Communications

Organic Structure Characterization by Natural-Abundance Nitrogen-15 Nuclear Magnetic Resonance Spectroscopy. Penicillin and Cephalosporin Derivatives[1]

Summary: The ^{15}N resonances of a series of penicillin and cephalosporin derivatives have been detected at the natural-abundance level; crude structure–chemical shift correlations can be ascertained.

Sir: We report here (Table I) preliminary results of an examination of structurally complex organic molecules by ^{15}N NMR spectroscopy at the natural-abundance level (0.36%) of this isotope. Our results demonstrate the practicability of the technique as a complement to other types of NMR studies and illustrate some operational requirements for its success. Previous natural-abundance spectra of the insensitive (0.1% that of ^{1}H for an equal number of nuclei) ^{15}N nucleus have been of small molecules as neat liquids or as highly concentrated solutions, or have required very long total accumulation times.[2] Very recently, the natural-abundance spectrum of an amino sugar has been reported.[3]

To optimize signal strength, soluble ester derivatives of the title compounds were chosen; concentrations were in the range of 1–2.5 *M*.[4] The large molecular sizes and the high solution viscosity were expected to increase molecular correlation times and shorten T_1 values.[5] No paramagnetic complexes were used to further shorten T_1.[6] Noteworthy is the fact that, under the conditions used,[7] no signals were obtained from small molecules which did not have proton-bearing nitrogen atoms (e.g., nitrobenzenes, pyridines), even as pure liquids, in the absence of a paramagnetic additive. Additionally, the phases of the nitrogen resonances in most of the compounds studied here had the same polarity. Since the nuclear Overhauser effect (NOE) inverts a ^{15}N resonance because of the negative magnetic moment of ^{15}N,[8] this observation suggests that even the nonprotonated lactam nitrogen experiences a NOE, although we do not know whether this leads to an intensity enhancement. In the most favorable cases, adequate signal-to-noise ratios were obtained in 2 hr (Figure 1), while at worst overnight runs were employed. Peak assignments were based on known ^{14}N shifts[9] and were confirmed for **1b** by increasing the pulse angle and repetition rate (Figure 2). As expected, the signal of the secondary amide nitrogen increased in intensity while that of the lactam disappeared, owing to its anticipated longer T_1 value.

Table I
^{15}N Chemical Shifts of Antibiotic Derivatives[a]

Compd	Concn, *M*	t,[b] hr	S/N[c]	δ_N, ppm Lactam	Amide	Other
1a	1.0	10.9	15	134.77	80.52	
1b	2.5[d]	1.8[e]	10[e]	134.30	85.13	
2	1.4	2.1	11	106.10	78.03	
3	1.4	12.5	19	121.56	74.04	
4	2.0[f]	10.2	12	130.78	85.89	185.95
5	1.7	10.9	22	117.29	78.91	
6	1.3[f]	14.3	25	129.87	83.84	9.68

[a] In parts per million downfield from external 2.9 *M* $^{15}NH_4Cl$ in 1 *M* HCl contained in a 2-mm capillary. Experimental error was ±0.1 ppm. Solvent was dioxane except as noted. [b] Total accumulation time. [c] Signal-to-noise ratio of strongest peak. [d] In benzene. [e] For t 16.9, S/N 38. [f] In D_2O.

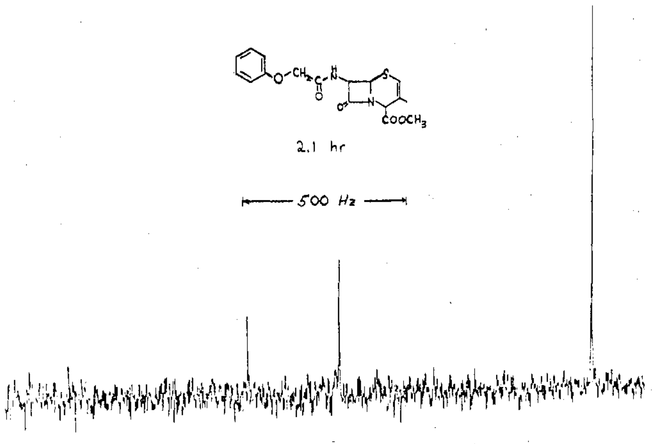


Figure 1. Natural-abundance ^{15}N spectrum of Δ^2-cephalosporin V methyl ester, obtained after 2597 transients with a 30° pulse width and 2.9-sec repetition rate. The highest field signal is that of the ammonium chloride reference.

