Potentiometric and Spectroscopic Study of Nickel(II) and Cobalt(II) Complexes of Histamine-Containing Dipeptides

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The formation and structure of cobalt(II) and nickel(II) complexes of glycylhistamine (Gly-Hist) and sarcosylhistamine (Sar-Hist) have been studied by pH-metric and ^{1}H-NMR methods. An EPR study of the analogous, diamagnetically diluted copper(II) complex is also included. In the pH range 3–9, monomeric 1:1 and 1:2, more or less deprotonated complexes are formed (MLH, ML, MLH_{-1}, $MLH_{-1}(OH)$, ML_2, ML_2H_{-1}, M = Co(II), Ni(II)) depending on the pH and metal-to-ligand ratio. The interaction between ligands and nickel(II) at pH > 9 was characterized by the formation of imidazole-bridged (through both N^1 and N^3 nitrogens) polynuclear species $(NiLH_{-2})_n$, with {4N} coordinated metal centers. In the case of Gly-Hist, the ^{1}H-NMR study showed the existence of a single oligomeric (probably tetrameric, $n = 4$) species, while the steric hindrance of the N-CH_3 group in Sar-Hist produces a series of coexisting oligomers, with $n \geq 4$.

Introduction

The complex-formation processes in peptide chemistry are greatly influenced by the coordination of side-chain donor groups. In this respect, the imidazole moiety seems to be one of the most important, since it offers a particularly stable metal-binding site in the physiological pH range. The imidazole ring of histidyl residues can be found in a large number of metalloproteins as binding sites for various metal ions (Mn(II), Fe(II)/(III), Co(II), Cu(I)/(II), and Zn(II)). A good example is the copper(II)- and zinc(II)- containing superoxide dismutase (SOD), in which the imidazole moieties, besides exhibiting monodentate coordination, can also act as bridging ligands between the two metal centers.[1] Such complexes, containing bidentate, bridging imidazole ligands are therefore of current interest from a biomimetic point of view.

Although copper(II) complexes of histidine-containing di- and tripeptides have been extensively studied,[2–12] less attention has been given to other metal ions such as nickel(II) and cobalt(II).[3,13,14] In the physiological pH range, {3N} coordinated monomeric complexes are predominant with X-His and X-His-Y peptides, and {4N} coordinated monomeric complexes with X-Y-His. An interesting and much discussed feature of copper(II) and nickel(II)—unlike cobalt(II)—is the ability to form oligomeric species at higher pH (>9). These oligomeric complexes $(MLH_{-2})_n$ are believed to be in equilibrium with the monomeric $MLH_{-1}(OH)$ and are generally considered as {4N} coordinated polynuclear species with the imidazole rings as bridging bidentate units through coordination of both N^1 and N^3 nitrogens. According to earlier suggestions for the nickel(II)– and copper(II)–Gly-His systems, the association of four (or more) monomeric units is preferred on steric grounds.[2] A cyclic tetrameric structure was determined recently by X-ray crystallography[15] for the solid complex $[\{Au^{III}(Gly\text{-}His)\}_4]\cdot 10H_2O$. In solution, however, no structural study supports this picture.

Recently we reported the formation of copper(II) complexes of β-alanylhistamine (carcinine),[16] glycylhistamine (Gly-Hist), and sarcosylhistamine (Sar-Hist).[17] These compounds were compared with the analogous histidine-containing dipeptides, and some noteworthy differences were found, mostly in the physiological pH range. At pH > 9, oligomeric species are formed in these systems too. We proposed a microscopic deprotonation pathway between $CuLH_{-1}$ and $CuLH_{-2}(OH)$ by the formation of $CuLH_{-1}(OH)$ and $CuLH_{-2}$ alternative microspecies.[17] The latter complex—in which N^1 is deprotonated instead of the equatorially coordinated water molecule—is also in equilibrium with the oligomeric $(CuLH_{-2})_n$ complex, due to the favored substitution of the water molecule in the fourth position of $CuLH_{-2}$ species by an N^1-pyrrolic nitrogen of another $CuLH_{-2}$ complex.

In the present paper, we report the results of pH-metric and spectroscopic (EPR and ^{1}H-NMR) studies of nickel(II) and cobalt(II) complexes of Gly-Hist and Sar-Hist, with special attention to the oligomerization processes in alkaline solution.

