66. Formation and dissociation mechanism of amide complexes III[1]), Water substitution as the rate limiting factor for the interconversion of Cu^{2+} complexes with neutral and deprotonated amide groups

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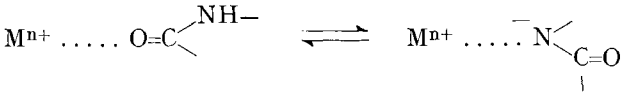
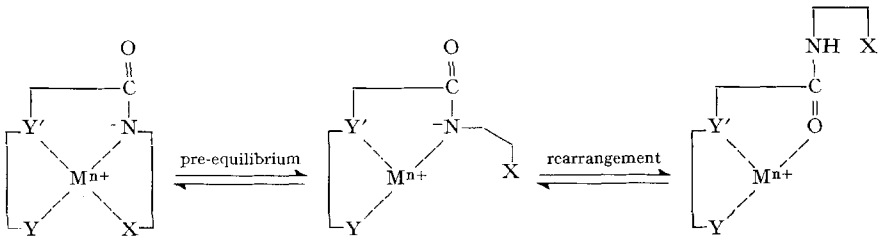
Summary. The protonation and deprotonation rates of the coordinated amide group in the Cu^{2+} complexes with N^{α}-(2-pyridyl-methyl)-glycinamide (I) and N^{α}-(2-pyridyl-methyl)-glycine-ethylamide (II) have been studied by stopped flow techniques. It is shown that the rate determining step of the formation of the complex with the deprotonated amide group is given by the rate of water dissociation from Cu^{2+}. Weaker bases than OH^- or stronger acids than water can react by a different path, in which the proton transfer and/or the rotation from the O-co-ordinated into the N-co-ordinated form and *vice versa* is rate determining.

In aqueous solution amides form two different types of complexes with heavy metal ions. The neutral amide group is bound through its carbonyl oxygen [2], whereas the deprotonated group co-ordinates *via* the nitrogen atom as in (1) [3].



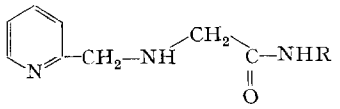
(1)

For Cu^{2+} and Ni^{2+} the formation and dissociation of the deprotonated complex is comparatively slow with oligopeptides such as triglycine [4] [5] and similar ligands like N,N'-bis-glycyl-1,3-diaminopropane [1] [6]. The kinetics can be directly followed by stopped flow or even classical techniques. Little is known, however, about the rate of the actual rearrangement (1) since pre-equilibria (2) with unknown influence were involved in the systems studied so far.



(2)

[1]) For communication II: see [1].



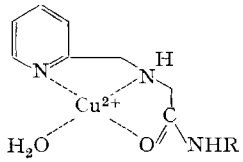
I: R = –H
II: R = $-C_2H_5$

In order to study reaction (1) without such interference, two ligands, N^{α}-(2-pyridyl-methyl)-glycinamide (I) and N^{α}-(2-pyridyl-methyl)-glycine-ethylamide (II) [2], in which the amide group is at one end of the molecule, have been chosen.

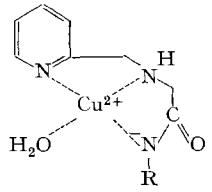
Experimental. – N^{α}-(2-Pyridyl-methyl)-glycinamide (I) and N^{α}-(2-pyridyl-methyl)-glycine-ethylamide (II) were synthesized as described [2]. Kinetic runs were followed spectrophotometrically at 591 nm for I and at 606 nm for II on a *Durrum* D110 stopped flow instrument with a 2 cm KelF cell thermostated at 25°. The deprotonation of the amide group was studied by mixing solutions containing 10^{-3} M $CuSO_4$ and $1.1 \cdot 10^{-3}$ M ligand dihydrochloride with 2,4,6-collidine (pH 7–8.2) or borate (pH 8.1–9) buffers. Protonation was studied by mixing fresh[2]) solutions of 10^{-3} M $CuSO_4$ and $1.1 \cdot 10^{-3}$ M ligand of pH > 9.5 with 2,4,6-collidine (pH 6.5–7.2), α-picoline (pH 5.1–6.6) or *m*-phenylene-diamine (pH 3.8–5) buffers. Experiments in which the initial pH of the complex solution was varied from 9–11 gave identical results.

