

New Multidentate Ligands. II. Amino Acids Containing α -Pyridyl Groups^{1,2}

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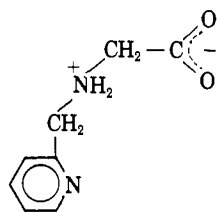
The synthesis of 2-(aminomethyl)pyridine-*N*-acetic acid (AMPA) and of ethylenebis-*N,N'*-(2-aminomethyl)pyridine-*N,N'*-diacetic acid (EDAMPDA) is described. The chelating tendencies of these new ligands for divalent metal ions, and for Fe^{3+} , are determined and are compared with those of the analogous aliphatic polyamines and with ethylenediaminetetraacetic acid. Although the replacement of acetic acid groups by α -pyridyl groups lowers the metal ion affinities slightly, the lowered hydrogen ion affinities of the α -pyridyl groups renders the new ligand EDAMPDA more effective than EDTA in the binding of transition metal ions.

In a recent publication⁴ it was reported that the substitution of an α -pyridyl group for an alkylamino group of an aliphatic polyamine results in the lowering of the metal ion binding tendencies of the polyamine because the aromatic nitrogen atoms are less basic than aliphatic nitrogen atoms when they are similarly positioned in polydentate ligands. However, the lowered basicity of the aromatic nitrogens also involved lowered competition with hydrogen ions in solutions having pH values below 9.5–10, making the aromatic amines more effective in the binding of metal ions.

The purpose of the present investigation was to synthesize and study metal binding by aminopolycarboxylic acid analogs in which one or more α -acetic acid groups are replaced by α -pyridyl donor groups. Such studies would be useful for the design of new and possibly more effective ligands for the complexing of metal ions in aqueous solution.

Experimental

Synthesis of Ligands. 2-(Aminomethyl)pyridine-*N*-monoacetic Acid (AMPA). Nine grams (0.087 mole) of glycine ethyl ester, dissolved in 100 ml. of absolute alcohol, was treated with 8.9 g. (0.087 mole) of 2-pyridinecarboxaldehyde and heated on a steam bath



I
2-(aminomethyl)pyridine-*N*-acetic acid, HL

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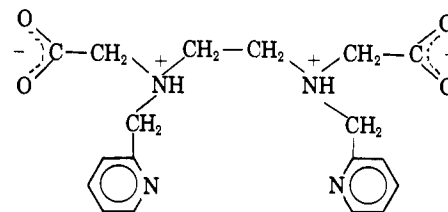
(4) R. G. Lacoste and A. E. Martell, *Inorg. Chem.*, **3**, 881 (1964).

for 1 hr. The reaction mixture was reduced in a low pressure Parr hydrogenation apparatus in the presence of 1.0 g. of 10% Pd-C catalyst. Distillation of the reaction mixture resulted in the isolation of the product as a pale yellow oil boiling in the range 165–167° (19 mm.). The yield of ethyl 2-(aminomethyl)pyridine-*N*-monoacetate was 5.4 g. or 32% of the theoretical amount.

The ester was saponified by boiling for 2 hr. in a 25% solution of sodium hydroxide. The solvent was removed, the acidified solution was evaporated to dryness, and organic materials were taken up in hot ethanol. Treatment of the solution with HCl gas resulted in the formation of a crystalline hydrochloride, which was recrystallized from aqueous alcohol. The yield of purified 2-(aminomethyl)pyridine-*N*-monoacetic acid dihydrochloride from the ester was 25% of the theoretical amount. The pure product was obtained as colorless crystals, m.p. 168–169° dec.

Anal. Calcd. for $C_8H_{10}N_2O_2$: C, 40.34; H, 5.04; N, 11.75; neut. equiv., 238. Found: C, 40.40; H, 4.94; N, 11.6; neut. equiv., 253.

