noraporphines, another to a dehydroaporphine, and the last to a 7-hydroxyaporphine. Considering all these structural variations with regard to the aporphine bases tabulated in their

TABLE 1. ^{13}C-nmr Chemical Shifts of Compounds **1-6** and of the Hydrochlorides of **1** and **4**

Atom	Compounds							
	5[a]	**6**[a]	**1**[b]	**2**[b]	**3**[b]	**4**[b]	**1**-HCl[c]	**4**-HCl[c]
C-1	141.6	144.3	141.5	145.0	141.4	145.1	142.9	147.3
C-2	146.5	151.3	145.9	152.0	145.7	152.0	148.1	152.9
C-3	110.2	111.6	110.1	111.8	109.5	111.3	110.9	112.2
C-3a	122.9	128.6[d]	123.9	128.8[d]	123.8	128.4[d]	121.2[d]	126.7[d]
C-1a	119.4	125.9	119.1	126.4	119.3	126.8	119.0	125.9[d]
C-1b	127.5	127.7[d]	129.1[d]	127.6[d]	128.2[d]	127.5[d]	121.9	118.3
C-4	28.4	28.7	28.9	29.0	28.8	28.9	24.7	25.6
C-5	52.8	52.5	43.2	43.0	53.3	53.1	—[g]	51.4
C-6a	62.5	62.3	53.8	53.7	62.6	62.5	52.2	61.4
C-7	33.6	33.5	36.5	36.4	34.0	34.8	32.3	30.0
C-7a	126.0	126.6	128.7[d]	128.2[d]	127.9[d]	127.8[d]	125.1	125.9[d]
C-8	127.9	128.4	128.1	128.2	128.2	128.4	128.7	129.3
C-9	113.2[e]	114.0[e]	112.4[e]	113.2[e]	112.4[e]	113.1[e]	112.1[e]	113.5[e]
C-10	155.3	155.7	159.2	158.4	158.2	158.5	158.2	158.6
C-11	115.4[e]	114.5[e]	114.1[e]	113.5[e]	114.0[e]	113.7[e]	115.1[e]	113.5[e]
C-11a	133.0	132.1	133.3	132.9	133.1	132.8	132.9	131.9
N-Me	43.5	43.5	—	—	43.8	43.6	—	—[g]
1-OMe	—	59.6	—	60.1	—	60.2	—	60.9
2-OMe	55.7	55.5	56.1[f]	55.7[f]	56.0[f]	55.7[f]	56.2[f]	56.0[f]
10-OMe	—	—	55.2[f]	55.2[f]	55.2[f]	55.3[f]	55.2[f]	55.2[f]

[a]Data from Severini Ricca and Casagrande (3), revised; 25.2 MHz, DMSO.
[b]20 MHz, $CDCl_3$.
[c]20 MHz, DMSO-d_6.
[d,e,f]Assignments interchangeable within columns.
[g]Obscured by solvent signal.

study, we were unable to find any clear-cut distinction between the chemical shifts of C-1b and C-3a. On the other hand, Jackman *et al.* (5) used relaxation time measurements, selective irradiation experiments, and comparisons of the spectra of aporphines with different substitution patterns to obtain a coherent set of assignments that has generally been taken as a standard by the authors of more recent papers.

We have recorded the ^{13}C-nmr spectra of the hydrochlorides of **1** and **4** (Table 1) as examples of a 1-hydroxy-2-methoxynoraporphine and a 1,2-dimethoxyaporphine. The assignment of the C-7a resonance is reached unambiguously by considering relaxation times (5), and this atom, like C-3a, lies three bonds away from the nitrogen and belongs to an aromatic ring. Although both situations are not strictly identical, it seems reasonable to assume that the ^{13}C-nmr signals of C-3a and C-7a should be displaced to a similar extent by *N*-protonation. In the cases of **1** and **4** we found that *N*-protonation shifts the C-7a resonance upfield by 3.6 and 1.9 ppm, respectively. Basing our assignments on reference (5), the C-3a signals appear shifted upfield by 2.7 and 1.7 ppm, in agreement with our assumption, while the C-1b peaks are much more strongly displaced 7.2 and 9.2 ppm upfield. If we use the work of Severini Ricca and Casagrande (3) as the basis of our assignments, the C-3a resonances undergo large upfield shifts and the signals attributed to C-1b are only weakly displaced. The report of Jackman *et al.* (5), therefore, leads to a much more reasonable distribution of protonation shifts. If the C-1 phenolic function is methylated, Jackman *et al.* (5) indicate that the *meta*-carbon signals (C-1b and C-3) are hardly displaced at all, and the *para*-carbon peak (C-3a) undergoes a downfield shift of 4.9 to 5.0 ppm. Severini Ricca and

