TABLE I
ANALYTICAL DATA OF COMPLEXES IIa, b AND IIIa–c

Complex	Mp, °C	Empirical formula	Mol wt, Calcd	Mol wt, Mass spec[c]	% C, Calcd	% C, Found	% H, Calcd	% H, Found	Optical rot.,[a] deg	Yield, %
$(CO)_2Rh(tfacCam)$ (IIa)	131	$RhC_{14}H_{14}F_3O_4$	406.162	406	41.40	42.04	3.47	3.50	+106	92
$(C_2H_4)_2Rh(tfacCam)$ (IIb)	145 dec	$RhC_{16}H_{22}F_3O_2$	406.246	406	47.30	48.18	5.46	5.58	+102	75
(COD)Rh(tfacCam) (IIIa)	163	$RhC_{20}H_{26}F_3O_2$	458.318	458	52.41	52.51	5.72	5.59	+80	Quant
(DCP)Rh(tfacCam) (IIIb)	130–142[b]	$RhC_{22}H_{26}F_3O_2$	482.338	482	54.78	55.07	5.43	5.56	+87[b]	Quant
(NOR)Rh(tfacCam) (IIIc)	87	$RhC_{19}H_{22}F_3O_2$	442.276	442	51.59	52.28	5.01	5.29	+82	Quant

[a] Complex solutions 10^{-2} *M* in chloroform (23°). [b] Sublimed sample. [c] Molecular ion peak.

$Pd(C_6H_5CN)_2Cl_2$[16] was added. The mixture was refluxed for 4 hr, stirred for another 8 hr at room temperature, and then filtered. The solvent was evaporated *in vacuo* and the residue was extracted with boiling *n*-hexane. The hexane solution was filtered, concentrated, and left at 0° for a prolonged period, whereby large red-brown crystals of the palladium chelate were formed; mp 237° dec; yield 77%; $[\alpha]^{23}D$ +182° (10^{-2} *M* in chloroform). *Anal.* Calcd for $PdC_{24}H_{28}F_6O_4$: C, 47.95; H, 4.69. Found: C, 48.32; H, 4.72.

Preparation of Nickel(II) *d*-Bis(3-trifluoroacetylcamphorate), Ni(tfacCam)$_2$.—A 632-mg sample (1 mmol) of $Ba(tfacCam)_2$ was dissolved in 150 ml of boiling ethanol, and 238 mg (1 mmol) of $NiCl_2 \cdot 6H_2O$, dissolved in 25 ml of ethanol, was filtered in. The mixture was stirred for 10 min at 60° and then the barium chloride formed was filtered off. The solvent was removed *in vacuo*, the residue was taken up in 15 ml of *n*-hexane, and the extract was filtered. The hexane was removed *in vacuo* and the green residue was dissolved in 5 ml of ethanol. The ethanol was allowed to evaporate slowly, whereby large green crystals were formed. The crystals are efflorescent and should therefore be stored under ethanol. The green chelate had no melting point up to 300° but loses water at 100°. It can be completely dehydrated in an oven at 100° and high vacuum and is very soluble in common organic solvents; yield, quantitative; $[\alpha]^{23}D$ +215° (10^{-2} *M* in chloroform). *Anal.* Calcd for $NiC_{24}H_{28}F_6O_4$: C, 52.11; H, 5.10; Ni, 10.61; mol wt 553. Found: C, 51.37; H, 5.29; Ni, 10.58; mol wt 562 (vaporimetric in DMF).

(16) M. S. Kharasch, R. C. Seyler, and F. R. Mayo, *J. Amer. Chem. Soc.*, **60**, 882 (1938).

