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Potentiometric Investigation on the System Lead(II)−Arginine

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ABSTRACT: Arginine is an amino acid playing an important role in human physiology because it is involved in the production of NO, which is a vasodilatator and is the precursor of urea. Although the effect of arginine on tissue lead(II) mobilization in plumbism has been studied, its property as a ligand toward cations has not been adequately investigated. In this paper the behavior of arginine as a ligand toward lead(II) is studied in two different ionic media: 1.00 mol dm^{-3} NaCl and 1.00 mol dm^{-3} $NaClO_4$. Galvanic cells involving a lead amalgam and glass electrodes were employed to study the behavior of L-arginine as a ligand toward lead(II). The experimental data, obtained at 25 °C and in 1.00 mol dm^{-3} NaCl are explained by assuming the presence of the species PbH_2L, PbHL, and PbH_2L_2 with the relative stability constants. The data obtained in $NaClO_4$ are explained by assuming the formation of the species PbH_2L, PbHL, PbH_2L_2, and PbL with the relative stability constants. In all of the above species, the symbol L indicates arginine completely deprotonated. The role played by the ionic medium is discussed.

INTRODUCTION

Among amino acids, arginine plays an important role from a physiological point of view because, as a precursor of urea, it is found to be involved in the production of NO, which seems to be a vein dilatator, and the presence of symmetric and asymmetric dimethylarginine, together with homocysteine, has been the subject of important studies correlated to disease evaluation.[1−3]

Amino acids, in particular arginine and several cations are present together in the human body. The behavior of arginine toward cations represents an important and useful subject of research.

Oskarsson et al.,[4] studying behavioral and neurochemical effects after combined perinatal treatment of rats with lead and disulfiran, observed an increase of arginine corresponding to the treatment with lead and disulfiram. Ding et al.[5] investigated the influence of infusions of L-arginine on the hypertension induced by lead. Malvezzi et al.[6] studied the effect of L-arginine and other compounds on tissue lead mobilization and blood pressure levels in plumbism.

In other fields, Wilke[7] evaluated the effects of soil properties on the microbial toxicity of lead.

However, in the literature, few investigations are reported on the properties of arginine as a ligand.[8−10]

L-Arginine, (*S*)-2-amino-5-guanidinopentanoic acid, also identified as α-amine-δ-guanidinvalerianic acid, in the deprotonated form (L = $C_6H_{13}N_4O_2^-$) has one carboxylic, one amino, and a guanidine group.

Recently, all three-protonation constants of arginine were accurately determined at 25 °C and 1.00 mol dm^{-3} NaCl and 1.00 mol dm^{-3} $NaClO_4$, as ionic media, by means of electromotive force (emf) measurements by using a hydrogen electrode. In the same paper, silver and glass electrodes were employed to study the behavior of L-arginine as a ligand toward silver(I).[11]

Knowledge of the protolytic equilibria of arginine and the complex formation between arginine and silver(I) at 25 °C and 1.00 mol dm^{-3} NaCl and 1.00 mol dm^{-3} $NaClO_4$, as ionic media, was necessary to investigate the behavior of arginine toward calcium(II) and magnesium(II) in the same experimental conditions.[12]

Few papers studying arginine as a ligand for heavy and in particular toxic metal cations can be found.[8−10]

The most representative research on complex formation between lead(II) and arginine was performed by Pelletier.[13] He explained his potentiometric data (glass electrode at 25 °C) by assuming the presence of two complexes with constants log $K_1 = 3.27$ and log $K_2 = 3.18$, but the ionic strength used to obtain the potentiometric data was not given.

More recently, Clarke and Martell[14] in studying the behavior of arginine as a ligand toward several cations, proposed the formation of complexes with ratios 1:1 and 1:2 between the cation and ligand. However, these researchers did not investigate the complex formation between lead(II) and arginine.

In all literature research relative to arginine, by taking into account the protolytic properties, the ligand was considered acting as HL.

