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Schiff Bases of Pyridoxal 5′-Phosphate and 5′-Deoxypyridoxal with Phenylglycine Derivatives and Their Metal Complexes

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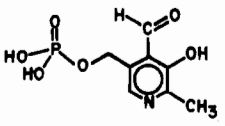
Received May 31, 1989

Potentiometric p[H] measurements are employed to evaluate protonation constants, metal complex stabilities, and Schiff base (SB) formation constants in solutions containing pyridoxal 5′-phosphate (PLP) 5′-deoxypyridoxal (DPL), phenylglycine and its derivatives, and divalent transition-metal ions at 25 °C and at an ionic strength of 0.100 M (KNO_3). Protonation constants for phenylglycine (PG), (4-methoxyphenyl)glycine (MPG), and (4-sulfophenyl)glycine (SPG) indicate that these synthetic amino acids are more acidic than glycine and other natural amino acids. Stability constants are reported for 1:1 and 1:2 complexes of Mn^{2+}, Co^{2+}, Ni^{2+}, Cu^{2+}, and Zn^{2+} with PG, MPG, and SPG. Hydrolysis of these metal ions complicates the study of the nature of the metal complex formed at high p[H]. The vitamin B_6 derivatives, pyridoxal 5′-phosphate and 5′-deoxypyridoxal, were found to form only 1:1 metal complexes. The equilibrium constants for the formation of Schiff bases by PLP and DPL with PG, MPG, and SPG are reported. The monoprotonated form of the Schiff base was found to be the most stable species in each of the systems studied. The equilibrium constants are reported for protonated 1:1 Cu(II)–Schiff base complexes, $CuSBH_x^{n+}$ and for 1:1 and 2:1 Schiff base complexes of Mn(II), Co(II), Ni(II), and Zn(II). The formation of a number of hydroxometal Schiff base chelates in alkaline solutions is observed. The equilibrium constants determined are employed to calculate the distribution of complex species as a function of p[H] in solutions containing these ligands and the metal ions identified above.

Introduction

Pyridoxal 5′-phosphate (PLP, **1**), the form in which vitamin B_6 exists in nature, is known to catalyze important biochemical reactions such as transamination in physiological as well as model systems.[1-4] Vitamin B_6 catalysis involves formation of Schiff base intermediates through condensation of the carbonyl group of the coenzyme and the α-amino group of amino acids.



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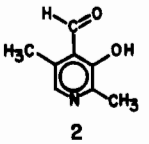

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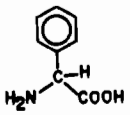

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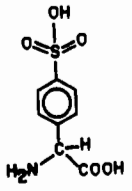

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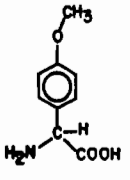

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For some time, oxidative deamination of α-amino acids mediated by copper-containing deaminases was thought to involve pyridoxal 5′-phosphate.[5-8] Although the absorption spectra of a number of copper-containing deaminases show[9-12] similarities to that of PLP (**1**), attempts to isolate this compound from deaminases have been unsuccessful.[12-14] Recent studies[15-19] indicate the presence of a benzoquinone derivative, pyroquinolinequinone (PQQ), in amine oxidases from various sources, and it was suggested that this compound is the active species in the Cu(II)-containing amine oxidases. Also, Mondovi and co-workers[20] suggested that the PQQ is modified by interactions with the proteic matrix so that its reactivity is similar to that of **1**.

Earlier model studies[21-23] have shown that vitamin B_6 catalyzes the deamination of amino acids in the presence of transition-metal ions and dioxygen. Hamilton and Revesz[22] proposed a mechanism for this reaction whereby a transition-metal ion forms a chelate with the terdentate Schiff base (SB) and transfers a pair of electrons to end-on bound dioxygen to maintain the oxidized form of the coenzyme. An alternative pathway was proposed by Tatsumoto et al.,[23] in which dioxygen was considered bound directly to the SB in the metal–Schiff base chelate in order to oxidize the amino acid. The present research was undertaken to provide information on the complexes formed anaerobically in vitamin B_6–amino acid systems as the initial phase in the subsequent study of oxidative deamination.

For this investigation, the following compounds were selected: PLP (**1**) and 5′-deoxypyridoxal (DPL, **2**) as forms of the coenzyme, phenylglycine (PG, **3**), (4-methoxyphenyl)glycine (MPG, **4**), and (4-sulfophenyl)glycine (SPG, **5**) as amino acid substrates, and Mn(II), Co(II), Ni(II), Cu(II), and Zn(II) as transition-metal catalysts.

In order to understand the kinetic behavior of amino acid substrates, vitamin B_6 catalysts, and their Schiff bases in the presence and absence of metal ions in aqueous solutions, it is necessary to determine the molecular species formed under various solution conditions. Thus the first phase of this investigation consists of detailed equilibrium studies involving all of the solution

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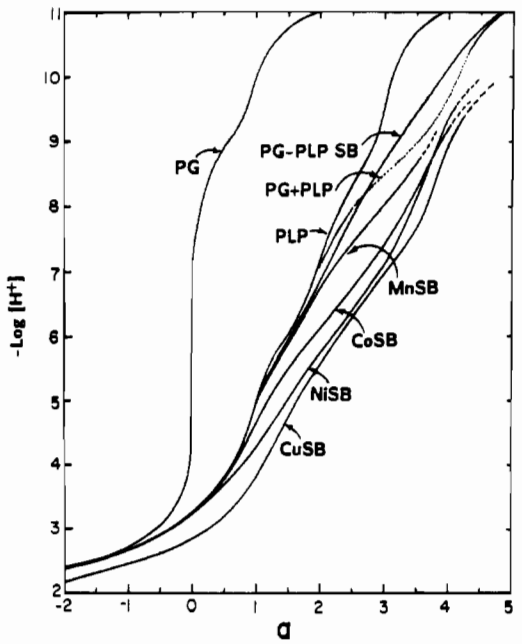

Figure 1. Potentiometric titration curves (PG–PLP systems) for the measurements made at 25 °C, μ = 0.100 M (KNO_3), titrant (KOH) concentration = 0.1033 M, a = mol of base/mol of ligand, and total initial volume V_i = 50.0 mL for PG, PLP, SB, and PG + PLP and 55.0 mL for MnSB, CoSB, NiSB, CuSB: PG, 2.00×10^{-3} M PG + 3.00×10^{-3} M HNO_3; PLP, 2.00×10^{-3} M PLP + 3.00×10^{-3} M HNO_3; SB, 2.00×10^{-3} M PG + 2.00×10^{-3} M PLP + 3.00×10^{-3} M HNO_3; PG + PLP, calculated curve for 2.00×10^{-3} M PG + 2.00×10^{-3} M PLP + 3.00×10^{-3} M HNO_3 assuming no Schiff base formation; MnSB, 9.95×10^{-4} M Mn(II) + 2.00×10^{-3} M PG + 2.00×10^{-3} M PLP + 3.00×10^{-3} M HNO_3; CoSB, 1.011×10^{-3} M Co(II) + 2.00×10^{-3} M PG + 2.00×10^{-3} M PLP + 3.00×10^{-3} M HNO_3; NiSB, 9.86×10^{-4} M Ni(II) + 2.00×10^{-3} M PG + 2.00×10^{-3} M PLP + 3.00×10^{-3} M HNO_3; CuSB, 1.003×10^{-3} M Co(II) + 2.00×10^{-3} M PG + 2.00×10^{-3} M PLP + 3.00×10^{-3} M HNO_3.

