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Ultrasonic Absorption Studies of the Nickel(II) Amino Acid Complexes in Aqueous Solutions

Shoji Harada, Kazumi Higashi, Kiyoshi Tamura, Michinori Hiraishi,
and Tatsuya Yasunaga*

Department of Chemistry, Faculty of Science, Hiroshima University, Higashisenda-machi, Hiroshima 730

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The ultrasonic absorption has been measured in aqueous solutions of Ni(II) complexes of glycine, β-alanine, 4-aminobutyric acid, and 6-aminohexanoic acid. The relaxation absorption could not be ascribed to one of the steps of the ordinary step-by-step complex formation mechanism. Kinetic data were obtained as functions of the initial concentrations of Ni(II) and amino acids, the pH, and the temperature. The relaxation absorption was attributed to the formation-breaking equilibrium of an intramolecular hydrogen bond due to the carboxylato group of amino acid coordinated to the Ni(II) ion. The rate constant of the hydrogen bond-breaking process has been obtained. The relationship between the relaxation parameters and the acid dissociation constant of the carboxyl group of the amino acid ligands is discussed with reference to the analogous result for the carboxylato ligands reported previously. Supplementary work is also carried out for Ni(II)-aspartato and glutamato complexes.

With the help of the relaxation techniques, the kinetics of the metal complex formation has been widely studied, and a step-by-step complex formation mechanism involving the rate-determining release of the solvent molecule from the inner-sphere of the metal ion has been proposed.1) Most of the ultrasonic relaxation absorptions in the metal complex solutions have been interpreted in terms of the above mechanism.2–4) In a previous paper,5) the present authors have reported that the ultrasonic relaxation absorption in the Ni(II)–carboxylato solutions can be ascribed to a reaction different from one of the steps of the above mechanism. Recently, the same kind of relaxation absorption has also been found in the Ni(II)–amino acid complex solutions.

The metal–amino acid complexation is not only interesting in itself, but is also important as the model system for the metal–protein interaction, and so it has been extensively studied from the static6) and the kinetic7–11) points of view. The purposes of the present investigation are to assign the relaxation absorption in the MHz frequency range, to obtain the kinetic parameters of the very fast reaction related to this absorption, and, moreover, to obtain information concerning the details of the Ni(II)–amino acid complex formation reaction. Studies were carried out for the Ni(II) complexes of glycine(gly), β-alanine(β-ala), 4-aminobutyric acid(ABA), and 6-aminohexanoic acid(AHA). Supplementary work was also performed for the complexes of aspartic acid(asp) and glutamic acid(glu).

Experimental

All the chemicals used were of a reagent grade and were used without further purification. Deionized and distilled water was used for the preparation of all solutions. Sample solutions were freshly prepared by dissolving $Ni(NO_3)_2$ and amino acid in water and by adjusting the pH by the drop-by-drop of dilute NaOH and/or HNO_3. The pH was adjusted to an appropriate value by taking into account the dissociation constant, K_a, of carboxyl group of amino acid and the hydrolysis of the Ni(II) ion. The ionic strength was kept constant at 2, with some exceptions, by addition of $NaNO_3$.

The ultrasonic absorption was measured by the pulse method12) in the frequency range from 3.5 to 95 MHz. The sound velocity was measured at 1.92 MHz by the sing-around method. All the measurements were carried out at 25.0± 0.05 °C. All of the absorptions were expressed by the following single-relaxation equation for the excess absorption per wavelength $(\alpha'\lambda)$:13)

$$(\alpha'\lambda) = 2(\alpha'\lambda)_{\max}\cdot\frac{(f/f_r)}{1+(f/f_r)^2} \qquad (1)$$

were α' is the excess absorption coefficient;14) λ, the wavelength; f, the frequency, and f_r, the relaxation frequency; the subscript, max, indicates the maximum value.