Casagrande (3) suggest that the *para*-resonance should be practically unchanged as a result of *O*-methylation, and the two *meta*-carbon signals should be displaced downfield to very different degrees: C-3 by 0.9 to 1.8 ppm and C-1b by 5.1 to 6.0 ppm. We view such a situation as quite unlikely. Taking both *N*-protonation and *O*-1-methylation shifts into account, we believe that all the C-1b and C-3a assignments in Severini Ricca and Casagrande (3) for 1-hydroxy-2-methoxyaporphines (caaverine, lirinidine, apoglaziovine, isoboldine, and bracteoline) ought to be inverted. As apoglaziovine [**5**] and nuciferoline [**6**] share the 1,2,10-trioxygenation pattern with compounds **1** to **4**, we have included the pertinent ^{13}C-nmr data in Table 1 for comparison.

It now becomes possible to analyze the changes produced in the ^{13}C-nmr spectra of this group of alkaloids by methylation of the phenol group at C-10. The *ipso*-(C-10) peak moves downfield by 2.8 to 2.9 ppm, both *ortho*-(C-9 and C-11) signals undergo upfield shifts of less than 2 ppm, the *meta*-(C-8 and C-11a) resonances are virtually unchanged, and the *para*-(C-7a) peak is displaced downfield by 0.9 to 2.2 ppm. A similar analysis of the *O*-methylation displacements in the ^{13}C-nmr spectra of aporphines with a single ring-D oxygen atom at C-9, although necessarily based on less data (6), suggests that the chemical shifts of these compounds are also rather insensitive to this structural change. It is worth noting that the *O*-methylation shifts predicted empirically for simple phenols are quite small, particularly at the *meta*- and *para*-positions (11,12). The ^{13}C-nmr behavior of the aporphines with ring-D monooxygenation at C-9 or C-10 would, thus, seem to be in line with that of relatively uncrowded phenols, as *O*-methylation should not introduce any major change in the conjugation of the oxygen atom with the aromatic ring. A similar situation can be predicted for aporphines with a single oxygen atom on ring D at C-8 or C-11. On the contrary, downfield *O*-methylation shifts of several ppm should be expected for the ^{13}C resonances of the atoms *para* with regard to the hydroxyl group in the cases of 8-hydroxy-9-substituted, 3-hydroxy-2-substituted, and ring A- and D-trisubstituted aporphines. Here, replacement of the phenol hydrogen atom should result in torsion of the aryl-oxygen bond, and as a result of this, greater localization of the oxygen lone electron pairs, decreased electron density at the ring positions, and, therefore, relative deshielding of the carbon nuclei, particularly *ortho* and *para* with regard to the modified phenol function, as has already been suggested for 1-hydroxy-2-methoxy- and 11-hydroxy-10-methoxyaporphines (5). This hypothesis should clearly be checked experimentally and, in view of the pharmacological importance of some aporphine derivatives, points to the potential interest of quantum-chemical studies of the electronic structure of these compounds.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Analytical tlc was carried out on silica-gel precoated foils; preparative chromatography was effected on silica gel, either with precoated plates (0.5 mm thickness) or on "flash" columns; CH_2Cl_2-MeOH-NH_4OH (90:9:1) was used as eluent in every case, saturating the tlc chamber with NH_3 vapor. ^{1}H-nmr spectra were recorded at 60 MHz in $CDCl_3$ with TMS as internal reference. ^{13}C-nmr spectra were recorded at 20 MHz in $CDCl_3$ or DMSO-d_6 (for the salts).

(−)ZENKERINE [**1**].—Amorphous, purple-red solid, $[\alpha]^{23}D$ −99° (c=0.10 MeOH); ^{1}H nmr δ 3.80 s (3H, MeO-2 or -10), 3.86 s (3H, MeO-10 or -2), 6.57 s (1H, H-3), 6.73 dd J=8.0; 2.5 Hz (1H, H-9), 7.13 d J=8.0 Hz (1H, H-8), 8.02 d J=2.5 Hz (1H, H-11); ^{13}C nmr δ in Table 1.