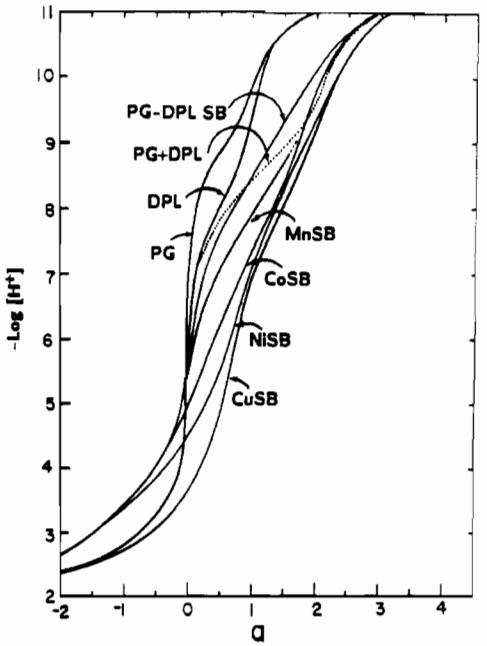

Figure 2. Potentiometric titration curves (PG–DPL systems) for the measurements made at 25 °C, μ = 0.100 M (KNO_3), titrant (KOH) concentration = 0.1033 M, a = mol of base/mol of ligand, and total initial volume V_i = 50.0 mL for PG, DPL, SB, and PG + DPL and 55.0 mL for MnSB, CoSB, NiSB, and CuSB: PG, 2.00×10^{-3} M PG + 3.00×10^{-3} M HNO_3; DPL, 2.00×10^{-3} M DPL + 3.00×10^{-3} M HNO_3; SB, 2.00×10^{-3} M PG + 2.00×10^{-3} M DPL + 3.00×10^{-3} M HNO_3; PG + DPL, calculated curve for 2.00×10^{-3} M PG + 2.00×10^{-3} M DPL + 3.00×10^{-3} M HNO_3 assuming no Schiff base formation; MnSB, 9.95×10^{-4} M Mn(II) + 2.00×10^{-3} M PG + 2.00×10^{-3} M DPL + 3.00×10^{-3} M HNO_3; CoSB, 1.011×10^{-3} M Co(II) + 2.00×10^{-3} M PG + 2.00×10^{-3} M DPL + 3.00×10^{-3} M HNO_3; NiSB, 9.86×10^{-4} M Ni(II) + 2.00×10^{-3} M PG + 2.00×10^{-3} M DPL + 3.00×10^{-3} M HNO_3; CuSB, 1.003×10^{-3} M Cu(II) + 2.00×10^{-3} M PG + 2.00×10^{-3} M DPL + 3.00×10^{-3} M HNO_3.

components described above, including the complexes formed by metal ions with Schiff bases as well as with the individual amino acids and the forms of coenzymes employed.

Experimental Section

Materials. Pyridoxal 5′-phosphate was purchased from ICN Biomedicals and the extent of hydration was determined by the potentiometric method.[24] D-(−)phenylglycine was obtained from Sigma Chemical Co. and recrystallized from an ethanol–water mixture at p[H] 4.6. 5′-Deoxypyridoxal was prepared by the method of Iwata.[25] (4-Methoxyphenyl)glycine and (4-sulfophenyl)glycine were synthesized. A carbonate-free concentrate of potassium hydroxide (Dilut-it) was purchased from J. T. Baker Chemical Co. Analytical reagent grade disodium salt of H_2EDTA and acidimetric standard grade potassium hydrogen phthalate were obtained from MCB and Fisher. Analytical reagent grade nitrate salts of metal ions Mn(II), Co(II), Ni(II), Cu(II), and Zn(II) were obtained from Mallinckrodt Chemical Co.

(4-Sulfophenyl)glycine. First, 5.0 g of phenylglycine was mixed thoroughly with 8.0 mL of fuming sulfuric acid (37% SO_3) at room temperature and stirred for about 48 h. This mixture was then diluted with 10 mL of distilled water and cooled to room temperature. The compound that precipitated was filtered through a sintered glass funnel and was washed repeatedly with small volumes of distilled water to remove the excess acid. Colorless (4-sulfophenyl)glycine was recrystallized from hot water. Yield: 4.0g (52%). Anal. Calcd for $C_8H_9NO_5S$: C, 41.56; H, 3.92; N, 6.06; S, 13.87. Found: C, 40.98; H, 4.32; N, 5.71; S, 13.64.

(4-Methoxyphenyl)glycine. For the synthesis of (4-methoxyphenyl)-glycine, the method described for the synthesis of phenylglycine[26] was employed with modifications. 4-Methoxybenzaldehyde was used in place of benzaldehyde as the starting material; the rest of the procedure was unchanged. Anal. Calcd for $C_9H_{11}NO_3$: C, 59.66; H, 6.12; N, 7.73. Found: C, 59.56; H, 6.12; N, 7.69.

Preparation of Solutions. Metal ion solutions (0.01 M) were prepared from their nitrate salts. The ionic strength of these solutions was adjusted to 0.100 M by the addition of the required amount of KNO_3. These solutions were standardized by complexometric titration with a standard solution of Na_2H_2EDTA according to the procedure of Schwarzenbach and Flaschka.[27]

Amino acid solutions (0.0020 M) were prepared by mixing accurately weighed samples into a known volume of doubly distilled water. A known amount of the standard solution of HNO_3 was added when necessary. Pyridoxal derivatives were weighed out and added directly to the experimental solution to avoid decomposition.

A carbonate-free aqueous solution of 0.10 M potassium hydroxide was prepared from a "Dilut-it" ampule and was stored in a serum bottle connected with a Metrohm piston buret capable of delivering solution with an accuracy of 0.0005 mL. This solution was standardized by titration with standard potassium acid phthalate. The accuracy of standardization and the amount of dissolved carbonate in potassium hydroxide were regularly checked by a standard GRAN analysis.[28]

Potentiometric Measurements. Potentiometric equilibrium measurements were carried out with a Corning Research p[H]/ion meter fitted with glass (Sargent Welch) and calomel reference (Fisher) electrodes and calibrated to read −log [H^+] directly prior to each experiment with a freshly prepared solution of HNO_3 (p[H] = 2.000, μ = 0.100 M). The temperature was maintained at 25.00 ± 0.05 °C with a thermostated cell, and the ionic strength was adjusted to 0.100 M by the addition of KNO_3 as supporting electrolyte. Typical concentrations of experimental solutions were 0.0020 M in ligand with the molar concentration of metal ions equivalent to or half of that of ligand for the study of metal complexation. Oxygen and carbon dioxide were excluded from the reaction mixture by maintaining a slight positive pressure of purified nitrogen in the reaction cell. The last traces of oxygen were removed from the nitrogen by

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Table I. Protonation Constants[a,b]

ligand	log K^{H}_{HL}[c]	log $K^{H}_{H_2L}$	log $K^{H}_{H_3L}$
PG (3)	8.92	1.86	
MPG (4)	9.07	1.95	
SPG (5)	8.66	1.77	
glycine[d]	9.56	2.36	
phenylalanine[d]	9.09	2.19	
PLP (1)	8.27	6.09	3.60
	8.33[e]	6.10[e]	3.62[e]
DPL (2)	8.04	4.09	
	8.03[e]	4.14[e]	

[a] μ = 0.100 M (KNO_3); t = 25.0 °C. [b] σ_{fit} = 0.002. [c] $K^{H}_{H_nL} = [H_nL]/[H_{n-1}L][H^+]$. [d] From ref 30. [e] From ref 31.

passing it through an alkaline pyrogallol solution.

Equilibrium p[H] values were determined at every incremental addition of KOH to the experimental solutions. Each titration was generally continued until the p[H] of the solution increased to 11 and then it was back-titrated with a standard solution of HNO_3. The experimental p[H] values were plotted as a function of a values to obtain p[H] profiles for each systems (Figures 1 and 2). An a value is the ratio of moles of base added to moles of ligand.