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Table 1. Stability Constants and Derived Data for Co(II) and Ni(II) Complexes of Histamine- and Histidine-Containing Dipeptides (as Their Logarithms), $\beta_{pqr} = [M_pL_qH_r]/[M]^p[L]^q[H]^r$, M = Ni or Co, I = 0.1 mol dm^{-3} ($NaClO_4$), T = 298, with Estimated Errors in Parentheses (Last Two Digits) (Calculation of Derived Data: $pK_{NH} = \log \beta_{110} - \log \beta_{pqr}$; $pK^{ML_2}_{ML_2H_{-1}} = \log \beta_{120} - \log \beta_{12\bar{1}}$; $\log K^4_{olig} = {}^1/_4 \log \beta_{44\bar{8}} - \log \beta_{11\bar{1}}$)

	Gly-Hist[a]		Sar-Hist[a]		Gly-His			
	Ni(II)	Co(II)	Ni(II)	Co(II)	Ni(II)		Co(II)	
$\log \beta_{pqr}$								
111	10.46(02)	10.10(02)	10.64(02)	10.17(02)	11.34[b]	11.07[c]	10.61[b]	
110	4.20(02)	3.13(03)	3.86(02)	2.84(02)	4.68[b]	3.9[c]	3.44[b]	3.32[d]
120	7.73(02)	5.65(04)	7.46(04)	4.78(04)	9.64[b]	8.82[c]	6.57[b]	
$11\bar{1}$	−2.69(01)	−5.18(02)	−3.03(01)	−5.19(02)	−1.35[b]	−1.50[c]	−3.96[b]	−3.92[d]
$11\bar{2}$	[−12.22][e]	−15.41(05)	−12.51(10)	−15.66(02)		−12.48[c]	−15.45[b]	
$12\bar{1}$	−0.16(01)	−2.26(04)	−0.48(02)	−3.07(06)	2.07[b]	0.92[c]	−1.49[b]	
$44\bar{8}$	[−37.68][e]		−38.45(05)					
pK_{NH}	6.89	8.31	6.89	8.03	6.03[b]	5.4[c]	7.40[b]	7.24[d]
$pK^{ML_2M}_{L_2H_{-1}}$	7.89	7.91	7.94	7.85	7.57[b]	7.90[c]	8.06[b]	
$\log K^4_{olig}$	−6.73		−6.58					

[a] This work. [b] From ref 13 (T = 298 K, I = 0.2 mol dm^{-3} (KCl)). [c] From ref 29 (T = 298 K, I = 0.1 mol dm^{-3} (KNO_3)). [d] From ref 30 (T = 298 K, I = 0.1 mol dm^{-3})). [e] Values with lesser degree of precision (see Experimental Section).

Experimental Section

Materials. Glycyl- and sarcosylhistamine were synthesized as described earlier.[18] Their purity was checked by NMR measurements, elemental analysis, and determinations of molecular weights MW from acid-base titrations (MW = 241 (calc 241.12) and 255.5 (calc 255.15) for Gly-Hist and Sar-Hist, respectively).

Stock solutions of metal perchlorates (Alfa Ventron products) were standardized complexometrically.

Polynuclear species of Gly-Hist ($Ni(LH_{-2})_n$ and $Cu(LH_{-2})_n$) have a relatively low solubility in water; thus they slowly precipitate from a *ca.* 0.03 M solution at pH 10.6. The pure complex is obtained by washing the filtered precipitate with water. Due to the similar stabilities of $Ni(LH_{-2})_n$ and $Cu(LH_{-2})_n$ species, the diamagnetically diluted copper(II) complex (Cu:Ni = 1:49; see below) can be prepared in a similar way.

pH-Metric Measurements. The coordination equilibria were investigated by potentiometric titrations at 298.15 ± 0.1 K under nitrogen atmosphere at constant ionic strength (0.1 mol dm^{-3} $NaClO_4$). In the case of cobalt(II), nitrogen was bubbled through the alkaline solution of pyrogallol to eliminate O_2 traces. The titrant solution (NaOH) was also prepared and stored in a closed box, under nitrogen atmosphere. Changes in pH were followed by using an Orion (Cat alogue No. 91-03) combined glass electrode and an Orion 710 pH-meter (precision 0.1 mV). For the quantitative evaluation of the data, eq 1 was used with the experimental electromotive force values (E)

$$E = E_0 + \frac{RT}{F} \ln [H^+] + j_H[H^+] + j_{OH}[H^+]^{-1}K_W \qquad (1)$$

and the equilibrium hydrogen ion concentration [H$^+$], where j_H and j_{OH} are fitting parameters in acidic and alkaline media for the correction of experimental errors, mainly arising from the liquid-junction potential and from the possible alkaline and acidic deviations of the glass electrode,[19] and $K_w = 10^{-13.75}$ is the autoprotolysis constant of water.[20]

The complex formation constants were calculated as the averages of 10 independent titrations. The metal ion to ligand ratios were varied from 1:1 (1:2 for Co(II)) to 1:6, with metal ion concentrations between 10^{-3} and 10^{-2} mol dm^{-3}. In the case of the nickel(II)−Gly-Hist system, precipitation occurs above pH 9 due to the low solubility of the oligomeric ($(NiLH_{-2})_n$) complex (~0.002 M); thus the experimental data for this species are limited to 20−80% formation of complex in question, depending on the concentration of nickel(II).

