

¹H-NMR and NOE Studies of 2-Cephems and 3-Cephems

Chaeuk Im¹, Chul Bu Yim¹, Ronald G. Micetich² and Mohsen Daneshtalab²

¹College of Pharmacy, Chung-ang University, Heuksuk-Dong, Dongjak-Ku, Seoul 156-756, Korea and ²Synphar Laboratories Inc., 4290-91A street, Edmonton, Alberta, Canada T6E 5V2

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The ¹H-NMR signals of 2-cephems and 3-cephems have been assigned and the Nuclear Overhauser Effect (NOE) study of these compounds was undertaken.

Key words : 2-Cephem, 3-Cephem, ¹H-NMR, Nuclear overhauser effect

INTRODUCTION

In our research for elastase inhibitors, we have synthesized cephalosporin derivatives and studied their biological activity. We prepared 2-cephems and 3-cephems as intermediates (Scheme 1) and their NOE studies were undertaken to help the interpretation of ¹H-NMR spectra, especially for the C2-H, C4-H and C3-CH₂ in 2-cephems and C2 α -H, C2 β -H and C3-CH₂ in 3-cephems. In this paper, we report the ¹H-NMR assignments of title compounds and also discuss their NOE data.

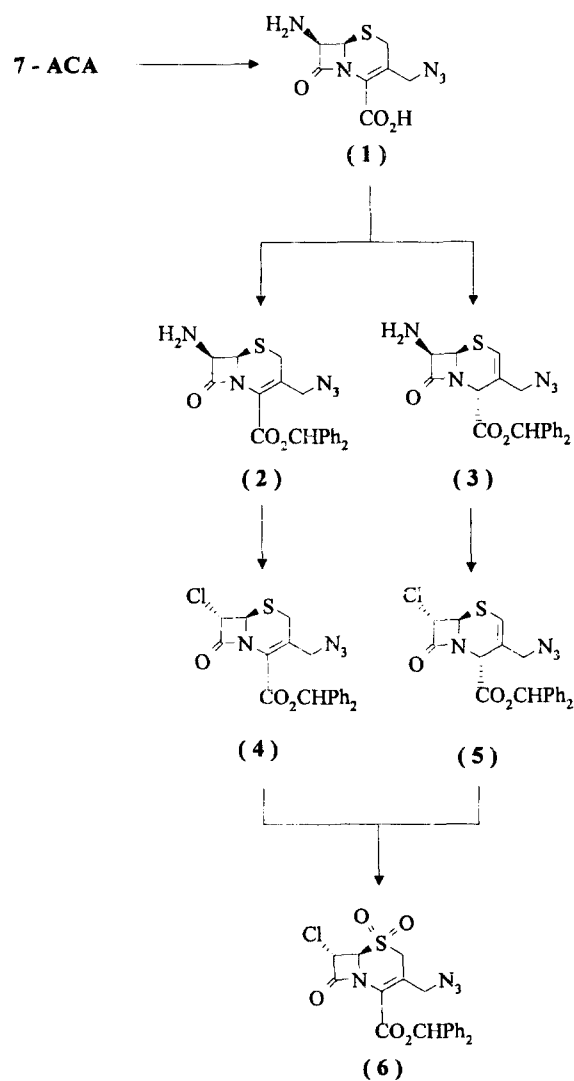
MATERIALS AND METHODS

The ¹H-NMR spectra were recorded on a Bruker AC-200E spectrometer at 25°C in CDCl₃ with tetramethylsilane (TMS) as an internal standard. Coupling constants (J) of the ¹H-NMR spectra are reported in Hertz (Hz). NOE data were collected using the multiple irradiation method (Neuhaus, 1983) with preirradiation times varying from six to ten seconds. NOE data are reported as percentage increases over the nonirradiated peak integration values.

RESULTS AND DISCUSSION

The coupling constants between the C6-H and C7-H (J_{6,7}) in the compounds (1, 2, and 3) are 5.1, 5.2, and 4.1 Hz, respectively, which indicated that these protons are in a *cis* relationship, i. e., C6 α -H and C7 α -H. However, this coupling constant in compound (4) is 1.3 Hz and in compound (5 and 6), they are so small that the peaks for C7-H become singlets. It sug-

gested that these protons are in a *trans* relationship to each other, i. e., C6 α -H and C7 β -H. The NMR data of



Scheme 1. Synthetic route to 2-Cephems and 3-Cephems.