Ethylenebis-*N,N'*-(2-aminomethyl)pyridine-*N,N'*-diacetic Acid (EDAMPDA). To 28.5 g. (0.074 mole) of ethylenebis-*N,N'*-(2-aminomethyl)pyridine tetrahydrochloride,⁴ was added 7.61 g. (0.074 mole) of 50% sulfuric acid, with cooling to 10°. This temperature was maintained during the dropwise addition of 13 ml. (0.184 mole) of a 36% solution of formaldehyde.



II

ethylenebis-*N,N'*-(2-aminomethyl)pyridine-*N,N'*-diacetic acid

Finally, a 30% solution containing 7.61 g. (0.148 mole) of sodium cyanide was added over a period of 2 hr., while the temperature was maintained between 5 and 10°. After all the reactants were added, the reaction solution was brought to room temperature. After 24 hr. the reaction mixture was evaporated to dryness and the organic material was taken up in hot absolute ethanol. The crude nitrile was obtained as a viscous pale yellow oil.

The nitrile was hydrolyzed by refluxing with concentrated hydrochloric acid for 10 hr. Ammonia was removed by adding excess NaOH and passing air through the reaction mixture. The solution was then acidified with HCl. The sodium chloride formed was removed by repeated evaporation of the solution and extraction with hot absolute ethanol. The alcoholic

extracts were then evaporated to dryness, and the colorless monosodium salt of the amino acid was obtained by recrystallization from aqueous ethanol; yield, 6.0 g., representing 22% of the over-all theoretical yield. The product melted at 210–215° with decomposition.

Anal. Calcd. for $C_{18}H_{21}N_4O_4Na \cdot H_2O$: C, 54.3; H, 6.0; N, 14.7; Na, 6.0; neut. equiv., 380. Found: C, 54.5; H, 5.5; N, 14.7; Na, 5.6; neut. equiv., 394.

In addition to the elemental analyses, the structures of the ligands were confirmed by careful infrared spectral determinations.

Procedure. Potentiometric equilibrium measurements were made with a glass-calomel electrode system as described previously.⁴ The ionic strength of all solutions was maintained at 0.10 by the addition of KNO_3 , and the temperature was maintained at $25 \pm 0.02^\circ$.

The potentiometric measurements of solutions involving the direct combination of the metal ion with acid forms of the ligand were satisfactory for the determination of the stability constants of the alkaline earth chelates; however, the high stabilities of transition metal chelates of EDAMPDA and AMPA necessitated the use of a competing ligand. For this purpose the polyamine triaminotriethylamine ("tren") was employed in the presence of excess Ca(II) ion, as first suggested by Schwarzenbach.⁵ The use of Ca(II) as a secondary metal ion was feasible for EDAMPDA because of the relatively high stability of the Ca(II)–EDAMPDA chelate. Direct titration of the system containing the two ligands and the two metal ions gave a buffer region in the pH range 6–9 corresponding to the conversion of the metal–EDAMPDA chelate to the metal–tren chelate and the Ca(II) chelate of EDAMPDA. The titration curves gave only approximate results since equilibrium was attained slowly. Therefore a number of solutions were made up having neutralization values corresponding to different points in the buffer region of interest. These solutions were kept under nitrogen in closed containers. Measurements of hydrogen ion concentrations over a period of days gave the required equilibrium values.

The same procedure was used for the determination of the stabilities of the transition metal chelates of AMPA, except that the competing metal ion, Ca^{2+} , was not employed because of the low affinity of this ligand for the alkaline earth ions.

Calculations. The equilibrium constants for acid dissociation and metal complex formation were calculated after establishment of the stoichiometry of the reactions on the basis of the titration curves (see Results) and of related experimental observations.