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Properties of Palladium(II) Complexes of Peptides and Histidine in Basic Solutions[1]

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By comparison with properties of a related series of ligands, the change in net circular dichroism (CD) sign from negative to positive occurring upon addition of base with pK = 11.7 in the quadridentate Pd(II) complex of GlyGly-L-Ala is identified as replacement of bound carboxylate by hydroxide in the tetragonal coordination plane. A similar transformation occurs in the Cu(II) complex with pK = 13.1. Instead of the usual negative, a net positive CD is observed for several tetragonal peptide complexes where the carboxyl terminal L-alanyl residue is bound only *via* the deprotonated amide nitrogen with an unbound carboxylate group. Nmr chemical shift nonequivalence of glycyl methylene protons observed in the zwitterion form of L-AlaGly does not occur in the terdentate Pd(II) complex but appears upon displacement of the bound carboxylate group by hydroxide and in Pd(en)(AlaGly)^0, where there are four nitrogen donor atoms about Pd(II). Absorption and CD spectra are reported for complexes of Pd(en)(L ligand) where the optically active ligand possesses two nitrogen donors. Comparisons from among the compounds permit assignment of the transformation occurring upon addition of base to Pd(en)(L-His)$^+$ (His = histidine) with pK = 10.83 to ionization of the pyrrole hydrogen.

Introduction

Tetragonal transition metal ions form complexes with peptides the structures of which are well understood. For example, after the addition of 3 equiv of base to a solution containing equimolar amounts of $PdCl_4^{2-}$ and tripeptide, an end point is reached by pH 6. At this point the ligand is tetradentate, being bound in the coordination plane by one amino, two deprotonated amide, and one carboxylate oxygen donor atoms to yield a complex of net unit negative charge. The complex exhibits an absorption maximum at 300 nm with $\epsilon \sim 1300$, and the circular dichroism (CD) values of a variety of peptide complexes of Pd(II) have been reported.[2] Ni(II) and Cu(II) form similar tetragonal complexes.[3,4] In this paper we study the behavior of tripeptide complexes of these transition metal ions upon the addition of a fourth equivalent of base. There has been uncertainty concerning the structures of the complexes under these conditions. Absorption, CD, and proton magnetic resonance (pmr) spectra of carefully chosen complexes are utilized to define the nature of the transformations taking place. The Pd(II) com-

(1) This paper is abstracted from the Ph.D. Thesis (1971) of T. P. Pitner, who was holder of a NASA predoctoral fellowship. The research was supported by a grant from the National Science Foundation.

(2) E. W. Wilson, Jr., and R. B. Martin, *Inorg. Chem.*, **9**, 528 (1970).
(3) J. W. Chang and R. B. Martin, *J. Phys. Chem.*, **73**, 4277 (1969).
(4) J. M. Tsangaris and R. B. Martin, *J. Amer. Chem. Soc.*, **92**, 4255 (1970).

plexes proved most tractable but enough results were obtained with the other metal ions to permit comparisons. The pH region of ionizations from unbound pyrrole nitrogens in metal ion bound imidazole groups such as occur in histidine complexes has also been uncertain for many years. In this paper we report the pK_a value of a pyrrole ionization from a Pd(II)-bound histidine and compare it with values recently determined for complexes of other metal ions.

Experimental Section

All amino acid and peptide ligands were high grade commercial products. Their equivalent weights were checked by titration and molar absorptivities corrected in some instances. The preparation of peptide complexes from Na_2PdCl_4 (Alfa Inorganics, Inc.) has been described.[2] Pd(dien)(L-Ala)$^+$ was prepared by first combining equimolar amounts of Na_2PdCl_4 and diethylenetriamine (dien) to yield a solution of Pd(dien)Cl^+ which absorbed at 332 nm with ϵ 430. Upon addition of one equivalent of anionic L-alanine, the absorption maximum shifts to 298 nm. Complexes containing ethylenediamine (en) were prepared from orange-yellow Pd(en)Cl_2 by a procedure already described.[5] Circular dichroism spectra were recorded on a Jasco J-10B instrument and proton magnetic resonance spectra on a Hitachi Perkin-Elmer R-20 spectrometer. Other instrumentation has been mentioned.[2] No ionic strength controls were employed in this work and the experiments were conducted at room temperature, about 24°.