The ionic strength was adjusted to 0.5 with KCl in all solutions. The organic buffer bases were freshly distilled before use. The other reagents were of analytical grade and used without further purification. Doubly distilled water was used throughout. In all cases the buffer concentration was varied from $2 \cdot 10^{-2}$ to $2 \cdot 10^{-1}$ M and the reaction rates were extrapolated to zero buffer concentration. Activation parameters were obtained from measurements at different pH values, each at various temperatures between 20° and 40°.

Results. – Equimolar mixtures of Cu^{2+} with I or II form complexes of structure III in weakly acidic solution, whereas above pH 8 deprotonation and rearrangement of the amide group to give IV occurs [2].



III



IV

The kinetic measurements were therefore evaluated assuming a reversible equilibrium between III and IV (3) with k_+ and k_- as rate constants for the forward

$$\text{III} \underset{k_-}{\overset{k_+}{\rightleftharpoons}} \text{IV} + \text{H}^+ \qquad (3)$$

and the reverse reactions. In all kinetic runs $[H^+]$ was kept constant and hence equilibrium was reached by a first order process with an effective rate constant $k_{obs} = k_+ + k_-$ [8]. The pH-dependence of k_{obs} is given in Fig. 1. Three distinct parts A, B and C can be seen:

[2]) Alkaline solutions of Cu^{2+} and I or II turn yellow-green after 2–3 days at room temperature. Probably an oxidation of the ligand takes place, similar to that observed by *Margerum et al.* for the Cu^{2+} and Ni^{2+} complexes with tetraglycine [7].

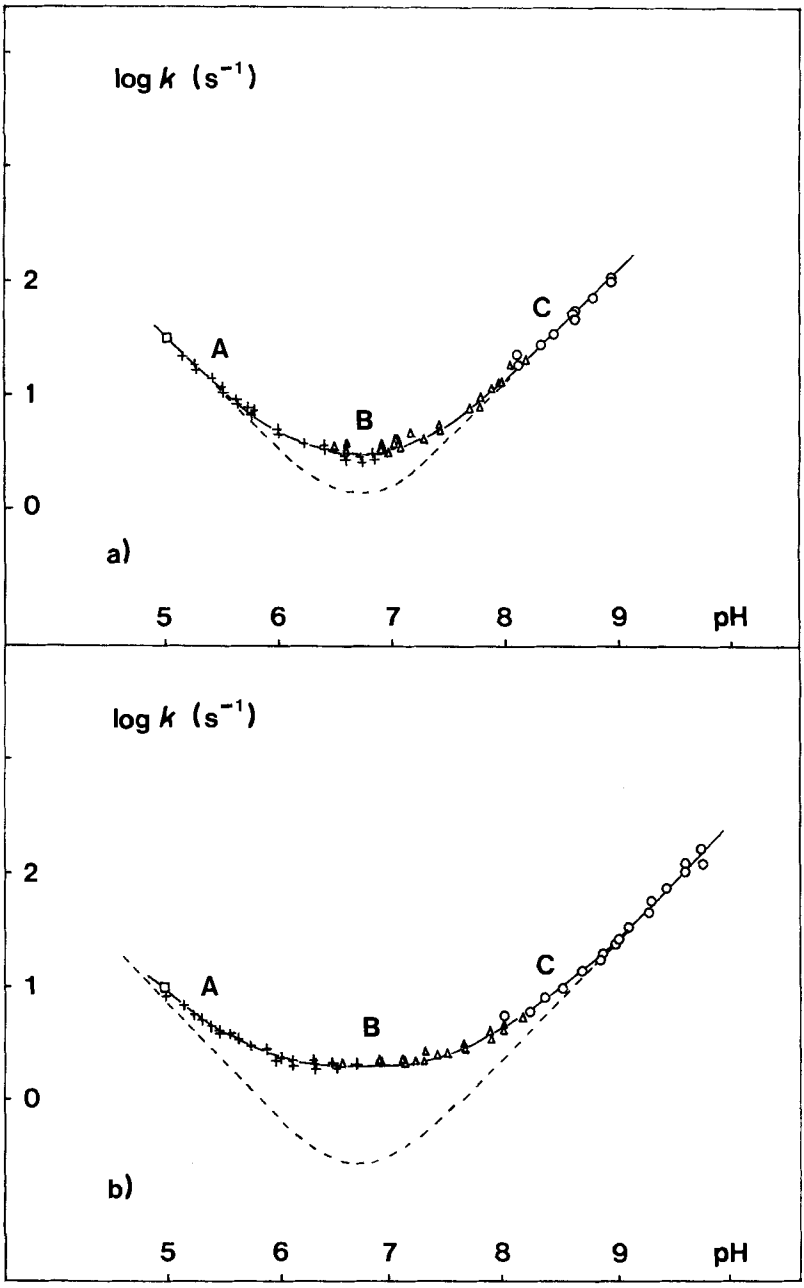


Fig. 1. *pH-dependence of the experimental rate constants k_{obs} for formation and dissociation of the deprotonated amide complex IV*