The species formed in the investigated systems can be characterized by the general equilibrium process (2) (charges omitted). The formation

$$pM + qL + rH \rightleftharpoons M_pL_qH_r \qquad (2)$$

constants (β_{pqr}) for this generalized reaction were evaluated from the pH-metric titration data with the PSEQUAD computer program.[21]

Spectroscopic Measurements. The visible absorption spectra were recorded on a Varian DMS 100 UV/vis spectrophotometer. For the EPR measurements, a diluted powder sample of 2% Cu(II)−Gly-Hist in diamagnetic Ni(II)−Gly-Hist was used to obtain the spectrum on a Bruker ER-200 D spectrometer at room temperature and 9.45 GHz. Proton NMR spectra were recorded on a Bruker AM-400 spectrometer with dioxane as internal standard (3.7 ppm), mostly using aqueous solution (H_2O) and presaturation to eliminate the solvent signal. Chemical exchange was probed by the standard EXSY[22] (EXchange SpectroscopY) pulse sequence D−90−t_1−90−t_m−90−t_2(AQ) in phase-sensitive (TPPI) mode.[23,24] A 10% random variation of mixing time (t_m = 0.4, 0.8, 1.6, and 4s) was applied to cancel scalar correlation effects and minimize the t_1 noise. A total of 512 experiments with 16 scans of 2K data points were collected and zero-filled to obtain a 2K × 2K data matrix.

Results and Discussion

Potentiometric Measurements. The stability constants determined for nickel(II) and cobalt(II) complexes are given in Table 1. The protonation constants of ligands studied in this paper have been already determined[17] at the macroscopic level (pK_1 = 6.78 and 6.77 and pK_2 = 8.04 and 8.31 for Gly-Hist and Sar-Hist, respectively) and also at the so-called microscopic level.[25] The first macroscopic deprotonation can be approximately assigned to the imidazole ring and the second one to the terminal amino nitrogen.

The titration curves of systems containing Ni(II) and L in the molar ratio 1:1 show a characteristic inflection point at pH ~ 10 after consuming 4 equiv of base per metal (another weakly developed inflection point can be detected at pH ~ 8 after adding *ca.* 3 base equiv of base). In the case of 1:2 Co(II):L systems, the only inflection point was found at pH 9 after consumption of 5 equiv of base per metal. At higher pH, however, further base consumption was measured (*ca.* 5.7−5.8 equiv up to pH 11.2). These facts suggest two successive deprotonations of coordinated ligand(s) or water starting from ML species. The second deprotonation takes place at much higher pH in the case of Co(II) complexes.

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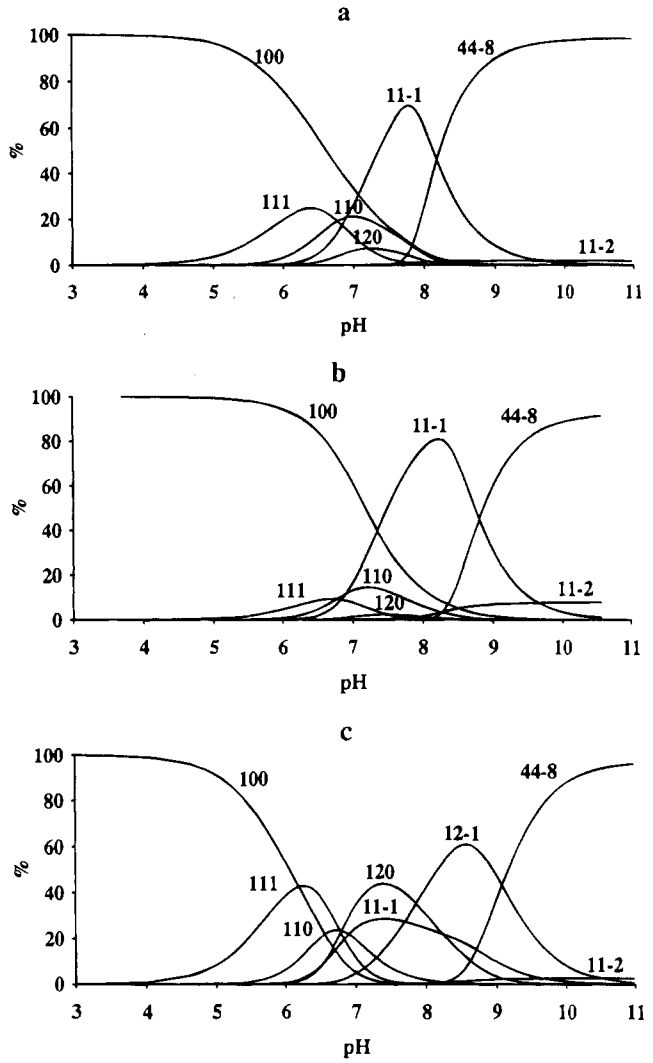

Figure 1. Concentration distribution of complexes in the Ni(II)−Sar-Hist system as a function of pH. [Ni] = 0.01 (a), 0.002 (b), 0.006 (c) mol dm^{-3}. [M]:[L] = 1:1 (a, b), 1:4 (c).

