155. Metal-Ion-Coordinating Properties of a Viral Inhibitor, a Pyrophosphate Analogue, and a Herbicide Metabolite, a Glycinate Analogue: The Solution Properties of the Potentially Five-Membered Chelates Derived from Phosphonoformic Acid and (Aminomethyl)phosphonic Acid

by **Bin Song**[1])[a]), **Dong Chen**[1])[a]), **Matthias Bastian**[a]), **R. Bruce Martin**[2])[b])*, and **Helmut Sigel**[a])*

[a]) Institute of Inorganic Chemistry, University of Basel, Spitalstrasse 51, CH-4056 Basel
[b]) Chemistry Department, University of Virginia, Charlottesville, VA 22903, USA

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The pyrophosphoric-acid-analogue phosphonoformic acid (pfa) and the amino-acid-analogue (aminomethyl)phosphonic acid (ampa) both form, in the deprotonated state, *i.e.*, as $^{-}OOC{-}PO_3^{2-}$ and $H_2N{-}CH_2{-}PO_3^{2-}$, respectively, five-membered chelate rings with metal ions. pfa inhibits both phosphate transport and virus replication, while ampa is a metabolic product of the common herbicide glyphosate (= *N*-(phosphonomethyl)glycine). The acidity constants of H_2pfa^- and $H_2ampa^{\pm}$ as well as the stability constants of the [M(Hpfa)], $[M(pfa)]^-$, $[M(Hampa)]^+$, and [M(ampa)] complexes, where $M^{2+} = Mg^{2+}$, Ca^{2+}, Sr^{2+}, Ba^{2+}, Mn^{2+}, Co^{2+}, Ni^{2+}, Cu^{2+}, Cu(2,2′-bipyridyl)$^{2+}$, Cu(1,10-phenanthroline)$^{2+}$, Zn^{2+}, or Cd^{2+}, have been determined by potentiometric pH titrations in aqueous solution at 25° and $I = 0.1$ M ($NaNO_3$). The structures of isomeric complexes and the connected intramolecular equilibria are deduced and evaluated based on the equilibrium constants measured and those calculated *via* the pK_a values of the above mentioned ligands and previously established log *K vs.* pK_a straight-line plots (*H. Sigel et al., Helv. Chim. Acta* **1992**, *75*, 2634) for a simple phosphonate-M^{2+} coordination. pfa forms stronger complexes than ampa with all the above mentioned metal ions, with the single exception of [Cu(ampa)] which is slightly more stable than $[Cu(pfa)]^-$. In neutral solutions, more precisely at pH of *ca.* 6, pfa complexes of alkaline-earth-metal ions retain one phosphonate-bound proton, [M(Hpfa)], while those of the transition-metal ions chelate with the trianionic ligand, pfa^{3-}. In accord with increasing ligand-basicity, the stability-constant order for all metal-ion complexes is oxalate < pfa < pyrophosphate but, owing to proton competition in pyrophosphate, in neutral solutions metal-ion complexation of pfa^{3-} competes with $P_2O_7^{4-}$. With ampa alkaline-earth-metal ions interact only with the phosphonate group of even the dianionic ligand (though Mg^{2+} appears to form a low fraction of a [Mg(ampa)] chelate) while transition-metal ions form chelates which are comparable in stability to those of glycinate.

Introduction. – Contraction and relaxation of skeletal muscle depend on the release and reaccumulation of Ca^{2+} by the sarcoplasmic reticulum (SR). The amount of Ca^{2+} taken up by isolated SR vesicles is relatively low in the absence of anions such as inorganic phosphate, oxalate, or pyrophosphate, but increases several-fold in the presence of these ions [1]. Indeed, the sarcoplasmic reticulum of skeletal muscle contains a phosphate transporter which may be inhibited by phosphonoformic acid (pfa)[3]), which itself is transported into the SR increasing the level of Ca^{2+} accumulation [2].

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[1]) Work performed at the University of Basel during study leaves from the Zhongshan (Sun Yatsen) University in Guangzhou (*B. S.*) and the University of Nanjing (*D. C.*), People's Republic of China.

[2]) Performed in Basel[a]) during the summer of 1993.

[3]) Abbreviations used: ampa, (aminomethyl)phosphonic acid (protonation degree undefined), $ampa^{2-}$, (aminomethyl)phosphonate; Arm, heteroaromatic nitrogen base; bpy, 2,2′-bipyridyl; L, general ligand; M^{2+}, divalent metal ion; pfa, phosphonoformic acid (protonation degree undefined); pfa^{3-}, phosphonoformate; phen, 1,10-phenanthroline; RPO_3, any phosphonate or phosphate-monoester ligand (mostly with a non-interacting organic residue).

Phosphonoformate (pfa^{3-})[3]), $^{-}OOC-PO_3^{2-}$, often considered as a pyrophosphate or diphosphate analogue, even though it forms five-membered chelates, whereas diphosphate gives rise to six-membered rings, is quite generally a rather bioactive ligand. For example, it both inhibits [3] and stimulates [4] the Na^+-inorganic phosphate co-transport in mammalian brush-border membranes. It also inhibits prolidase, a Mn^{2+}-dependent hydrolase, that cleaves dipeptides involving the N-atom of proline, presumably by complex formation at the active site [5]. This enzyme is present in microorganisms and many mammalian tissues as well; in humans, a deficiency in prolidase results in a complex clinical syndrome involving mental retardation [6]. Furthermore, pfa inhibits replication of viruses including the African swine fever virus [7], the herpes simplex virus (HSV) [8], where pfa directly inhibits HSV DNA polymerase [9], and the human immunodeficiency virus (HIV), where it selectively inhibits the DNA polymerase activity, but not the RNase H activity of HIV reverse transcriptase [10]. All DNA polymerases require a divalent cation for activity, Mg^{2+} being considered as the physiological activator [11] [12]; many of them also contain an intrinsic Zn^{2+} [12–14].

The other ligand in this study, the anion of the amino-acid analogue (aminomethyl)phosphonic acid (ampa)[3]), $^{+}H_3N-CH_2-PO_3H^{-}$, which corresponds in its structure to glycine ($^{+}H_3N-CH_2-COO^{-}$), is the main metabolic product of the common highly effective broad-spectrum herbicide glyphosate, *N*-(phosphonomethyl)glycine [15]. It is thus not surprising that many metabolic studies of glyphosate and ampa have been conducted [16], and that especially great efforts are being made to detect and to quantify ampa in plant, food, water, and soil samples [17]. Furthermore, the enzyme-inhibiting properties of aminophosphonates have been tested [18]; *e.g.*, ampa is a weak inhibitor of leucine aminopeptidase [19], a Zn^{2+}-containing metalloenzyme [19].

From the examples given above, it is evident that pfa and ampa are biologically active in systems which depend on the presence of metal ions; hence, the metal-ion binding-properties of these two phosphonate derivatives are of interest. However, in this respect, to the best of our knowledge, so far only little information exists; *i.e.*, some stability constant data are available for the Mg^{2+}, Ca^{2+} [20] [21], Mn^{2+} [22], and Zn^{2+} [21] complexes of pfa, as well as for the Mg^{2+}, Ca^{2+}, Co^{2+}, Ni^{2+}, Cu^{2+}, and Zn^{2+} complexes of ampa [23]. Now, we describe the complexes formed by four alkaline earth and six dipositive transition-metal ions (M^{2+}) with both potentially five-membered chelate ligands, pfa^{3-} and $ampa^{2-}$; the corresponding monoprotonated complexes are also considered. The experiments in this study were conducted under conditions of excess metal ion so that only 1:1 complexes need to be considered. In addition, the stability is quantified of some mixed ligand complexes formed by Cu^{2+}, a heteroaromatic amine (Arm), *i.e.*, 2,2′-bipyridyl (bpy) or 1,10-phenanthroline (phen), and either pfa or ampa.

Experimental. – *Materials.* The trisodium salt of phosphonoformic acid hexahydrate (purity 98%) and (aminomethyl)phosphonic acid (purity 99%) were obtained from *Aldrich-Chemie*, Buchs, Switzerland. The aq. stock solns. of these phosphonates (which were used as purchased) were freshly prepared daily, and, each time, the exact concentration was newly established from the potentiometric pH titrations and the curve-fit procedure for the determination of the pK_a values (see below).

The heteroaromatic amines (= Arm), *i.e.*, 2,2′-bipyridyl or 1,10-phenanthroline monohydrate (both *pro analysi*), the disodium salt of 1,2-diaminoethane-*N,N,N′,N′*-tetraacetic acid (Na_2H_2edta), potassium hydrogen phthalate, HNO_3, NaOH (*Titrisol*), and the nitrate salts of Na^+, Mg^{2+}, Ca^{2+}, Sr^{2+}, Ba^{2+}, Mn^{2+}, Co^{2+}, Ni^{2+}, Cu^{2+}, Zn^{2+}, and Cd^{2+} (all *pro analysi*) were obtained from *Merck AG*, Darmstadt, FRG. All solns. were prepared using distilled CO_2-free water.

The titer of the NaOH used for the titrations was determined with potassium hydrogen phthalate. The exact concentrations of the ligand stock solns. used in the experiments with metal ions (titrated in the presence of an excess of HNO_3; see below) were measured with NaOH, and the concentrations of the stock solns. of the divalent metal ions were determined *via* their edta complexes.

Potentiometric pH Titrations. The pH titrations were carried out with a *Metrohm E536* potentiograph equipped with a *E655* dosimat and *6.0202.100* (*NB*) combined macro glass electrodes. The buffer solns. (pH 4.64, 7.00, 9.00) used for calibration were also from *Metrohm*. The direct pH-meter readings were used in the calculations of the acidity constants; *i.e.*, these constants are so-called practical, mixed or *Brønsted* constants [24]. The negative logarithms of these practical acidity constants given for aqueous solns. at $I = 0.1$M ($NaNO_3$) and 25° may be converted into the corresponding concentration constants by subtracting 0.02 pK units [24].

It should be emphasized that the ionic product of water (K_w) and the mentioned conversion term do *not* enter into our calculation procedures, because we evaluate the *differences* in NaOH consumption between the corresponding solns., *i.e.*, always solns. with and without ligand are titrated (see also below and [24] [25]).