Results

As indicated in Table I, increasing the pH from neutral solutions shifts the absorption maxima of the tripeptide complexes of Pd(II) about 10 nm to longer wavelength and alters the CD spectra. The change in CD upon adding base to neutral solutions of Pd(GlyGly-L-Ala)$^-$ is shown in Figure 1. Increasing the pH inverts the CD sign pattern and changes the net CD sign over the entire d–d manifold from negative to positive. The spectral results show no significant concentration dependence from 10^{-4} to 10^{-2} M and are reversible with respect to pH. A tight isosbestic point is observed at 322 nm suggesting an equilibrium between two major species in solution. From an analysis of the two extreme curves in Figure 1 and three intermediate ones, the apparent pK = 11.7. One of the purposes of this study was to determine to what structural change this acidity constant refers.

Table I
Spectral Properties of Palladium(II) Complexes

		Absorption		Circular dichroism	
	pH	nm	ϵ	nm	$\Delta\epsilon$
Pd(GlyGly-L-Ala)$^-$	7.0	299	1250	337	−0.99
				290	+0.41
	12.8	311	720	356	+1.22
				310	−0.85
+1 equiv of NH_3	10.0	297	840	350	−0.14
				297	+0.39
Pd(Gly-L-AlaGly)$^-$	7.4	301	1150	339	−1.40
				290	+0.15
	12.1	309	760	343	−1.12
				290	+0.18
Pd(GlyGly-L-AlaNH)$^-$	8.4, 12.7	282	1340	323	−1.20
				278	+0.52
Pd(en)(Gly-L-Ala)^0	8.0, 12.1	293	240	332	+0.12
				284	−0.06
Pd(dien)(L-Ala)$^+$	7.0	298	390	278	−0.08

Addition of 1 equivalent of NH_3 to Pd(GlyGly-L-Ala)$^-$ at pH 10.0 shifts the absorption maximum to the blue, suggesting four nitrogen donors, and markedly reduces the magnitude of the negative CD extremum without altering the positive one so that a net positive CD is observed through the d–d manifold. In contrast to the preceding tripeptide complex, the CD of Pd(Gly-L-AlaGly)$^-$ exhibits no inversions and only weakens slightly in magnitude upon addition of base to pH 12.1.

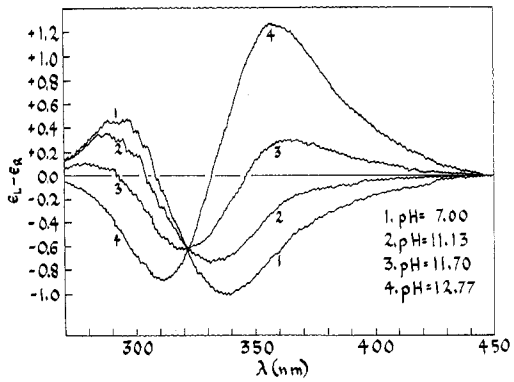

Figure 1.—Circular dichroism spectra of the Pd(II) complex of GlyGly-L-Ala at several pH values in the d–d absorption region.

Titration of a solution containing equimolar amounts of $PdCl_4^{2-}$ and glycylglycyl-L-alaninamide yields an end point after the addition of 4 equiv of base at pH 8.4. At this pH the complex formed absorbs maximally at 282 nm consistent with four nitrogen donors about the coordination plane. No uv or CD spectral changes are observed upon addition of base to pH 12.7. CD requires addition of the fourth equivalent of base for significant development and is then similar to that of Pd(GlyGly-L-Ala)$^-$. This result is consistent with the noncooperativeness of amide hydrogen ionizations in palladium(II)–peptide complexes in contrast to nickel(II)–tripeptide complexes where a cooperative interaction occurs in the change from an octahedral to a tetragonal complex.[6,7]