Computations. The proton association constants, formation and protonation constants of Schiff bases, and formation constants of 1:1 and 2:1 ligand-to-metal complexes were calculated by the use of the Fortran program BEST.[29] The program refines stability constants by iterative nonlinear least-squares fit of potentiometric equilibrium curves through a set of simultaneous mass balance equations for all the components expressed in terms of known and unknown equilibrium constants. For example, the p[H] profile of a metal ion Schiff base system is analyzed with the protonation constants of individual ligands, stability constants for metal complexes that may form, the constants for the formation and protonation of Schiff bases as known constants (determined previously by separate experiments) and a series of metal–Schiff base chelate formation, protonation, and deprotonation constants as unknowns (but with reasonable initial estimated values). Stability constants of 1:1 and 2:1 ligand-to-metal complexes were evaluated from the p[H] profiles for the systems containing 1:1 and 2:1 molar stoichiometry of ligand to metal ion, respectively. The errors reported (σ_{fit}) are based on the differences between the calculated and experimental hydrogen ion concentrations over the entire equilibrium curve for each system. The species considered present in the experimental solutions were those that one would expect to form according to established principles of coordination chemistry. Care was taken not to formulate additional complex species merely for the purpose of improving the fit of the calculated results to the experimental curves. Species distribution curves were calculated and plotted with the Fortran program SPE[24] and the Basic program GENPLOT developed in this laboratory.

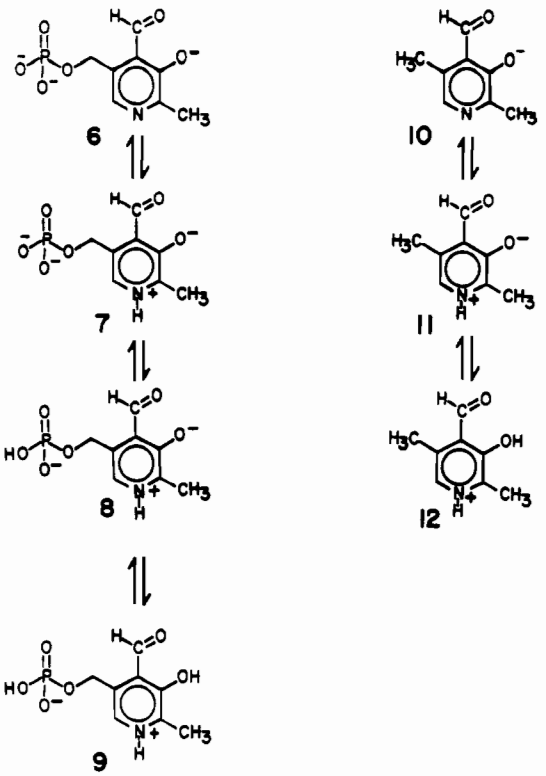
Results and Discussion

Protonation. Protonation constants defined by eq 1 are listed in Table I.

$$H_{n-1}L^{x-1} + H^+ \rightleftharpoons H_nL^x \qquad K^{H}_{H_nL} = \frac{[H_nL^x]}{[H_{n-1}L^{x-1}][H^+]} \qquad (1)$$

Amino Acids. Protonation constants for amino acids with aromatic substituent on the α-carbon had not been reported previously. The proton affinities of the carboxylate group were determined by the addition of known excess of acid in the beginning of the potentiometric determination. The data in Table I show that aromatic substitution on the α-carbon decreases the basicity of both the amino groups and carboxylate groups compared to β-substituted amino acids such as phenylalanine. The protonation constant of SPG shows that sulfonation of the phenyl ring further decreases the basicity of both the amino and carboxylate groups. In the case of MPG, on the other hand, the electronic effect of the methoxy substituent counteracts that of the phenyl ring, and an increase is therefore noticed in protonation constants, compared to those of PG. In general, synthetic amino acids analyzed in the present study are less basic than natural amino acids.

Scheme I



Pyridoxal Derivatives. Pyridoxal (PL), a common form of the vitamin B_6 group of coenzymes, exists predominantly as a hemiacetal in aqueous solutions. Harris and co-workers[31] reported that the concentration of the hemiacetal form is over 90% of the total pyridoxal concentration. The choice of PLP and DPL for this study eliminates hemiacetal formation, thus providing more active carbonyl groups and greatly enhanced tendency toward SB formation with amino acids.

Various techniques have been utilized to evaluate protonation constants of vitamin B_6 and its analogues in various media, temperatures, and ionic strengths.[32-36] The first protonation of trianionic PLP occurs at the pyridine nitrogen with an equilibrium constant of $10^{8.27}$. The second protonation occurs at the phosphate group, with higher proton affinity than that of phenoxide, with protonation constants of $10^{6.09}$ and $10^{3.60}$, respectively. The fourth protonation occurs only in very acidic media, and its equilibrium constant was not determined. The major species formed in this protonation sequence are shown in Scheme I. Equilibrium constants for various microspecies in equilibrium with each other over a range of p[H] values have been reported.[3,30,32]

The protonation constants of DPL are considerably lower than those of PLP; thus, it appears that the negative charge of the phosphate ester group effectively increases the basicity of the pyridine nitrogen.

Metal Complexes. Chelate stability constants of 1:1 and 2:1 stoichiometry of a ligand and a metal ion are defined by eq 2 and 3. See **13** for an illustration of 2:1 complexes.

$$M^{2+} + L^{n-} \rightleftharpoons ML^{2-n} \qquad \beta_{ML} = \frac{[ML^{2-n}]}{[M^{2+}][L^{n-}]} \qquad (2)$$

$$M^{2+} + 2L^{n-} \rightleftharpoons ML_2^{2-2n} \qquad \beta_{ML_2} = \frac{[ML_2^{2-2n}]}{[M^{2+}][L^{n-}]^2} \qquad (3)$$

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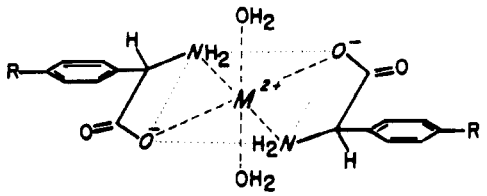

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Stability constants for PG, MPG, and SPG with the divalent metal ions selected are listed in Table II. The presence of a bulky phenyl ring on the α-carbon atom of the amino acid seems to cause steric hindrance, which deters the formation of 3:1 amino acid–metal complexes, which are known for some amino acids that do not have large α-substituents.[30,37] Substituents in the 4-position of the benzene ring of phenylglycine produce only a marginal difference in the stabilities of the metal complexes formed. MPG, being the most basic of the amino acids studied, forms the most stable complexes. This is especially true of the 2:1 complex formed with Cu(II). The assessment of the formation of hydroxo-coordinated metal complexes in basic solution was often complicated by the hydrolysis of the metal ion, and equilibrium constants for the formation of hydroxometal chelates are not reported. Stabilities for complexes of aromatic amino acids studied followed the order for analogous compounds in the literature.[30] The decrease in the basicity of the amino acids studied seems to have affected only to a small extent their ability to complex divalent metal ions.

Pyridoxal derivatives, PLP and DPL, form stable chelate compounds with divalent metal ions by coordination through the phenoxide and formyl groups. There are some indications[38,39] of phosphate–formyl coordination in the monoprotonated form of PLP, but the chelate ring is too large to significantly contribute to the stability of such a species. Moreover, the monoprotonated species exists only in a narrow p[H] region due to the protonation of phosphate to form the diprotonated species, seriously diminishing its coordination tendency. Equilibria governing the protonations of metal complexes of PLP and DPL are represented by eq 4.

$$H_{n-1}ML^{x} + H^{+} \rightleftharpoons H_nML^{x+1}$$

$$K^{H}_{H_nML} = \frac{[H_nML^{x+1}]}{[H_{n-1}ML^{x}][H^{+}]} \qquad (4)$$

Stability constants and protonation constants for metal complexes of PLP and DPL are listed in Table III. As expected, completely deprotonated forms of the coenzyme have better electron-donor ability to form 1:1 complexes with metal ions than their protonated analogues, mainly because protonation of the ligand at the pyridine nitrogen greatly decreases basicities of the chelating functional groups. A comparison of stability constants for protonated forms of DPL and PLP with various metal ions suggests that the ease of protonation on a metal complex is inversely proportional to its stability. This is a well-known relationship and is due to the differences in the polarization of the electron density toward the metal ion and away from the protonation site. The reported values[3] for Cu(II) and Zn(II) complexes of PLP agree with those evaluated in this study. Metal complexes of DPL have not been reported in the literature thus far. DPL was found to be a weaker ligand for transition metals than PLP. Both PLP and DPL do not have a tendency toward the formation of ML_2 complexes under the experimental conditions employed. Hydroxo-coordinated complex species do not seem to be stable enough to form in the experimental p[H] region (2.2–9.0).