Determination of Equilibrium Constants. The acidity constants K^H_{H2ampa} and K^H_{Hampa} for $H_2ampa^{\pm}$ were determined by titrating 50 ml of aq. 0.4 mM HNO_3 and $NaNO_3$ ($I = 0.1$M; 25°) in the presence and absence of 0.3 mM $^+H_3NCH_2PO_3^{2-}$ under N_2 with 2 ml of 0.03M NaOH and by using the differences in NaOH consumption between two such titrations for the calculations. For the determination of K^H_{H2pfa} and K^H_{Hpfa} of H_2pfa^-, the concentrations of HNO_3 and $^-OOCPO_3^{2-}$ were 0.55 mM and 0.3 mM, respectively, and 1 ml of 0.03M NaOH was used in the titrations. K^H_{H2pfa} and K^H_{Hpfa} were calculated with a *Hewlett-Packard Vectra 60PC* desk computer (connected with a *Brother M1509* printer and a *Hewlett-Packard 7475A* plotter) by a curve-fit procedure using a *Newton-Gauss* nonlinear-least-squares program within the pH range determined by the lowest point of neutralization reached under the experimental conditions (*ca.* 30% neutralization for the equilibrium $H_2pfa^-/Hpfa^{2-}$) and *ca.* 90% neutralization for the equilibrium $Hpfa^{2-}/pfa^{3-}$. The calculations for K^H_{H2ampa} and K^H_{Hampa} were carried out correspondingly in the range between 3% neutralization for the equilibrium $H_2ampa^{\pm}/Hampa^-$ and *ca.* 70% for the equilibrium $Hampa^-/ampa^{2-}$. For the acidity constants of $H_2ampa^{\pm}$ and H_2pfa^-, each 32 pairs of independent titrations were made and the results averaged.

The stability constants $K^M_{[M(Hampa)]}$, $K^M_{[M(ampa)]}$ and $K^M_{[M(Hpfa)]}$, $K^M_{[M(pfa)]}$ were determined under the same conditions as the corresponding acidity constants, but $NaNO_3$ was partly or fully replaced by $M(NO_3)_2$ ($I = 0.1$M, 25°). The $Hampa^-/M^{2+}$ ratios were 1:111 (Mg^{2+}, Ca^{2+}, Sr^{2+}, Ba^{2+}), 1:89 (Mg^{2+}, Ca^{2+}), 1:56 (Mn^{2+}, Co^{2+}, Ni^{2+}, Zn^{2+}, Cd^{2+}), 1:44 (Co^{2+}, Ni^{2+}), 1:28 (Mn^{2+}, Zn^{2+}, Cd^{2+}), and 1:11 and 1:5.6 (Cu^{2+}). The ratios pfa^{3-}/M^{2+} were 1:56 (Mg^{2+}, Ca^{2+}, Sr^{2+}, Ba^{2+}), 1:28 (Mg^{2+}, Ca^{2+}, Sr^{2+}, Ba^{2+}), 1:14 (Mg^{2+}, Ca^{2+}, Sr^{2+}, Ba^{2+}, Mn^{2+}, Co^{2+}, Ni^{2+}, Zn^{2+}, Cd^{2+}), 1:7 (Ba^{2+}, Mn^{2+}, Co^{2+}, Ni^{2+}, Zn^{2+}, Cd^{2+}), 1:5.6 (Mn^{2+}, Zn^{2+}, Cd^{2+}), 1:2.8 (Mn^{2+}, Cu^{2+}), and 1:1.4 (Cu^{2+}). $K^M_{[M(HL)]}$ and $K^M_{[ML]}$, where $L = ampa^{2-}$ or pfa^{3-}, were computed for each pair of titrations with a curve-fitting procedure by taking into account the species H^+, H_2L, HL, L, M^{2+}, [M(HL)], and [ML]. The data were collected every 0.1 pH unit from *ca.* 5% complex formation to a neutralization degree of *ca.* 90% or to the beginning of the hydrolysis of $[M(aq)]^{2+}$, which was evident from the titrations without ligand.

The stability constants, $K^{Cu(Arm)}_{[Cu(Arm)(HL)]}$ and $K^{Cu(Arm)}_{[Cu(Arm)L]}$ of the ternary complexes, where Arm = bpy or phen and $L = ampa^{2-}$ or pfa^{3-}, were determined under exactly the same conditions and in the same way as given above for the corresponding binary complexes, but now a 1:1 ratio of Arm/Cu^{2+} was employed. Under these conditions the formation of the $[Cu(Arm)]^{2+}$ complexes is practically complete in the pH range used for the evaluation, as was evident from the titrations in the absence of ampa or pfa; this agrees with the well known high stability of the $[Cu(bpy)]^{2+}$ and $[Cu(phen)]^{2+}$ complexes [26].

None of the stability constants calculated individually showed any dependence on pH or on the excess amount of M^{2+} or Cu^{2+}/Arm. The results listed in *Table 1* (*vide infra*) for the various complexes are the averages of the evaluations of at least eight independent pairs of titration curves.

Results and Discussion. – 1. *Acidity Constants of H_3pfa.* Phosphonoformic acid, H_3pfa, is a three-proton donor: the first proton is released from the phosphorous-acid residue in the low pH range (pK^H_{H3pfa} = 0.78 [21] and 1.7 [27] at I = 0.1 and 0.05, respectively, at 25°) which is not of interest in the present context; however, the other two deprotonation steps need to be considered:

$$HOOCPO_3H^- \rightleftharpoons H^+ + {}^-OOCPO_3H^- \quad (1a)$$

$$K^H_{H2pfa} = [H^+][Hpfa^{2-}]/[H_2pfa^-] \quad (1b)$$

$$^{-}OOCPO_3H^{-} \rightleftharpoons H^{+} + {}^{-}OOCPO_3^{2-} \tag{2a}$$

$$K^{H}_{Hpfa} = [H^{+}][pfa^{3-}]/[Hpfa^{2-}] \tag{2b}$$

The carboxylic acid ionization (*Eqn. 1*) occurs with $pK^{H}_{H_2pfa} = 3.61 \pm 0.01$ and the final phosphonate deprotonation (*Eqn. 2*) with $pK^{H}_{Hpfa} = 7.57 \pm 0.01$ in aqueous solution at $I = 0.1$M ($NaNO_3$) and 25°. These results are in excellent agreement with previous determinations [21] [23c] [27].

Substitution of the C-bound H-atom in HCOOH ($pK_a = 3.58$ [28]), by the monoprotonated phosphonate group, PO_3H^-, does not affect the carboxylic-acid ionization. This means, that the electron-withdrawing effect of the PO_3H^- unit and the influence of the now also present negative charge cancel each other. This observation contrasts with the result of the substitution of the P-bound H-atom of phosphorous acid, H_3PO_3 ($pK_a \approx 6.5$; average from [29–31]), by a carboxylate group, COO^-; in this case the acidity of the monoprotonated phosphonate group is decreased by *ca.* 1.1 pK units. Here, the retarding effect of the additional negative charge of the COO^- group on the H^+ removal is clearly seen. However, the COO^- unit and a CH_3 group exercise the same effect: the acidity constant of monoprotonated methyl phosphonate, $CH_3PO_3H^-$ ($pK_a = 7.53$ [32]), is practically identical with the one for $Hpfa^{2-}$, $^{-}OOCPO_3H^-$ ($pK_a = 7.57$).

2. *Acidity Constants of H_3ampa^+.* The net zero-charge form of (aminomethyl)phosphonic acid, $H_2ampa^{\pm}$, undergoes two deprotonations:

$$^{+}H_3NCH_2PO_2OH^{-} \rightleftharpoons H^{+} + {}^{+}H_3NCH_2PO_3^{2-} \tag{3a}$$

$$K^{H}_{H_2ampa} = [H^{+}][Hampa^{-}]/[H_2ampa^{\pm}] \tag{3b}$$

$$^{+}H_3NCH_2PO_3^{2-} \rightleftharpoons H^{+} + H_2NCH_2PO_3^{2-} \tag{4a}$$

$$K^{H}_{Hampa} = [H^{+}][ampa^{2-}]/[Hampa^{-}] \tag{4b}$$

Clearly, there is also a H_3ampa^+ form, but this exists only at a very low pH ($pK^{H}_{H_3ampa} \approx 0.4$ [23c]) and is not considered here. The first of the two other deprotonations with $pK^{H}_{H_2ampa} = 5.42 \pm 0.01$ may be assigned to the phosphonate deprotonation (*Eqn. 3*) and the second, with $pK^{H}_{Hampa} = 10.08 \pm 0.01$ ($I = 0.1$M, $NaNO_3$; 25°), to the ammonium-group deprotonation (*Eqn. 4*). Both constants are in perfect accord with values given in [23c] as well as with those determined by *Hägele et al.* [21].

Substitution of a C-bound H-atom in methylammonium ion, $CH_3NH_3^+$ ($pK_a = 10.7$ ([23]) by a PO_3^{2-} group acidifies the ammonium group by *ca.* 0.6 pK units. Here the electron-withdrawing properties of the PO_3^{2-} unit are clearly dominating over the charge effect which should operate into the opposite direction. Substitution of the P-bound H-atom of phosphorous acid, H_3PO_3 ($pK_a \approx 6.5$ [29–31]), by the $CH_2NH_3^+$ group acidifies the phosphonate deprotonation by *ca.* 1.1 pK_a units, while the carboxylate-group substitution mentioned at the end of the previous section (*Sect. 1*) *decrease* the acidity by the same magnitude. The mentioned acidification is thus understandable as a consequence of the positive charge of the $CH_2NH_3^+$ group.