For metal ion promoted peptide hydrogen ionizations in tetraglycine only Ni(II) exhibits a cooperative interaction, accompanied by a change in spin state, while Cu(II)[6] and Pd(II) do not. Titration of a solution containing equimolar amounts (5 mM) of tetraglycine and $PdCl_4^{4-}$ yields a sharp end point after the addition of only 3 equiv of base at pH 5.5, where the absorption maximum occurs at 311 nm (ϵ 800). Addition of a fourth equivalent of base occurs separately with a second sharp end point appearing by pH 9.0, where the absorption maximum occurs at 285 nm (ϵ 1300). From the analyses of the titration curve and absorptivity changes at 280 nm upon addition of the fourth equivalent of base, we calculate pK_a = 7.50 ± 0.02. The absorption maximum at 285 nm for the pH 9 species indicates four nitrogen donor atoms in the Pd(II) complex. The titration results require three nitrogen donors for the pH 5.5 species and the relatively long wavelength of 311 nm suggests chloride ion in the fourth coordination position. The apparent acidity constant then refers to replacement of the chloride ion with metal ion promoted ionization and complexation of the carboxyl terminal amide nitrogen of tetraglycine.

The last two complexes in Table I contain four nitrogen donor atoms with the L-alanine residue bound to

(5) T. P. Pitner and R. B. Martin, *J. Amer. Chem. Soc.*, **93**, 4400 (1971).

(6) R. B. Martin, M. Chamberlin, and J. T. Edsall, *ibid.*, **82**, 495 (1960).

(7) R. Mathur and R. B. Martin, *J. Phys. Chem.*, **69**, 668 (1965); **75**, 4066 (1971).

Pd(II) only through the nitrogen atom with the carboxylate group unattached. The last complex demonstrates that an unchelated L-alanine coordinated through the tetrahedral amino nitrogen gives rise to small but significant negative optical activity in the ligand field bands of Pd(II).

The effect of adding base upon the d–d absorption and CD spectra was also investigated for tripeptide complexes of Cu(II) and Ni(II). For Cu(GlyGly-L-Ala)$^-$ both spectra are pH independent from neutral solutions up to pH 11. In this pH region an absorption maximum occurs at 550 nm (ϵ 134) and a CD minimum at 574 nm ($\Delta\epsilon = -0.35$). As more base is added both spectra undergo reversible, concentration-independent changes so that by pH 14 the absorption maximum shifts to 590 nm (ϵ 112) and three CD extrema appear, at 475 nm ($\Delta\epsilon = +0.02$), at 552 nm ($\Delta\epsilon = -0.25$), and at 654 nm ($\Delta\epsilon = +0.40$). An analysis of the CD spectra at 640 nm at intermediate pH values yields an apparent $pK = 13.1 \pm 0.1$.

The CD spectrum of Ni(GlyGly-L-Ala)$^-$ consists of a minimum at 478 nm ($\Delta\epsilon = -0.85$) with a shoulder near 415 nm ($\Delta\epsilon = -0.2$) and remains constant upon addition of base to pH 13. A solution containing equimolar amounts of tripeptide and Ni(II) to which only 2 equiv of base was added yields CD magnitudes half those given above for the addition of 3 equiv of base. Thus the cooperative nature of the transition in the formation of tetragonal Ni(II) complexes of tripeptides, already indicated by absorption spectra and magnetic susceptibility measurements,[6,7] is confirmed by CD of the GlyGly-L-Ala complex. After the addition of 2 equiv of base half of the Ni(II) is present as yellow, diamagnetic, tetragonal NiL$^-$ and half as blue, paramagnetic, octahedral NiL$^+$, where only the amino group of the tripeptide is coordinated.