The third acid dissociation constant of AMPA and the third and fourth dissociation constants of EDAMPDA were calculated in the usual manner as single dissociation steps. The first two dissociation reactions of both ligands were not separated in the titration curves and were calculated by the graphical method described by Schwarzenbach and Ackermann.⁶

Except for the cases noted below, the formation constants of the 1:1 metal complexes were calculated

algebraically in the usual manner.⁷ Details of the calculations have been described by Lacoste.⁸

Use of a Competing Ligand. The equilibrium constant of the reaction occurring in the presence of a competing ligand and a competing metal ion described above may be expressed by the relationship

$$K' = \frac{[CaL][MB^{2+}][H^+]^3}{[ML][H_3B^{3+}][Ca^{2+}]} \quad (1)$$

where H_2L represents EDAMPDA and H_3B^{3+} represents the trihydrochloride (or triprotonated) form of β, β', β'' -triaminotriethylamine. The constant K' may be calculated algebraically from the data obtained in the buffer region of the reaction, the stoichiometric relations which apply to the reaction system being the same as described by Schwarzenbach and Freitag.⁵ The formation constant of ML may be calculated from K' and other known equilibrium constants from

$$K_{ML} = \frac{K_{CaL}K_{MB}K_1K_2K_3}{K'} \quad (2)$$

where K_{CaL} and K_{MB} are the formation constants of CaL and MB, and K_1 , K_2 , and K_3 are first, second, and third acid dissociation constants of H_3B .

The calculation of the stability constants of the transition metal–AMPA chelates with a competing ligand, but without a competing metal ion, was carried out as described previously⁴ for the Ni(II) and Cu(II) chelates of ethylenebis-N,N'-(2-aminomethyl)pyridine.

Results

AMPA. The potentiometric equilibrium curves of the dihydrochloride of AMPA in Figure 1 indicate two strongly acidic groups, taken as the carboxyl and pyridinium groups, and one very weakly acidic group, the secondary aliphatic α -amino group. For Zn(II), Cd(II), and all of the transition metals shown, complex formation occurs with (or after) the displacement of all three protons from the ligand, and the relative formation constants are indicated to have the order $Cu(II) > Ni(II) > Co(II) > Zn(II) > Cd(II) > Mn(II)$.

The additional neutralization step at high pH for Cu(II) indicates the formation of a hydroxo metal chelate in which a hydroxide ion is coordinated to the metal ion. The pK of this reaction is 9.2. Similar secondary coordination steps cannot be suggested for Co(II), Zn(II), and Cd(II), even though similar high pH neutralization steps are apparent in the potentiometric curves because of the appearance of a solid phase at the beginning of the buffer region in which this reaction would be presumed to take place.

The equilibrium constants and metal complex formation constants for AMPA, calculated as described above, are given in Table I.

EDAMPDA. The potentiometric equilibrium curves of EDAMPDA, shown in Figure 2, indicate two strongly acidic groups and two weakly acidic groups, one of which is much weaker than the other. The very acidic groups in the tetra- and triprotonated forms are the carboxyl and pyridinium groups. The available protons are probably distributed between these groups because their acidities are of the same order of magnitude.

(5) G. Schwarzenbach and E. Freitag, *Helv. Chim. Acta*, **34**, 1503 (1951).

(6) G. Schwarzenbach and H. Ackermann, *ibid.*, **31**, 1029 (1948).

(7) A. E. Martell and M. Calvin, "Chemistry of the Metal Chelate Compounds," Prentice-Hall, Inc., Englewood Cliffs, N. J., 1952, Chapter 3.

(8) R. G. Lacoste, Dissertation, Clark University, 1957.

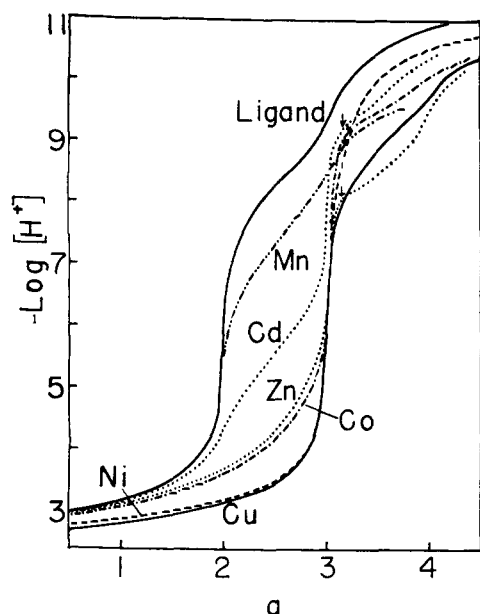


Figure 1. Potentiometric equilibrium curves for the reaction of the dihydrochloride of 0.0010 *M* 2-(aminomethyl)pyridine-*N*-acetic acid (AMPA) as the free ligand, and in the presence of equimolar concentrations of Cu(II), Ni(II), Co(II), Zn(II), Cd(II), and Mn(II) ions; 25°, $\mu = 0.10$ *M* (KNO₃).