We shall also find use for the rare microform of $Hampa^-$, *i.e.*, for $NH_2CH_2P(O)_2OH)^-$, in *Sect. 8* and *9* and, therefore, derive now its properties. Of the same overall 1– charge as the more common form, $^{+}H_3NCH_2PO_3^{2-}$, the rare microform is generated by the alternative deprotonation pathway expressed in *Equilibria 5a* and *6a*; the corresponding micro-acidity constants, $k_{a/1}$ and $k_{a/2}$, are defined in *Eqns. 5b* and *6b*:

$$^{+}H_3NCH_2P(O)_2(OH)^- \rightleftharpoons H^+ + H_2NCH_2P(O)_2(OH)^- \tag{5a}$$

$$k_{a/1} = [H^+][H_2NCH_2PO_3H^-]/[^{+}H_3NCH_2PO_3H^-] \tag{5b}$$

$$H_2NCH_2P(O)_2(OH)^- \rightleftharpoons H^+ + H_2NCH_2PO_3^{2-} \tag{6a}$$

$$k_{a/2} = [H^+][H_2NCH_2PO_3^{2-}]/[H_2NCH_2PO_3H^-] \tag{6b}$$

We set the difference $pk_{a/2} - pK^H_{H_2ampa} = pK^H_{Hampa} - pk_{a/1} = 2.0$, corresponding to the glycine system ($^{+}H_3NCH_2COO^- \rightleftharpoons H_2NCH_2COOH$) for which this value has been derived earlier [33] based on the pK_a of the monoprotonated glycine methyl ester, $^{+}H_3NCH_2C(O)OCH_3$, as a model for the ammonium-group deprotonation from the glycine cation, $^{+}H_3NCH_2COOH$. With the given specification, we obtain $pk_{a/1} = 8.1$ and $pk_{a/2} = 7.4$. The ratio of the common to the rare microform of the same net charge is then given [33] by *Eqn. 7*:

$$\frac{[^{+}H_3NCH_2PO_3^{2-}]}{[H_2NCH_2PO_3H^-]} = \frac{K^H_{H_2ampa}}{k_{a/1}} = \frac{10^{-5.42}}{10^{-8.1}} = 10^{2.7} = 500 \tag{7}$$

This result means, only once within 500 $Hampa^-$ species is the proton located at the phosphonate residue and not, as usual, at the amino group.

3. *Stability Constants of pfa and ampa Metal-Ion Complexes.* Metal ions interact with both the monoprotonated and completely deprotonated forms of each ligand, L:

$$M^{2+} + HL \rightleftharpoons [M(HL)] \tag{8a}$$

$$K^M_{[M(HL)]} = [[M(HL)]]/([M^{2+}][HL]) \tag{8b}$$

$$M^{2+} + L \rightleftharpoons ML \tag{9a}$$

$$K^M_{[ML]} = [[ML]]/([M^{2+}][L]) \tag{9b}$$

Deprotonation in the monoprotonated complex, [M(HL)],

$$[M(HL)] \rightleftharpoons H^+ + ML \tag{10a}$$

$$K^H_{[M(HL)]} = [H^+][[ML]]/[[M(HL)]] \tag{10b}$$

is linked to the stability constants (*Eqns. 8* and *9*) and deprotonation in the free monoprotonated ligand, K^H_{HL} (*Eqns. 2* and *4*), by *Eqn. 11*:

$$pK^H_{[M(HL)]} = \log K^M_{[M(HL)]} - \log K^M_{[ML]} + pK^H_{HL} \tag{11}$$

Table 1 shows the results for the stability constants as $\log K^M_{[M(HL)]}$, $\log K^M_{[ML]}$, and $pK^H_{[M(HL)]}$ for each ligand and ten metal ions together with the corresponding values for two ternary [Cu(Arm)(L)] complexes[3]).

Some literature comparisons are possible with pfa. Reported successive pK_a values for H_2pfa^- of 3.59 and 7.56 [27] agree excellently with those of *Table 1* (see *Footnote a*; *cf.* also *Sect. 1*). Log-stability constants reported from the same laboratory [20] are for Mg^{2+} 1.7

Table 1. *Logarithms of the Stability Constants of [M(HL)] (Eqn. 8) and [ML] (Eqn. 9) Complexes with pfa and ampa[3]) as Determined by Potentiometric pH Titrations in Aqueous Solution, Together with the Negative Logarithms of the Acidity Constants (Eqns. 10* and *11) of the Corresponding [M(HL)] Complexes* (I = 0.1 M, $NaNO_3$; 25°)[a])[b])

M^{2+}	L = pfa^{3-}			L = $ampa^{2-}$		
	$\log K^{M}_{[M(HL)]}$	$\log K^{M}_{[ML]}$	$pK^{H}_{[M(HL)]}$[c])	$\log K^{M}_{[M(HL)]}$	$\log K^{M}_{[ML]}$	$pK^{H}_{[M(HL)]}$[c])
Mg^{2+}	1.61 ± 0.10	4.11 ± 0.07	5.07 ± 0.12	1.22 ± 0.05	1.94 ± 0.06	9.36 ± 0.08
Ca^{2+}	1.71 ± 0.15	3.57 ± 0.09	5.71 ± 0.18	1.03 ± 0.03	1.62 ± 0.04	9.49 ± 0.05
Sr^{2+}	1.45 ± 0.16	2.94 ± 0.07	6.08 ± 0.17	0.79 ± 0.06	1.34 ± 0.04	9.53 ± 0.07
Ba^{2+}	1.42 ± 0.12	2.73 ± 0.05	6.26 ± 0.13	0.67 ± 0.04	1.17 ± 0.05	9.58 ± 0.06
Mn^{2+}	2.44 ± 0.16	5.10 ± 0.13	4.91 ± 0.21	1.77 ± 0.04	3.62 ± 0.08	8.23 ± 0.09
Co^{2+}	2.41 ± 0.05	5.31 ± 0.05	4.67 ± 0.07	1.52 ± 0.04	4.58 ± 0.04	7.02 ± 0.06
Ni^{2+}	2.45 ± 0.08	5.39 ± 0.07	4.63 ± 0.11	1.47 ± 0.03	5.30 ± 0.04	6.25 ± 0.05
Cu^{2+}	3.69 ± 0.15	7.78 ± 0.06	3.48 ± 0.16	2.67 ± 0.03	8.09 ± 0.02	4.66 ± 0.04
$Cu(bpy)^{2+}$	3.97 ± 0.10	7.94 ± 0.05	3.60 ± 0.11	2.45 ± 0.06	7.52 ± 0.02	5.01 ± 0.06
$Cu(phen)^{2+}$	4.02 ± 0.19	7.99 ± 0.05	3.60 ± 0.20	2.43 ± 0.06	7.60 ± 0.04	4.91 ± 0.07
Zn^{2+}	2.57 ± 0.03	5.64 ± 0.03	4.50 ± 0.04	1.72 ± 0.07	4.91 ± 0.07	6.89 ± 0.10
Cd^{2+}	2.57 ± 0.12	5.45 ± 0.10	4.69 ± 0.16	2.02 ± 0.03	5.14 ± 0.06	6.96 ± 0.07

[a]) Acidity constants of the ligands $pK^{H}_{H_2pfa}$ = 3.61 ± 0.01, pK^{H}_{Hpfa} = 7.57 ± 0.01; $pK^{H}_{H_2ampa}$ = 5.42 ± 0.01, pK^{H}_{Hampa} = 10.08 ± 0.01.

[b]) The errors given are *three times* the standard error of the mean value or the sum of the probable systematic errors, whichever is larger. The error limits of the derived data, in the present case for $pK^{H}_{[M(HL)]}$, were calculated according to the error propagation after *Gauss*.

[c]) These values were calculated according to *Eqn. 11* by using the acidity constants given in *Footnote a* and the stability constants listed above in *Columns 2, 3,* and *5, 6.*

([M(HL)]) and 3.59 ([ML]), and for Ca^{2+} 1.84 ([M(HL)]) and 3.55 ([ML]). Of the four values, only that of 3.59 disagrees with our results in *Table 1*; however, our value is in agreement with the general trends (see. *e.g. Table 2,* in *Sect. 4*). An ESR study [22] reports Mn^{2+}-binding constants also in close accordance with those in *Table 1*, 2.57 ([M(HL)]) and 5.34 ([ML]). In addition, there is good agreement with the data of *Hägele et al.* [21] determined *via* potentiometric pH titrations: $\log K^{M}_{[M(Hpfa)]}$ = 1.66 (Mg^{2+}), 1.81 (Ca^{2+}), 2.42 (Zn^{2+}), and $\log K^{M}_{[M(pfa)]}$ = 3.84 (Mg^{2+}), 3.57 (Ca^{2+}), 5.58 (Zn^{2+}). Overall, the agreement between the eight known constants for pfa complexes and the present results is satisfying.

Several comparisons with literature values [23] are possible for ampa complexes. Excellent agreement occurs with pK_a values for $H_2ampa^{\pm}$ and $\log K$ values for [M(HL)] and [ML] complexes of Mg^{2+}, Ca^{2+}, Co^{2+}, Ni^{2+}, Cu^{2+}, and Zn^{2+} (I = 0.1 M, KNO_3; 25°) [34]. On the other hand, a second study [35], also under the same conditions as ours (I = 0.1 M, $NaClO_4$; 25°), reports lower pK_a values for the deprotonation of $H_2ampa^{\pm}$ (0.13 and 0.22 pK units), significantly greater (0.7 to 1.4 log units) log-stability constants for [M(HL)] with Co^{2+}, Ni^{2+}, Cu^{2+}, Zn^{2+}, and Cd^{2+}, but somewhat closer agreement (within 0.4 log units) for [ML] complexes of these metal ions and Mn^{2+}. Finally, the closely related CH_3-substituted ligand, (1-aminoethyl)phosphonate, $H_2N(CH_3)CHPO_3^{2-}$ [36], exhibits for the diprotonated state comparable pK_a values (I = 0.2 M, KNO_3; 25°), and closely similar stability constants for the [ML] complexes of Co^{2+}, Ni^{2+}, and Cu^{2+}, but a value for Zn^{2+} that appears high by 1.1 log units.

In general, there is good agreement between those systems studied earlier with values available in the literature and our results; in the case of a discrepancy, we consider our values as more reliable, because they are consistent with each other as well as with the general trends (see *Table 2* in *Sect. 4* and the evaluations in *Sect. 6–10*).