(−)-*O*-METHYLZENKERINE [**2**].—Zenkerine, dissolved in MeOH, was methylated with CH_2N_2 in Et_2O at 5°. The major product was separated by column chromatography: dark brown, glassy solid; ^{1}H nmr δ 3.68 s (3H, MeO-1), 3.79 s (3H, MeO-2 or -10), 3.83 s (3H, MeO-10 or -2), 6.63 s (1H, H-3), 6.78 dd J=8.0; 2.5 Hz (1H, H-9), 7.16 d J=8.0 Hz (1H, H-8),

8.10 d J=2.5 Hz (1H, H-11); ^{13}C nmr δ in Table 1.

(−)-PULCHINE [**3**].—Zenkerine was *N*-methylated with HCHO-$NaBH_4$ in MeOH. The dark, glassy product was purified by column chromatography: $[\alpha]^{21}D$ −130° (c=0.10, MeOH); ^{1}H nmr δ 2.52 s (3H, N-Me), 3.81 s (3H, MeO-2 or -10), 3.84 s (3H, MeO-10 or -2), 6.59 s (1H, H-3), 6.79 dd J=8.0; 2.5 Hz (1H, H-9), 7.20 d J=8.0 Hz (1H, H-8), 8.06 d J=2.5 Hz (1H, H-11); ^{13}C nmr δ in Table 1.

(−)-1,2,10-TRIMETHOXYAPORPHINE [**4**]. *O*-Methylzenkerine was *N*-methylated with HCHO-$NaBH_4$ in MeOH. Glassy solid, $[\alpha]^{23}D$ −169° (c=0.10, MeOH); ^{1}H nmr δ 2.54 s (3H, N-Me), 3.68 s (3H, MeO-1), 3.82 s (3H, MeO-2 or -10), 3.87 s (3H, MeO-10 or -2), 6.63 s (1H, H-3), 6.81 dd J=8.0; 2.5 Hz (1H, H-9), 7.18 d J=8.0 Hz (1H, H-8), 8.03 d J=2.5 Hz (1H, H-11); ^{13}C nmr δ in Table 1.

LITERATURE CITED

1. E. Wenkert, B.L. Buckwalter, I.R. Burfitt, M.J. Gasić, H.E. Gottlieb, E.W. Hagaman, F.M. Schull, and P.M. Wovkulich, in: "Topics in Carbon-13 NMR Spectroscopy," vol. 2. Ed. by G.C. Levy, Wiley, New York, 1976.
2. S. Kano, Y. Takahagi, E. Komiyama, T. Yokomatsu, and S. Shibuya, *Heterocycles*, **4**, 1013 (1976).
3. G. Severini Ricca and C. Casagrande, *Gazz. Chim. Ital.*, **109**, 1 (1979).
4. A.J. Marsaioli, F. de A.M. Reis, A.F. Magalhães, E.A. Rúveda, and A.M. Kuck, *Phytochemistry*, **18**, 165 (1979).
5. L.M. Jackman, J.C. Trewella, J.L. Moniot, M. Shamma, R.L. Stephens, E. Wenkert, M. Leboeuf, and A. Cavé, *J. Nat. Prod.*, **42**, 437 (1979).
6. H. Guinaudeau, M. Leboeuf, and A. Cavé, *J. Nat. Prod.*, **46**, 761 (1983).
7. R. Hocquemiller, P. Cabalion, A. Bouquet, and A. Cavé, *C.R. Acad. Sci. Paris, Ser. C*, **285**, 447 (1977).
8. D.L. Edie, *Diss. Abstr. Intern. B*, **35**, 3827 (1975).
9. D.S. Bhakuni and R.S. Singh, *J. Nat. Prod.*, **43**, 252 (1982).
10. M. Shamma, *Experientia*, **18**, 64 (1962).
11. M. Hesse, H. Meier, and B. Zeeh, "Spektroskopische Methoden in der organischen Chemie," 2nd ed., Thieme, Stuttgart, 1984, p. 228.
12. H.O. Kalinowski, S. Berger, and S. Braun, "^{13}C-NMR-Spektroskopie," Thieme, Stuttgart, 1984, p. 286.

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