Chemical shifts observed in pmr spectra for glycyl methylene protons are listed in Table II. Peak identifications in neutral solutions of Pd(GlyGlyGly)$^-$ are assigned by comparison with the preceding complexes in Table II containing alanyl residues. The spectrum at pH 12.8 was assigned by observing the disappearance and appearance of peaks at intermediate pH values. The sum of the chemical shifts for all three kinds of methylene protons is 0.17 ppm greater in the anionic form of GlyGlyGly than in the Pd(II) complex of the same net negative charge. The upfield shift brought about by Pd(II) is comparable to those observed for Zn(II) and Cd(II) complexes and is less than upfield shifts induced by tetragonal Ni(II).[7,8] No glycyl methylene nonequivalence is observed for any of the cases listed in Table II.

TABLE II
CHEMICAL SHIFTS OF GLYCYL METHYLENE PROTONS[a]

	pH	Amino	Central	Carboxyl
Pd(GlyGly-L-Ala)$^-$	7.0	3.48	3.84	...
Pd(L-AlaGlyGly)$^-$	7.0	...	3.80	3.64
Pd(GlyGlyGly)$^-$	7.0	3.53	3.82	3.65
	12.8	3.53	3.74	3.53
GlyGlyGly	<1	3.92	4.06	4.04
	5.4	3.91	4.03	3.78
	>13	3.40	3.99	3.78

[a] In ppm downfield from internal DSS in D_2O solutions.

Glycyl methylene nonequivalence of 0.24 ppm does occur in the pmr spectrum of Pd(en)(AlaGly)^0 for which the absorption maximum at 290 nm indicates four nitrogen donors so that the carboxylate group is unbound. The Pd(II) complexes of GlyAla or AlaGly do not exhibit nonequivalence in neutral solutions. As the basicity of the solution containing the latter complex is raised to pH 13, the glycyl methylene protons undergo a large upfield shift of 0.57 ppm and become nonequivalent to the extent of 0.16 ppm. The large upfield shift for the glycyl protons is consistent with substitution of the bound carboxylate group by hydroxide resulting in greater shielding by the negatively charged carboxylate that is no longer neutralized by the positively charged Pd(II). A greater nonequivalence of 0.73 ppm is observed in a 1:1 Pd(II) chelate of sarcosine (*N*-methylglycine). Finally a solution containing a 2:1 molar ratio of glycylglycine to Pd(II) exhibits at pH 12 only two major peaks in the pmr spectrum suggesting two ligands chelated in a single kind of symmetrical complex with four nitrogen donors.

Upon increasing the pH from about 7 to near 13 the complex Pd(en)(His)$^+$ undergoes a small change in absorption and a larger alteration of CD spectra as shown in Table III and Figure 2. The results are reversible and unaffected by a tenfold dilution from 4 to 0.4 m*M*. Tight isosbestic points are observed at 277 nm in absorption and at 302 nm in CD indicating the presence of only two major species. From the results of seven intermediate curves including those shown in Figure 2, we calculate $pK_a = 10.83 \pm 0.03$ at 275 nm and a simi-

TABLE III
ABSORPTION AND CD EXTREMA FOR Pd(II) COMPLEXES CONTAINING L-AMINO ACIDS AND DIAMINOETHANE

		Absorption		Circular dichroism	
	pH	nm	ϵ	nm	$\Delta\epsilon$
Pd(en)(His)$^+$	7	288	400	321	−0.14
				277	+0.60
	13	291	430	300	+0.20
Pd(en)(histidinol)$^{2+}$	7	288	440	307	+0.48
				255	−0.27
	13	292	455	325	+0.25
				288	−0.18
Pd(en)(DABA)$^+$	7–13	290	400	280	+0.39
Pd(en)(DAPA)$^+$	8–13	287	300	282	+0.29
Pd(en)(3-MeHis)$^+$	7–13	288	395	323	−0.14
				280	+0.63
Pd(en)[Pd(en)(His)]$_2^{2+}$	7–12	292	375	325	−0.23
				275	+0.58

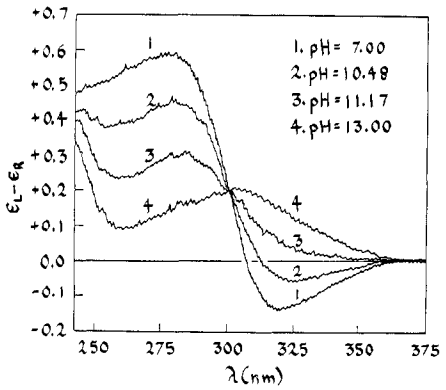

Figure 2.—Circular dichroism spectra of Pd(en)(L-His) at several pH values.