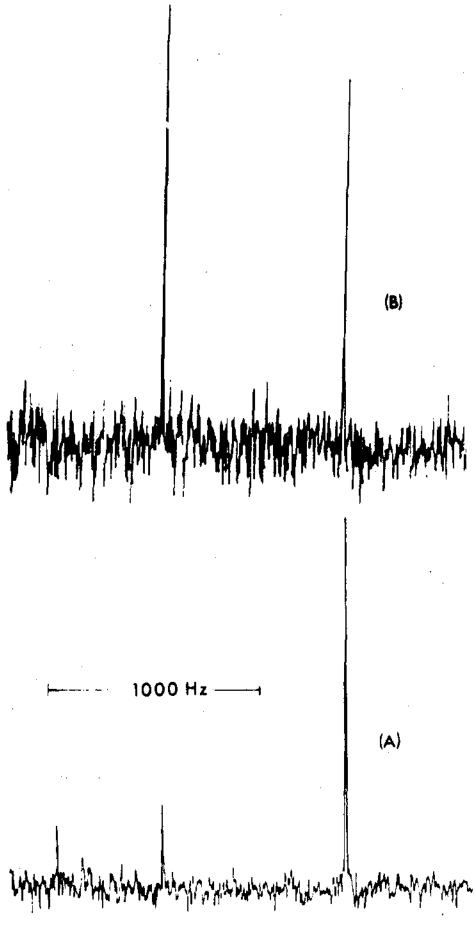
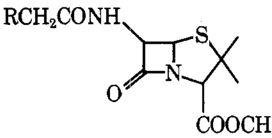
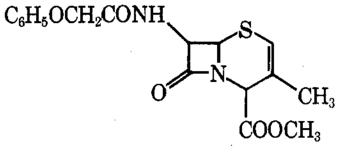


Figure 2. Natural-abundance ^{15}N spectra of penicillin G methyl ester (**1b**), obtained after 2000 transients with (a) a 30° pulse width and 3.2-sec repetition rate and (b) a 45° pulse width and 1.2-sec repetition rate. The highest field signal in each case is that of the ammonium chloride reference.

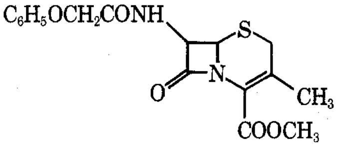
Structure-induced changes in the chemical shifts are consistent with observations on small molecules.[2a] Thus, introduction of an oxygen γ to a nitrogen causes an upfield shift (**1a** vs. **1b**) comparable with that observed in analogous ^{13}C spectra.[10] When **1a** is converted to its sulfoxide (**5**) both nitrogens experience upfield shifts but to different extents, probably reflecting both geometrical (cis vs. trans) and structural (sulfonyl vs. oxygen) contributions. Insofar as **3** and **6** are appropriate models, ionization of the 4-carboxyl group appears to have a larger effect on the chemical shift of the lactam nitrogen than would be observed for a carbon at the analogous position.[11]



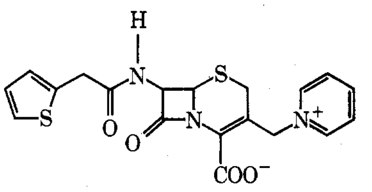
1a, R = C_6H_5O
1b, R = C_6H_5



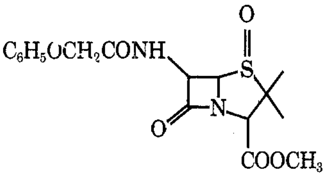
2



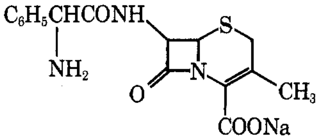
3



4



5



6

Of special interest are the lactam nitrogen resonance positions in **1–3**. The 30-ppm change in conversion from the five- to the six-membered ring (**1** vs. **2**) is not readily explicable. Because the lactam nitrogen in **2** is essentially planar,[12] increased amide conjugation would be expected to have induced a downfield shift. The corresponding nitrogen in **3** is less planar than in **2**, with concomitant decreased amide conjugation. Here, however, enamine-type conjugation is possible, and indeed the difference in resonance positions between **2** and **3** is comparable with that between cyclohexylamine and aniline.[2a] It may be that the nitrogen resonance position is insensitive to minor changes in the degree of amide delocalization.

Work is in progress to determine if the nitrogen chemical shifts of this class of compounds, including commercially available ones such as **4** and **6**, may be correlated with biological and pharmacological properties.

Acknowledgments. This work was supported by CUNY Faculty Research Award 10588 and by the U.S. Public Health Service, Grant 21148 from the Division of General Medical Sciences. Funds for the spectrometer were supplied in part by National Science Foundation Grant GP-37025. We are pleased to acknowledge the encouragement, advice, and samples provided by several research personnel of the Lilly Research Laboratories.

References and Notes

(1) Reported in part at the 16th Annual Experimental NMR Conference, Asilomar, Pacific Grove, Calif., April 23, 1975, and at the 169th National Meeting of the American Chemical Society, Philadelphia, Pa., April 11, 1975.