The aim of this investigation was to study the equilibria occurring between lead(II) and arginine in a wide reagent concentration range. In this paper, arginine in the deprotonated form (L = $C_6H_{13}N_4O_2^-$), protons and lead(II) are considered independent reagents. The target is to find the ratio of reagents, to verify what species with different ratios between lead(II) and arginine or polynuclear complexes, with participation of protons, could be formed.

A wide variation of reagent concentration in the investigated solutions provokes a remarkable variation of the activity coefficients of the reagents. To minimize the activity coefficient variation in spite of the change of the concentration of the reagents, the constant medium method proposed by Biedermann and Sillèn[15] was adopted. The investigation proposed here was carried out in two ionic media, i.e., 1.00 mol dm^{-3} NaCl and 1.00 mol dm^{-3} $NaClO_4$. The adoption of a constant ionic medium allows substitution until 15% the ions of the ionic medium with those of the reagents without any appreciable variation of the activity coefficients. On this basis, concentrations can substitute activities in all calculations.

Furthermore, it will be interesting to compare the behavior of the reagents in the two different ionic media.

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EXPERIMENTAL PART

2.1. Method of Investigation. In aqueous solutions, where lead(II) and arginine are present as independent reagents, without any preliminary hypothesis, the general equilibrium can be written

$$q\mathrm{Pb} + p\mathrm{H} + rL \leftrightarrow \mathrm{Pb}q\mathrm{H}pLr \qquad (1)$$

In eq 1, charges are omitted, $q \geq 1$; $p \leq \geq 0$ and $r \geq 1$. When $q > 1$, polynuclear complexes in lead(II) are formed, when $p \neq 0$, complexes with participation of hydrogen ions are formed and, in particular, $p > 0$ indicates the formation of protonated species, whereas $p < 0$ corresponds to species with loss of protons. The free concentrations of the reagents are indicated by small c, while capital C_x indicates the total concentration of the species x.

In the constant ionic medium method, concentrations can substitute activities, equilibrium 1 can be defined by the constant:

$$\beta_{q,p,r} = c_{\mathrm{Pb}q\mathrm{H}pLr}({c_{\mathrm{Pb}}}^{q}{c_{\mathrm{H}}}^{p}{c_{L}}^{r})^{-1} \qquad (2)$$

The main target of this investigation is to determine, over a wide reagent concentration range, the prevailing values of q, p, and r and the corresponding values of the constants $\beta_{q,p,r}$.

The electromotive force (e.mf) of the following galvanic cells was measured:

$$\mathrm{RE/Solution\ S/GE} \qquad (\mathrm{I})$$

$$\mathrm{Pb(Hg)/Solution\ S/RE} \qquad (\mathrm{II})$$

where GE, is a glass electrode, RE is a reference electrode, and Pb(Hg) is a lead amalgam electrode, which will be described in the Experimental Part.

Solution S has the following general composition:

C_{H} M in H^+; C_{Pb} M in Pb^{2+}; $(1 - C_{\mathrm{H}} - 2C_{\mathrm{Pb}})$ M in Na^+, and 1 M in μ, where μ represents the anion of the ionic medium, alternatively, Cl^- or $\mathrm{ClO_4}^-$.

At 25 °C and in millivolts, in a constant ionic medium, the emf of cells I and II can be expressed, respectively, as follows:

$$E_{\mathrm{I}} = E^{\circ}{}_{\mathrm{I}} + 59.16 \log c_{\mathrm{H}} + Ej \qquad (3)$$

$$E_{\mathrm{II}} = E^{\circ}{}_{\mathrm{II}} - 29.58 \log c_{\mathrm{Pb}} - Ej \qquad (4)$$

$E^{\circ}{}_{\mathrm{I}}$ and $E^{\circ}{}_{\mathrm{II}}$, two constants, and Ej, the liquid junction potential depending only on c_{H}, were determined in the first part of each measurement, when $C_{\mathrm{Pb}} = c_{\mathrm{Pb}}$ and $C_{\mathrm{H}} = c_{\mathrm{H}}$, i.e., in the absence of arginine. In the second part of the measurement, a solution containing arginine, in the selected ionic medium, was added gradually, so that the ligand concentration increased gradually, while C_{Pb} and C_{H} were kept constant. For each addition, from the measurements of E_{I} and E_{II}, the free concentrations of lead(II) c_{Pb} and hydrogen c_{H} ion can be calculated.