Schiff Base Formation and Protonation. The equilibrium p[H] profiles for the systems containing equimolar quantities of PG and PLP and equimolar quantities of PG and DPL are shown in Figures 1 and 2, respectively. The curves labeled SB are different from the p[H] profiles calculated by assuming no interaction between two Schiff base forming components, indicated by the dotted curve. Although the differences seem small, the computational analysis indicated the formation of a substantial amount of HSB and H_2SB species (and H_3SB in PLP systems) and a relatively smaller amount of SB. The Schiff base formation from vitamin B_6 coenzymes involves a short-lived carbinolamine intermediate that undergoes dehydration in a subsequent step.[2,3] It has been suggested that, in acidic solutions, the formation of carbinolamine is generally the rate-determining step while, in neutral and alkaline solutions, it is the elimination of the water molecule of the carbinolamine that governs the rate. However, the relative concentration of the carbinolamine in aqueous solutions is suggested[40] to be insignificant to be considered in the evaluation

Table II. Formation Constants for Metal Complexes of PG, MPG, and SPG[a,b]

ligand	metal ion	log β_{ML}[c]	log β_{ML_2}[d]
PG	Mn^{2+}	2.58	4.50
	Co^{2+}	3.89	7.04
	Ni^{2+}	4.98	9.08
	Cu^{2+}	7.45	13.65
	Zn^{2+}	4.13	7.85
MPG	Mn^{2+}	2.65	4.86
	Co^{2+}	3.96	7.11
	Ni^{2+}	5.05	9.20
	Cu^{2+}	7.59	14.16
	Zn^{2+}	4.08	7.68
SPG	Mn^{2+}	2.29	3.97
	Co^{2+}	4.01	6.78
	Ni^{2+}	5.11	8.83
	Cu^{2+}	7.45	13.13
	Zn^{2+}	4.28	7.49

[a] μ = 0.100 M (KNO_3); t = 25.0 °C. [b] σ_{fit} = 0.003. [c] β_{ML} = [ML]/[M][L]. [d] β_{ML_2} = $[ML_2]/[M][L]^2$.

Table III. Formation and Protonation Constants for Metal Complexes of PLP and DPL[a,b]

vitamin B_6	metal ion	log K_{ML}[c]	log K^{H}_{MHL}[d]	log $K^{H}_{MH_2L}$
PLP	Mn^{2+}	3.25	7.73	5.6
	Co^{2+}	3.88	6.96	5.2
	Ni^{2+}	3.99	6.64	4.3
	Cu^{2+}	6.21	5.46	4.6
	Zn^{2+}	3.75	6.53	
DPL	Mn^{2+}	2.91	7.51	
	Co^{2+}	2.90	6.26	
	Ni^{2+}	3.39	6.66	
	Cu^{2+}	5.04	5.07	
	Zn^{2+}	2.40	6.09	

[a] μ = 0.100 M (KNO_3); t = 25.0 °C. [b] σ_{fit} = 0.003. [c] K_{ML} = [ML]/[M][L]. [d] $K^{H}_{MH_nL}$ = $[MH_nL]/[MH_{n-1}L][H^+]$.

Table IV. Schiff Base Formation and Protonation[a,b]

Schiff base m–n[c]	log β_{SB}[d]	log K^{H}_{HSB}[e]	log $K^{H}_{H_2SB}$	log $K^{H}_{H_3SB}$	log K'_{HSB}[g]
PLP–PG	0.9	10.97	7.13	5.7	3.67
PLP–MPG	1.4	10.63	7.10	5.5	3.77
PLP–SPG	1.0	10.49	7.33	5.8	3.14
DPL–PG	0.9	11.00	6.11		3.90
DPL–MPG	1.0	11.00	6.26		4.01
DPL–SPG	1.1	10.62	6.69		3.72
PLP–Gly[f]	1.0	11.88	6.65		

[a] μ = 0.100 M (KNO_3); t = 25.0 °C. [b] σ_{fit} = 0.007. [c] Schiff base forming components: pyridoxal derivative (m) and amino acid (n). [d] $\beta_{SB_{m-n}}$ = [SB]/[m][n]. [e] $K^{H}_{H_nSB}$ = $[H_nSB]/[H_{n-1}SB][H^+]$. [f] From ref 34. [g] K'_{HSB} = [HSB]/[mH][n].

(37) Smith, R. M.; Martell, A. E. *Critical Stability Constants*; Plenum: New York, 1980; Vols. 3 and 5.
(38) Vishwanathan, T. S.; Swift, T. J. *Can. J. Chem.* **1979**, *57*, 1050.
(39) Vishwanathan, T. S.; Swift, T. J. *Can. J. Chem.* **1980**, *58*, 1118.
(40) Gout, E.; Zadour, M.; Beguin, C. C. *Nouv. J. Chem.* **1984**, *8*, 243.
(41) Capasso, S.; Giordano, F.; Mattia, C.; Mazzarella, L.; Ripamonti, A. *J. Chem. Soc., Dalton Trans.* **1973**, 2228.

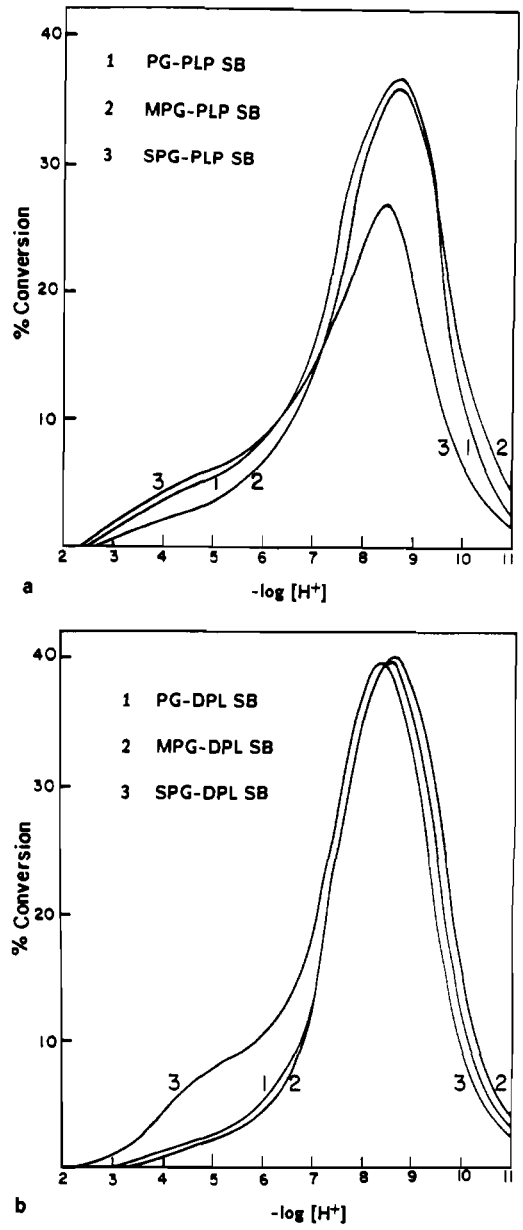


Figure 3. Schiff base formation in PLP (a) and DPL (b) systems. Percent conversion = total concentration of Schiff bases/initial concentration of the amino acid × 100. Total concentration of Schiff base species = $[H_2SB] + [HSB] + [SB]$. In each case, total concentration of amino acid = 2.000×10^{-3} M, [PLP] = 2.000×10^{-3} M, [DPL] = 2.000×10^{-3} M; V_i = 50.0 mL, t = 25 °C, and μ = 0.100 M (KNO_3). PG–PLP stands for Schiff base formation with components PG and PLP, etc.

of macroscopic equilibrium constants for Schiff base formation.