(2) CW mode: (a) R. L. Lichter and J. D. Roberts, *J. Am. Chem. Soc.*, **94**, 2495, 4904 (1972); (b) R. L. Lichter and J. D. Roberts, *Org. Magn. Reson.*, **6**, 636 (1974). FT mode: (c) J. M. Briggs, L. F. Farnell, and E. W. Randall, *J. Chem. Soc. D*, 680 (1971); (d) P. S. Pregosin, A. I. White, and E. W. Randall, *ibid.*, 1602 (1971); (e) T. Suzuki, T. Yamaguchi, and M. Imamari, *Tetrahedron Lett.*, 1809 (1974); (f) G. Adler and R. L. Lichter, *J. Org. Chem.*, **39**, 3547 (1974); (g) D. W. Moore and W. M. Tolles, 15th Annual Experimental NMR Conference, Raleigh, N.C., April 29, 1974; (h) A. J. DiGioia, E. H. Cole, and R. L. Lichter, 15th Annual Experimental NMR Conference, Raleigh, N.C., 29 April 1974; (i) R. L. Lichter, *J. Magn. Reson.*, **18**, 367 (1975).

(3) B. Coxon, *Carbohydr. Res.*, **35**, C1 (1974).

(4) In the case of **6** the concentration in fact is not known because the substance precipitated during the course of the experiment.

(5) E.g., R. A. Goodman, E. Oldfield, and A. Allerhand, *J. Am. Chem. Soc.*, **95**, 7553 (1973).

(6) L. F. Farnell, E. W. Randall, and A. I. White, *J. Chem. Soc., Chem. Commun.*, 1159 (1972).

(7) Proton noise-decoupled spectra were obtained in 10-mm o.d. tubes using a JEOL PS/PFT-100 spectrometer equipped with the EC-100 data system and fitted with a probe insert designed to improve sensitivity.[2h] In general, a frequency range of 2 kHz with 8K words for FID accumulation was employed, using 30–45° pulses and repetition rates of 3–4.5 sec. No attempts were made to optimize these conditions, which were chosen because spectra could be obtained reliably.

(8) R. L. Lichter and J. D. Roberts, *J. Am. Chem. Soc.*, **93**, 3200 (1971).

(9) P. Hampson and A. Mathias, *Mol. Phys.*, **11**, 541 (1966).

(10) (a) R. A. Archer, R. D. G. Cooper, P. V. DeMarco, and L. F. Johnson, *Chem. Commun.*, 1291 (1970); (b) E. L. Eliel, W. F. Bailey, L. D. Kopp, R. L. Willer, D. M. Grant, R. Bertrand, K. A. Christensen, D. K. Dalling, M. W. Duch, E. Wenkert, F. M. Schell, and D. W. Cochran, *J. Am. Chem. Soc.*, **97**, 322 (1975).

(11) J. B. Stothers, "Carbon-13 NMR Spectroscopy", Academic Press, New York, N.Y., 1972, pp 147, 150.

(12) R. M. Sweet, in "Penicillins and Cephalosporins", E. H. Flynn, Ed., Academic Press, New York, N.Y., 1972, Chapter 7.

Robert L. Lichter*
Department of Chemistry
Hunter College of the City University of New York
New York, N.Y. 10021

Douglas E. Dorman
Lilly Research Laboratories, Eli Lilly and Company
Indianapolis, Indiana 46202

Received August 11, 1975

Silver(I)-Promoted Reactions of Strained Hydrocarbons. Oxidation vs. Rearrangement

Summary: The reaction between silver trifluoroacetate and quadricyclene in methanol has been studied with the aid of an interfaced GC–MS data acquisition system. In addition to a Bronsted acid and silver metal, the products include norbornadiene (**2**), 3-methoxy-5-norbornene (**3**), 3-methoxynortricyclene (**4**), 3-trifluoroacetoxy-5-norbornene (**5**), 3-trifluoroacetoxynortricyclene (**6**), three isomeric 3,5-dimethoxynortricyclenes (**7**, **8**, and **9**), 3-methoxy-5-trifluoroacetoxynortricyclene (**10**), 3-methoxy-5-hydroxynortricyclene (**11**), and methyl trifluoroacetate. The formation of the various products as a function of time was monitored by gas chromatography. Finally, the formation of **7**, **8**, **9**, **10** and **11** along with acid and Ag^0 is interpreted as a two-electron redox reaction.

Sir: We wish to present evidence that oxidation occurs when certain strained hydrocarbons are treated with Ag(I) salts. Such a process was first proposed in 1971 by Kaiser, Childs, and Maitlis, who studied the action of various Lewis acids on tri-*tert*-butylprismane,[1] but their suggestion has since received little attention. We have been investigating some Ag(I)-promoted reactions of various quadri-