A blank solution of either $Ni(NO_3)_2$ or amino acid showed no discernible relaxation absorption. These results suggest that the present relaxation absorption is ascribed to the Ni(II)–amino acid complex formation reaction, but to neither the hydrolysis of the Ni(II) ion nor to the protonation of amino acid.15,16)

Results and Discussion

The relaxation absorption was studied as functions of Ni(II) and amino acid concentrations by keeping one constant and varying the other. The results are summarized in Table 1, where three features may be noted. The first is the independence of the relaxation frequency of both the Ni(II) and amino acid concent-

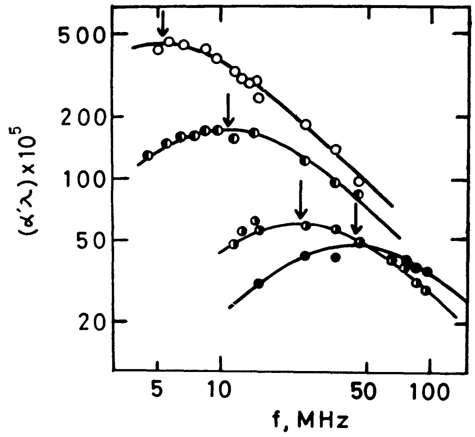


Fig. 1. Ultrasonic absorptions of the Ni(II)–amino acid complex solutions at 25 °C: ∑Ni=1.0 M, ∑amino acid=2.0 M.
(●): Ni(II)-gly, (◑): Ni(II)-β-ala,
(◐): Ni(II)-ABA, (○): Ni(II)-AHA.

Table 1. Ultrasonic parameters for the Ni(II)-amino acid complex solutions at 25 °C

$\sum$Ni (M)	$\sum$L (M)	pH	f_r (MHz)	$(\alpha'\lambda)_{\max}$ $\times 10^5$
		Glycine		
0.5	2.0	5.4	45	39
1.0	2.0	5.1	44	48
		β-Alanine		
1.0	1.0	5.3	25	40
1.0	1.5	5.3	23	53
1.0	2.0	5.3	25	61
1.0	2.5	5.3	23	70
		4-Aminobutyric acid		
0.5	0.5	5.3	11	17
0.5	1.0	5.9	12	55
1.0	0.5	5.5	12	40
1.0	2.0	6.4	11	173
1.5	0.5	5.2	11	53
		6-Aminohexanoic acid		
0.2	1.0	6.4	4.5	140
0.4	1.0	6.4	5.0	152
1.0	0.5	5.8	4.8	163
1.0	2.0	6.4	5.2	461
		Aspartic acid		
0.5	1.0	5.2	22	20
		Glutamic acid		
0.5	0.5	5.2	18	60
0.5	1.0	5.3	19	150

Table 2. pH dependence of the ultrasonic parameters for the Ni(II)-glutamic acid complex solutions at 25 °C

$\sum$Ni (M)	$\sum$Glu (M)	pH	f_r (MHz)	$(\alpha'\lambda)_{\max}$ $\times 10^5$
1.0	0.5	3.7	16	73
1.0	0.5	3.5	23	66
1.0	0.5	3.3	28	64
1.0	0.5	3.0	32	51

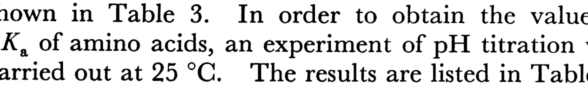
Table 3. Temperature dependence of the ultrasonic parameters for the Ni(II)-4-aminobutyric acid complex solutions

$\sum$Ni (M)	$\sum$ABA (M)	pH	t (°C)	f_r (MHz)	$(\alpha'\lambda)_{\max}$ $\times 10^5$
1.0	2.0	5.9	10	8.0	139
1.0	2.0	5.9	20	10.8	162
1.0	2.0	5.9	25	11.5	173
1.0	2.0	5.9	30	13.5	196

rations. The second is the increase in the absorption with the increases in both the Ni(II) and amino acid concentrations. The third is the specificity of the ligand effect on the relaxation frequency and the excess absorption. Some representative absorptions of the Ni(II)-amino acid solutions are shown in Fig. 1.

The effect of the pH on the relaxation absorption was studied for the Ni(II)-glu system. As may be seen in Table 2, a decrease in pH causes an increase in f_r and a decrease in the excess absorption. The temperature dependence of the relaxation absorption was studied for the Ni(II)-ABA system; the results are shown in Table 3. In order to obtain the value of pK_a of amino acids, an experiment of pH titration was carried out at 25 °C. The results are listed in Table 4.

The complex formation reactions of Ni(II) with amino acids (*e.g.*, gly[7] or β-ala[8]) have been expressed as follows:



$$\underset{①}{\mathrm{NiL}_{n-1} + \mathrm{L}} \underset{k_{21}}{\overset{k_{12}}{\rightleftharpoons}} \underset{②}{\mathrm{NiL}_{n-1}\mathrm{O}\genfrac{}{}{0pt}{}{\diagup\mathrm{H}}{\diagdown\mathrm{H}}\ \mathrm{L}} \underset{k_{32}}{\overset{k_{23}}{\rightleftharpoons}} \underset{③}{\mathrm{NiL}_n} \quad (n=1,\ 2,\ \text{or } 3) \qquad (2)$$

Here ① is the free ions, ② the outer-sphere complex, and ③ the inner-sphere complex; L is the ligand complexing with the metal (the charges have been neglected in this paper). To begin with, the present relaxation absorption was investigated by assigning it to one of these steps.

