Pergamon

Tetrahedron Letters, Vol. 37, No. 10, pp. 1655-1658, 1996
Copyright © 1996 Elsevier Science Ltd
Printed in Great Britain. All rights reserved
0040-4039/96 $15.00 + 0.00

S0040-4039(96)00084-6

Determination of a Symmetrical Dimer Structure in Benzo[c]phenanthridine Alkaloids by Pulsed-Field-Gradient HMBC

Radek Marek and Vladimír Sklenář[*]

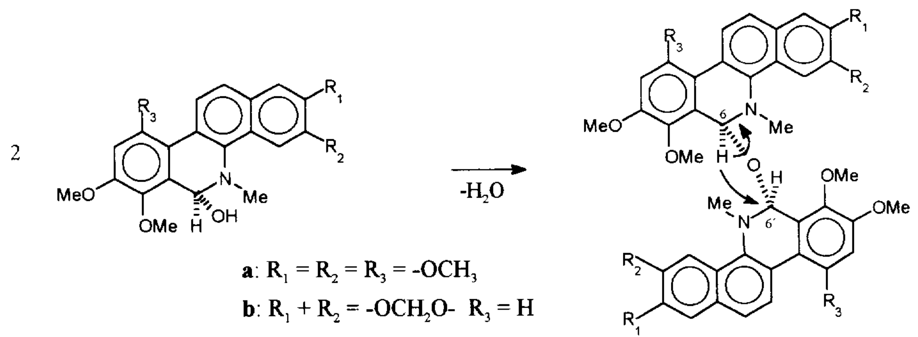
Department of Chemistry, Faculty of Science, Masaryk University,
Kotlářská 2, CZ-611 37 Brno, Czech Republic

Jiří Dostál and Jiří Slavík

Department of Biochemistry, Faculty of Medicine, Masaryk University,
Komenského nám. 2, CZ-662 43 Brno, Czech Republic

Abstract: The pulsed-field-gradient HMBC - based determination of symmetrical benzo[c]phenanthridine dimers is reported. The determination is based on the observation of two different scalar interactions between H6 and C6 atoms.

Quaternary benzo[c]phenanthridine alkaloids reacting with hydroxide anion in aqueous media are converted to the species called pseudobases.[1-4] Until recently the pseudobase formation in benzophenanthridines remainded an unanswered part of alkaloid chemistry. We have found[5] that alkaloid sanguilutine chloride, for the first time isolated from *Sanguinaria canadensis* L. (Papaveraceae)[6,7], gives the relatively stable 6-hydroxyadduct **1a**.



a: $R_1 = R_2 = R_3 = -OCH_3$

b: $R_1 + R_2 = -OCH_2O-$ $R_3 = H$

1

2

Figure 1

Under favourable conditions in non-polar environment the free hydroxy adduct **1a** spontaneously transforms to an oxygen bridged dimer **2a** (Figure 1).[5]

The formation of analogous dimers in chelerythrine and sanguinarine was detected by MS and elemental analysis,[8,9] but it was never exactly proved by other methods including NMR spectroscopy. This is the first report on the unequivocal NMR confirmation of a symmetrical benzo[c]phenanthridine dimer.

All ^{1}H- and ^{13}C-NMR signals of **2a** were assigned[5] on the basis of NOESY,[10] HSQC[11] and HMBC[12,13] experiments. The atoms H6 and C6 exhibiting for monomer **1a** the chemical shifts of 6.00 (^{1}H) and 79.03 (^{13}C) ppm are slightly changed in the dimer **2a** and the chemical shifts of 6.55 and 77.72 ppm have been observed.

Bis[6-(5,6-dihydrosanguilutinyl)]ether **2a** contains two benzo[c]phenanthridine moieties linked by oxygen atom in the position 6 as was for the first time proposed by Slavík.[6]

The carbon atoms 6 and 6′ in the dimer structure are separated by two bonds. As a result, the hydrogen atom H6 should show not only one-bond scalar coupling to C6 but also a long-range interaction across three bonds to C6′ (H6-C6-O-C6′). One-bond interaction to C6′and three-bond coupling to C6 should also be observed for the hydrogen H6′. Both scalar interactions can be clearly identified in 2D NMR experiment which correlates simultaneously single- and long-range ^{1}H-^{13}C scalar interactions.