4. *Comparison of the Experimentally Determined Stability Constants of the Complexes with Calculated Values Derived from the Phosphonate-Group Basicity.* It is well known that for families of structurally related ligands plots of $\log K^{M}_{[ML]}$ *vs.* pK^{H}_{HL} result in straight lines [37]. This is also true for $[M(RPO_3)]$ complexes, where RPO_3^{2-} respresents a simple unidentate phosphonate (like methyl or ethyl phosphonate) or phosphate-monoester ligand (like phenyl phosphate), as is demonstrated for two examples in the *Figure.*

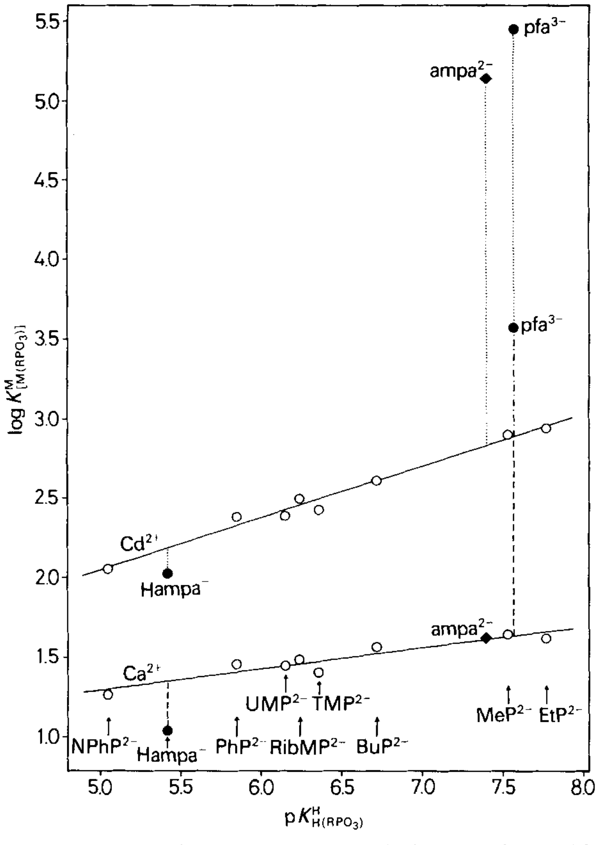


Figure. *Evidence for an enhanced stability of the $[Ca(pfa)]^-$ and $[Cd(pfa)]^-$ complexes and for a reduced stability of the $[Ca(Hampa)]^+$ and $[Cd(Hampa)]^+$ complexes* (●), *based on the relationship between* $\log K^{M}_{[M(RPO_3)]}$ *and* $pK^{H}_{H(RPO_3)}$ *for the 1:1 complexes of* Ca^{2+} *and* Cd^{2+} *with some simple phosphate monoester or phosphonate ligands* (RPO_3^{2-}): *4-nitrophenyl phosphate* ($NPhP^{2-}$), *phenyl phosphate* (PhP^{2-}), *uridine 5′-monophosphate* (UMP^{2-}), D-*ribose 5-monophosphate* ($RibMP^{2-}$), *thymidine* (= *2′-deoxyribosylthymine*) *5′-monophosphate* (TMP^{2-}), *butyl phosphate* (BuP^{2-}), *methanephosphonate* (MeP^{2-}), *and ethanephosphonate* (EtP^{2-}) (from left to right) (○). The least-squares lines are drawn through the corresponding eight data sets, which are taken for the phosphate monoesters from [38] and for the phosphonates from [32]; the equations for these base lines are given in Table 5 of [32]. The points due to the equilibrium constants for the $[M(pfa)]^-$ and $[M(Hampa)]^+$ systems (●) are based on the data listed in *Table 1*. The vertical *broken* (Ca^{2+}) and *dotted* (Cd^{2+}) lines emphasize the stability differences to the corresponding reference lines; these differences equal $\log \Delta_{[M(RPO_3)]}$ as defined in *Section 4* by *Eqn. 12*. The log stability constants of the [M(ampa)] complexes (*Table 1*) (◆) are plotted *vs.* the micro-acidity constant $pk_{a/2} = 7.4$ (see *Sect. 2* and *9*). All the plotted equilibrium-constant values refer to aqueous solutions at $I = 0.1$ M ($NaNO_3$) and 25°.

Recently, in comprehensive studies [32] [38] [39], well defined slopes and intercepts for the base-line equation for each metal ion considered in the present study have been elaborated. With these straight-line equations [32] [39] and the acidity constants of the monoprotonated phosphonate groups (pK_a = 7.57 for $Hpfa^{2-}$ and 5.42 for $H_2ampa^{\pm}$) stability constants may be calculated (calc.) for $[M(pfa)]^-$ and $[M(Hampa)]^+$ complexes. It must be emphasized that these stability constants, which are listed in *Columns 3* and *6* of *Table 2*, refer to complex species with a sole phosphonate coordination of the metal ion.

Table 2. *Stability-Constant Comparisons for the* $[M(pfa)]^-$ *and* $[M(Hampa)]^+$ *Complexes between the Measured Stability Constants* (exper.) *and the Calculated Stability Constants* (calc.) *Based on the Basicity of the Phosphonate Group and the Base-Line Equations Determined Earlier* [32] [39] (I = 0.1 M, $NaNO_3$; 25°)[a])

	$\log K^{M}_{[M(pfa)]}$		$\log \Delta_{[M(pfa)]}$[d])	$\log K^{M}_{[M(Hampa)]}$		$\log \Delta_{[M(Hampa)]}$[d])
M^{2+}	exper.[b])	calc.[c])		exper.[e])	calc.[f])	
Mg^{2+}	4.11 ± 0.07	1.85 ± 0.03	2.26 ± 0.08	1.22 ± 0.05	1.40 ± 0.03	−0.18 ± 0.06
Ca^{2+}	3.57 ± 0.09	1.63 ± 0.05	1.94 ± 0.10	1.03 ± 0.03	1.35 ± 0.05	−0.32 ± 0.06
Sr^{2+}	2.94 ± 0.07	1.35 ± 0.04	1.59 ± 0.08	0.79 ± 0.06	1.18 ± 0.04	−0.39 ± 0.07
Ba^{2+}	2.73 ± 0.05	1.28 ± 0.04	1.45 ± 0.06	0.67 ± 0.04	1.09 ± 0.04	−0.42 ± 0.06
Mn^{2+}	5.10 ± 0.13	2.48 ± 0.05	2.62 ± 0.14	1.77 ± 0.04	1.97 ± 0.05	−0.20 ± 0.06
Co^{2+}	5.31 ± 0.05	2.24 ± 0.06	3.07 ± 0.08	1.52 ± 0.04	1.76 ± 0.06	−0.24 ± 0.07
Ni^{2+}	5.39 ± 0.07	2.28 ± 0.05	3.11 ± 0.09	1.47 ± 0.03	1.75 ± 0.05	−0.28 ± 0.06
Cu^{2+}	7.78 ± 0.06	3.51 ± 0.06	4.27 ± 0.08	2.67 ± 0.03	2.51 ± 0.06	0.16 ± 0.07
$Cu(bpy)^{2+}$	7.94 ± 0.05	3.53 ± 0.07	4.41 ± 0.09	2.45 ± 0.06	2.53 ± 0.07	−0.08 ± 0.09
$Cu(phen)^{2+}$	7.99 ± 0.05	3.54 ± 0.06	4.45 ± 0.08	2.43 ± 0.06	2.54 ± 0.06	−0.11 ± 0.08
Zn^{2+}	5.64 ± 0.03	2.59 ± 0.06	3.05 ± 0.07	1.72 ± 0.07	1.85 ± 0.06	−0.13 ± 0.09
Cd^{2+}	5.45 ± 0.10	2.89 ± 0.05	2.56 ± 0.11	2.02 ± 0.03	2.18 ± 0.05	−0.16 ± 0.06

[a]) All error limits correspond to *three times* the standard error; *cf.* also *Footnote b* of *Table 1*.

[b]) Values from *Column 3* in *Table 1* (*Eqn. 9*).

[c]) Calculated with pK^{H}_{Hpfa} = 7.57 (*Eqn. 2*; see *Sect. 1* and *Footnote a* in *Table 1*) and the base-line equations listed in *Table 5* of [32]; the error limits are *three times* the SD values given in Table 6 of [32]. The values for the two $[Cu(Arm)(pfa)]^-$ mixed-ligand complexes were calculated with the straight-line terms summarized in Table 5 of [39].

[d]) Calculated according to *Eqn. 12*.

[e]) Values from *Column 5* of *Table 1* (*Eqn. 8*).

[f]) Calculated with pK^{H}_{H2ampa} = 5.42 (*Eqn. 3*; see *Sect. 1* and *Footnote a* in *Table 1*); otherwise, the above given *Footnote c* applies.

For each phosphonate-metal-ion complex, a difference between the experimentally measured log-stability constant, $\log K_{exper.}$, and the calculated one, $\log K_{calc.}$, can now be obtained according to *Eqn. 12*:

$$\log \Delta_{[M(RPO_3)]} = \log K_{exper.} - \log K_{calc.} \qquad (12)$$

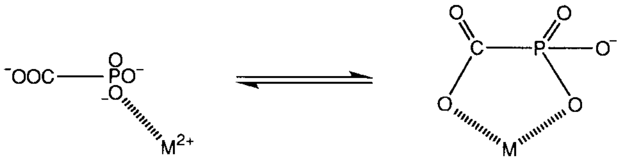
These differences correspond to the vertical dotted lines seen in the *Figure*; the actual values for $\log \Delta_{[M(RPO_3)]}$ are given in *Columns 4* and *7* of *Table 2*.

It is evident that any metal-ion interaction in a $[M(RPO_3)]$ complex in addition to that with the phosphonate group must be reflected in an increased stability constant [37]. Indeed, the high $\log \Delta_{[M(pfa)]}$ values in *Table 2* for the phosphonoformate complexes immediately demonstrate that metal ions hardly bind in a unidentate mode to pfa^{3-} but chelate to the phosphonate and carboxylate groups in $[M(pfa)]^-$ (see also *Sect. 5*). On the other hand, the values for the $[\mathrm{M(Hampa)}]^+$ complexes do also not rest on the reference lines (see *Fig.* and *Table 2*) but lie below the baseline (except for Cu^{2+}; see *Sect. 7*) indicating that the charged ammonium group in these complexes excercises a stronger repulsion effect on the binding of a twofold positively charged metal ion than on that of the proton which carries only a single charge (see also *Sect. 6*). This charge effect is of relevance in the evaluations carried out in the next section.

As expected, the charge effect in the various $[\mathrm{M(Hampa)}]^+$ complexes is rather independent of the kind of metal ion involved. This is quite different, again as expected, for the $\log \Delta$ values of the $[\mathrm{M(pfa)}]^-$ species, because the extent of chelate formation depends on the binding properties of the metal ion involved.

