Tetrahedron Letters No. 15, pp 1327 - 1328. 0040-4039/79/0408-1327$02.00/0

^{1}H-NMR STUDY ON ACYLAMINO ACIDS. RELATIONSHIP BETWEEN THE CHEMICAL SHIFT OF AMIDE PROTONS AND ELECTRONEGATIVITY OF ACYL GROUPS

Yasuyuki Shimohigashi,* Tetsuo Kato, Shinwon Kang, Yoshihiro Minematsu, Michinori Waki and Nobuo Izumiya

Laboratory of Biochemistry, Faculty of Science, Kyushu University 33, Higashi-ku, Fukuoka 812, Japan

Summary: The chemical shift of amide protons of acylamino acids showed a linear relation with electronegativity of the acyl groups. Prediction of the carbonyl group participated in intramolecular hydrogen bonding was discussed, gramicidin S being used as an example.

The relationship between the chemical shift of substituted hydrocarbons and electronegativity of the substituents is typically shown in Shoolery's rule,[1] however, no such a rule has been reported with respect to amide protons. Here we report the effect of the electronic character of carbonyl groups on the ^{1}H-NMR chemical shift of amide protons of amino acids acylated with variously electronegative acyl groups.

Formyl, acetyl and trifluoroacetyl-L-Phe were prepared from L-phenylalanine dissolved in appropriate acids and acid anhydrides. Monochloroacetyl, dichloroacetyl and trichloroacetyl-L-Phe were prepared from L-phenylalanine and the respective acid chlorides by Schotten-Baumann reaction. Three acyl($C_nH_{2n+1}CO$; n=2,3,4)-L-Phe were prepared from L-phenylalanine and the corresponding carboxylic acids by mixed anhydride method. Other amino acids (L-alanine, L-leucine and *O*-benzyl-L-serine) were acylated in a similar manner. All the melting points of crystalline products accorded with those shown in the literature. All the acylamino acids were dissolved in DMSO-d_6 and ^{1}H-NMR spectra were recorded on a JEOL JNM PS-100 spectrometer at 29°C, tetramethylsilane being used as an internal reference.

The amide proton chemical shift of acetyl-L-Phe was independent of concentration (from 24 mM to 1920 mM) in DMSO-d_6, suggesting the absence of intermolecular hydrogen bonding. Table 1 shows the coupling constants $J_{NH-C\alpha H}$ of several acyl-L-Phe. Similarities in the patterns of splitting, in the chemical shifts of $C\alpha H$, $C\beta H_2$ and aromatic protons and in the coupling con-

Table 1. Vicinal coupling constants of R-CO-L-Phe.

R	CH_3CH_2	CH_3	H	$ClCH_2$	Cl_2CH	Cl_3C	F_3C
$J_{NH-C\alpha H}$ (Hz)	8.2	8.1	8.0	8.0	8.0	8.2	8.8

stants among most acyl-L-Phe indicate various acyl-L-Phe are in a similar conformation. Therefore, wide distribution in the chemical shifts of amide protons observed from 8.0 ppm to 9.74 ppm is attributed to the effect of different acyl groups. Ferguson proposed the use of pK_a value of carboxylic acid R-CH_2-COOH as the measure of relative electronegativity of group R.[2] In Fig. 1, the chemical shifts of amide protons are plotted against pK_a values of carboxylic

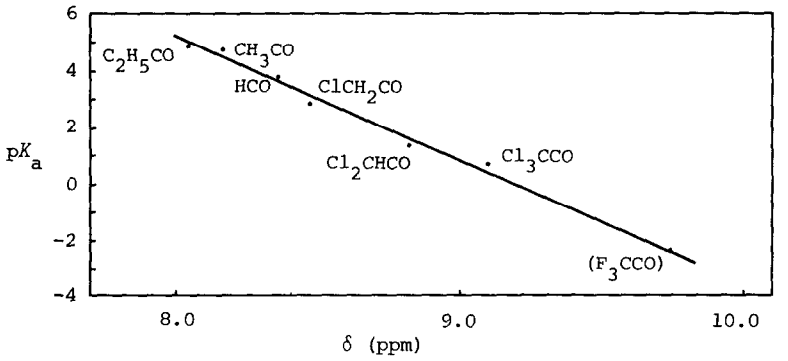

Fig. 1. Relationship between chemical shifts of amide protons and pK_a values used for acylation. In case of trifluoroacetic acid, acidity function was used instead of pK_a.[3]

acids used for acylation. A linear relationship exists between the chemical shifts and pK_a values or electronegativity of the acyl groups. Formula (1) shows the relation. The same relation was observed in ^{1}H-NMR spectra of other acylamino acids.

$$\delta_{(ppm)} = -0.22pK_a + 9.18 \qquad (1)$$

No methods have ever been developed to identify hydrogen bonded carbonyl with ^{1}H-NMR.[4] However, the results of this study can be applied for prediction of the carbonyl group participated in hydrogen bonding. For example, amide proton of the L-Leu residue in gramicidin S, adjacent to exposed carbonyl, appears at 8.35 ppm in DMSO-d_6, while, amide protons of the D-Phe and L-Leu residues, adjacent to the intramolecularly hydrogen bonded carbonyls, shift downfield to 9.15 ppm and 8.7 ppm, respectively.[5] Observation on tuberactinomycin N[6] can also be explained in terms of these results.

References

1. J.N. Shoolery in "Tech. Inform. Bull.", Vol. 2 (No. 3), Varian Assoc., Palo Alto, Calif. (1959).
2. L.N. Ferguson in "The Modern Structural Theory of Organic Chemistry", Chap. 2, Prentice-Hall Inc., Englewood Cliffs, N.J. (1963).
3. N. Tokura and M. Nojima, *J. Syn. Org. Chem. Jpn.*, 33, 854 (1975).
4. Y. Kyogoku and K. Kawano in "Biologically Active Peptides Produced by Microorganisms", pp. 280, H. Umezawa, T. Shiba and T. Takita, Eds., Kyoritsu Shuppan, Tokyo (1976).
5. A. Stern, W.A. Gibbons and L.C. Craig, *Proc. Natl. Acad. Sci. U.S.A.*, 61, 734 (1968); T. Kato and N. Izumiya in "Chemistry and Biochemistry of Amino Acids, Peptides, and Proteins", Vol. 2, pp. 1, B. Weinstein, Ed., Marcel Dekker, New York, N.Y. (1974).
6. T. Wakamiya and T. Shiba, *Bull. Chem. Soc. Jpn.*, 48, 2502 (1975).

(Received in Japan 16 January 1979)