The pH-metric curves between pH 4 and 11 were best fitted by considering the complexes listed in Table 1, including monomeric (1:1 and 1:2) and oligomeric complexes.

Equilibrium data on Gly-His complexes[13] are also included for comparison. Distribution curves of species in Ni(II)−Sar-Hist are shown in Figure 1. In both systems the complex formation begins at pH ~ 4, with the formation of {1N} and {2N} coordinated MLH and ML complexes, respectively (with the possibility of an additional carbonyl oxygen binding). In the free ligands, the deprotonations of the amino and imidazole groups overlap;[25] thus it can be assumed that the first metal-promoted deprotonation (the formation of MLH complex) can also occur at either nitrogen. In ML complexes, however, both (imidazole and amino) nitrogens are already coordinated. The subsequent deprotonation (ML ⇌ MLH_{-1} + H^+ , pK_{NH}) can be attributed to the deprotonation of peptide nitrogen, forming the {3N} coordinated MLH_{-1} complex, which is the predominant species at pH 8 in equimolar solution (Figure 1a). Although all complexes presently reported are slightly less stable compared to the analogous Gly-His complexes (Table 1), the difference in metal-promoted deprotonation of peptide nitrogen is noteworthy (pK_{NH} = 6.89 and 6.03 for Ni(II)−Gly-Hist and −Gly-His systems, respectively). This deviation may be assigned to the Coulombic effect of the nearly negative charge or to the further stabilization of an additional axial coordination by a carboxylate group in the case of Gly-His ({3N,COO^-}. In

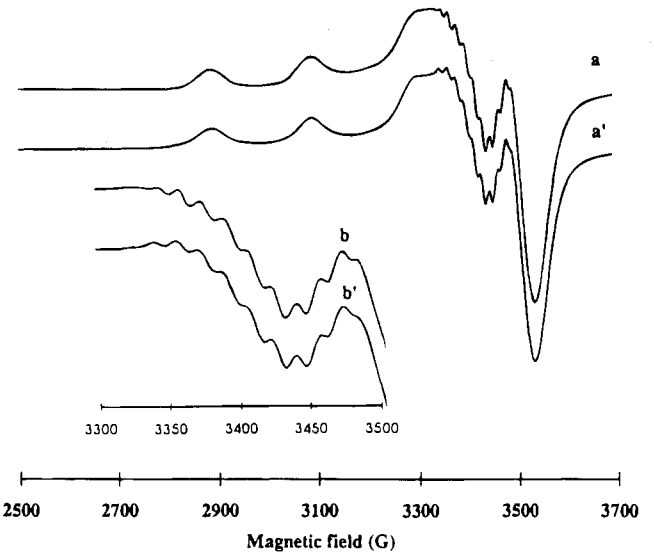

Figure 2. Experimental (a, b) and calculated (a′, b′) EPR spectra of the $(CuLH_{-2})_n$ complex solid (L = Gly-Hist) diluted diamagnetically by the analogous Ni(II) complex. Insert shows the enlarged g region (T = 295 K).

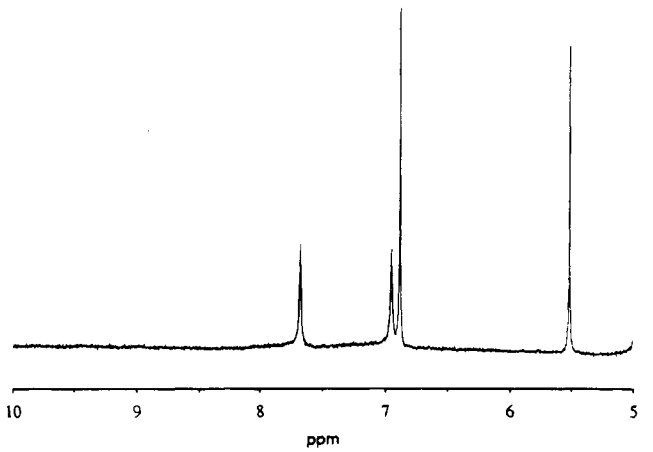

Figure 3. ^{1}H-NMR spectrum of imidazole protons of the Ni(II)−Gly-Hist system, T = 298 K, pH 11.0, [L] = 1.2[Ni] = 0.005 mol dm^{-3} in methanol-d_4 (see text).