According to this procedure c_{H}, c_{Pb}, experimental data and C_{Pb} and C_{H}, analytical data, are the basis of the treatment applied to find the prevailing values of q, p, and r and the relative constants.

Material and Analysis. A Sigma L-arginine p.a. reagent was twice crystallized from twice distilled water. The purified reagent was dried in oven at 120 °C until constant weight was reached. Its purity and absence of water was checked by thermogravimetric analysis.

Hydrochloric acid, sodium chloride, sodium perchlorate, perchloric acid, and sodium hydroxide were prepared and analyzed as previously[16,17] described.

Lead(II) chloride and perchlorate were prepared and analyzed as previously described.[17,18]

Lead amalgam was prepared by bringing in contact little pieces of metallic lead (Riedel-de Haën p.a.), surface cleaned immediately before use, with distilled mercury in such a ratio to obtain a lead amalgam with about 0.1% of lead. The amalgam was kept under 0.01 M HCl or $\mathrm{HClO_4}$, respectively,

Details on Experimental Apparatus. Emf measurements of cell (I) were performed with a Radiometer mod. PHM 64 pH meter equipped with a glass electrode G202B from the same firm. Those of cell (II) were performed by means of a multimeter (Keithley mod. 199 system DMM). The salt bridge was similar to that described by Forsling et al.[19]

The reference electrode, R.E. was prepared according to Brown.[20] For solutions in NaCl as ionic medium, it was R.E. = Ag, AgCl/1.00 M NaCl, saturated with AgCl/1.00 M NaCl, while when the ionic medium was $\mathrm{NaClO_4}$, it was R.E. = Ag, AgCl/0.01 M Cl^-, 0.99 M $\mathrm{ClO_4}^-$ 1.00 M Na^+/ 1.00 M $\mathrm{NaClO_4}$.

Constant values of $\mathrm{E_I}$ were obtained a few minutes after each addition, whereas it was necessary to wait 30 − 45 min to obtain constant values of $\mathrm{E_{II}}$. The values of $\mathrm{E_{II}} - \mathrm{E}^{\circ}{}_{\mathrm{II}}$ were reproducible within ±0.1 mV, those of $\mathrm{E_I} - \mathrm{E}^{\circ}{}_{\mathrm{I}}$ were reproducible within ±0.2 mV.

The agreement of data obtained from direct and back emf measurements allows the assumption that measurements were carried out in solutions at real equilibrium.

Nitrogen (99.995%) from a cylinder, further purified by passing through 10% NaOH, 10% $\mathrm{H_2SO_4}$ and the selected ionic medium, was bubbled through the solution S to eliminate oxygen and $\mathrm{CO_2}$ from the solutions.

RESULTS AND DISCUSSION

The experimental data c_{Pb} and c_{H}, obtained from the emf measurements in the range $1 \leq -\log c_{\mathrm{H}} \leq 8$ and the analytical concentrations, C_{Pb}, C_{H}, and C_{L} are the basis used to find the prevailing q, p, and r, values and the relative constants, both in NaCl and in $\mathrm{NaClO_4}$.

The general procedure to obtain the q, p, and r values and the constants started from the material balance of lead(II) so that the dependence of its total concentration on the measured variables (c_{Pb} and c_{H}) and the calculated (c_{L}) was studied. This procedure was suitable because many experimental points were obtained at different C_{Pb}, C_{H}, and C_{L} so that the presence of polynuclear complexes or complexes with participation of protons could be investigated.