a) With N^{α}-(2-pyridyl-methyl)-glycinamide as ligand. b) With N^{α}-(2-pyridyl-methyl)-glycine-ethylamide as ligand. – – – calculated with k_H (4) and k_{OH} (5); —— calculated with k_H, k_{OH} and k_0. Buffers: □ *m*-phenylenediamine; + α-picoline; △ 2,4,6-collidine; ○ borate

In A, dissociation of IV is predominant and the rate is proportional to $[H^+]$ (4). C describes the formation of IV with a rate proportional to $[OH^-]$ (5). In order to calculate k_{OH} from C, $K_w = 1.85 \cdot 10^{-14}$ for $I = 0.5$ was used [9].

$$-d[IV]/dt = k_H \cdot [IV][H^+] \quad (4)$$

$$+d[IV]/dt = k_{OH} \cdot [III][OH^-] = k_{OH} \cdot K_w[III]/[H^+] \quad (5)$$

B is a broad minimum, where both the forward and the reverse reactions contribute to the rate. k_H and k_{OH} alone cannot explain the observed pH profile in Fig. 1, however. A term k_0 independent of pH has to be introduced, so that the experimental and calculated curves agree. The rate constants k_H, k_{OH} and k_0 and their activation parameters are shown in Table 1, together with the corresponding values for the Cu^{2+}/triglycine system.

Table 1. *Rate constants and activation parameters for the Cu^{2+} complexes with I, II and triglycine*

	I	II	triglycine
k_{OH} (M^{-1} s^{-1})	$6.7 \cdot 10^6$	$1.3 \cdot 10^6$	$2.5 \cdot 10^4$
ΔE^*_{OH} (kcal/mole)	16.8	–	–
$\log A_{OH}$ (M^{-1} s^{-1})	13.8	–	–
k_H (M^{-1} s^{-1})	$3.3 \cdot 10^6$	$7.1 \cdot 10^5$	$4.9 \cdot 10^6$
ΔE^*_H (kcal/mole)	16.4	–	–
$\log A_H$ (M^{-1} s^{-1})	12.8	–	–
$k_0 = k_{H_2O} \cdot [H_2O]$ (s^{-1})	2.1	2.3	$1.8 \cdot 10^{-3}$
k'_{H_2O} (M^{-1} s^{-1})	$4.7 \cdot 10^{-3}$ [a]	$1.3 \cdot 10^{-4}$	$4.5 \cdot 10^{-2}$
k_{PDA} (M^{-1} s^{-1}) [b]	$3.6 \cdot 10^3$	$2.0 \cdot 10^3$	–
k_{borate} (M^{-1} s^{-1})	$5.5 \cdot 10^2$	$1.7 \cdot 10^2$	$2.0 \cdot 10^2$

a) calculated from K'_{pot} and k_H.
b) PDA = *m*-phenylenediamine.

k_0 may be assigned either to k_{H_2O} (6) or k'_{H_2O} (7).

$$IV + H_2O \underset{k_{OH}}{\overset{k_{H_2O}}{\rightleftharpoons}} III + OH^-: K_{pot} = \frac{[III][OH^-]}{[IV]} \quad (6)$$

$$III + H_2O \underset{k_H}{\overset{k'_{H_2O}}{\rightleftharpoons}} IV + H_3O^+: K'_{pot} = \frac{[IV][H^+]}{[III]} \quad (7)$$

Table 2. *Equilibrium constants, obtained potentiometrically and kinetically, for the deprotonation of the complexes III with the neutral amide group*

	$-\log K_{pot}$ [a]	$-\log K_{kin}$	$-\log K'_{pot}$ [a]	$-\log K'_{kin}$
I	6.62	6.45	7.11	6.34
II	5.80	5.73	7.93	5.49

a) *cf.* [2].