The outer-sphere complex formation process, ①$\rightleftharpoons$②, is considered to be very rapid, and the relaxation frequency of this process would lie, if observed, in a frequency range not lower than 200 MHz. Moreover, the ligand specificity should not be reflected in this process.[1,13] These characteristics were not, however, observed.

The inner-sphere complex formation, ②$\rightleftharpoons$③, is the rate-determining process, and the relaxation time $\tau(=(2\pi f_r)^{-1})$ is expressed by the following equation:

$$\tau^{-1} = 2\pi f_r = k_{23}\cdot\frac{K_{12}\{[\mathrm{NiL}_{n-1}]+[\mathrm{L}]\}}{1+K_{12}\{[\mathrm{NiL}_{n-1}]+[\mathrm{L}]\}} + k_{32}, \qquad (3)$$

where $K_{12}=k_{12}/k_{21}$.

When the following relations are introduced,

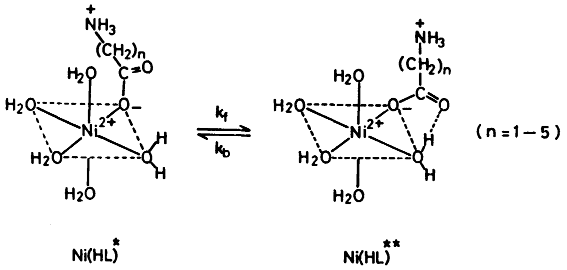
$$0 < \frac{K_{12}\{[\mathrm{NiL}_{n-1}]+[\mathrm{L}]\}}{1+K_{12}\{[\mathrm{NiL}_{n-1}]+[\mathrm{L}]\}} < 1, \qquad (4)$$

Eq. 3 gives

$$\tau^{-1} < k_{23} + k_{32}. \qquad (5)$$

The k_{23} values for the water release from NiL_{n-1} are: $3\times10^4\ \mathrm{s}^{-1}$ for $n=1$ (*i.e.*, Ni^{2+} ion), $1.2\times10^5\ \mathrm{s}^{-1}$ for $n=2$, and $4.0\times10^5\ \mathrm{s}^{-1}$ for $n=3$ when L=gly.[7] In the Ni(II)-amino acid system, $k_{32} \ll k_{23}$ is generally established. Then, the τ^{-1} value due to the ②$\rightleftharpoons$③ process cannot exceed $10^6\ \mathrm{s}^{-1}$ (or $f_r<0.2$ MHz), no matter how increased the ionic concentration is. When L=β-ala,[8] the chelate-ring closure step is known to be rate-determining. The rate constant of this process is smaller than that for the first coordination to the metal. The above discussion shows that the present relaxation absorption of $\tau^{-1}>3\times10^7\ \mathrm{s}^{-1}$ can not be ascribed to one of the steps of the inner-sphere complex-formation reaction where the release of the coordinated water molecule from Ni(II) is connected and rate-determining.

It is generally believed that the zwitterionic form of amino acid is unreactive and that only the anionic form is reactive for the complex formation.[7-11,17,18] Cassatt and Wilkins[10] discussed the unreactivity of the zwitterionic form of the amino acid and argued that even if the coordination is initiated by the carboxylato group, the bond breaks much faster than the deprotonation of the $-NH_3^+$ group, followed by a chelate formation. On the other hand, the results of the present relaxation absorption studies definitely show that relatively stable complexes are formed even in the case of the ABA and AHA systems, where the chelate complex is not conceivable. This implies that the zwitterionic form of amino acid can form a complex by the coordination of its carboxylato group to Ni(II), by analogy with the corresponding carboxylato ion. Some of the experimental evidence of the complex formation of the zwitterionic form has also been given by pH titration,[19] solubility,[20] and Raman spectral[21] investigations.



(6)

Fig. 2. Appropriate model of the intramolecular hydrogen bond equilibrium of the Ni(II)–amino acid complex.