3.1. Results Obtained in 1.00 M NaCl and Discussion. Experimental data were obtained for C_{H} = (0.050, 0.070, 0.100, and 0.150) mol dm^{-3} and C_{Pb} = (0.25, 0.50, 1.00, and 2.00) × 10^{-3} mol dm^{-3}. Taking into account the eq 1 and the mass action law yields the lead(II) material balance as follows:

$$C_{\mathrm{Pb}} = c_{\mathrm{Pb}} + \sum\sum\sum q\beta_{q,p,r}{c_{\mathrm{Pb}}}^{q}{c_{\mathrm{H}}}^{p}{c_{\mathrm{L}}}^{r}$$

As c_{Pb} was experimentally measured and C_{Pb} was analytically known, the following function can be calculated:

$$\begin{aligned}\eta &= \log(C_{\mathrm{Pb}}/c_{\mathrm{Pb}}) \\ &= \log(1 + \sum\sum\sum q\beta_{q,p,r}{c_{\mathrm{Pb}}}^{q-1}{c_{\mathrm{H}}}^{p}{c_{\mathrm{L}}}^{r})\end{aligned} \qquad (5)$$

In eq 5 the hydrolytic species of lead(II) can be neglected by considering the concentration range of the reagents in this work and the Carell and Olin data on lead(II) hydrolysis.[21,22]

The function η, directly obtained from experimental data, is plotted in Figure 1 versus $-\log c_{\mathrm{H}}$.

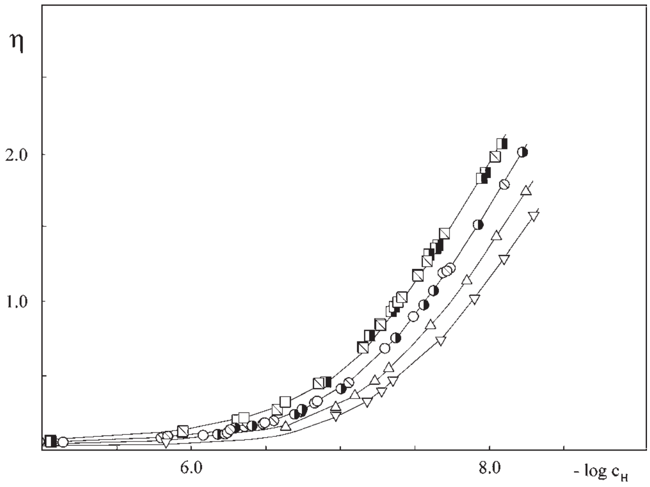

Figure 1. Dependence of η on $-\log c_H$ in NaCl. Points are plotted at $C_H = 0.150$ M and C_{Pb} = (□, ⧅, ◨ = 1, 0.5, 0.25) × 10^{-3} M; $C_H = 0.100$ M and C_{Pb} = (○, ⦶, ◑ = 2, 1, 0.5) × 10^{-3} M; $C_H = 0.070$ M and C_{Pb} = (△) 0.5 × 10^{-3} M; and $C_H = 0.050$ M and C_{Pb} = (▽) 0.25 × 10^{-3} M. At different C_{Pb} but at the same C_H, points fall on the same curve.