5. *Structure of the* $[M(pfa)]^-$ *and* $[M(Hpfa)]$ *Complexes in Solution: Intramolecular Equilibria.* Even the lowest $\log \Delta_{[\mathrm{M(pfa)}]}$ value of 1.45 in *Column 4* of *Table 2*, which refers to $[\mathrm{Ba(pfa)}]^-$, represents a considerable stability increase indicating a high degree of chelate formation. However, here the question arises: how large is the charge effect of the carboxylate group on the binding of a metal ion to the phosphonate group, *i.e.*, on the stability of the *open* $^-\mathrm{OOC{-}PO_3 \cdot M}$ species?

As the distance of a phosphonate-bound metal ion to the positively charged ammonium group in a $^+\mathrm{H_3NCH_2{-}PO_3 \cdot M}$ complex corresponds to that in $^-\mathrm{OOC{-}PO_3 \cdot M}$, one expects that the charge effect is comparable in both cases, though of opposite sign. As Ca^{2+}, Sr^{2+}, and Ba^{2+} are known to have a very low affinity for amino groups [40], no chelation is expected (see also *Sect. 6*), and the three values in *Column 7* of *Table 2* for these $[\mathrm{M(Hampa)}]^+$ complexes represent the negative charge effect certainly well: the average (considering also the internal and external error limits) equals -0.38 ± 0.09 log units. In the case of the $[\mathrm{M(pfa)}]^-$ complexes, one expects thus a *positive* contribution to the stability of the same size; hence, subtraction of 0.38 log units from the $\log \Delta_{[\mathrm{M(pfa)}]}$ values listed in *Column 4* of *Table 2* will result in $\log \Delta^*$ values which now solely represent the stability increase due to chelate formation of the phosphonate-bound metal ion with the carboxylate group. The corresponding values are listed in *Table 3* together with the dimensionless constant K_I for the intramolecular *Equilibrium 13*:



(13)

This constant may now be calculated (for details see, *e.g.*, [32] [37] [41]) according to *Eqn. 14*:

$$K_\mathrm{I} = [\text{chelate}]/[\text{open complex}] = 10^{\log \Delta^*} - 1 \qquad (14)$$

the percentage of the chelate occurring in *Equilibrium 13* follows from *Eqn. 15*:

$$\%\,[\text{chelate}] = 100 \cdot K_\mathrm{I}/(1 + K_\mathrm{I}) \qquad (15)$$

The results of *Table 3* confirm the assumptions indicated above: the formation degree of the chelate in *Equilibrium 13* is beyond 90% for all the $[\mathrm{M(pfa)}]^-$ complexes studied.

Table 3. *Extent of Chelate Formation (Equilibrium 13) in* $[M(pfa)]^-$ *Complexes as Quantified by the Dimensionless Equilibrium Constant* K_I *(Eqn. 14) and the Percentage of* $[M(pfa)]^-_{chelate}$ *(Eqn. 15) in Aqueous Solution* ($I = 0.1$ M, $NaNO_3$; 25°)[a])

M^{2+}	$\log \Delta_{[M(pfa)]}$ (*Eqn. 12*)[a])	$\log \Delta^*$ *cf.*[b])	K_I (*Eqn. 14*)	% $[M(pfa)]^-_{chelate}$ (*Eqns. 13, 15*)
Mg^{2+}	2.26 ± 0.08	1.88 ± 0.12	74.9 ± 21.0	98.7 ± 0.4
Ca^{2+}	1.94 ± 0.10	1.56 ± 0.13	35.3 ± 10.9	97.2 ± 0.8
Sr^{2+}	1.59 ± 0.08	1.21 ± 0.12	15.2 ± 4.5	93.8 ± 1.7
Ba^{2+}	1.45 ± 0.06	1.07 ± 0.11	10.7 ± 3.0	91.5 ± 2.2
Mn^{2+}	2.62 ± 0.14	2.24 ± 0.17	173 ± 68	99.4 ± 0.2
Co^{2+}	3.07 ± 0.08	2.69 ± 0.12	489 ± 135	99.8 ± 0.1
Ni^{2+}	3.11 ± 0.09	2.73 ± 0.13	536 ± 161	99.8 ± 0.1
Cu^{2+}	4.27 ± 0.08	3.89 ± 0.12	7760 ± 2140	99.99 ± 0.01
$Cu(bpy)^{2+}$	4.41 ± 0.09	4.03 ± 0.13	10700 ± 3200	99.99 ± 0.01
$Cu(phen)^{2+}$	4.45 ± 0.08	4.07 ± 0.12	11700 ± 3200	99.99 ± 0.01
Zn^{2+}	3.05 ± 0.07	2.67 ± 0.11	467 ± 118	99.8 ± 0.1
Cd^{2+}	2.56 ± 0.11	2.18 ± 0.14	150 ± 49	99.3 ± 0.2

[a]) The values for $\log \Delta_{[M(pfa)]}$ (*Eqn. 12*) and their error limits (3σ) are from *Table 2*; see also *Footnote b* of *Table 1*.

[b]) $\log \Delta^* = \log \Delta_{[M(pfa)]} - (0.38 \pm 0.09)$; this value represents the stability enhancement due to chelation (see *Sect. 5*).

Comparison of the $\log K^M_{[M(HL)]}$ stability constants (*Eqn. 8*) of $Hpfa^{2-}$ (*Table 1*) with $\log K^M_{[ML]}$ values of formate, $HCOO^-$ [28] [42], indicates greater stabilities of the [M(Hpfa)] complexes by > 1.3 log units. (Both carboxylate-protonated free ligand-species have closely similar pK_a values (see *Sect. 1*), allowing a direct comparison of the log-stability constants.) Considering that the overall charges of $(HCOO \cdot M)^+$ and $(M \cdot OOC{-}PO_3H)^{\pm}$ are different, a promoting charge effect of the PO_3H^- residue must be taken into account for the $^-OOC{-}PO_3H^-$ species. As here a negative charge is operating with regard to M^{2+} in the same distance as in the examples considered above, this effect will again be well represented by 0.38 log units. Hence, subtracting 0.4 log units from the total stability enhancement of > 1.3 log units leaves us with an enhancement of 0.9 log units or more which is due to chelation in the [M(Hpfa)] complexes. This then means that from the $[M(Hpfa)]^{\pm}$ species more than 87% exist as chelates.

Thus, both the $[M(pfa)]^-$ and [M(Hpfa)] complexes are to the largest part chelated and the $pK^H_{[M(Hpfa)]}$ values recorded in *Column 4* of *Table 1* refer to a deprotonation involving chelated ligands:

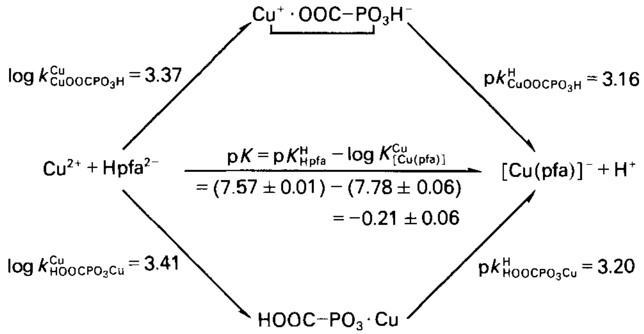
(16)

The $pK^H_{[M(Hpfa)]}$ values (*Table 1*) are at least 1.3 p*K* units less than the $pK^H_{Hpfa} = 7.57$ for the phosphonate deprotonation in the monoprotonated free ligand, indicating promotion of the phosphonate acidity by chelated metal ions. In the [M(Hpfa)] chelate, a phosphonate group binds both, a metal ion and a proton.

The above analysis as summarized in *Equilibrium 16* certainly applies to all [M(Hpfa)] complexes with $\mathrm{p}K^{\mathrm{H}}_{[\mathrm{M(HL)}]} > 4.5$ (*Table 1*), but it is for [Cu(Hpfa)] probably only partly correct because here $\mathrm{p}K^{\mathrm{H}}_{[\mathrm{Cu(HL)}]} = 3.48 \pm 0.16$, a value which is comparable to $\mathrm{p}K^{\mathrm{H}}_{\mathrm{H_2pfa}} = 3.61 \pm 0.01$, and which holds for the ionization of the carboxylic acid group in $\mathrm{HOOC{-}PO_3H^-}$. Hence, one may suspect that also a species $\mathrm{HOOC{-}PO_3 \cdot Cu}$ forms in which the metal ion is at the phosphonate residue and the proton at the carboxylate group. Indeed, the analysis *via* micro constants as summarized in *Fig. 2* proves this suspicion to be correct. The ratio, *R*, of the chelated isomer[4], $\underline{\mathrm{Cu^+ \cdot OOC{-}PO_3}}\mathrm{H^-}$ (which is depicted in *Eqn. 16*), to the phosphonate-bound species, $\mathrm{HOOC{-}PO_3 \cdot Cu}$, follows from the microstability constants given in the *Scheme* below:

$$R = \frac{[\underline{\mathrm{Cu^+ \cdot OOC - PO_3}}\mathrm{H^-}]}{[\mathrm{HOOC{-}PO_3 \cdot Cu}]} = \frac{k^{\mathrm{Cu}}_{\mathrm{CuOOCPO_3H}}}{k^{\mathrm{Cu}}_{\mathrm{HOOCPO_3Cu}}} = \frac{10^{3.37}}{10^{3.41}} = 10^{-0.04} \approx 10^{0} = \frac{1}{1}$$

Hence, both isomers occur for [Cu(Hpfa)] in a 1:1 ratio. A corresponding analysis for [Cu(bpy)(Hpfa)] and [Cu(phen)(Hpfa)] reveals that the chelated isomers occur to *ca.* 60% and the open phosphonate-bound species to 40%. As expected and already concluded above, for [Zn(Hpfa)], the chelated isomer (*Eqn. 16*), dominates with a formation degree of 95% the situation already strongly, and this is even more true for the other [M(Hpfa)] complexes listed in *Table 1*.