spite of above fact, the presently reported pK_{NH} values still reflect the strong preference of Co(II) to deprotonate peptide nitrogens compared with those of simple dipeptides, emphasizing to the role of the imidazole ring as an anchoring group. In the presence of excess ligand, bis complexes (ML_2 and ML_2H_{-1}) are also formed in the range pH 6−10. The fairly close values of $pK^{ML_2}_{ML_2H_{-1}}$ and pK^{HL}_{L} for nickel(II), cobalt(II), and even copper(II) ($pK^{CuL_2}_{CuL_2H_{-1}}$ = 7.60 and 7.98 in the case of Gly-Hist and Sar-Hist, respectively[17]) strongly suggest the deprotonation of a free NH_3^+ group, which remains uncoordinated after deprotonation.

$$ML_2 \rightleftharpoons ML_2H_{-1} + H^+$$

In this respect the latter complexes should be denoted as $M(LH_{-1})LH$ and $M(LH_{-1})L$, where the second ligand is coordinated monodentately through the imidazole ring.

Above pH 8, the color of the solution containing nickel(II) changes from light green to yellow (λ^{d-d}_{max} = 450 nm), indicating the formation of new complex(es) with square planar arrangements around nickel(II). By analogy with the copper-(II)-containing systems,[16,17] two microscopic deprotonation processes are responsible for this new base consumption: the

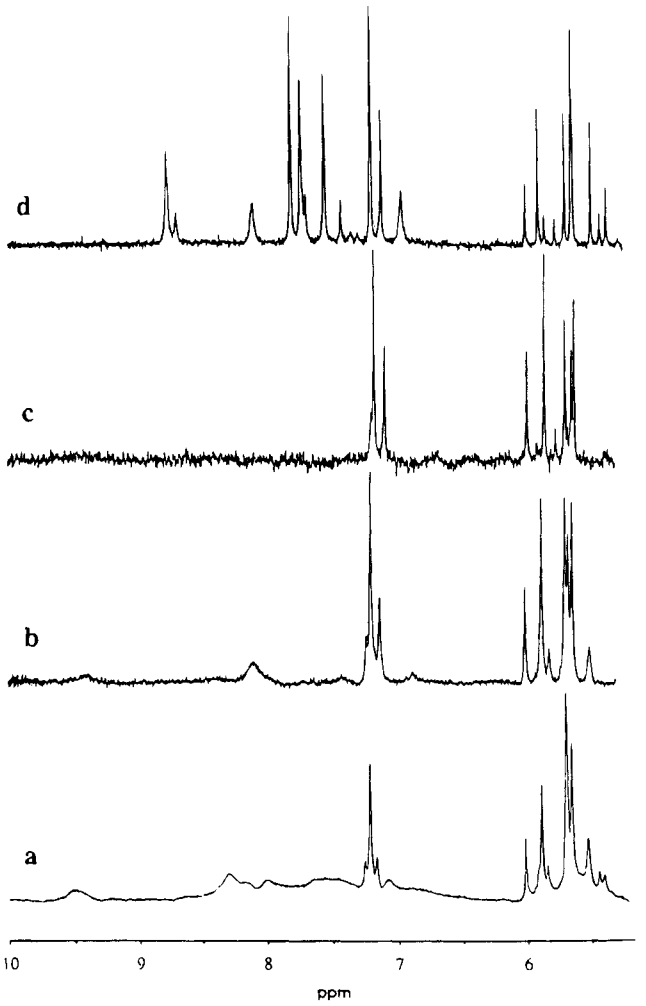

Figure 4. Parts of ^{1}H-NMR spectra of the Ni(II)−Sar-Hist system in aqueous solution, pH 11.0, [M]:[L] = 1:1. Spectra a−c: T = 298 K; [L] = 0.2, 0.1, 0.05 mol dm^{-3}, respectively. Spectrum d: T = 353 K; [L] = 0.05 mol dm^{-3}.

hydrolysis of the MLH_{-1} complex ($MLH_{-1} \rightleftharpoons MLH_{-1}(OH) + H^+$) and the deprotonation of the N^1 nitrogen in the imidazole ring ($MLH_{-1} \rightleftharpoons MLH_{-2} + H^+$). The latter species can readily oligomerize, forming an imidazole-bridged polynuclear structure. The overall equilibrium $MLH_{-1}(OH) \rightleftharpoons (MLH_{-2})_n$ in the present systems is significantly shifted toward the oligomeric species; thus the simultaneous presence of the monomeric complex could not be detected independently as for copper(II) complexes[16,17] (the formation of $NiLH_{-1}(OH)$ is less than 10% even in diluted solution (Figure 1b)). Introducing this complex, however, besides the oligomeric species, improves significantly the curve fitting of potentiometric data. In the calculation, we considered only the tetrameric species as the most simple among the possible oligomeric complexes. Although this is an oversimplified model of the real situation in the case of the nickel(II)−Sar-Hist system (see later), it can describe the titration curves correctly, since the log K^n_{olig} ($MLH_{-1} \rightleftharpoons (1/n)(MLH_{-2})_n + H^+$) equilibrium constants for different values of n ($\geq$4) would be practically identical.[2] The earlier published log K^4_{olig} values for copper(II) complexes (−7.46 (Gly-His-Gly),[7] −7.95 (Carcinine),[16] −7.62 and −7.44 (Gly-Hist and Sar-Hist)[17]) are significantly smaller than presently reported constants for nickel(II) (see Table 1). The higher stability of nickel(II) complexes results from the additional ligand stabilization, due to the formation of spin-paired planar nickel(II) complexes with four nitrogen donor atoms.