The reaction associated with the relaxation absorption has the following features: (1) it involves the Ni(II) complex of the zwitterionic form of amino acid unidentated by its carboxylato group; (2) it is not concerned with the water release from Ni(II), and (3) it occurs very rapidly and has a time constant of the order of 10^{-7}—10^{-8} s. An appropriate mechanism is Reaction 6 in Fig. 2. This is similar to that earlier proposed by the present authors for the interpretation of the relaxation absorption of the Ni(II)–carboxylato solutions.[5] Further support for the assignment is as follows: (1) For Reaction 6, the relaxation frequency is related to the rate constants by this equation:[22]

$$2\pi f_r = k_f + k_b. \tag{7}$$

As the concentration term is not involved in this equation, the independence of f_r of the ionic concentrations can be approved.

(2) The maximum excess absorption per wavelength, $(\alpha'\lambda)_{max}$, for Reaction 6 is given by[13]

$$\begin{aligned}(\alpha'\lambda)_{max} &= \frac{\pi(\Delta V)^2}{2\beta_0 RT}\cdot\left\{\frac{1}{[Ni(HL)^*]}+\frac{1}{[Ni(HL)^{**}]}\right\}^{-1}\\ &= \frac{\pi(\Delta V)^2}{2\beta_0 RT}\cdot[Ni(HL)]\cdot\frac{K}{(1+K)^2},\end{aligned} \tag{8}$$

where $[Ni(HL)]=[Ni(HL)^*]+[Ni(HL)^{**}]$, $K=k_f/k_b$, ΔV is the reaction volume change, and β_0 is the adiabatic compressibility. This equation shows that $(\alpha'\lambda)_{max}$ is proportional to the Ni(HL) complex concentration. Since the stability constant for the complex of the zwitterionic form of amino acid, K_{HL},

$$K_{HL} = \frac{[Ni(HL)]}{[Ni]\cdot[HL]} \tag{9}$$

has not been reported for Ni(II), the absolute value of the Ni(HL) complex concentration cannot be known. However, the relative change can be assumed by replacing with $K_{HL}\cdot[Ni]\cdot[HL]$ (see Eq. 9). Figure 3 shows the $(\alpha'\lambda)_{max}$ *vs.* $[Ni]\cdot[HL]$ plot for the Ni(II)–β-ala system, for which the literature values of the stability constants[23] enabled us to calculate the ionic concentrations. The plot gives a straight line which cuts through the origin of the coordinates. This result conforms to Eq. 8.

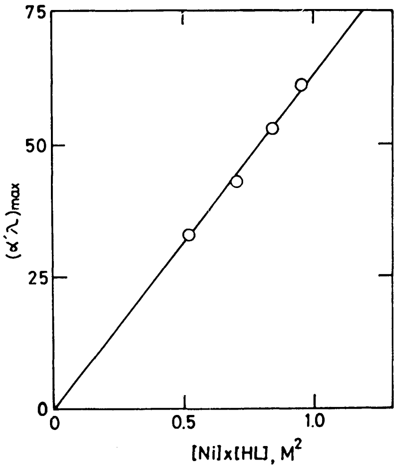


Fig. 3. $(\alpha'\lambda)_{max}$ *vs.* $[Ni]\cdot[HL]$ plot for the Ni(II)-β-ala system.

(3) As may be seen in the data of the Ni(II)–glu system in Table 2, the relaxation absorption decreases with the decrease in the pH. This result can be easily understood from the fact that the complex concentration decreases with the decrease in the concentration of the unprotonated ligand. On the other hand, however, the increase in f_r with the decrease in the pH cannot be easily understood because the decrease in the concentration of the unprotonated ligand generally induces a decrease in f_r. The conflict has now been resolved by considering the catalyzing effect of the hydrogen ion on the hydrogen-bond formation and breaking.[24] According to this idea, the increase in f_r can be considered to be due to that of the rate constant, k_b and/or k_f, induced by the increase in the proton concentration.