Scheme. *Equilibrium Scheme Showing the Interrelation between the Monoprotonated and Chelated [Cu(Hpfa)] Species*, $\underline{\mathrm{Cu^+ \cdot OOC{-}PO_3}}\mathrm{H^-}$ (*cf. Eqn. 16*; see at the top in the scheme), *and the Isomer Having the Proton at the Carboxylate and* $\mathit{Cu^{2+}}$ *at the Phosphonate Group*, $\mathrm{HOOC{-}PO_3 \cdot Cu}$ (bottom), *Together with the Other Species in Equilibrium with These Two Isomers.* The scheme defines also micro constants (*k*) and gives their interrelation with the macro constants (*K*); the arrows indicate the direction for which the constants are defined. The macro constants are known from the experiments (*Table 1*), and the micro constants have been derived on the following reasonings: The micro-acidity constant, $k^{\mathrm{H}}_{\mathrm{HOOCPO_3Cu}}$, for the deprotonation of the $\mathrm{HOOC{-}PO_3 \cdot Cu}$ species is estimated as $\mathrm{p}k^{\mathrm{H}}_{\mathrm{HOOCPO_3Cu}} = \mathrm{p}K^{\mathrm{H}}_{\mathrm{HCOOH}}$ [28] – (*charge effect*; see the second paragraph in *Sect. 5*) = (3.58 ± 0.01)–(0.38 ± 0.09) = 3.20 ± 0.09 (this constant is given on the right side in the lower part of the scheme). From this result and $1/K^{\mathrm{H}}_{[\mathrm{Cu(Hpfa)}]} = (1/k^{\mathrm{H}}_{\mathrm{CuOOCPO_3H}}) + (1/k^{\mathrm{H}}_{\mathrm{HOOCPO_3H}})$ follows $1/k^{\mathrm{H}}_{\mathrm{CuOOCPO_3H}} = (1/K^{\mathrm{H}}_{[\mathrm{Cu(Hpfa)}]}) - (1/k^{\mathrm{H}}_{\mathrm{HOOCPO_3Cu}}) = (1/10^{-3.48}) - (1/10^{-3.20}) = 10^{3.48} - 10^{3.20} = 10^{3.16}$ and, hence $\mathrm{p}k^{\mathrm{H}}_{\mathrm{CuOOCPO_3H}} = 3.16$ (*upper part*; *right*). The two micro-stability constants given at the left side in the *Scheme* follow now from the properties of cyclic systems.

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[4] The possible traces of an open species, $\mathrm{M^+ \cdot OOC - PO_3H^-}$, in which $\mathrm{M^{2+}}$ is solely bound to the carboxylate group, are ignored in this analysis (see also the limiting value discussed earlier in this section).

6. *Some Structural Considerations on [M(Hampa)]$^+$ Complexes in Solution.* For the [M(Hampa)]$^+$ complexes of monoprotonated (aminomethyl)phosphonate in *Table 1* an unusual stability order $Mn^{2+} > Co^{2+} > Ni^{2+}$ suggests primarily phosphonate binding. Indeed, this order contrasts with the *Irving-Williams* sequence, but it corresponds to observations made with phosphate-monoester ligands [38]. Hence, in the case of [M(Hampa)]$^+$ the primary coordination reaction refers to a simple phosphonate-coordination without chelation:

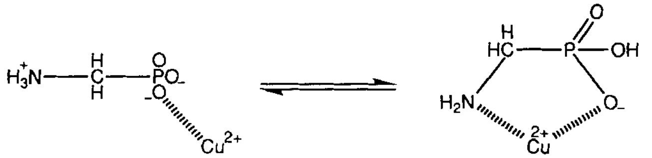
$$^{+}H_3NCH_2PO_3^{2-} + M^{2+} \rightleftharpoons {}^{+}H_3NCH_2PO_3 \cdot M \qquad (17)$$

The negative $\log \Delta_{[M(Hampa)]}$ values in *Table 2* are supporting the above view. Instead of the points due to the stability-constant logarithm resting on the standard line for each metal ion in the $\log K^{M}_{[M(RPO_3)]}$ *vs.* $pK^{H}_{H(RPO_3)}$ plot [32], for $pK^{H}_{H_2ampa} = 5.42$ (except for Cu^{2+}, see *Sect. 7*), the points lie below the reference line (see *Fig.*) by the $\log \Delta_{[M(Hampa)]}$ values listed in *Column 7* of *Table 2*. These negative $\log \Delta_{[M(Hampa)]}$ values indicate that the charged ammonium group reduces complex stability proportionately more than phosphonate basicity.

Indeed, the *N*-trimethylammonium group in *N*-trimethylammonium methylphosphonate also reduces complex stability more than predicted by its $pK^{H}_{H(RPO_3)} = 5.10$ [43]. The Ca^{2+} and Cu^{2+} complexes lie 0.34 and 0.26 log units, respectively, below straight reference-lines established for a series of substituted phosphonates [43]. These values compare favorably with $\log \Delta_{[M(Hampa)]} = -0.32$ ($\pm$0.06) for Ca^{2+} (*Table 2*), and an average of -0.26 ($\pm$0.09) for [Co(Hampa)]$^+$ and [Ni(Hampa)]$^+$; complexes of two transition-metal ions with a similar size as Cu^{2+}.

Thus, for complexation at the phosphonate group, charged ammonium or trimethylammonium groups in $^{+}X_3NCH_2PO_3^{2-}$ ligands (X = H or CH_3) result in weaker complex-stabilities than suggested by $\log K^{M}_{[M(RPO_3)]}$ *vs.* $pK^{H}_{H(RPO_3)}$ straight-line plots established with uncharged substituents of RPO_3^{2-} ligands. The definite establishment of this fact is important, because it implies that just as a positively charged substituent has a stability decreasing effect, a negatively charged one has to have a stability promoting effect that in absolute terms will be of the same size for an identical ligand geometry; this then is further justification for some of the evaluations presented in *Sect. 5* (see there, *e.g.*, in the second paragraph).

7. *A More Detailed Appraisal of the Structural Aspects for [Cu(Hampa)]$^+$ and [M(Hampa)]$^+$ Species in General.* Since a $\log \Delta_{[M(Hampa)]} = -0.26$ ($\pm$0.09) value is suggested for Cu^{2+} by the analysis presented in the second to the last paragraph in *Sect. 6*, the observed +0.16 ($\pm$0.07) value in *Table 2* for [Cu(Hampa)]$^+$ indicates an 0.42 ($\pm$0.11) log units augmented stability. This enhanced stability may be accounted for by the presence of a chelated form:



(18)

We define the equilibrium constant K_I between chelated and open forms again as given in *Eqn. 14*. The experimentally observed stability constant (*cf. Eqn. 8*) is then given by *Eqn. 19*,

$$K^{M}_{[M(Hampa)]} = \frac{([\text{open complex}] + [\text{chelate}])}{[M^{2+}][H_3^+NCH_2PO_3^{2-}]} = K^{M}_{open}(1 + K_I) \qquad (19)$$

where K^{M}_{open} is the stability constant expected for the open form [32] [37] [41], and the augmentation may be defined as in *Eqn. 20*:

$$\log \Delta^* = \log K^{M}_{[M(Hampa)]} - \log K^{M}_{open} \qquad (20a)$$

$$= \log \Delta_{[M(Hampa)]} - (-0.26 \pm 0.09) \qquad (20b)$$

From the above given augmented stability, $\log \Delta^* = 0.42 \pm 0.11$, and *Eqns. 14* and *15*, we obtain $K_I = 1.63 \pm 0.67$ and, for the formation degree of the chelate in *Equilibrium 18*, follows 62 ± 10%. Thus, the augmented stability resulting in a positive $\log \Delta_{[Cu(Hampa)]} = 0.16$ value in *Table 2* may be accounted for by occurrence of *ca.* 60% of a chelate form. In this chelate, the phosphonate group both bears a proton and coordinates to a metal ion, analogous to the protonated [M(Hpfa)] complex depicted above at the left in *Equilibrium 16*.

Of course, at this point one may argue that the use of −0.26 ± 0.09 (based on Co^{2+} and Ni^{2+}) for the open $[Cu(Hampa)]^+$ complex is not really appropriate[5]), and that the value of −0.38 ± 0.09 log units derived in *Sect. 5* from the complexes of $Hampa^-$ with Ca^{2+}, Sr^{2+}, and Ba^{2+} should be employed as a probably more justified basis for comparison[5]). In doing so, one obtains an augmentation of 0.54 ± 0.11 log units (= (0.16 ± 0.07) − (−0.38 ± 0.09); *Eqn. 20b*) for the stability of the $[Cu(Hampa)]^+$ complex and consequently (*via Eqn. 14*) $K_I = 2.47 \pm 0.88$ and, thus (*Eqn. 15*), for the formation degree of the chelate 71 ± 7%. These results may be considered as upper limits, yet it is satisfying to see that within the error limits they are identical with those given in the preceding paragraph.

The same chelate as depicted in *Equilibrium 18* for $[Cu(Hampa)]^+$ may also occur to a lesser extent with other metal ions in *Table 2*. However, the small differences between experimental and estimated stability constant logarithms (*Eqn. 20*) allow only some rough estimates for the chelate percentage occuring in other $[M(Hampa)]^+$ systems. In the following calculations, we use the value of −0.38 ± 0.09 log units for the open form; hence, the results represent rather upper limits. Application of the given value to the data of *Table 2* gives for $[Cu(bpy)(Hampa)]^+$ 50 ± 15% chelate ($\log \Delta^* = 0.30 \pm 0.13$; $K_I = 1.00 \pm 0.60$), for $[Cu(phen)(Hampa)]^+$ 46 ± 15% chelate ($\log \Delta^* = 0.27 \pm 0.12$; $K_I = 0.86 \pm 0.51$), for $[Zn(Hampa)]^+$ 44 ± 17% chelate ($\log \Delta^* = 0.25 \pm 0.13$; $K_I = 0.78 \pm 0.53$), and for $[Cd(Hampa)]^+$ 40 ± 15% chelate ($\log \Delta^* = 0.22 \pm 0.11$; $K_I = 0.66 \pm 0.42$). Clearly, Zn^{2+} and Cd^{2+} are those ions for which, next to Cu^{2+}, a significant affinity for amino groups is expected. In all the other cases in *Table 2*, there is

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[5]) Co^{2+} and Ni^{2+} may to some extent undergo the same reaction as described above for Cu^{2+} (see also *vide infra* in this section); for the alkaline-earth ions Ca^{2+}, Sr^{2+}, and Ba^{2+}, this is certainly not the case, as their affinity for amino groups is very low. Therefore, the use of their (averaged) $\log \Delta_{[M(Hampa)]}$ value as a basis in the evaluation may appear as more appropriate.

less than 35% chelation in the $[M(Hampa)]^+$ complexes; in fact, the calculations show that it is often even zero within the error limits.