Figure 3 shows the extent of formation of Schiff base species as a function of p[H]. It was observed that the amount of Schiff base formed gradually increases with p[H] until a maximum is reached around p[H] = 8.5. The total concentration of the Schiff base species rapidly drops above this p[H]. A maximum of 40% of the components reacted to form Schiff bases under the experimental conditions. The monoprotonated Schiff base is the predominant species above p[H] = 6.5. The species with additional protonation were found to form at lower p[H]. In contrast to PG and MPG Schiff base systems, SPG systems form higher concentrations of di- and triprotonated Schiff bases. A substantial difference in the extent of conversion among SPG–PLP and SPG–DPL Schiff base systems seems to be due to the 5′-phosphate, which lowers the overall Schiff base formation by reducing the electrophilicity of the 4-carbonyl group.

The stability constants β_{SB}, evaluated for the formation of unprotonated Schiff base from unprotonated forms of various amino acids and pyridoxal derivatives are listed in Table IV. The equilibria governing this reaction as well as subsequent protonation are expressed by eqs 5a, 5b, and 6, where HAA stands for PG, SPG (neglecting charge of the sulfo group), and MPG and H_xVB stands for PLP (x = 3) and DPL (x = 1), respectively.

$$AA^- + VB^{x-} \rightleftharpoons SB^{-1-x} \qquad \beta_{SB} = \frac{[SB^{-1-x}]}{[AA^-][VB^{x-}]} \tag{5a}$$

$$AA^- + HVB^{1-x} \rightleftharpoons HSB^{x-} \qquad K'_{HSB} = \frac{[HSB^{x-}]}{[AA^-][HVB^{1-x}]} \tag{5b}$$

$$H_{n-1}SB^{n-x-1} + H^+ \rightleftharpoons H_nSB^{n-x}$$

$$K^H_{H_nSB} = \frac{[H_nSB^{n-x}]}{[H_{n-1}SB^{n-x-1}][H^+]} \tag{6}$$

The low equilibrium constants for the formation of unprotonated SB suggest that the process of SB formation between unprotonated components is not strongly favored in aqueous solutions. The log β_{SB} values for various unprotonated Schiff bases reported[2,3] for vitamin B_6 coenzymes range between 0.5 and 2.0 in agreement with the constants reported here, which range from 0.9 to 1.4. The monoprotonated form of the SB, **15**, is the most stable and is predominant in aqueous solutions (see Section II). Therefore, it is more appropriate to express the Schiff base reaction as the formation of HSB. The proton in HSB is bound to the azomethine nitrogen and is stabilized by the H-bonding interaction with phenoxide and carboxylate groups. A constant K'_{HSB} is defined for the formation of this species, starting from the monoprotonated form of the coenzyme and unprotonated form of the amino acid (eq 5b). A comparison between β_{SB} and K'_{HSB} suggests that the formation of HSB is orders of magnitude more favorable than the formation of SB. The species distribution curves (not shown) developed from these equilibrium data indicate that the relative amount of unprotonated Schiff base does not exceed 5% under the experimental conditions while HSB accounts for over 90% of the total amount of the Schiff base present. The log K'_{HSB} values are smaller for the Schiff bases of PLP compared to those for the Schiff bases of DPL indicating that the 4′-carbonyl carbon of PLP is less electrophilic than that of DPL because of the presence of negatively charged 5′-phosphate. Also, it is observed that the value of K'_{HSB} increases with the basicity of the parent amino acid.

While the magnitude of K'_{HSB} indicates the equilibrium formation of monoprotonated SB, the magnitude of K^H_{HSB} is a measure of the basicity of the azomethine group. Further protonation of **15** occurs at pyridine nitrogen and phosphate oxygen. A comparison of $K^H_{H_2SB}$ and $K^H_{H_3SB}$ with K^H_{HPLP} and $K^H_{H_2PLP}$ suggests that formation of H_2SB involves the protonation of the pyridine nitrogen, as is the case with HPLP (see Scheme I and II). The decrease in the magnitude of the protonation constant of pyridine nitrogen upon Schiff base formation is expected because conjugation to the more strongly electron-withdrawing protonated 4′-imine nitrogen, together with the H-bonding interaction of phenoxide, renders pyridine nitrogen lower in basicity. This effect is enhanced in the case of DPL Schiff bases where protonation constant of pyridine nitrogen is decreased by about 2 log units upon Schiff base formation. The differences in the Schiff bases of PLP and DPL seem to be due to the charge on the phosphate group of PLP, which generally tends to increase the basicity of all donor groups in the adjacent ring. The relative concentrations of di- and triprotonated forms of the Schiff base are substantially lower than that of the monoprotonated form because of increased competition at lower p[H] with amino acid protonation, making amino acids less available for Schiff base formation. The second protonation constant[3] of PLP–glycine is $10^{6.65}$ and that of the PLP–alanine Schiff base is $10^{6.57}$. Corresponding protonation constants for the Schiff bases reported here range between $10^{7.10}$ and $10^{7.33}$. This difference can be attributed to the difference in the basicity of the parent amino acids. The presence of a phenyl ring on the carbon atom adjacent to the azomethine nitrogen decreases the basicity of the azomethine nitrogen, which in turn increases the basicity of the second protonation site, pyridine nitrogen.

Table V. Logarithms of Stability Constants[a,b] for Metal Complexes of PLP Schiff Bases

M^{2+}	β_{MSB}	K^{H}_{MSBH}	$K^{H}_{MSBH_2}$	$K_{MSB(OH)}$	$\beta_{M(MSB)2}$	$K^{H}_{M(SB)_2H}$	$K^{H}_{M(SB)_2H_2}$
				PG–PLP			
Mn	7.31	*c*	*c*	*c*	14.5	9.2	9.1
Co	9.70	7.80	5.9	*c*	17.3	9.3	*c*
Ni	10.41	7.75	6.0	*c*	17.7	9.6	6.8
Cu	16.51	6.78	5.5	−10.76	*c*	*c*	*c*
Zn	9.77	7.36	6.1	*c*	20.2	7.7	7.6
				MPG–PLP			
Mn	7.56	7.86	*c*	*c*	*c*	*c*	*c*
Co	9.68	7.83	5.9	*c*	16.7	9.1	7.7
Ni	10.56	7.81	6.1	*c*	17.3	9.4	7.8
Cu	16.34	6.86	5.6	−10.89	*c*	*c*	*c*
				SPG–PLP			
Mn	7.30	7.93	6.8	*c*	13.7	8.7	8.3
Co	9.00	8.07	6.1	*c*	16.6	9.0	*c*
Ni	10.12	7.92	6.1	*c*	16.2	9.6	7.6
Cu	15.85	6.91	5.6	−10.62	*c*	*c*	*c*
Zn	9.33	7.59	5.8	*c*	*c*	*c*	*c*

[a] See definitions in the text. [b] σ_{fit} = 0.007–0.018. [c] Species concentration < 1 ppm.