The third and fourth neutralization steps involve the aliphatic nitrogens of the substituted ethylenediamine group. Their relative basicities are quite analogous to

Table I. Log Formation Constants (Log K_{MA})^a

Metal ion	AMP ^b	AMPA ^c	Log K_{AMPA} - log K_{AMP}
H ⁺ pK ₁	2.14	2.19	
pK ₂	8.57	2.46	
pK ₃	...	8.18	
Cu ²⁺	9.5	11.8	2.3
Ni ²⁺	7.1	10.2	3.1
Co ²⁺	5.3	8.1	2.8
Zn ²⁺	5.2	7.6	2.4
Cd ²⁺	4.5	6.1	1.6
Mn ²⁺	...	4.2	...

^a $\mu = 0.10$ (KNO₃), 25°. ^b AMP = 2-aminomethylpyridine. ^c AMPA = 2-(aminomethyl)pyridine-*N*-monoacetic acid.

those of the well-known ligand EDTA, and to the parent polyamine EDAMP, described in a previous paper.⁴

The potentiometric curves indicate similar stoichiometry and high stability for the reaction of EDAMPDA with all the divalent ions shown. In all cases coordination occurs directly with the displacement of all the protons attached to the basic functional groups. The positions of the potentiometric curves indicate the following relative stabilities of the divalent metal complexes: Cu(II), Ni(II), Co(II), Zn(II), Cd(II) > Mn(II), Fe(II) > Ca(II) > Mg(II).

In the case of Fe(III), the reaction occurs directly with the neutralization of 5 equiv. of base, showing that a very stable hydroxo Fe(III) chelate is formed. The very high stability of this complex is evidenced by the lack of precipitation of ferric hydroxide at the highest pH (10–11) employed in the determination. The analogous hydroxo-Fe(III)-EDTA chelate decomposes at a pH of ~8.

The acid dissociation constants and metal chelate formation constants of EDAMPDA, determined as described above, are listed in Table II.

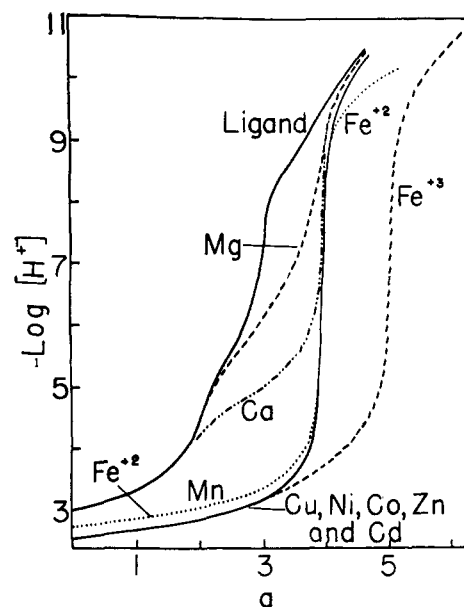


Figure 2. Potentiometric equilibrium curves for the reaction of the 0.0010 *M* dihydrochloride of ethylenebis-*N,N'*-(2-aminomethyl)pyridine-*N,N'*-diacetic acid (EDAMPDA) as the free ligand and in the presence of equimolar concentrations of Cu(II), Ni(II), Co(II), Zn(II), Cd(II), Mn(II), Fe(II), Fe(III), Ca(II), and Mg(II) ions; 25°, $\mu = 0.10$ *M* (KNO₃).