8. *Is there an Alternative Amino-Group Microform-Pathway for Metal-Ion Binding to Hampa$^-$?* To this point, the analysis has been based upon primary phosphonate-group coordination in the species $^+H_3NCH_2PO_3^{2-}$. There is also the possibility that metal-ion coordination should be viewed as occurring at the amino group in the tautomeric microform, $NH_2CH_2PO_3H^-$. In *Sect. 2*, we estimated the micro-acidity constant, $\mathrm{p}k_{a/1} = 8.1$, for ammonium group deprotonation to yield this microform (*Eqn. 5*).

The transition-metal ions, especially Cu^{2+}, are the most likely to favor amino-group coordination. The stability constant for Cu^{2+} with NH_3, $\log K^{Cu}_{[Cu(NH_3)]} = 4.1$ [31], is substantially greater than the experimental value of 2.67 in *Table 2* for $[Cu(Hampa)]^+$. The ammonia value needs to be reduced by *ca.* 0.7 log units[6]) because of the greater basicity of NH_3 ($\mathrm{p}K^H_{H(NH_3)} = 9.4$ [31]) compared to the estimated $\mathrm{p}k_{a/1} = 8.1$ in the rare microform. However, the major reduction is due to only 1/500 of the 1 – charged forms being the rare microform, $NH_2CH_2PO_3H^-$, with a free amino group.

Thus, we reduce the ammonia value for both lower basicity and abundance of the $NH_2CH_2PO_3H^-$ form to find $4.1 - 0.7 - 2.7 = 0.7$ as the estimated practical stability-constant logarithm for Cu^{2+} binding to the free amino group in the microform. Since this value is appreciably smaller than the experimental value of 2.67 for $[Cu(Hampa)]^+$ (*Table 2*), we conclude that the alternative amino-group microform pathway is not competitive with primary metal-ion binding at the phosphonate group of $^+H_3NCH_2PO_3^{2-}$ for any metal ion in this study. Thus, this analysis supports further the evaluations presented in *Sect. 7*.

9. *Isomeric Forms of the [M(ampa)] Compexes in Solution.* The $\log K^M_{[M(ampa)]}$ values in *Table 2* for [M(ampa)] refer to the reactions of the basic ligand-form (*Eqn. 21*),

$$M^{2+} + H_2NCH_2PO_3^{2-} \rightleftharpoons H_2NCH_2PO_3 \cdot M \qquad (21a)$$

$$K^M_{open} = [H_2NCH_2PO_3 \cdot M]/([M^{2+}][H_2NCH_2PO_3^{2-}]) \qquad (21b)$$

which is perhaps followed by chelation of the open form:

$$H_2NCH_2PO_3 \cdot M \rightleftharpoons H_2\overline{NCH_2PO_3 \cdot M} \qquad (22a)$$

$$K^*_{\mathrm{I}} = [H_2\overline{NCH_2PO_3 \cdot M}]/[H_2NCH_2PO_3 \cdot M] \qquad (22b)$$

The overall observed stability constant is then given by *Eqn. 23*:

$$K^M_{[M(ampa)]} = \frac{[\text{open complex}] + [\text{chelate}]}{[M^{2+}][ampa^{2-}]} = K^M_{open}(1 + K^*_{\mathrm{I}}) \qquad (23)$$

We may estimate $\log K^M_{open}$ from the linear $\log K^M_{[M(RPO_3)]}$ *vs.* $\mathrm{p}K^H_{H(RPO_3)}$ plots, discussed in *Sect. 4* [32] (see also *Fig.*), with $\mathrm{p}k_{a/2} = 7.4$, which was deduced in *Sect. 2*, for the rare $NH_2CH_2PO_3H^-$ microform. Tabulated results for $\log K^M_{open}$ and $\log \Delta_{[M(ampa)]}$ according to *Eqn. 24*,

$$\log \Delta_{[M(ampa)]} = \log K^M_{[M(ampa)]} - \log K^M_{open} = \log(1 + K^*_{\mathrm{I}}) \qquad (24)$$

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[6]) The $\mathrm{p}K_a$ difference, $\mathrm{p}K^H_{H(NH_3)} - \mathrm{p}k_{a/1} = 9.4 - 8.1 = 1.3$, leads to a reduction of the log-stability constant of *ca.* 0.7 log units under the reasonable assumption that the slope of the corresponding $\log K$ *vs.* $\mathrm{p}K_a$ plot is in the order of 0.5.

which is analogous to *Eqn. 12*, are given in *Table 4* together with the intramolecular equilibrium constant K_I^* (*Eqn. 22*) and the percentages of the chelated isomers (*Eqn. 15*).

Table 4. *Comparison of the Measured Stability,* $K^M_{[M(ampa)]}$ *(Eqn. 23), of the [M(ampa)] Complexes*a) *with the Calculated Stability,* K^M_{open} *(Eqn. 21b), for an Isomer with a Sole* M^{2+}*-Phosphonate Coordination (Eqn. 21a)*b)*, and Extent of the Intramolecular Chelate Formation (Eqn. 22a) in the [M(ampa)] Complexes* (I = 0.1M, $NaNO_3$; 25°)

M^{2+}	$\log K^M_{[M(ampa)]}$ (*Eqns. 9* and *23*)a)	$\log K^M_{open}$ (*Eqn. 21*)b)	$\log \Delta_{[M(ampa)]}$ (*Eqns. 12* and *24*)	K_I^* (*Eqn. 22*)	% $[M(ampa)]_{chelate}$ (*Eqns. 15* and *22a*)
Mg^{2+}	1.94 ± 0.06	1.81 ± 0.03	0.13 ± 0.07	0.35 ± 0.21	26 ± 11
Ca^{2+}	1.62 ± 0.04	1.61 ± 0.05	0.01 ± 0.06	~0	~0
Sr^{2+}	1.34 ± 0.04	1.34 ± 0.04	0.00 ± 0.06	~0	~0
Ba^{2+}	1.17 ± 0.05	1.27 ± 0.04	−0.10 ± 0.06	~0	~0
Mn^{2+}	3.62 ± 0.08	2.44 ± 0.05	1.18 ± 0.09	14.1 ± 3.3	93.4 ± 1.4
Co^{2+}	4.58 ± 0.04	2.20 ± 0.06	2.38 ± 0.07	239 ± 40	99.6 ± 0.1
Ni^{2+}	5.30 ± 0.04	2.24 ± 0.05	3.06 ± 0.06	1150 ± 170	99.9 ± 0.1
Cu^{2+}	8.09 ± 0.02	3.43 ± 0.06	4.66 ± 0.06	45700 ± 6700	100 ± 0.01
$Cu(bpy)^{2+}$	7.52 ± 0.02	3.45 ± 0.07	4.07 ± 0.07	11700 ± 2000	99.99 ± 0.01
$Cu(phen)^{2+}$	7.60 ± 0.04	3.46 ± 0.06	4.14 ± 0.07	13800 ± 2300	99.99 ± 0.01
Zn^{2+}	4.91 ± 0.07	2.54 ± 0.06	2.37 ± 0.09	233 ± 50	99.6 ± 0.1
Cd^{2+}	5.14 ± 0.06	2.83 ± 0.05	2.31 ± 0.08	203 ± 37	99.5 ± 0.1

a) These values are from *Column 6* of *Table 1*; they were determined by potentiometric pH titrations in aqueous solutions; see also *Footnote b* of *Table 1*.

b) Calculated with $pk_{a/2}$ = 7.4 (*Sect. 2*) and the reference-line equations of Table 5 in [32] and Table 5 in [39]; otherwise also here *Footnote c* of *Table 2* applies.

For the four alkaline-earth-metal ions in *Table 4*, the values of $\log \Delta_{[M(ampa)]}$ in *Column 4* average to zero indicating that no significant amounts of chelates occur. Indeed, this conclusion is certainly correct for the [M(ampa)] complexes of Ca^{2+}, Sr^{2+}, and Ba^{2+}. If the value of $\log \Delta_{[Mg(ampa)]} = 0.13$ for [Mg(ampa)] is real (*Column 4* of *Table 4*), then there is a formation degree of *ca.* 25% for the chelate (*Equilibrium 22a*). The uncertainty originates here mainly from the estimate of $pk_{a/2} = 7.4$ (*Sect. 2*) which leads to a larger error for the value of $\log K^M_{open}$ than that given in *Column 3* of *Table 4*. However, if one assumes for this micro-acidity constant the generous error limit of ±0.3 pK units, one obtains for [Mg(ampa)] an upper limit of 35 (±10)% ($\log \Delta_{[Mg(ampa)]} = 0.19 \pm 0.07$) and a lower limit of 15 (±13)% ($\log \Delta_{[Mg(ampa)]} = 0.07 \pm 0.07$) for chelate formation. This then indicates that, with [Mg(ampa)], some chelate is formed.

On the other hand, all the transition-metal ions exhibit significant amounts of chelated forms in their [M(ampa)] complexes, these being 93% for Mn^{2+} and > 99% for all the others (*Table 4*). It may be emphasized that, for these metal ions, the uncertainty in $pk_{a/2} = 7.4$ is irrelevant; a change of ±0.3 pK units alters the formation degree of the [Mn(ampa)] chelate only by ±1%. Thus, for all the transition elements, *Equilibrium 22a* is certainly far on its right side.

In this context, it is interesting to replace the amino substituent in the phosphonate ligand by an OH group; *i.e.*, to compare the metal-ion binding properties of $H_2NCH_2PO_3^{2-}$ with those of $HOCH_2PO_3^{2-}$. (Hydroxymethyl)phosphonate ($HOMP^{2-}$) has been studied ($pK^H_{H(HOMP)} = 6.97 \pm 0.01$ for $H(HOMP)^-$) [43], and the stability constants of its Ca^{2+} ($\log K^{Ca}_{[Ca(HOMP)]} = 1.68 \pm 0.01$) and Cu^{2+} complexes ($\log K^{Cu}_{[Cu(HOMP)]} = 3.53 \pm 0.05$) have been determined in aqueous solution at 25° and I = 0.1M (KNO_3) [43]. Use of these equilibrium constants and application of the $K^{M}_{[M(RPO3)]}$ *vs.* $pK^{H}_{H(RPO3)}$ plots [32] for the calculation of the log-stability constants for the open isomer with a sole phosphonate metal-ion coordination gives a formation degree of the chelate in the Ca^{2+} system of $26 \pm 9\%$ ($\log \Delta_{[Ca(HOMP)]} = 0.13 \pm 0.05$; $K_I = 0.35 \pm 0.16$) and in the Cu^{2+} system of $50 \pm 9\%$ ($\log \Delta_{[Cu(HOMP)]} = 0.30 \pm 0.08$; $K_I = 1.00 \pm 0.37$). This result together with those given in *Table 4* confirm the well known experiences *i*) that alkaline-earth ions have a higher affinity to O binding sites than to N sites, and also *ii*) that the stability differences between complexes formed with these ions on the one hand and 3d transition ions on the other are much less significant with O donors than with N donors [40].