Table VI. Logarithms of Stability Constants[a,b] for Metal Complexes of DPL Schiff Bases

M^{2+}	β_{MSB}	K^{H}_{MSBH}	$K_{MSB(OH)}$	$\beta_{M(SB)2}$	$K^{H}_{M(SB)_2H}$	$K^{H}_{M(SB)_2H_2}$
			PG–DPL			
Mn	7.69	7.75	*c*	14.2	8.2	*c*
Co	9.94	7.46	*c*	17.9	9.1	7.1
Ni	12.54	6.06	−11.08	*c*	*c*	*c*
Cu	16.76	6.46	−10.00	*c*	*c*	*c*
Zn	9.88	7.00	*c*	16.7	8.1	*c*
			MPG–DPL			
Mn	7.74	7.82	*c*	14.3	7.9	*c*
Co	10.33	7.08	*c*	17.0	9.1	7.2
Ni	11.70	6.95	−10.75	*c*	*c*	*c*
Cu	17.18	6.16	−11.49	*c*	*c*	*c*
Zn	9.90	7.10	*c*	17.4	8.5	7.2
			SPG–DPL			
Mn	7.53	7.80	*c*	13.6	*c*	*c*
CO	9.63	7.71	*c*	17.4	9.3	6.9
Ni	12.21	6.27	−12.81	*c*	*c*	*c*
Cu	16.08	6.99	−10.64	*c*	*c*	*c*
Zn	9.81	7.02	*c*	15.8	*c*	*c*

[a] See definitions in the text. [b] σ_{fit} = 0.010–0.019. [c] Species concentration < 1 ppm.

Metal Complexes of Schiff Bases. The Schiff bases formed in this research serve as terdentate ligands by the coordination of the imino, phenoxide, and carboxylate donor groups with the metal ion. It has been suggested that for such complexes the metal ion is more strongly coordinated to the imino nitrogen and phenoxide oxygen than to the carboxylate oxygen.[42] The extension of the conjugation of the pyridine ring to the Schiff base nitrogen requires that the six-membered chelate ring containing the metal ion, phenoxide, and imino nitrogen coincide with the plane of the pyridine ring. Coordination of the carboxylate group to the metal ion also orients the carboxylate oxygen close to that plane.

The potentiometric titration curve for a Schiff base system is modified in the presence of transition-metal ions (see Figures 1 and 2). Inspection of the (metal ion)–SB curves indicates that the coordination of Cu(II) occurs well below p[H] = 3, while for other metal ions coordination occurs at higher p[H] values (note that calculations are required to demonstrate that the Cu(II) complex formed is a 1:1 Cu(II)–SB complex). The p[H] measurements were carried out for systems containing 1:1:1 and 1:1:0.5 molar ratios of amino acid to pyridoxal derivative to metal ion, respectively. The appearance of precipitate even in weakly acidic solutions was a characteristic feature of the majority of the 1:1:1 systems. More useful information was obtained from the metal–Schiff base systems with the lower stoichiometric ratio of metal ions (Figures 1 and 2), whereby the complex species formed were soluble over a larger range of p[H]. The time intervals required between equilibrium p[H] measurements were quite large, especially for the Ni(II)–Schiff base systems, requiring as much as an hour for each equilibrium point in some p[H] ranges. The metal–Schiff base complex equilibria in aqueous solutions are expressed by eq 7–11 for PLP and PG as representative precursors.

$$M^{2+} + PG^{-} + PLP^{3-} \rightleftharpoons MSB^{2-}$$

$$\beta_{MSB} = \frac{[MSB^{2-}]}{[M^{2+}][PG^{-}][PLP^{3-}]} \tag{7}$$

$$MSBH_{n-1}^{n-3} + H^{+} \rightleftharpoons MSBH_n^{n-2}$$

$$K^{H}_{MSBH_n} = \frac{[MSBH_n^{n-2}]}{[MSBH_{n-1}^{n-3}][H^{+}]} \tag{8}$$

$$M^{2+} + 2PG^{-} + 2PLP^{3-} \rightleftharpoons M(SB)_2^{6-}$$

$$\beta_{M(SB)_2} = \frac{[M(SB)_2^{6-}]}{[M^{2+}]^2[PG^{-}]^2[PLP^{3-}]} \tag{9}$$

$$M(SB)_2H_{n-1}^{n-7} + H^{+} \rightleftharpoons M(SB)_2H_n^{n-6}$$

$$K^{H}_{M(SB)_2H_n} = \frac{[M(SB)_2H_n^{n-6}]}{[M(SB)_2H_{n-1}^{n-7}][H^{+}]} \tag{10}$$

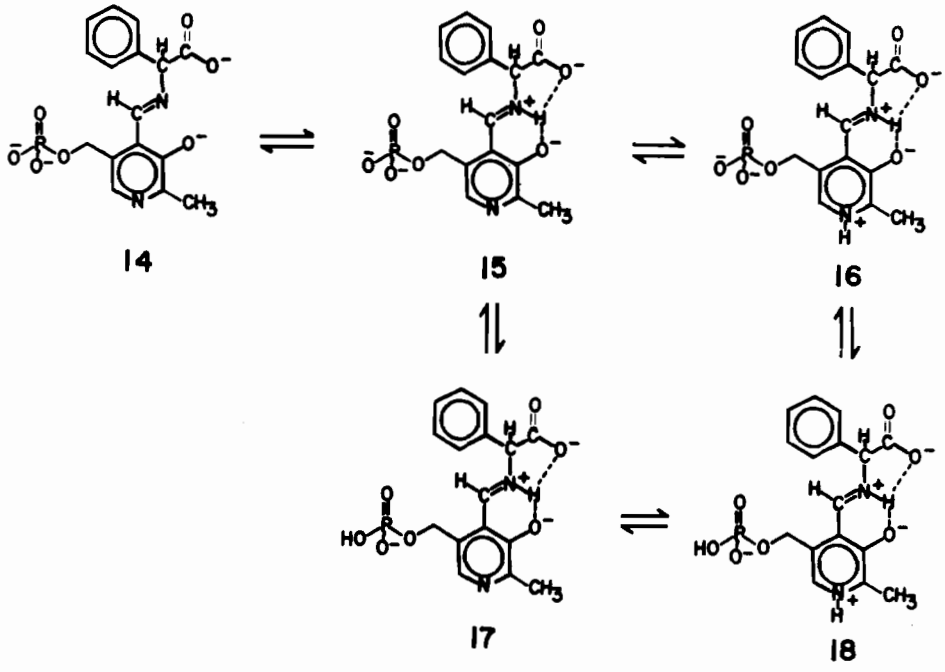
$$MSB(H_2O)^{2-} \rightleftharpoons MSB(OH)^{3-} + H^{+}$$

$$K_{MSB(OH)} = \frac{[MSB(OH)^{3-}][H^{+}]}{[MSB^{2-}]} \tag{11}$$

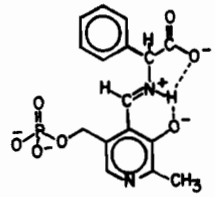
β_{MSB} designates the equilibrium constant for metal–Schiff base chelate formation between metal ion and unprotonated forms of PLP and DPL, and amino acids. The formation of a hydroxo-metal–Schiff base species is viewed as the deprotonation of a coordinated water molecule. Stability constants evaluated for these equilibria are listed in Tables V and VI. It is noted that the log β_{MSB} values for metal chelates vary considerably with the metal ion, however, for a given metal ion; variation of the Schiff base system involving the components employed in this work produces only a small change in the stability of the metal–Schiff base chelate formed. The change in the basicity of the imine nitrogen of the Schiff base by a change in the amino acid precursor is reflected by its affinity for metal ions. In general, Schiff base chelates derived from MPG were the most stable among those studied. The hydrophobic nature of this amino acid, which tends to promote precipitation, together with a strong tendency of Zn(II) to hydrolyze, complicated the study of the Zn–MPG–PLP system; thus, equilibrium constants for this system are not reported.

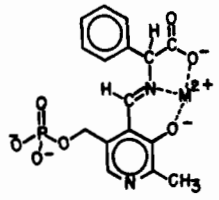
(42) Weng, S. H.; Leussing, D. L. *J. Am. Chem. Soc.* **1983**, *105*, 4082.

