Stability Constants of Copper(II), Zinc, Manganese(II), Calcium, and Magnesium Complexes of *N*-(Phosphonomethyl)glycine (Glyphosate)

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The stability constants of several 1:1 metal complexes of *N*-(phosphonomethyl)glycine (glyphosate) at $I=0.1$ (KNO_3) and 25 °C have been determined by potentiometric pH titration. The following values for log K_{ML} were found: Cu(II) 11.92, Zn 8.4, Mn(II) 5.53, Ca 3.25, and Mg 3.25. The pK values of glyphosate are: p$K_1=2.27\pm0.02$, p$K_2=5.57\pm0.004$, and p$K_3=10.25\pm0.02$; these are mixed constants, valid for $I=0.1$ (KNO_3).

A large number of important chelating agents belong to the group of aminopolyacids; two of the best known are nitrilotriacetic acid and ethylenediaminetetraacetic acid, and others contain methylphosphonic instead of one or more of the acetic acid groups. Numerous determinations of stability constants of metal complexes with these ligands have been carried out;[1] one interesting exception, for which no data are available, is *N*-(phosphonomethyl)glycine (trivial name: glyphosate), $^-HO_3PCH_2NH_2^+CH_2COOH$, marketed since 1971 as a herbicide.[2] A study of the chelating properties of this substance is justified not only by its chemical interest, but also because this information is essential for understanding the behaviour of the herbicide in soil and plants.

In order to ensure consistent results, we have determined the dissociation constants of glyphosate at $I=0.1$ (KNO_3), the medium used for the determinations of stability constants of the metal complexes.

EXPERIMENTAL

Apparatus. pH was measured at 25 °C with a Radiometer digital pH meter model 64 using a combined glass/calomel electrode type C.

Materials. All solutions were prepared with boiled demineralized water. Standard buffers for calibration were prepared from analytical grade chemicals. Potassium hydroxide, 0.05 M, was prepared from May and Baker concentrated volumetric solution. Metal nitrate stock solutions, 0.1 M, were standardized by complexometric titration with EDTA, using standard methods.[5]

Glyphosate was prepared from the commercial product Roundup® by ion-exchange chromatography on Dowex 1. Formic acid, 2 M, was used for elution, and the crude product was recrystallized three times from water/ethanol.

All solutions, except standard buffers, were adjusted to an ionic strength of 0.1 with KNO_3.

pH calibrations. Three standard buffers were used for calibrations: phthalate (pH=4.008), phosphate (pH=7.413), and borax (pH=9.180). Calibration was rechecked after each run of a titration curve; no deviation ever exceeded 0.005 pH units.

In order to be able to calculate $[H^+]$ and $[OH^-]$ from pH readings, we measured pH in a series of HNO_3 solutions from 1 mM to 10 mM and in a series of KOH solutions from 0.25 mM to 1.25 mM. We found pH $+\log[H^+]=0.067\pm0.005$ and pH $-\log[OH^-]=13.76\pm0.06$.

Recording a titration curve. The initial volume of the titrand solution was 30.0 cm³, and it was 2.67 mM with respect to glyphosate. Metal concentration was equal to ligand concentration. A total of 5 cm³ 0.05 M KOH was added, and about 50 points of the titration curve were recorded. Stirring was stopped at each reading, to avoid a streaming potential at the reference electrode.

CALCULATIONS

Titration curves without metal. The degree of neutralization, n, is given by

$$n = \frac{B - A + [H^+] - [OH^-]}{C_L} \quad (1)$$

where C_L is the total molar concentration of glyphosate, A is the excess of strong acid added prior to titration ($A = 0$ in the actual case), and B is the amount of strong base added, in equiv./dm^3. $[H^+]$ and $[OH^-]$ were calculated from measured pH values and the results of measurements on dilute nitric acid and potassium hydroxide.

Preliminary measurements showed that, although glyphosate should be able to take up a proton to form a positive ion, the corresponding pK value is negative. Hence we disregard this possibility in the sequel.

As the pK values are widely spaced, they can be calculated simply as

$$pK_1 = pH + \log \frac{1-n}{n} \quad 0 < n < 1 \quad (2)$$

$$pK_2 = pH + \log \frac{2-n}{n-1} \quad 1 < n < 2 \quad (3)$$

$$pK_3 = pH + \log \frac{3-n}{n-2} \quad 2 < n < 3 \quad (4)$$

These values are so-called mixed constants (hydrogen ion activity, but concentration of acid and base).