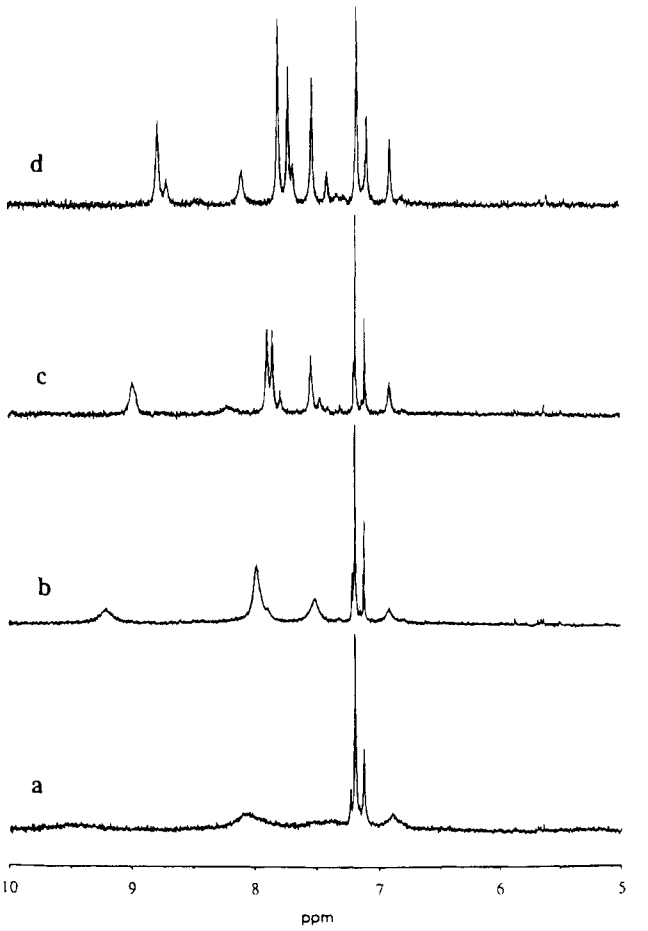

Figure 5. Parts of ^{1}H-NMR spectra of the Ni(II)−Sar-Hist system deuterated at C^2−H in D_2O, pH 11.4, [Ni] = [L] = 0.05 mol dm^{-3}. T = 298 K (a), 318 K (b), 338 K (c), and 353 K (d).

In the case of cobalt(II), the formation of oligomeric complexes was not detected in the pH range 2−11.4. The cobalt(II)-promoted deprotonation of the N^1 nitrogen of the imidazole ring probably takes place at higher pH.[26]

Spectroscopic Measurements. Due to the structural identity and similar stabilities of nickel(II)− and copper(II)−Gly-Hist complexes, the copper(II) ions disperse statistically among the oligomeric sequences in the powder sample obtained from a solution containing copper(II), nickel(II), and Gly-Hist in the molar ratio 1:49:50 at pH = 10.6. Indeed, the EPR spectra in Figure 2 show isolated copper(II) centers in this powder sample. (Without diamagnetic dilution, antiferromagnetic interaction was found for the solid complex by magnetic measurements.[27]) At room temperature, a resonance with $g_{||}$ = 2.1944(7), $g_\perp$ = 2.0396(4), $A_{||}$ = 196.3(6) G, and $A_\perp$ = 10.0(4) G was observed. The superhyperfine structure of the parallel region and the parameters determined show the coupling of four equatorial nearly equivalent nitrogen atoms ($A_{N\perp}$=16.2(4) G). The results are entirely compatible with the square-planar structure around each copper(II) center and present the first evidence for the expected {4N} coordination in such oligomeric species. The fourth nitrogen donor should be the deprotonated N^1-pyrrolic nitrogen from another monomeric unit, thus allowing the formation of polynuclear species.