(8) R. B. Martin and R. Mathur, *J. Amer. Chem. Soc.*, **87**, 1065 (1965).

lar value at 320 nm. In order to determine the structural alteration responsible for the ionization with pK_a = 10.83 in the Pd(en)(His)$^+$ complex, absorption and CD properties of several model complexes were investigated.

For all the complexes listed in Table III, the wavelength of the absorption maximum indicates four nitrogen donor atoms about each Pd(II). The absorption and CD spectra of the corresponding complex with L-histidinol, where the $-COO^-$ group of histidine is replaced by $-CH_2OH$, also exhibit a pH dependence in the pH 7–13 region. No buffering action on the addition of base or pH dependence of spectra is displayed by the complexes containing L-2,4-diaminobutyrate (DABA) or L-2,3-diaminopropionate (DAPA) as ligands. The results of these three model complexes suggest that the carboxylate group of histidine is not involved in the pH-dependent transition and implicate the imidazole ring. To test this hypothesis the complex with L-3-methylhistidine was prepared. With this ligand the same six-membered chelate ring involving two nitrogen donor atoms as for histidine is possible, but the remote, unbound pyrrole nitrogen of the imidazole ring is substituted with a methyl group instead of a hydrogen atom. As shown in Table III the 3-methylated ligand yields a complex that gives absorption and CD spectra nearly identical with those of the histidine complex in neutral solutions, without exhibiting the pH dependence of the latter complex. In addition no titratable hydrogens appear below pH 12. Therefore, the transition occurring with pK_a = 10.83 in Pd(en)(His)$^+$ corresponds to ionization of the hydrogen at the unbound pyrrole nitrogen of the imidazole ring.

The ionization occurring at the unbound pyrrole nitrogen of histidine in Pd(en)(His)$^+$ may also be blocked by adding 0.5 equiv of Pd(en)Cl_2 and 1 equiv of NaOH to a solution containing the histidine complex. That the resulting complex is trinuclear is suggested by the absorption maximum at 292 nm indicating four nitrogen donors about all Pd(II) present and the lack of pH dependence of the spectra suggesting that the pyrrole hydrogens are already displaced in neutral solutions by the added Pd(II). Molar absorptivities reported in Table III for this complex are based on the total amount of Pd(II) present. The similar CD sign pattern and magnitudes of the simple and trinuclear complexes suggest that the original configuration of ligands about Pd(II) in Pd(en)(His)$^+$ is not appreciably altered by the addition of Pd(en)Cl_2 to form the trinuclear complex.

The pmr spectrum due to 1,2-diaminopropionate in Pd(en)(DAPA)$^+$ exhibits 12 lines and was subjected to a complete ABX analysis. For the methylene protons with proton B at higher field than A, $\Delta\nu$ = 0.19 ppm and J_{AX} = 4.5 and J_{BX} = 8.7 cps. If the dihedral angles are near the normal values of 60 and 180°, this result is inconsistent with a pseudoaxial carboxylate group where it is expected that $J_{AX} = J_{BX} \approx 3$ cps and indicates instead a pseudoequatorial disposition where unequal couplings of about the observed magnitude are predicted. Furthermore the analysis indicates that in the favored conformer the high-field methylene proton is axial and the low-field equatorial. Though complicated by extensive coupling a zeroth-order analysis of the X part of the pmr spectrum of the complex Pd(en)(DABA)$^+$ yields J_{AX} = 3.5 and J_{BX} = 7.2 cps. This result also suggests a predominantly equatorial carboxylate group. These conclusions ensure that the positive CD reported in Table III for these complexes is not to be ascribed to apical chelation of the carboxylate group.