(4) The apparent activation energy can be obtained from the temperature dependence of f_r. For the Ni(II)–ABA system, $\Delta H^* = 4$ kcal mol^{-1} was obtained. This value is relatively small compared to those of the ordinary complex formations, but it is appropriate for the formation of a hydrogen bond.[25] The same value was also obtained in the case of the Ni(II)–carboxylato system.[5]

(5) One of the features of the relaxation absorption of the amino acid complexes is the intimate correlation between the pK_a value of the carboxyl group and the relaxation parameters, f_r and $(\alpha'\lambda)_{max}$. As is shown in Table 1 and Fig. 1, f_r increases and $(\alpha'\lambda)_{max}$ decreases with the decrease in the pK_a of the amino acid ligand. The same tendency was observed in the case of the Ni(II)–carboxylato complexes.[5] Moreover, as is shown in Fig. 4, the overlapping of the f_r *vs.* pK_a plots for the two groups suggests that the present absorption is caused

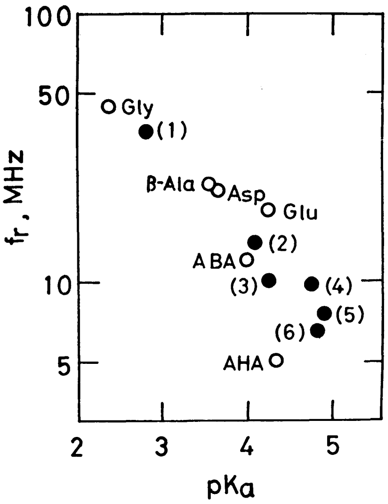


Fig. 4. f_r *vs.* pK_a plot for a series of Ni(II)–amino acid complexes(○) and Ni(II)–carboxylato complexes(●) at 25 °C: (1) chloroacetate, (2) 3-chloropropionate, (3) acrylate, (4) acetate, (5) propionate, (6) butyrate.

by the interactions between Ni(II) and the carboxylato group. If one assumes the $k_f \ll k_b$ relation, Eq. 7 can be reduced to

$$2\pi f_r = k_b. \tag{10}$$

The lower the value of pK_a of the carboxyl group is, the more labile the complex becomes. Analogously, the decrease of the pK_a value labilizes the hydrogen bond[26) in Ni(HL)**, which will induce the increase of the k_b value. Therefore the values of pK_a may be inversely proportional to f_r, as is shown in Fig. 4. The k_b values for the series of Ni(II)–amino acid complexes were obtained by means of Eq. 10: they are listed in Table 4. The magnitude of $(\alpha'\lambda)_{max}$ was in the order of the AHA>ABA>β-ala>gly systems under the same metal-ligand concentration, as is shown in Fig. 1. For a series of complexes, the ΔV value can be assumed to be the same. Therefore, from Eq. 8, the factors which contribute mainly to the value of $(\alpha'\lambda)_{max}$ seem to be the concentration and the K terms. Since the stability constant K_{HL} of Eq. 9 depends on the pK_a of the carboxyl group[26) and the electrostatic repulsion between the metal and the $-NH_3^+$ group,[27) the magnitude of the concentrations of Ni(HL) would seem to be in the order of: AHA>ABA>β-ala>gly. On the other hand, the larger the pK_a of a ligand is, the stronger will the intramolecular interaction between the carboxylato ion and the water molecule become. Therefore, the values of K in Eq. 8 would be in the order of: AHA>ABA>β-ala>gly. Whether the contribution of the concentration of Ni(HL) is predominant or that of the value of K, the ligand specificity observed in $(\alpha'\lambda)_{max}$ may, then, be well interpreted by the present mechanism.

Table 4. Rate constants of the intramolecular hydrogen bond-breaking process for the Ni(II)–amino acid complexes and the pK_a values of the carboxyl group of the amino acids

Ligand	pK_a	$k_b \times 10^{-7}$ (s^{-1})
Glycine	2.45[a)	28
β-Alanine	3.70[a)	15
4-Aminobutyric acid	3.94[b)	7.5
6-Aminohexanoic acid	4.34[b)	3.1
Aspartic acid	3.78[a)	14
Glutamic acid	4.25[a)	11

a) R. C. Weast, "Handbook of Chemistry and Physics," ed by J. Lewis and R. G. Wilkins, Interscience, New York, N. Y. (1960). b) This work.

The kinetic information obtained in this study of the Ni(II)–amino acid complexes, together with that reported previously on the Ni(II)–carboxylato complexes, provides a new type of equilibrium involving the coordination of the carboxylato group to the metal, as is shown in Reaction 6. As the relaxation effect is due to the motion of the carboxylato groups, the same kind of absorption can be expected to be observed in other divalent metal complexes of carboxylic acids and amino acids. Unfortunately, however, in other divalent metal systems, the relaxation effect based on the inner-sphere complex formation reaction falls into nearly the same time range as that of the present relaxation effect, and so it becomes rather difficult to study the latter effect separately. Studies of some of these systems are now in progess, however, and will be reported on it due course.

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