Table II. Log Formation Constants (Log K_{ML})^a

Metal ion	EDTA ^b	EDAMP ^c	EDAMPDA ^d	Log K_{EDTA} - log $K_{EDAMPDA}$
H ⁺ pK ₁	2.0	1.62	2.34	
pK ₂	2.67	1.81	3.02	
pK ₃	6.13	5.45	5.63	
pK ₄	10.26	8.23	8.84	
Cu ²⁺	18.3	16.3	20.4	-2.1
Ni ²⁺	18.4	12.7	16.6	1.8
Co ²⁺	16.1	12.8	14.0	2.1
Zn ²⁺	16.1	11.5	15.2	0.9
Cd ²⁺	16.4	9.9	14.6	1.8
Mn ²⁺	13.4	5.9	12.7	0.7
Fe ²⁺	14.5	...	12.4	2.1
Ca ²⁺	10.6	...	7.9	2.7
Mg ²⁺	8.7	...	5.5	3.2

^a $\mu = 0.10$ (KNO₃), 25°. ^b EDTA = ethylenediaminetetraacetic acid. ^c EDAMP = ethylenebis-*N,N'*-(2-aminomethyl)pyridine. ^d EDAMPDA = ethylenebis-*N,N'*-(2-aminomethyl)pyridine-*N,N'*-diacetic acid.

Discussion

The stability constants of the 1:1 chelates of AMPA are compared in Table I with those of the parent amine, AMP. It is noted that the additional acetate group increases the stabilities of the 1:1 transition metal chelates by about 2–3 orders of magnitude, this in spite of the fact that the completely dissociated form of AMPA is less basic than AMP. With respect to the equilibria taking place in solution, this difference in basicity renders the AMPA chelates a factor of 3 higher in stability than that reflected by the formation constants, for all pH values ranging from about 2.5 to 8.

The lowered basicity of the *N*-acetate derivative relative to the parent diamine is similar to what takes place in the substitution of other polyamines with acetate groups (*i.e.*, *N,N'*-ethylenediaminediacetic acid vs. ethylenediamine, etc.). No explanation has ever been offered for this effect, but it may be due to electro-

Table III. Comparison of Metal-Binding Tendencies of Protonated Ligands

Metal ion	Log equilibrium constant ^a					
	K'_{EDTA}^b	K'_{EDAMPDA}^b	$K'_{\text{EDTA-EDAMPDA}}^b$	K''_{EDTA}^c	K''_{EDAMPDA}^c	$K''_{\text{EDTA-EDAMPDA}}^c$
Cu ²⁺	1.9	5.9	-4.0	8.0	11.6	-3.6
Ni ²⁺	2.0	2.1	-0.1	8.1	7.8	+0.3
Co ²⁺	-0.3	-0.5	+0.2	5.8	5.2	+0.6
Zn ²⁺	-0.3	0.7	-1.0	5.8	6.4	-0.6
Cd ²⁺	0.0	0.1	-0.1	6.1	5.8	+0.3
Mn ²⁺	-3.0	-1.8	-1.2	3.1	3.9	-0.8
Fe ²⁺	-1.9	-2.1	+0.2	4.2	3.6	+0.6

^a K' is the equilibrium constant for combination of the metal ion with diprotonated ligands, H_2L^n , where $n = 2-$ for EDTA and 0 for EDAMPDA ($K' = [ML^n][H^+]^2/[M^{2+}][H_2L^n]$); K'' is the equilibrium constant for combination of the metal ion with the monoprotinated ligand, HL^m , where $m = 3-$ for EDTA and 1- for EDAMPDA ($K'' = [ML^{m+1}][H^+]/[M^{2+}][HL^m]$). ^b K' may be applied unambiguously to the pH range 3.0-5.0. ^c K'' may be similarly applied to the pH range 6.5-8.0, for both ligands.

static interaction of the negative carboxylate oxygens with the proton attached to the nitrogen (through a hydrogen-bonded solvent molecule), or in part to the weak inductive effect of the carbonyl carbon atom of the carboxylate group, or to a combination of both effects. It should be noted, however, that EDAMPDA is an exception to this general behavior in that it is more basic than the parent ligand, EDAMP.