Titration curves with metal. The method of Chaberek and Martell[6] had to be modified to take into account the acid-base properties of the complex. The calculations were carried out as follows:

The total concentrations of metal (M) and of ligand (L) are given by

$$C_M = [M^{2+}] + [ML^-] + [MLH] + [MLOH^{2-}] \quad (5)$$

$$C_L = [L^{3-}] + [HL^{2-}] + [H_2L^-] + [H_3L] + [ML^-] + [MLH] + [MLOH^{2-}] \quad (6)$$

where $MLOH^{2-}$ denotes a complex ion, where a proton has been removed from one of the coordinated water molecules. As in all experiments $C_M = C_L$, we have from (5) and (6)

$$[L^{3-}] + [HL^{2-}] + [H_2L^-] + [H_3L] = [M^{2+}]$$
$$[L^{3-}]\alpha_{L(H)} = [M^{2+}] \quad (7)$$

with the side-reaction coefficient $\alpha_{L(H)}$ given by

$$\alpha_{L(H)} = 1 + a(H^+)/K_3 + a(H^+)^2/K_2K_3 + a(H^+)^3/K_1K_2K_3 \quad (8)$$

where $a(H^+)$ is defined as 10^{-pH}. A further equation is the proton-balance equation, which yields

$$B + [H^+] - [OH^-] = [H_2L^-] + 2[HL^{2-}] + 3[L^{3-}] + 3[ML^-] + 2[MLH] + 4[MLOH^{2-}] \quad (9)$$

If we multiply (6) by 3 and subtract (9), we find

$$[L^{3-}] = \frac{3C_L - B - [H^+] + [OH^-] - [MLH] + [MLOH^{2-}]}{a(H^+)\alpha'_{L(H)}} \quad (10)$$

where

$$\alpha'_{L(H)} = 1/K_3 + 2a(H^+)/K_2K_3 + 3a(H^+)^2/K_1K_2K_3 \quad (11)$$

$[L^{3-}]$ may be calculated directly from (10) when $[MLH] - [MLOH^{2-}]$ is negligible; otherwise, we have

$$\frac{[ML^-]}{[M^{2+}][L^{3-}]} = K_{ML} \quad (12)$$

$$\frac{[MLH]}{a(H^+)[ML^-]} = K_{H(ML)} \quad (13)$$

$$\frac{[MLOH^{2-}]}{a(OH^-)[ML^-]} = K_{OH(ML)} \quad (14)$$

which yields, using (7)

$$[MLH] - [MLOH^{2-}] = K_{ML}\alpha_{L(H)}\alpha'_{ML(H)}a(H^+)[L^{3-}]^2 \quad (15)$$

where

$$\alpha'_{ML(H)} = K_{H(ML)} - K_{OH(ML)}Kw/a(H^+)^2 \quad (16)$$

$[Kw = a(H^+)a(OH^-)]$.

When (15) is inserted in (10), the following quadratic equation for $[L^{3-}]$ results:

$$K_{ML}\alpha_{L(H)}\alpha'_{ML(H)}a(H^+)[L^{3-}]^2+a(H^+)\alpha'_{L(H)}[L^{3-}]$$
$$-3C_L+B+[H^+]-[OH^-]=0 \qquad (17)$$

As the coefficient of $[L^{3-}]^2$ is given in terms of the unknown stability constants, the solution must be found by iteration.

When $[L^{3-}]$ has been found, $[M^{2+}]$ is calculated from (7) and $[ML^-]$ from

$$[ML^-]=(C_M-[M^{2+}])/\alpha_{ML(H)} \qquad (18)$$

where

$$\alpha_{ML(H)}=1+K_{H(ML)}a(H^+)+K_{OH(ML)}a(OH^-) \qquad (19)$$

is the side-reaction coefficient of the complex.

Finally, K_{ML} is calculated, and its logarithm plotted against pH. Different values of $K_{H(ML)}$ and $K_{OH(ML)}$ are tried, in order to find those giving the least possible variation of log K_{ML} with pH.

In the pH interval where nearly all the ligand is bound in complex, it is sometimes possible to calculate $K_{H(ML)}$ and $K_{OH(ML)}$ in the same manner as for the free ligand [eqns. (1−4)]. In this case, we may obtain $[L^{3-}]$, and hence K_{ML}, directly.

RESULTS

Five titration curves were recorded for the free ligand and two for each of the metal complexes
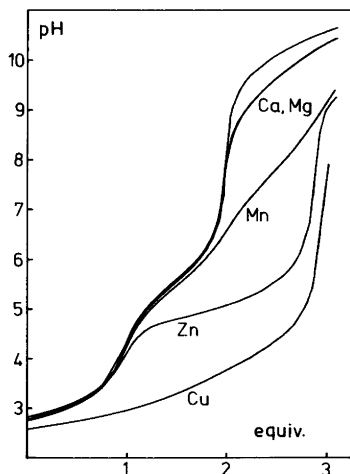

Fig. 1. Titration curves of glyphosate (upper curve) and of five solutions with metal. The abscissa is the number of equivalents of KOH added per mol of glyphosate.

Table 1. Dissociation constants of glyphosate at $I=0.1$ (KNO$_3$) and 25 °C.