Because of ^{13}C natural abundance in most cases only one of the two positions C6 and C6′ is occupied by the ^{13}C isotope. If ^{13}C is located at the position C6 the one-bond scalar interaction gives rise to a doublet for H6 signal at frequencies f_{H6}-0.5*($^1J_{H6,C6}$) and f_{H6}+0.5*($^1J_{H6,C6}$). The signals at frequencies f_{H6}-0.5*($^3J_{H6,C6'}$) and f_{H6}+0.5*($^3J_{H6,C6'}$) are expected for the three-bond scalar interaction in isotopomers with ^{13}C at the position C6′.

The ^{1}H-^{13}C interactions outlined above were detected by the HMBC[12] experiment with pulsed field gradients[13] for selection of coherence transfer pathways to improve suppression of proton signals from ^{1}H-^{12}C moieties. No low-pass J-filter has been included to remove single bond connectivities. The delay for evolution of long range couplings was set to 66.6 ms and the gradient pulses with the ratios of 50:30:40 were applied.[14]

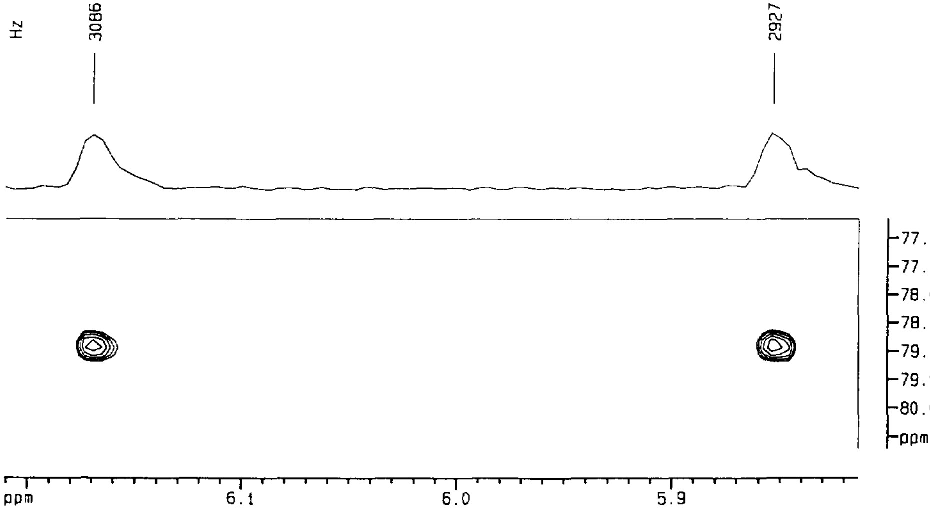


Figure 2a (partial HMBC spectrum of monomer structure **1a**)

Figure 2a shows the part of the HMBC spectrum of 6-hydroxy-5,6-dihydrosanguilutine **1a** (monomer). The signal of H6 is split by one-bond coupling $^1J = 159$ Hz.

In the HMBC spectrum of the dimer **2a** a clear splitting of the H6 signal by $^1J = 160$ Hz is observed. However, an additional correlation peak to C6 is also detected due to the three-bond interactions C6-H6′ and C6′-H6 (Fig. 2b). Because of a small value of the corresponding coupling constant $^3J_{H6,C6}$ unresolved multiplet is positioned at the proton chemical shift of H6.

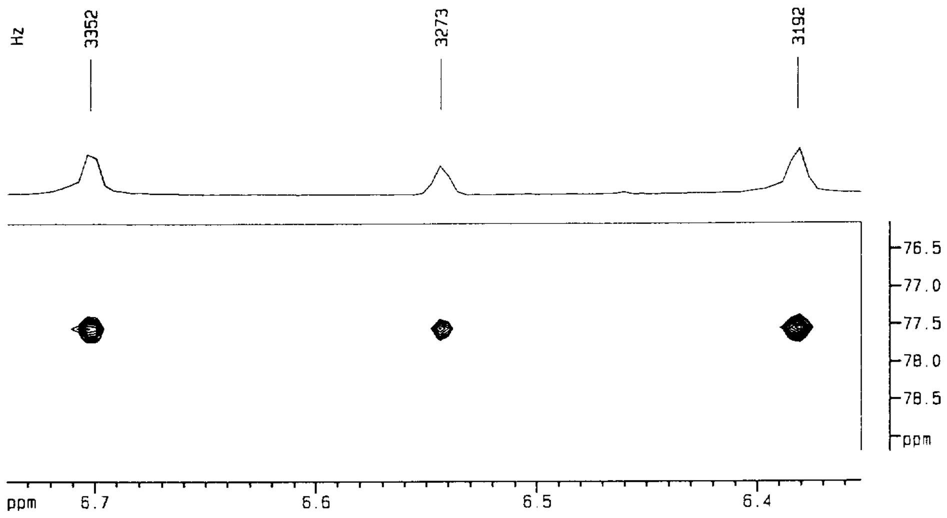


Figure 2b (partial HMBC spectrum of dimeric structure **2a**)