All the L-α-amino acid ligands of Table III are bound to Pd(II) through two nitrogen donor atoms and yield a net positive CD for the sum of all transitions over the entire d–d manifold. This result is also found for 2:1 Pd(II) complexes and 1:1 and 2:1 Cu(II) complexes of these ligands when bound through two nitrogen donors and contrasts with the net negative sign found for complexes with L ligands bound through a substituted glycine mode.[2,9]

Discussion

Comparison of the results obtained upon adding base to Pd(GlyGly-L-Ala)$^-$ as shown in Figure 1 with those of the other compounds reported in Table I suggests that replacement of the bound carboxylate by hydroxide is responsible for the changes observed. Polymer formation is excluded, only a carboxyl terminal alanine tripeptide complex exhibits the CD inversion, and the corresponding tripeptide amide complex with four nitrogen donors about the nitrogen-liking Pd(II) displays no spectral changes with increased pH. In addition, the methylene hydrogens of the carboxyl terminal residue in Pd(GlyGlyGly)$^-$ shift more upfield than the other two pairs upon addition of base. This result is consistent with a now unbound, negatively charged carboxylate group. The apparent pK = 11.7 then refers to the midpoint pH for substitution of carboxylate by hydroxide ion in the Pd(II) coordination plane.

By analogy with the Pd(II) complex, production of a net positive CD with pK = 13.1 upon addition of base to Cu(GlyGly-L-Ala)$^-$ also suggests substitution of carboxylate by hydroxide in the chelate plane. The carboxylate group is unbound at high pH. As for Pd(II) only the tripeptide complex with a carboxyl terminal L-alanyl residue yields a net positive CD at high pH. Further evidence for hydroxide ion in the coordination plane and not in an axial position even for Cu(II) is provided by a CD comparison of dipeptide and tripeptide complexes. The one negative extremum CD curve of complexes such as Cu(Gly-L-Ala)^0 undergoes a splitting into two negative extrema upon ionization to hydroxide at about pH 9.5 of water coordinated in the fourth position.[10] The glycyl-L-alaninamide complex behaves similarly.[4] A similar splitting is also observed when a solution containing Cu(Gly-L-Ala-Gly)$^-$ is made 1 M in NaOH to yield a dinegatively charged complex.[11] Substitution of the carboxylate group by hydroxide results in a similar disposition of donor groups and the asymmetric center in the di- and tripeptide complexes.

From a comparison of the GlyGly-L-Ala complexes substitution of carboxylate by hydroxide ion in tetragonal complexes occurs with increasing difficulty in the order Pd(II) < Cu(II) < Ni(II) with pK values of

(9) E. W. Wilson, Jr., M. H. Kasperian, and R. B. Martin, *J. Amer. Chem. Soc.*, **92**, 5365 (1970).
(10) E. W. Wilson, Jr., and R. B. Martin, *Inorg. Chem.*, **10**, 1197 (1971).
(11) G. F. Bryce and F. R. N. Gurd, *J. Biol. Chem.*, **241**, 1439 (1966).

11.7, 13.1, and >14, respectively. For triglycine the pK values appear to be 12.0 and 12.8 for the Cu(II) and Ni(II) complexes, respectively.[12] These comparisons indicate that the methyl side chain in a carboxyl terminal residue inhibits substitution of carboxylate by hydroxide ion. This conclusion is consistent with the more restricted conformation available to an unbound carboxylate side chain with substituents larger than hydrogen.