Expt. No.	pK_1	pK_2	pK_3
1	2.22	5.56	−
2	2.26	5.58	10.29
3	2.27	5.58	10.28
4	2.29	5.58	10.21
5	2.31	5.58	10.22
Mean	2.27(2)	5.576(4)	10.25(2)

The numbers in parentheses give the standard deviation on the last digit shown.

(three for Mg). The shapes of the curves are shown in Fig. 1, the calculated dissociation constants are given in Table 1, and the stability constants of metal complexes are given in Table 2.

DISCUSSION

The results for the three dissociation constants of glyphosate agree qualitatively with previously reported values.[3,4] In order to test the quantitative agreement with Wauchope's results,[4] we may calculate the thermodynamic dissociation constants with the aid of the Debye-Hückel equation for activity coefficients:

$$\log \gamma_i = -\frac{Az_i^2\sqrt{I}}{1+Ba\sqrt{I}} \qquad (20)$$

Wauchope takes $a=5.5$ Å for H$_2$L$^-$, 6.0 Å for HL^{2-}, and 6.5 Å for L^{3-}. With these values, we have for the thermodynamic constants:

	I	pK_1	pK_2	pK_3
Wauchope[4]	~0.05	2.32	5.86	10.86
Present study	0.1	2.37	5.872	10.72

The agreement is satisfactory in view of the experimental uncertainty in glass electrode measurements at low and high pH values and the limitations of the Debye-Hückel law.

The values of the stability constants of the metal complexes place the metals in the order to be expected from a knowledge of the stabilities of complexes with related ligands.[1] It is particularly interesting to compare the chelating power of glyphosate with that of iminodiacetic (IDA) acid

Table 2. Stability constants K_{ML}, $K_{H(ML)}$, and $K_{OH(ML)}$ of the metal complexes of glyphosate at $I = 0.1$ (KNO$_3$) and 25 °C.

Metal	Expt. No.	log K_{ML}	log $K_{H(ML)}$	log $K_{OH(ML)}$
Cu	1	11.93	4.05	—
	2	11.91	4.05	—
	Mean	11.92	4.05	—
Cu	1	11.71	4.33	—
(2nd method)	2	11.71	4.36	—
	Mean	11.71	4.35	—
Zn	1	8.4	—	—
	2	8.4	—	—
	Mean	8.4	—	—
Mn	1	5.55	6.93	4.37
	2	5.52	6.91	4.23
	Mean	5.53	6.92	4.30
Ca	1	3.25	—	2.7
	2	3.25	—	2.9
	Mean	3.25	—	2.8
Mg	1	3.25	—	2.7
	2	3.24	—	.2.9
	3	3.25	—	2.9
	Mean	3.25	—	2.8

Table 3. Stability constants (log K_{ML}) of metal complexes of iminodiacetic acid and glyphosate at $I = 0.1$ (KNO$_3$) and 25 °C.

	Cu	Zn	Mn	Ca	Mg
IDA	10.55[6]	7.0[6]	—	2.59[a]	2.94[a]
Glyphosate	11.92	8.4	5.53	3.25	3.25

[a] 20 °C.[1]

(Table 3). We see that the substitution of a phosphonic for a carboxylic group increases the chelating power. This effect does not, however, appear to represent a general trend for the aminopolyacids.[7]

Glyphosate is probably a tridentate ligand. At present, no precise statements can be made with respect to the detailed structure of the complexes or to the interpretation of the values of $K_{H(ML)}$ and $K_{OH(ML)}$.

The pH interval useful for calculation of stability constants from pH titration curves is the one in which the concentrations of free and complexed metal are comparable. This interval should not include any of the pK values of the ligand, because of the strong buffering action of the ligand around this value, nor should it lie above the highest pK value of the ligand, as in this region pH will hardly be affected by complexing.

Fig. 2 shows the variation of the fraction of metal complexed as a function of pH. We see that the conditions for a precise determination of stability
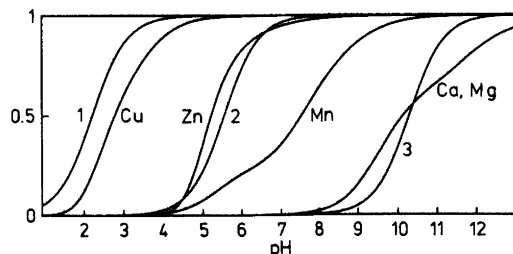

Fig. 2. Fractions of metal complexed as functions of pH at $C_M = C_L = 0.0025$ M and $I = 0.1$ (KNO$_3$). The curves marked 1, 2, and 3 represent the base fractions at the three dissociation steps of glyphosate.

constants are fulfilled for Cu and Mn, but not for Zn, Ca, and Mg. Indeed, this was what we experienced when performing the calculations. Hence, the situation calls for a check of the results by other methods, even for Cu due to the discrepancy between results of the two methods of calculation. Experiments with ion-selective electrodes, polarography, and gel chromatography are in progress at our laboratory.

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