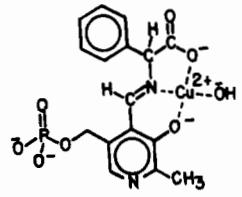
Scheme II



A comparison can be made between log β_{MSB} (Tables V and VI) and log K''_{HSB} [log β_{SB} (eq 5a) + log K^{H}_{HSB} (eq 6); see Table IV] because of the similarity in species **19** and **20** formed by these







equilibria, respectively. Equations 12 and 13, in which AA and VB represent amino acid and vitamin B_6 coenzyme, define K''_{HSB} and β_{MSB}.

$$K''_{HSB} = \frac{[HSB^n]}{[H^+][AA][VB]} = \beta_{SB}K^{H}_{HSB} \qquad (12)$$

$$\beta_{MSB} = \frac{[MSB^{n+1}]}{[M^{2+}][AA][VB]} \qquad (13)$$

Replacement of a proton bound to azomethine nitrogen of a Schiff base by a divalent metal ion, except for Cu(II), does not seem to make a remarkable difference in the stability. Some of the chelates investigated have even lower formation constants than HSB. The formation of Cu(II) chelates of greater stability than those of the other metal ions reported reflects the well-recognized higher affinity of Cu(II) for nitrogen and oxygen donors. A comparison of stability constants for the complexes derived from PLP with those of DPL chelates indicates that, with the exception of the Cu(II)–SPG Schiff base chelate, the presence of 5′-phosphate significantly decreases the stabilities of the metal–Schiff base chelates formed. This is an unexpected result because of the lower basicity of azomethine nitrogens in the PLP Schiff bases and the higher overall negative charge of the PLP Schiff base ligand. The fact that the trend in the metal chelate stabilities run counter to the trend in ligand stabilities must be a steric effect that operates in the coordination sphere of the metal ion, but does not interfere with the protonation reactions. The fact that this effect is only minimal with Cu(II), which involves nearly planar coordination, is in accord with this interpretation. Data in Tables IV–VI also show that the electron-withdrawing effect of the sulfonate of SPG also substantially lowers the stabilities.

Stability constants for metal chelates of DPL Schiff bases and the Mn(II), Co(II), and Ni(II) chelates of PLP Schiff bases have not been reported previously. Stability constants available in the literature[2,3,42] for Cu(II) and Zn(II) chelates of PLP Schiff base agree well with those reported here.

Protonation of a metal–Schiff base complex derived from PLP can occur at pyridine nitrogen and phosphate oxygens under the experimental p[H] conditions employed. The coordination sites of MSB are analogous to those of the monoprotonated Schiff base, HSB; thus, the protonation sequence of MSB may be expected to be the same as that for HSB (see Scheme II). Except for Cu(II), replacement of the proton in HSB by a divalent metal results in an increase in the protonation constant of the pyridine ring. This is again due to the greater coordinate bonding interaction of imine nitrogen with a proton than with a divalent metal ion.[43] In general, the first protonation constant of MSB derived from PLP is higher by a factor of $10^{0.6}$–$10^{0.7}$ than that of the analogous HSB. With few exceptions, the second protonation constant for the DPL metal–Schiff base chelates are lower than those of the corresponding PLP complexes, and this is in accord with the relative stabilities of the metal chelates (i.e. the greater coordinative interaction with the metal ion, the lower the tendency of the ligand to be protonated at a non-metal-coordinated donor site). The magnitude of variation in the second protonation constant of PLP Schiff base metal chelates also suggests that the second protonation must occur at phosphate oxygens.

The potentiometric data for the Schiff base systems containing Cu(II) indicates an insignificant tendency toward the formation of $M(SB)_2$ species while that for Co(II) and Ni(II) systems suggested the formation of considerable amount of these species. The differences in the equilibrium behavior (Figure 3) of Ni–

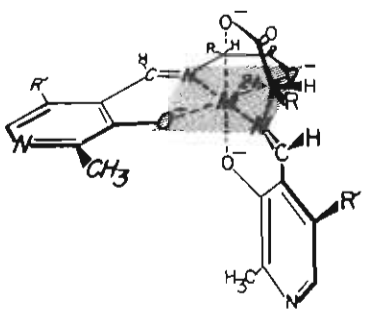
(43) Martell, A. E. In *Proceedings of the Symposium on Chemical and Biological Aspects of Pyridoxal Catalysis (Rome 1962)*; Snell, E. E., Fasella, P. M., Braunstein, A., Rossi Fanelli, A., Eds.; Pergamon Press: Oxford, England, 1963; p 13.

PLP–AA systems from those of Ni–DPL–AA are due mainly to the formation of $Ni(SB)_2$ species only with PLP Schiff bases and Ni(SB)OH species only with DPL Schiff bases. The formation of hydroxo complexes of metal–Schiff base chelate **21** is also a characteristic of Cu(II) systems.

Species Distribution Curves. The stability constants evaluated for all the Schiff base equilibria are utilized to calculate the distribution of Schiff base species over a p[H] range of 2–12. Inspection of these curves for Cu(II) systems (see Figure 4) indicates that the Schiff base complex formation reaches completion below p[H] 4, and a diprotonated species, $CuSBH_2$, predominates in concentration up to a p[H] of 5.5. Significant deprotonation of this species starts at p[H] = 3.5 and the CuSBH species becomes the predominant species above p[H] = 5.5, however, for a narrow p[H] region because the formation of CuSB is more favored above p[H] = 6.5. The CuSB species is formed to an extent of 98% and exists as a principal species over a p[H] range of 6.5–11. In more alkaline solutions, CuSB(OH) predominates. A more complicated distribution of species is observed for the Schiff base systems of other metal ions where the precursors of the Schiff base (amino acid and coenzyme) also compete with Schiff bases to coordinate the metal ion (see Figure 5a).

The species distributions for Ni(II)–Schiff base systems (Figure 5) are quite different for PLP and DPL. The total amount of the chelate formed at p[H] = 5 is less than 10% in PLP systems, while, at the same p[H], as much as 60% total Schiff base chelate species are formed in DPL systems. Also, chelates of Ni–PLP–PG predominate only above p[H] = 8 in contrast with the predominance of Ni–DPL–PG chelates above p[H] = 5. The PLP–PG Schiff base forms a $Ni(SB)_2$ species to the extent of 10%, on the other hand, DPL Schiff bases form NiSB(OH), exclusively.

The inspection of stability constants in Tables V and VI also indicates the coordination geometry of chelates. Metal ions, such as Mn^{2+} and Co^{2+} invariably form 1:2 chelates with considerable stability, whereby two terdentate Schiff bases seem to coordinate in mutually perpendicular planes and provide octahedral geometry around the metal ion as shown in **22**.



22

The coordination sphere for these metal ions in 1:1 chelates seems to contain three water molecules. For the 1:2 Schiff base chelates of Ni(II), one can also expect an octahedral arrangement of donor sites around the metal ion in a manner similar to the structure reported by Capasso and co-workers[41] for Ni[HPL–VAL]$_2$. The preference of Cu(II) for a square-planar geometry because of the Jahn–Teller distortion[44] is reflected by its inability to form 1:2 chelates of the Schiff bases under the conditions employed. The formation of a hydroxo complex seems to further stabilize the square planar geometry. The X-ray crystal structure of Cu[H_2PLP–PHE] (PHE = phenylalanine) has also demonstrated such a geometry around Cu(II).[45]

Concluding Remarks

The oxidative deamination reaction, which is to be investigated with these systems, is considered to proceed at a significant rate only in alkaline solutions.[22,23] In the systems selected for the study of this reaction, the species presumed to be least active catalytically, $M(SB)_2$, are formed in the neutral and weakly basic region, but generally to a small extent. The basic solutions of most of the Schiff base chelate systems studied have either MSB or