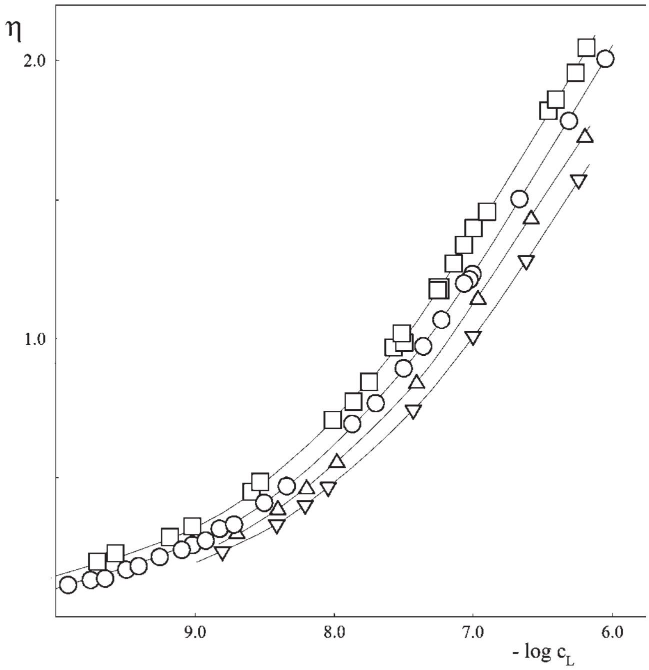

Figure 2. Points at different C_H (□ = 0.150, ○ = 0.100, △ = 0.070, and ▽ = 0.050 M) obtained in NaCl are plotted. Dependence of η on C_H is evident.

In the plot η versus $-\log c_H$, experimental points obtained at different total concentrations of lead(II), but at the same C_H, fall on the same curve independently of C_{Pb}. As η is not a function of C_{Pb}, polynuclear species of lead(II) are not present and then $q = 1$. It is necessary to study the dependence of η on the hydrogen ion concentration (value of p) and on the ligand concentration (value of r).

To find the prevailing values of p and r and the constants $\beta_{1,p,r}$, the dependence of η on the free concentration of the ligand, c_L has to be studied.

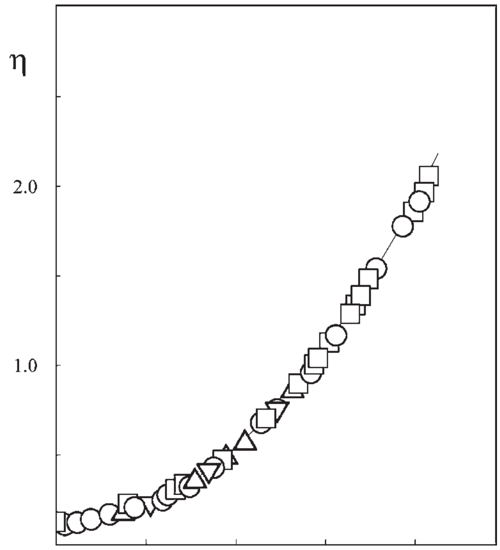

Figure 3. Dependence of η on $-\log c_{HL}$ in NaCl. In the considered $-\log c_{HL}$ range all points even at different C_H (□, ○, △, ▽ = 0.150, 0.100, 0.070, 0.050 M) fall on the same curve.

The free concentration of arginine, c_L, can be calculated from the material balance of the analytical excess of hydrogen, C_H:

$$C_H = c_H + \sum n k_n c_H{}^n c_L + \sum\sum p \beta_{1,p,r} c_{Pb} c_H{}^p c_L{}^r \qquad (6)$$

In eq 6, the k_n values are the protonation constants of arginine, determined in the same experimental conditions.[11] To calculate c_L, the last term of eq 6, relative to complexes formed with the participation of protons, can be neglected in the first approximation, because in all the investigated solutions $C_{Pb} \leq 0.02C_H$. From the same eq 6 also $c_{HL} = k_1 c_H c_L$ and $c_{H2L} = k_1 k_2 c_H{}^2 c_L$ can be calculated.

Figure 2 shows the dependence of η on $-\log c_L$. It can be observed that points obtained at different C_H fall on different curves. This evidence proves the existence of complexes with the participation of protons because η is a function of the hydrogen ion concentration. As η is an increasing function of C_H, complexes with the assumption of protons ($p \geq 0$) are formed.