10. *Some Considerations on the Stability of the Ternary [Cu(Arm)(RPO₃)] Complexes.* Some further insights into the properties of pfa and ampa as ligands may be gained by considering the stabilities of the various mixed ligand complexes listed in *Table 1*. The relative stability of ternary complexes toward their binary parent complexes is best quantified by considering *Equilibrium 25* [44] [45]:

$$[Cu(Arm)]^{2+} + [Cu(RPO_3)] \rightleftharpoons [Cu(Arm)(RPO_3)] + Cu^{2+} \tag{25}$$

Both sides of this equilibrium contain species of the same charge type, minimizing any electrostatic contribution to the corresponding equilibrium constant, which is defined by *Eqn. 26* and calculated with *Eqn. 27*:

$$10^{\Delta \log K_{Cu}} = \frac{[[Cu(Arm)(RPO_3)]][Cu^{2+}]}{[[Cu(Arm)]^{2+}][[Cu(RPO_3)]]} \tag{26}$$

$$\begin{aligned}\Delta \log K_{Cu} &= \log K^{[Cu(Arm)]}_{[Cu(Arm)(RPO3)]} - \log K^{Cu}_{[Cu(RPO3)]} \\ &= \log K^{[Cu(RPO3)]}_{[Cu(RPO3)(Arm)]} - \log K^{Cu}_{[Cu(Arm)]}\end{aligned} \tag{27}$$

In general, the position of *Equilibrium 25* is expected [44–46] to lie on the left side with negative values for $\Delta \log K_{Cu}$ due to the general rule that $K^{M}_{[ML]} > K^{M}_{[ML2]}$ [23]. Indeed, statistical considerations for $\Delta \log K_{Cu}$ assuming a tetragonal or *Jahn-Teller* distorted octahedral coordination sphere for Cu^{2+} lead for the binding of a bidentate and a monodentate ligand to $\Delta \log K_{Cu/statis./2,1} \approx -0.5$ [47] and for the coordination of two bidentate ligands to $\Delta \log K_{Cu/statis./2,2} \approx -0.9$ [44].

There is now a long-standing experience that enhanced complex stabilities and ligand-discriminations are observed for mixed ligand complexes containing a divalent metal ion of the second half of the 3d series and, aside from a further ligand, a heteroaromatic N base (Arm) such as 2,2-bipyridyl [44–46] [48], 1,10-phenanthroline [49], or imidazole [50], its π-accepting properties being crucial [45] [51]. This M^{2+}/Arm combination favors O over N donors with regard to the second ligand to be bound [44–46] [48] [50]. For example, the $\Delta \log K_{Cu}$ values for the ternary systems bpy/Cu^{2+}/oxalate, bpy/Cu^{2+}/glycinate, and bpy/Cu^{2+}/ethylene-1,2-diamine are *ca.* +0.7, −0.35, and −1.29, respectively [52]; *i.e.*, with oxalate, *Equilibrium 25* is displaced to the right, oxalate having a larger affinity for $[Cu(bpy)]^{2+}$ than for Cu^{2+}_{aq} (see *Eqn. 27*), while with ethylene-1,2-diamine it is far on the left side.

With these summarized previous observations in mind, it is revealing to view the results listed in *Table 5*. First, it needs to be emphasized that the $[Cu(Arm)]^{2+}$ complexes with the bidentate $Hpfa^{2-}$ (see also the last paragraph in *Sect. 5*) and pfa^{3-} ligands

(*Eqn. 16*; *cf. Sect. 5*) are by *ca.* 1.1 log units ($\approx 0.2 - (-0.9)$) more stable than statistically expected, if the equilibrium constant $\Delta \log K_{Cu}$ (*Eqn. 26*) is considered. *Equilibrium 25* is for all four cases (No. *1* and *2* in *Table 5*) clearly on the side of the mixed ligand complex, and there is no significant difference between the complexes containing bpy (series *a* in *Table 5*) and those containing phen (series *b*). The fact, that the $\Delta \log K_{Cu}$ values are *positive* proves that $Hpfa^{2-}$ and pfa^{3-} are typical O donors which are well suited to participate in mixed-ligand complex formation. With regard to biological systems, this is an important observation.

Table 5. *Comparison of the Relative Stabilities of Some Mixed Ligand Complexes in Analogy to* Equilibrium 25 *for Aqueous Solutions* ($I = 0.1$ M, $NaNO_3$; 25°)

No.	Equilibrium	$\Delta \log K_{Cu}$ [a])
1a	$[Cu(bpy)]^{2+} + [Cu(Hpfa)] \rightleftharpoons [Cu(bpy)(Hpfa)] + Cu^{2+}$	0.28 ± 0.18
1b	$[Cu(phen)]^{2+} + [Cu(Hpfa)] \rightleftharpoons [Cu(phen)(Hpfa)] + Cu^{2+}$	0.33 ± 0.24
2a	$[Cu(bpy)]^{2+} + [Cu(pfa)]^{-} \rightleftharpoons [Cu(bpy)(pfa)]^{-} + Cu^{2+}$	0.16 ± 0.08
2b	$[Cu(phen)]^{2+} + [Cu(pfa)]^{-} \rightleftharpoons [Cu(phen)(pfa)]^{-} + Cu^{2+}$	0.21 ± 0.08
3a	$[Cu(bpy)]^{2+} + [Cu(Hampa)]^{+} \rightleftharpoons [Cu(bpy)(Hampa)]^{+} + Cu^{2+}$	-0.22 ± 0.07
3b	$[Cu(phen)]^{2+} + [Cu(Hampa)]^{+} \rightleftharpoons [Cu(phen)(Hampa)]^{+} + Cu^{2+}$	-0.24 ± 0.07
4a	$[Cu(bpy)]^{2+} + [Cu(ampa)] \rightleftharpoons [Cu(bpy)(ampa)] + Cu^{2+}$	-0.57 ± 0.03
4b	$[Cu(phen)]^{2+} + [Cu(ampa)] \rightleftharpoons [Cu(phen)(ampa)] + Cu^{2+}$	-0.49 ± 0.04

[a]) Calculated according to *Eqn. 27* with the stability constants listed in *Table 1*. The error limits (3σ) were calculated according to the error propagation after *Gauss* by using the error limits determined for the stability constants of the binary and ternary complexes.

The ternary [Cu(Arm)(ampa)] complexes with the bidentate $H_2NCH_2PO_3^{2-}$ are by *ca.* 0.4 log units ($\approx -0.5 - (-0.9)$) more stable than is expected on the statistical basis for $\Delta \log K_{Cu/statist./2,2}$; however, the $\Delta \log K_{Cu}$ values for these equilibria (No. *4* in *Table 5*) are *negative*, and with *ca.* −0.5 log units in the order expected for binding of bidentate N,O donors. For the Cu^{2+}/Arm/$Hampa^{-}$ systems (No. *3*) it is interesting to observe that the $\Delta \log K_{Cu}$ values are also negative yet in addition about half-way between those observed for the pure O donor pfa (No. *1* and *2*) and the N,O chelator $ampa^{2-}$ (No. *4*); this finding agrees excellently with the conclusion (see *Sect. 7*) that only about half of the $[Cu(Arm)(Hampa)]^{+}$ species are N,O-chelated (*Eqn. 18*), whereas in the other half $[Cu(Arm)]^{2+}$ is only O-, *i.e.*, phosponate-coordinated.

Conclusions. – In comparing the two ligands pfa and ampa, we find for both, the [ML] and [M(HL)] stability constants in *Table 1*, except for the [ML] complex of Cu^{2+}, higher numerical values for the pfa complexes. These differences may become even larger in neutral and acidic solutions where metal ions face greater difficulties in competing with protons [40] for sites on the more basic ampa. Below a pH corresponding to the $pK^{H}_{[M(HL)]}$ values in *Columns 4* and *7* of *Table 1* complexes exist mainly in the [M(HL)] form, and only above this pH does the [ML] form dominate. Thus, while neutral solutions of pfa contain the strong [ML] complex, for ampa, this complex does not dominate for the alkaline-earth-metal ions until pH > 9.3.

For pfa, both [M(HL)] and [ML] complexes form five-membered chelate rings, with the exception of [Cu(Hpfa)] for which in equilibrium also some phosphonate-Cu^{2+}-only binding occurs, the proton being at the carboxylate end (see last paragraph in *Sect. 5*).

However, in general the phosphonate group of the [M(HL)] chelates interacts with both, a proton and a metal ion. All the $[M(pfa)]^-$ complexes are more stable than those of oxalate, $[M(C_2O_4)]$ [42], but the latter ligand is also considerably less basic.

The five-member-chelator, pfa^{3-}, may also be considered an analogue of pyrophosphate, $P_2O_7^{4-}$ (= PP_i), which forms a six-membered chelate ring with metal ions. Stability constants are larger for PP_i [31] than for pfa, but as PP_i contains a more basic group with $pK^H_{H(P_2O_7)} = 8.4$ [31], in neutral solutions, at equal concentrations, pfa^{3-} effectively competes with $P_2O_7^{4-}$ for most metal ions.

For ampa in the [M(HL)] complexes, only Cu^{2+} forms a significant amount of five-membered ring chelates (*cf. Sect. 7*), the other metal ions interact predominantly only with the phosphonate group. Upon deprotonation of the ammonium group to give [ML] complexes, the alkaline-earth-metal ions still mainly interact only with the phosphonate group (Mg^{2+} appears to form a low fraction of chelates; *cf. Sect. 9*), while transition-metal ions form chelates to more than 93%. These chelates are comparable in stability to those of glycinate [53], which is also somewhat less basic.

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