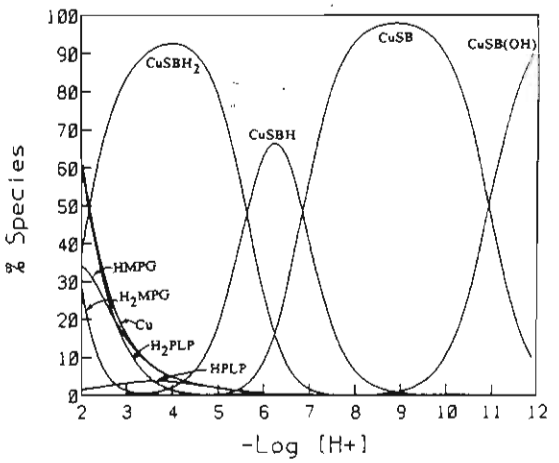


Figure 4. Species distribution curves for the Cu–MPG–PLP system. Molar ratio of the reactants Cu(II), MPG, and PLP is 1:1:1 at an initial concentration of 2.00×10^{-3} M. CuSB, CuSBH, $CuSBH_2$ and CuSBOH represent unprotonated, monoprotonated, diprotonated, and monohydroxo forms of the SB chelate, respectively. HPG and H_2PG are protonated forms of PG; HPLP and H_2PLP are protonated forms of PLP. Cu represents the uncomplexed Cu(II).

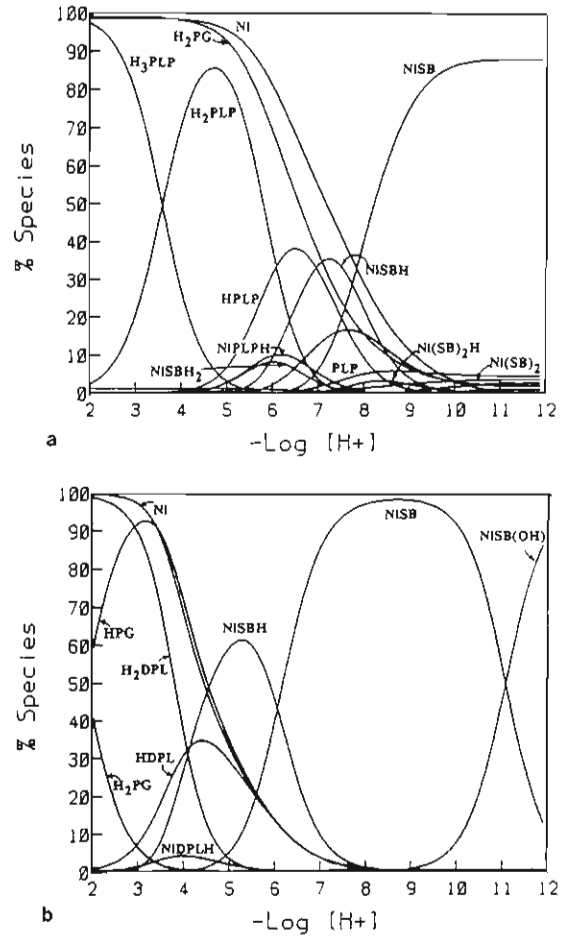


Figure 5. Species distribution curves for Ni–PG–PLP (a) and Ni–PG–DPL (b) systems. Molar ratio of Ni, PG, and PLP or DPL is 1:1:1 at an initial concentration of each component = 2.00×10^{-3} M. SB stands for Schiff base; NiSB, NiSBH, $NiSHB_2$, and NiSBOH represent unprotonated, monoprotonated, diprotonated and monohydroxo forms of the Schiff base chelate, respectively. $Ni(SB)_2$ and $Ni(SB)_2H$ are 2:1 Schiff base–Ni(II) chelates. HPG and H_2PG are protohated forms of PG, HPLP, H_2PLP, and H_3PLP are protonated forms of PLP, and HDPL and H_2DPL are protonated forms of DPL. Ni stands for uncomplexed Ni^{2+}; NiPLP, NiPLPH, and NiDPL are the respective complexes.

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MSB(OH) as the predominant species, and the metal chelates present under these conditions are considered good candidates as active intermediates in oxidative deamination and are thus easy to identify. The α-deprotonated intermediate of the metal–Schiff base chelates formed from phenylglycine and its derivatives are expected to be stabilized by conjugation of the imine group with the phenyl ring on the α-carbon, and this property is one of the reason for selecting phenylglycine Schiff bases for oxidative deamination studies.

Acknowledgment. This research was supported by Grant AM-11694 from the National Institute of Arthritis, Diabetes, Digestive, and Kidney Diseases, U.S. Public Health Service, and by Grant A-259 from the Robert A. Welch Foundation. V.M.S. thanks Dr. R. J. Motekaitis for computational assistance.

Registry No. **3**, 69-91-0; **4**, 124755-49-3; **5**, 2540-53-6; PLP, 54-47-7; DPL, 1849-49-6; PLP-PG, 124755-50-6; PLP-MPG, 124755-51-7; PLP-SPG, 124755-52-8; DPL-PG, 124755-53-9; DPL-MPG, 124755-54-0; DPL-SPG, 124755-55-1.

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Electrochemical and Spectroelectrochemical Characterization of Intermolecular Nitrosyl Transfer between Iron and Cobalt Porphyrins

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Received March 10, 1989

The transfer of a nitrosyl ligand between neutral and oxidized iron and cobalt metalloporphyrins in dichloromethane solutions was investigated by electrochemistry and FTIR or ESR spectroelectrochemistry. The transfer of NO from (P)Co(NO) to (P)Fe, from $[(P)Co(NO)]^+$ to $(P)FeClO_4$, and from $[(P)Fe(NO)]^+$ to (P)Co was demonstrated for complexes where P = the dianion of tetraphenylporphyrin (TPP), *meso*-tetrakis(2,4,6-trimethylphenyl)porphyrin (TMP), or octaethylporphyrin (OEP). The driving force in these reactions is related to both the nature and oxidation state of the central metal in (P)M(NO) or $[(P)M(NO)]^+$, where M = Fe or Co, and follows the order $(P)Fe(NO) > (P)Co(NO) > [(P)Fe(NO)]^+ > [(P)Co(NO)]^+$.

Introduction

Numerous spectroscopic,[1-11] structural,[12-18] and electrochemical[19-32] studies of neutral and oxidized iron and cobalt nitrosyl porphyrins have been reported in the literature. These compounds are represented by (P)M(NO) and $[(P)M(NO)]^+$, where M = Co(II), Fe(II), or Fe(III) and P is the dianion of a given porphyrin ring.

Intermolecular NO transfer between two coordinatively unsaturated transition-metal complexes is of interest in both inorganic and bioinorganic chemistry[33-35] and has been shown to occur through formation of a transient μ-bridged nitrosyl complex. The transfer of NO from a non-porphyrin complex to hemoglobin was reported by Doyle,[35] but an NO transfer between two metalloporphyrins has never been demonstrated.

In this paper, we present the first examples for NO-transfer reactions between Fe and Co metalloporphyrins. The investigated reactions are given by eqs 1–3, where P is the dianion of tetraphenylporphyrin (TPP), *meso*-tetrakis(2,4,6-trimethylphenyl)-porphyrin (TMP), or octaethylporphyrin (OEP). Each of the above reactions was monitored in CH_2Cl_2 by thin-layer and conventional electrochemistry as well as by FTIR or ESR spectroelectrochemistry.

$$(P)Fe + (P)Co(NO) \rightarrow (P)Fe(NO) + (P)Co \quad (1)$$

$$(P)Co + [(P)Fe(NO)]^+ \rightarrow (P)Co(NO) + (P)FeClO_4 \quad (2)$$

$$(P)FeClO_4 + [(P)Co(NO)]^+ \rightarrow [(P)Fe(NO)]^+ + (P)CoClO_4 \quad (3)$$

To date, there are only two reports in the literature involving the transfer of a nitrogen atom between two metalloporphyrins.

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