Correspondence to: Chaeuk Im, College of Pharmacy, Chung-ang University, Heuksuk-Dong, Dongjak-Ku, Seoul 156-756, Korea

Table I. ¹H-NMR data of compounds (1, 2, 4 and 6) (δ)

Compounds	C2α-H	C2β-H	CO ₂ CH	C6-H	C7-H	C3-CH ₂ N ₃
1 (DMSO-d ₆)	3.63 (d, 18.1 Hz)	3.42 (3, 18.1 Hz)	6.98 6.97	4.99 (d, 5.1 Hz)	4.81 (d, 5.1 Hz)	3.89, 4.37 (two d, 13.3 Hz)
2 (CDCl ₃)	3.57 (d, 18.4 Hz)	3.41 (d, 18.4 Hz)	6.96	4.96 (d, 5.2 Hz)	4.87~4.72 (m)	3.96, 4.23 (two d, 13.9 Hz)
4 (CDCl ₃)	3.51 (d, 18.1 Hz)	3.35 (d, 18.1 Hz)		4.76 (d, 1.3 Hz)	4.69 (d, 1.3 Hz)	3.92, 4.18 (two d, 14.1 Hz)
6 (CDCl ₃)	4.03 (d, 18.1 Hz)	3.81 (d, 18.1 Hz)		5.33 (d, 1.7 Hz)	4.79 (br. s)	4.15, 4.27 (two d, 14.9 Hz)

Table II. ¹H-NMR data of compounds (3 and 5) (δ)

Compounds	C4-H	C3-CH ₂ N ₃	C2-H	CO ₂ CH	C6-H	C7-H
3 (CDCl ₃)	6.38 (br. s)	3.85 3.87	5.10 (br. s)	6.89 6.90	5.18 (d, 4.1 Hz)	4.60 (m)
5 (CDCl ₃)	6.30 (d, 0.9 Hz)		5.14 (d, 0.9 Hz)		4.77 (br. s)	5.03 (br. s)

Table III. NOE data of compounds (2, 3, 4, 5 and 6)^a

Compounds	Irradiated protons	Observed protons (δ)	Increased intensity (%) ^b
2	C3-CH ₂ N ₃	High-field C2-H (3.41)	4
		CO ₂ CH (6.98)	1
2	C6-H	Low-field C2-H (3.57)	2
3	C3-CH ₂ N ₃	C2-H (5.10)	21
		C4-H (6.38)	12
	C6-H	C2-H (5.10)	Nil
		C4-H (6.38)	Nil
4	C3-CH ₂ N ₃	High-field C2-H (3.35)	4.3
		Low-field C2-H (3.51)	3.2
	C6-H	CO ₂ CH (6.97)	1
		Low-field C2-H (3.51)	2
5	C3-CH ₂ N ₃	C2-H (5.14)	13
		C4-H (6.30)	7
	C6-H	C2-H (5.14)	Nil
		C4-H (6.30)	Nil
6	C3-CH ₂ N ₃	High-field C2-H (3.81)	3
		CO ₂ CH (6.96)	1
		Low-field C2-H (4.03)	2

^aSample concentration was 5% w/v with TMS as internal field frequency lock in CDCl₃^bGiven as the percentage increase in integrated intensity on irradiation

compounds (1, 2, 4 and 6) are presented in Table I and that of compounds (3 and 5) are shown in Table II.

In the compound (6), there are two doublets for the C2-H₂ at δ 3.81 and 4.03. The higher field doublet (δ 3.81) could be assigned for the C2β-H and the lower field doublet (δ 4.03) for C2α-H. This assignment is supported by the following observations. First, the lower field doublet has a lower height and broader peaks than that of the higher field doublet. It may be due to small coupling (1.7 Hz) between the C2α-H and C6α-H, which is in a 1,3-cis-diaxial relationship. This unique long range coupling has been also reported to exist in 3-cephem-β-sulfoxides. Although the origin of this coupling is not apparent, it is believed that the β-sulfoxide bond is in some way responsible for

the transfer of spin coupling information from the C2 α-H to C6α-H (Cooper *et al.*, 1970). Second, the C6 α-H shows a doublet instead of a singlet due to this coupling. In compounds (1, 2 and 4), similar lower height and broader peaks of the lower field doublets were also found, even though apparent coupling between C2α-H and C6α-H were not observed in 200 MHz spectra.

In compounds (3 and 5), the C3-CH₂ peaks are singlets, but the compounds (1, 2, 4, and 6) show two doublet peaks for these protons. These observations may be explained as follows.

Cooper proposed that C2, C3, C4, N5 and C4-carbonyl group were laid in a plane for 3-cephem compounds (Cooper *et al.*, 1970). In this stereochemical

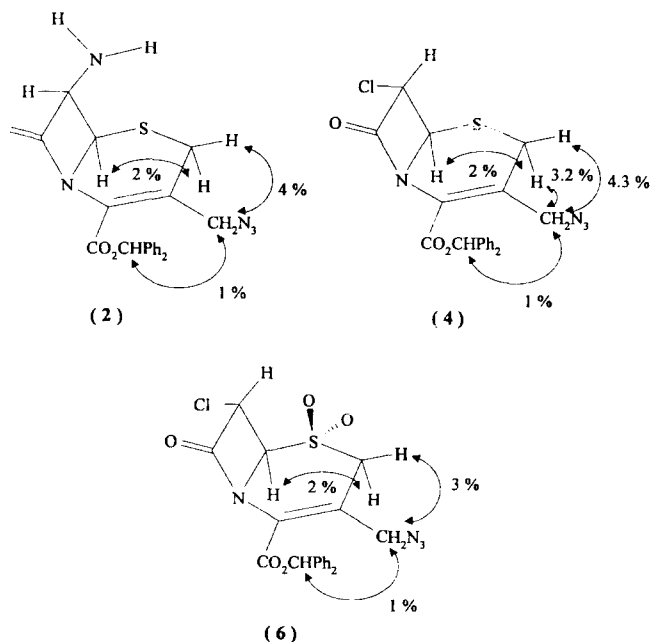


Fig. 1. Dihydrothiazine ring conformation and NOE data of 3-Cephems.