Comparison of the equilibrium constants obtained kinetically $K_{kin} = k_{H_2O}/k_{OH}$ and $K'_{kin} = k'_{H_2O}/k_H$ with the corresponding values of K_{pot} and K'_{pot} [2] shows unambiguously (*cf.* Table 2) that k_0 describes the forward reaction of equilibrium (6). The rate constant k'_{H_2O}, kinetically not observable, may be calculated from k_H and K'_{pot}. For ligands I and II we obtain $k'_{H_2O} = 0.26\ s^{-1}$ and $k'_{H_2O} = 0.072\ s^{-1}$, or $4.7 \cdot 10^{-3}$ $M^{-1}\ s^{-1}$ and $1.3 \cdot 10^{-4}\ M^{-1}\ s^{-1}$ as bimolecular rate constants.

The effect of some buffers on k_{obs} is shown in Fig. 2. In the case of I, 2,4,6-collidine and α-picoline have no effect on the reaction rate. With the ethyl derivative II as ligand inhibition by these two buffers is observed. Optical spectra indicate that the pyridine buffer bases co-ordinate to III and IV whether derived from I or II. The inhibition observed for II may be explained by steric hindrance of the N-ethyl group with the co-ordinated pyridine base.

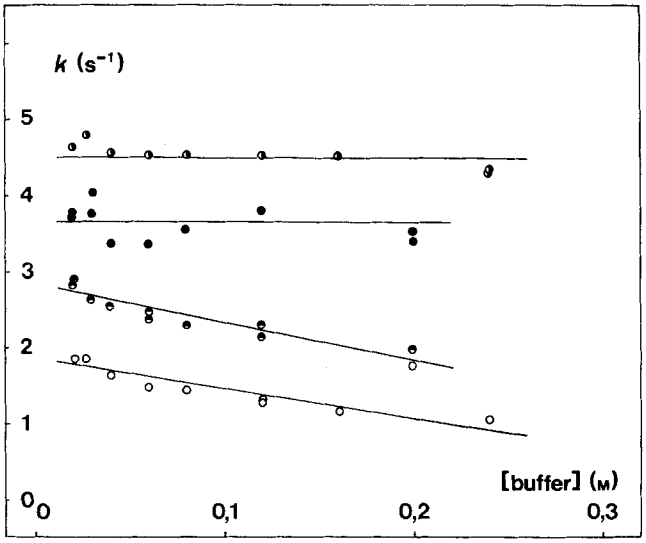


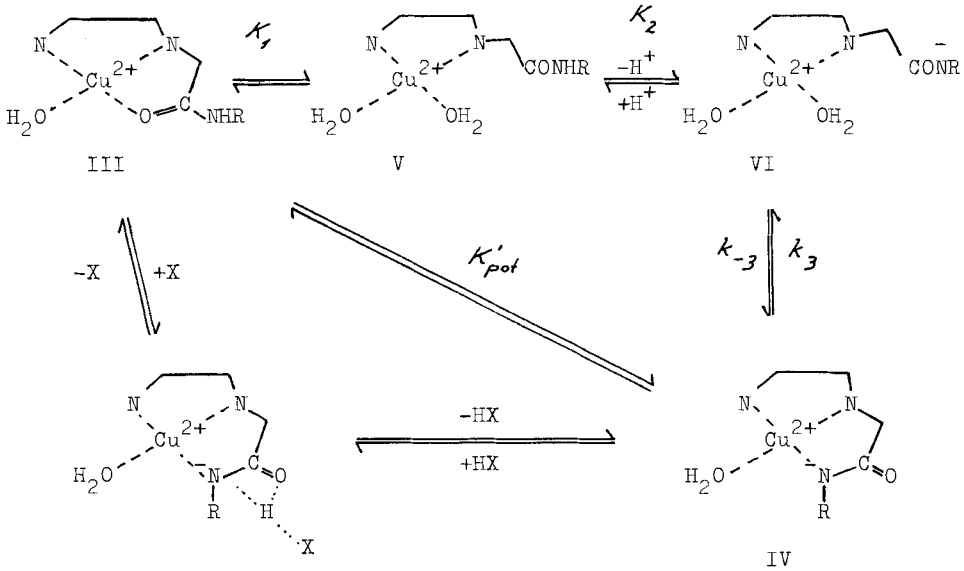
Fig. 2. *Influence of some buffers on the rate of reaction (3)*

◑ N^{α}-(2-Pyridyl-methyl)-glycinamide, α-picoline, pH = 6,0; ● N^{α}-(2-pyridyl-methyl)-glycinamide, 2,4,6-collidine, pH = 7,0; ◓ N^{α}-(2-pyridyl-methyl)-glycine-ethylamide, 2,4,6-collidine, pH = 7,0; ○ N^{α}-(2-pyridyl-methyl)-glycine-ethylamide, α-picoline, pH = 6.0