A comparison of the formation constants for EDAMPDA with those of the parent polyamine shows a quite variable increment of stability resulting from the addition of two acetate donor groups to the ligand. With the exception of Mn(II), the effect is about double, or nearly double, the effect of adding one acetate group to AMP, as discussed above.

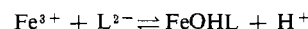
The formation constants of EDAMPDA are also compared with those of EDTA since the latter is so widely known and commonly used as a sequestering agent and metal buffering reagent. It is noted that the EDTA chelates of the transition metal ions are only slightly more stable (1-2 log K units) than those of EDAMPDA. Further, the much lower basicity (toward protons) of the anions of EDAMPDA would render it a more effective metal-binding reagent than EDTA. This fact is illustrated in Table III for two pH ranges in which the two ligands have the same degree of protona-

tion. It is seen that in the pH range where both ligands exist primarily in the monoprotinated form, the metal-binding tendencies of the two ligands are about the same (*i.e.*, the scatter of the differences in log K is nearly symmetrical about zero) except for copper(II).

For the pH range 3.0-5.0, where both ligands exist primarily in the diprotonated form, EDAMPDA definitely has the edge, although the scatter is such that two complexes of EDTA (those of Co²⁺ and Fe²⁺) are listed as slightly more stable.

Of the metal ions listed in Tables II and III, the stability of the Cu(II) complex of the ligand containing two α -pyridyl groups is quite exceptional. This difference is probably related to the high affinity of the Cu(II) ion for basic nitrogen atoms, and the limitation of its strong coordination tendencies in aqueous solution to four donor groups arranged in a planar fashion about the metal ion. It would, therefore, be reasonable to expect stronger bonding for IV, in which four nitrogen atoms are coordinated to the Cu(II) ion.

Although it was not possible to obtain an accurate value of the formation constant for the Fe(III) from potentiometric data, some qualitative comparisons can be made. From the experimental results it is apparent that the over-all reaction of the Fe(III) ion with EDAMPDA is



This reaction may take place in one or two steps; the curvature of the titration curve (Figure 2) between $a = 4$ and $a = 5$ indicates that some of the unhydrolyzed metal chelate may be present in the equilibrium mixture. The analogous complexes of EDTA (H_4L) have been reported for similar experimental conditions by Schwarzenbach and Heller.⁹ They are

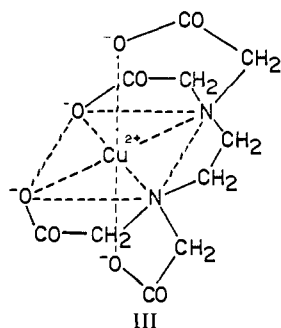
$$\frac{[FeL^-]}{[Fe^{3+}][L^{4-}]} = 10^{25.1}$$

$$\frac{[FeOHL^{2-}][H^+]}{[FeL]} = 10^{-7.5}$$

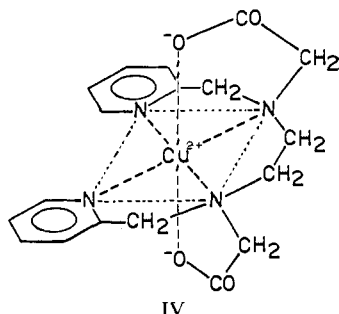
$$\frac{[FeOHL^{2-}][H^+]}{[Fe^{3+}][L^{4-}]} = 10^{17.6}$$

The stability of the hydroxo iron(III) chelate of EDAMPDA is apparently considerably higher than that of EDTA. From the Fe(III) titration curve in Figure 2 the pK of the normal (unhydrolyzed) iron chelate is no more than ~ 4 . Therefore it seems that the formation constant of the 1:1 Fe(III)-EDAMPDA chelate must be considerably in excess of $10^{21.6}$.

(9) G. Schwarzenbach and J. Heller, *Helv. Chim. Acta*, **34**, 576 (1951).



III
Cu(II) chelate of EDTA, CuL^{2-}



IV
Cu(II) chelate of EDAMPDA, CuL