L-α-Amino acids or peptides composed of L-amino acid residues when bound to transition metal ions in a normal way without binding of side chains usually display a net negative CD for d–d transitions.[2-4,10] Three Pd(II) complexes in Table I containing L-amino acid residues exhibit a net positive CD as does the Cu(II) complex of GlyGly-L-Ala at high pH. The common feature of all and only these cases is an L-amino acid residue bound to the metal ion by a deprotonated, trigonal amide nitrogen with an unbound carboxylate group. These results may be accommodated by any sector rule that describes the chelate plane as a nodal surface such as the hexadecant rule suggested as applicable to tetragonal transition metal ion complexes of Pd(II),[2] Cu(II),[4] and Ni(II).[3] L-Amino acid side chains lie in hexadecants assigned a negative sign. Examination of space-filling molecular models reveals that the least sterically crowded conformation of the carboxyl terminal residue with an unbound carboxylate group occurs upon rotation of the side chain out of a negative and into a positive sector below the plane of the chelate ring. In the same rotation the negative carboxylate group enters the negative hexadecant formerly occupied by the side chain in the fully chelated ligand. In the carboxyl terminal rotated conformation the negatively charged carboxylate group in a negative sector augments the activity of the side chain now in a positive sector so that substitution of carboxylate by hydroxide inverts the net sign of the optical activity over the d–d manifold. Of the Pd(II) complexes exhibiting net positive CD in Table I, significantly greater component magnitudes are observed for the first case where the coordinated hydroxide may yield a lesser cancelation of oppositely signed CD components than in the other cases where nitrogen donors occupy the four coordination sites.

Nonequivalence of glycyl methylene hydrogens has been observed in the pmr spectra of peptides containing a side chain in the amino terminal residue.[13,14] For aliphatic side chains nonequivalence occurs in the zwitterion form of L-AlaGly and in the central residue of L-AlaGlyGly and L-LeuGlyGly. Observable nonequivalence does not appear in the Pd(II) complexes of these peptides in neutral solutions. Nonequivalence does occur at high pH after substitution of the carboxylate group by hydroxide ion in the Pd(II) complex of L-AlaGly. Glycyl methylene nonequivalence is also observed in Pd(en)(AlaGly)^0 where the carboxylate group is unbound. Thus despite a more restricted conformation in the fully chelated complexes, nonequivalence appears only when the carboxylate group is unbound and hence negatively charged as in the zwitterion or hydroxide-substituted Pd(II) complex. These observations support the view that the field gradient provided by oppositely charged groups[13] is comparable in importance to restricted rotamer conformations[14] in leading to nonequivalence.

Pyrrole ionizations from simple 2:1 histidine complexes occur with average pK_a = 12.5 for Co(II) and 11.7 for Cu(II).[15] On the other hand, for the 1:1 glycyl-L-histidine complexes of Ni(II), Cu(II), and Pd(II) an apparent ionization occurring near pH 9.6 has been shown to be due to displacement of the pyrrole hydrogen by the free coordination site on the tetragonal metal ion to yield a polymer, probably a tetramer.[16] These metal ion promoted pyrrole hydrogen displacements occur in a significantly lower pH region (<10) than the simple pyrrole ionizations that take place with pK_a > 10. Thus assignment of the ionization with pK_a = 10.83 in Pd(en)(His)$^+$ to a simple pyrrole ionization with no new complexation is consistent with the stronger interaction of Pd(II) compared to Co(II) and Cu(II).

(12) E. J. Billo and D. W. Margerum, *J. Amer. Chem. Soc.*, **92**, 6811 (1970).

(13) V. J. Morlino and R. B. Martin, *ibid.*, **89**, 3107 (1967); *J. Phys. Chem.*, **72**, 2661 (1968).

(14) A. Nakamura and O. Jardetzky, *Proc. Nat. Acad. Sci. U. S.*, **58**, 2212 (1967).

(15) P. J. Morris and R. B. Martin, *J. Amer. Chem. Soc.*, **92**, 1543 (1970).

(16) P. J. Morris and R. B. Martin, *J. Inorg. Nucl. Chem.*, **33**, 2913 (1971).