The chemical behaviour of a related alkaloid chelerythrine appeared to be different[8] but the NMR assignment of the H6 and C6 signals and following determination of other signals was completed using the same method. A substance isolated after reaction of chelerythrine chloride with hydroxy anion in aqueous enviroment and dissolved in deuteriochloroform was determined as a mixture of bis[6-(5,6-dihydrochelerythrinyl)]ether **2b** and 6-hydroxy-5,6-dihydrochelerythrine **1b** (70% : 30%).

Also in this case only one bond splitting ($^1J = 160$ Hz) of signal H6 at 6.04 ppm was observed for **1b** in C6 chemical shift line at 79.31 ppm. The dimer **2b** (6.62 and 77.78 ppm) exhibits one-bond interaction of 161 Hz as well as the long-range coupling giving rise to the correlation peak with H6 and C6 chemical shifts.

Further studies will be conducted in order to confirm the dimeric structures of other benzo[c]phenanthridine alkaloid derivatives and to show general applicability of this method for the determination of -NH- bridged dimers as well as its utilization for study of other types of symmetrical dimeric structures.

REFERENCES AND NOTES

1. Šimánek, V. *Alkaloids* (N.Y.) **1985**, *26*, 185-240

2. Dostál, J.; Potáček, M. *Collect. Czech. Chem. Commun.* **1990**, *55*, 2840-2873

3. Šimánek, V.; Preininger, V. *Heterocycles* **1977**, *6*, 475-497

4. Bunting, J.W. *Adv. Heterocycl. Chem.* **1979**, *25*, 1-82

5. Dostál, J.; Slavík, J.; Potáček, M.; Marek, R.; Hoffmann, E. to be published

6. Slavík, J.; Slavíková, L. *Collect. Czech. Chem. Commun.* **1960**, *25*, 1667-1675

7. Krane, B.D; Fagbule, M.O.; Shamma, M.; Gozler, B. *J. Nat. Prod.* **1984**, *47*, 1-43

8. Dostál, J.; Táborská, E.; Slavík, J.; Potáček, M.; Hoffmann, E. *J. Nat. Prod.* **1995**, *58*, 723-729

9. Dostál, J.; Bochořáková, H.; Táborská, E.; Slavík, J.; Potáček, M.; Buděšínský, M.; Hoffmann, E. *J. Nat. Prod.* under revision

10. Jeener, J.; Meier, B.H.; Bachmann, P.; Ernst, R.R. *J. Chem. Phys.* **1979**, *71*, 4546-4553

11. Bodenhausen, G.; Ruben, D.J. *Chem. Phys. Lett.* **1980**, *69*, 185-187

12. Bax, A.; Summers, M.F. *J. Am. Chem. Soc.* **1986**, *108*, 2093-2094

13. Rinaldi, P.L.; Keifer, P.A. *J. Magn. Reson.* **1994**, *108*, 259-262

14. The spectra were obtained on a 500 MHz Bruker DRX spectrometer with 5 mm tripleresonance probehead equipped with a gradient coil using the pulse sequence: D1-90°(^{1}H)-D6-90°(^{13}C)-t_1/2-GP1--D16-180°(^{1}H)-GP2-D16-t_1/2-90°(^{13}C)-GP3-D16-ACQ(t_2) with following parameters: D1 = 2.3 s, D6 = 66.6 ms, D16 = 100 μs, GP = 1 ms, GP1:GP2:GP3 = 30G/cm:18G/cm:24G/cm, a 4085 Hz f_2 spectral window and an 16.35 kHz f_1 spectral window, block size 4k x 2k. The ^{13}C and ^{1}H spectra were referenced to the solvent signals of $CDCl_3$ and of residual $CHCl_3$ at 77.00 ppm (^{13}C) and 7.26 ppm (^{1}H), respectively.

(*Received in UK* 16 *November* 1995; *revised* 9 *January* 1996; *accepted* 12 *January* 1996)