The ^{1}H-NMR spectra of nickel(II)-containing systems show very broad lines between pH 4 and 9, due to the paramagnetic line broadening. However, the diamagnetic character of nickel-

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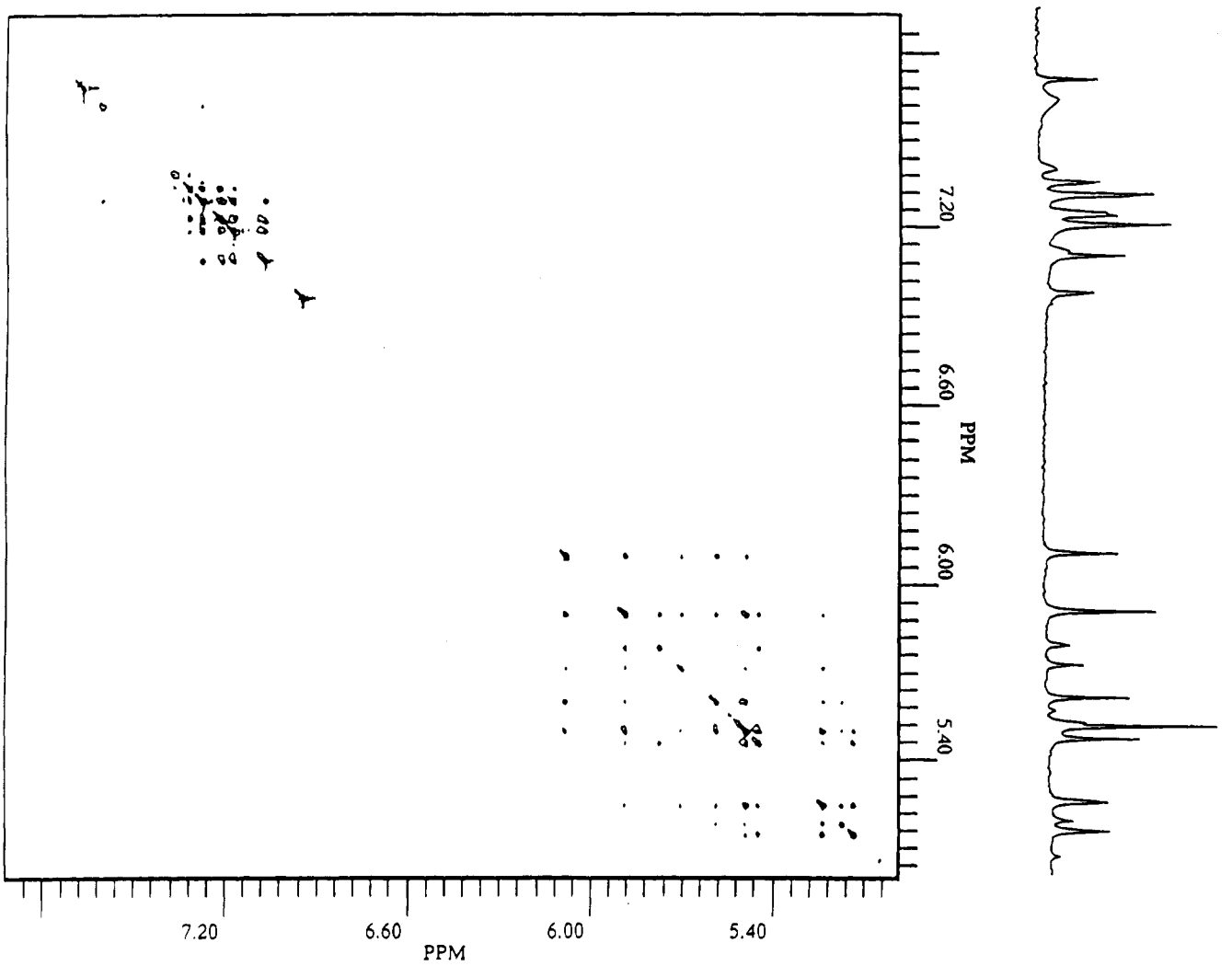

Figure 6. $^1H-^1H$ chemical exchange spectroscopy (NOESY pulse sequence) of the Ni(II)−Sar-Hist system in methanol-d_4, [Ni]:[L] = 0.05 mol dm^{-3}, T = 298 K, mixing time 0.8 s.

(II) multinuclear complexes affords an excellent opportunity to study their structure by NMR spectroscopy. The imidazole region of ^1H-NMR spectrum of the Ni(II)−Gly-Hist oligomer in the presence of a slight excess of Gly-Hist is shown in Figure 3. The spectrum reveals the formation of only one oligomeric species, which is kinetically stable on the NMR time scale. In the free ligand the carbon-bound C^2-H proton of the imidazole ring exchanges easily for deuterium in D_2O; this can help us to assign the signals. We found a small shift for the C^5-H signal and a very important upfield shift for the C^2-H proton of multinuclear species (5.52 ppm) compared to the free ligand (7.70 ppm). This large upfield shift is surprising since, for example, in the case of the diamagnetic nickel(II)−Pyr-His complex, much smaller upfield shift was found even after deprotonation of the N^1-pyrrolic[28] nitrogen. Such an important shift may provide further reasonable proof for metal coordination of both imidazole nitrogens. The NMR equivalence of all imidazole moieties in the polynuclear species $(NiLH_{-2})_n$ strongly supports the existence of cyclic oligomers, presumably cyclic tetramers, as already suggested for the analogous copper(II) complexes.