conformation, the C3-CH₂ and C4-carbonyl group are close in co-plane and may interact sterically and/or electronically. So it may prevent free rotation of the C3-CH₂ bond. This results in magnetically non-equivalent protons for C3-CH₂, which show two doublet peaks in their NMR spectra. A similar observation was observed in compounds (1, 2, 4 and 6).

NOE data of compounds (2, 4 and 6) support the above conformation (Table III). From the NOE data of compounds (2 and 4), the C₆-H would be close to the low field C2-H and CH₂N₃ would be close to high field C2-H. These observations suggest that the conformation of compounds (2 and 4) may be similar to the conformation in Fig. 1. The assignment of two C2-H doublets is based on the fact that the C₆-H should be close to C2 α -H and the CH₂N₃ should be close to C2 β -H. 2% NOE between the C₆-H and the lowfield C2-H and 4 and 4.3% NOE between CH₂N₃ and the high field C2-H indicate that the lowfield C2-H could be assigned to C2 α -H and the high field C2-H to C2 β -H. A similar assignment was previously reported (Demarco *et al.*, 1972). The NOE data of compound (6) were similar to compounds (2 and 4), so that the conformation and assignment of the C2-H should be similar. The small coupling (1.7 Hz) between the C2 α -H and the C6 α -H also support this conformation.

Cooper suggested that the 2-cephem compounds had a different conformation similar to Fig. 2 (Cooper *et al.*, 1970). In this model, the dihydrothiazine ring does not lie in a plane, so that C3-CH₂ group would have less steric and/or electronical interaction with C4-carbonyl group than in the 3-cephems. This may

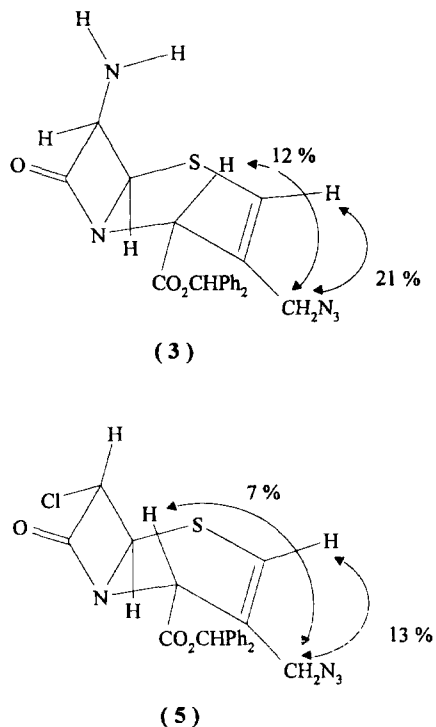


Fig. 2. Dihydrothiazine ring conformation and NOE data of 2-Cephems.

permit free rotation of the C3-CH₂ bond to give singlets for the C3-CH₂ in compounds (3 and 5). The NOE data of compounds (3 and 5) not only support above conformation, but also confirm the β -configuration of the C4-H, i.e., C4 β -H (Table III). There are 21 and 13% NOE between the C2-H and C3-CH₂, 12 and 7% NOE between C3-CH₂ and C₄-H and no NOE between the C₆-H and C2-H, C₄-H.

Another strong support for this chair form conformation comes from the coupling constant between the C2-H and the C4-H. Since the magnitude of this coupling constant is dependent on the angle between the plane of the double bond (i.e., H-C2=C3-C4 plane) and adjacent C4-H bond, the allylic coupling constant involving the C2-H and C4-H in 2-cephems would be stereospecific. In allylic coupling, the π electrons of the double bond apparently help to transmit the spin information from one nucleus to the other. When all nuclei are co-planar, there is no interaction of the allylic C-H bond orbital with the π system so that the allylic coupling constant become close to 0 Hz. However, when the allylic C-H bond is perpendicular to the C=C plane, the interaction between the π electrons and the C-H bond orbital would reach the maximum value, 3 Hz (Pavia *et al.*, 1996). The coupling constant ($J_{2,4}$) for compound (5) is 0.9 Hz but in compound (3), it is so small that the peaks for C2-H and C4-H become broad singlets. This result correspond with NOE data. The larger NOE between CH₂N₃

and C4-H of compound (3) than that of compound (5) suggests that C4-H in compound (3) come closer to CH₂N₃ and become co-planar to C2-H. It may result in smaller allylic coupling constant than in compound (5). The similar allylic couplings have been also reported by Cooper (Cooper *et al.*, 1970).

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