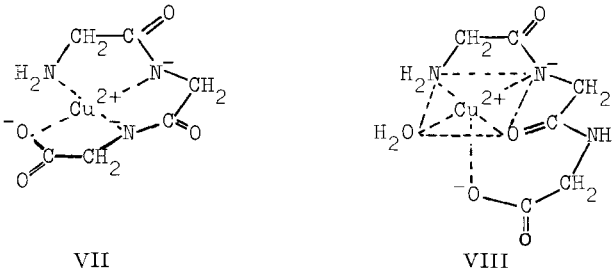
Discussion. – For rearrangement (1) a simple mechanism with the intermediates V and VI can be postulated (see Scheme). $K_1 = [III]/[V]$ has been estimated to be close to 10^3 for both I and II [2]. $K_2 = [VI][H^+]/[V]$ can be taken as approximately 10^{-14} since the positive charge on the complex will make the amide group slightly more acid than acetamide with $pK_a = 15.1$ [10]. With these values $K_3 = [IV]/[VI] = K'_{pot} \cdot K_1/K_2$ can be calculated to be 10^9–10^{10}.

The rate of dissociation of IV is determined by k_3 since steps VI → V and V → III are both very fast, the first being a simple acid-base reaction and the second a replacement of water in the co-ordination sphere of Cu^{2+}. Therefore $k_3 = k_0 = 2.15\ s^{-1}$ and $2.35\ s^{-1}$ resp. for I and II. That triglycine reacts about 10^3 times more

Scheme



slowly is expected, since, as shown by VII, binding of the carboxylate group [11] will make the formation of VI more difficult.



The formation of IV is given by (8). By comparison of (8) with (5) we obtain $k_{-3} = 10^9\ s^{-1}$, which is in the right order of magnitude for the water exchange rate

$$d[IV]/dt = \frac{K_2 \cdot k_{-3} \cdot [III]}{K_1 \cdot [H^+]} \tag{8}$$

of the Cu^{2+} complex [12]. Again triglycine reacts more slowly than I or II because the equilibrium III $\rightleftharpoons$ V is shifted considerably to the left by co-ordination of the carboxylate group (VIII) [4]. The formation of complexes with the deprotonated amide group thus can be explained by the well established general mechanism, in which the dissociation of a co-ordinated solvent molecule is rate determining. The only reason for the relatively slow reaction is the very low concentration of the intermediate VI.

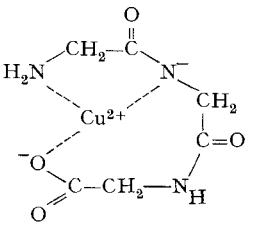
Besides the mechanism just discussed, a second path (see Scheme) must exist, since general acid and base catalysis is observed in our system as well as that of Cu^{2+}/triglycine [4] (see Table 1). The rate determining step must involve proton

transfer to or from the amide group and/or rotation of one of its co-ordinated forms into the other. Whether the acid HX reacts with the deprotonated nitrogen or the oxygen of the amide group, cannot be said. It has been shown that, in the bis-glycyl-glycinato-cobalt(III) cation, the carbonyl oxygen is the most basic site [13], however protonation of the co-ordinated nitrogen could be kinetically more important.

Comparison of k_H for I, II and triglycine must take into consideration: a) co-ordination of the carboxylate group of triglycine in VII, and b) that the charges of the deprotonated complexes are +1 for I and II, and −1 for triglycine. As discussed above, a) will slow down dissociation; because of b), ion pair formation, mostly due to electrostatic interaction, is more likely for triglycine than for I or II and hence the rate of protonation should be enhanced in the first case. These two opposing effects apparently nearly cancel each other, so that the rates of protonation for the three complexes IV are much the same (*cf.* Table 1).

Surprisingly, reaction of water as a base with III is more rapid for triglycine ($k'_{H_2O} = 4.5 \cdot 10^{-2}$ M^{-1} s^{-1}) than for I or II ($k'_{H_2O} = 4.7 \cdot 10^{-3}$ M^{-1} s^{-1}, and $1.3 \cdot 10^{-4}$ M^{-1} s^{-1}, resp.) by 1–2 orders of magnitude. This can only be understood by assuming equilibrium (9) for triglycine, in which IX is a suitable structure for the abstraction of the peptide proton.

VIII ⇌ IX (9)



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