In the case of the nickel(II)−Sar-Hist system the ^1H-NMR spectra are more complicated (Figures 4 and 5). The spectra a−c in Figure 4 show the concentration dependence (0.05−0.2 mol dm^{-3}) of imidazole proton signals of oligomeric complexes in aqueous solution at pH 11 ([M]:[L] = 1:1). The main difference compared to Figure 3 is the appearance of multiple signals. Fundamentally there are two possibilities for obtaining such multiplication of signals: (i) inequivalence of imidazole protons in the species, due to the conformational differences of monomeric units, or (ii) the presence of several oligomers, containing different numbers of monomeric units, in (slow) equilibrium. Increasing the concentration changes not only the chemical shifts but also the number of signals and the relative intensity. The great number of signals observed (mainly all at high temperature, Figure 4d) makes their assignment almost impossible. After deuteration of C^2-H (and all exchangeable) protons, only the C^5-H signals appear in the spectrum, as showed in Figure 5. Comparison of the spectra in Figures 4d and 5d reveals a shift of the imidazole protons during complexation similar to that mentioned for the nickel(II)−Gly-Hist complex. Figure 5 shows the temperature dependence of ^1H-NMR spectra of nickel(II)−Sar-Hist oligomeric complexes. It was observed that the number of signals increases on raising the temperature; this is inconsistent with the presence of different conformations, since at higher temperature, the faster molecular rearrangement should lead to the disappearance of conformational inequivalence (thus decreasing of the number of peaks). Thus, the only possibility is that a variable number of oligomeric complexes are in equilibrium, in slow mutual exchange on the

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NMR time scale. However this explanation cannot account for the evolution of spectra with increasing temperature. Several assumptions can be made at this point: either (i) at room temperature, due to the lack of fast and isotropic molecular motion of planar complexes, the residual dipolar interactions broaden the signals, while upon an increase in temperature they become narrower in function of the size of oligomers, or (ii) at any pH, there is a fast equilibrium between diamagnetic polynuclear species and traces of paramagnetic mononuclear complexes. At 298 K, the concentration of these monomeric complexes may be high enough for paramagnetic line broadening, while at higher temperature (a) the equilibrium can be entirely shifted toward the oligomerization or (b) the faster reorientation of the paramagnetic complex results in decreased transverse relaxation times and consequently in lines sharpening.

The three-pulse NOESY sequence allows the determinations of both NOE and chemical exchange of nuclei by two-dimensional spectroscopy. In phase-sensitive mode, for relatively small molecules, the exchange correlation signals have the same phase characteristics as the diagonal autocorrelation peaks, while NOE effects have opposite sign. In water, certain ^{1}H-NMR signals for the Ni(II)−Sar-Hist system are broad at room temperature (Figure 4); however, in methanol-d_4, they are relatively narrow, probably because of the decrease of solvent viscosity. It can be seen from Figures 4d and 6, that the spectra of the Ni(II)−Sar-Hist system have similar features in water and methanol-d_4 (the same number of species are formed in both solvents) with however some slight differences in chemical shifts, this allowed us to use the latter solvent for exchange spectroscopy. Due to the great number of exchanging species, there is no ideal mixing time to obtain maximal intensity for all cross-peaks. The recorded spectrum for a mixing time of 0.8 s is shown in Figure 6. It can be seen that cross-peaks were observed among (practically) all C^2−H signals and also among C^5−H signals, but there is no connectivity between C^2−H and C^5−H protons, as expected. This also supports the assumption that the connectivities observed are due to chemical exchange, not to signal exchange or NOE. The spectrum in Figure 6 confirms the former assumption that the multiple signals of imidazole protons can be assigned to different oligomeric complexes undergoing mutual ligand exchange.

Conclusions

Our study confirms and extends the results obtained earlier for the metal ion complexation of X-His and related peptides. We have shown that, after the formation of monomeric affiliated complexes (MLH, ML, MLH_{-1}, $MLH_{-1}(OH)$, ML_2, ML_2H_{-1}), polynuclear species are also formed in alkaline solutions (pH > 9) of nickel(II)-containing systems. In the case of Gly-Hist, only one multinuclear, diamagnetic complex (according to earlier suggestions[2,7] probably a tetramer) was observed with a planar 4N coordination. However, in the Ni(II)−Sar-Hist system, a number of oligomeric $(NiLH_{-2})_n$ complexes—with $n \geq 4$—were found. The probable explanation of this basic difference is the steric hindrance of the N−CH_3 group in Sar-Hist, which may decrease the salient stability of tetrameric species as compared to the higher homologues. The results also emphasize the sensitivity of self-assembling metal clusters to weak structural differences.

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