The inspection of Figure 2 shows that the complex formation depends on two variables: hydrogen ion and arginine concentrations. A preliminary investigation of different ranges of concentration was selected to find the prevailing p and r values. On this basis the investigation started from the range $4 \leq -\log c_H \leq 8$, by considering that in this range the prevailing form of arginine is the protonated one, i.e., HL. The hypothesis was formulated that the ligand could act as HL. To verify the hypothesis, a graphical approach was performed in Figure 3, by plotting η versus $-\log c_{HL}$. The range $4 \leq -\log c_H \leq 8$ corresponds to the range $-\log c_{HL} \leq 4$, for the concentration of HL.

From Figure 3, it can be observed that in the range $-\log c_{HL} \leq 4$, all the points fall on the same curve independently of C_{Pb} and C_H. The hypothesis that in this range the ligand acts in the form HL is confirmed. As the slope of the trend of the points is higher than 1, the formation of PbHL and $Pb(HL)_2$ can be assumed i.e., $p = r$, which can be 1 or 2. Equation 5 in this range becomes

$$\eta = \log(1 + \beta_{1,1} c_{HL} + \beta_{1,2} c_{HL}{}^2) \qquad (7)$$

To verify this hypothesis, the graphical procedure proposed by Sillén[23] is applied.

Table 1. Species Assumed To Explain the Experimental Data for the System Lead(II)−Arginine and Values of Relative Constants at 25 °C and 1.00 M NaCl

species	graphic method	PC elaboration	proposed value
PbH_2L	0.4 (21.58) ± 0.15	0.35 (21.53) ± 0.10	0.35 (21.53) ± 0.10
PbHL	3.20 (15.55) ± 0.10	3.19 (15.54) ± 0.10	3.19 (15.54) ± 0.10
PbH_2L_2	5.75 (30.45) ± 0.10	5.78 (30.48) ± 0.10	5.78 (30.48) ± 0.10

Equation 7 can be normalized by means of the following two mathematical positions:

$$\log u = \log c_{HL} + \frac{1}{2}\log \beta_{1,2} \qquad \text{and}$$

$$\log \beta_{1,1} = \log \alpha + \frac{1}{2}\log \beta_{1,2}$$

By combining the positions with eq 7, we obtain the following family of curves:

$$Y = \log(1 + \alpha u + u^2) \tag{7N}$$

The family of the normalized curves is superimposed on the experimental points of Figure 3 and the two plots are moved parallel to the abscissa, until the best fit is reached. As in eq 7N the position and form of the curve are relative to u and α, respectively, the best fit can be obtained only for a curve with a determined value of α. In the position of best fit, $\log u = 0$, in the normalized curve, corresponds to a value of $\log c_{HL}$ in the plot of experimental data. This value of $-\log c_{HL}$ provides $^1/_2 \log \beta_{1,2}$, which, combined with the value of α, gives also $\log \beta_{1,1}$. These values can be considered a first approximation because only a part of the points was considered.

In the more acidic range ($1 \le -\log c_H \le 4$) even if η is plotted versus $-\log c_{HL}$, points fall on different curves, showing that in this range, η is still an increasing function of C_H.

It seems reasonable to suppose that the ligand can act both as HL and as H_2L.

To verify this hypothesis, in the range $1 \le -\log c_H \le 4$, the contribution of PbHL and $Pb(HL)_2$, obtained above by using the first approximation constants, is subtracted from the total concentration of lead, C_{Pb}, to have $C'_{Pb} = C_{Pb} - \beta_{1,1}c_{Pb}c_{HL} - \beta_{1,2}c_{Pb}c_{HL}{}^2$.

By supposing that the lone form under which lead(II) could be present is PbH_2L, it is possible to write $C'_{Pb} = \beta_{1,2,1}c_{Pb}c_H{}^2c_L$. The validation of this hypothesis is supported by the constancy of the following expression: $C'_{Pb}(c_{Pb}c_H{}^2c_L)^{-1}$ = constant = $\beta_{1,2,1}$. The value $\log \beta_{1,2,1} = 0.4 \pm 0.15$ is obtained.

The refined values of $\log \beta_{1,2,1}$, $\log \beta_{1,2}$, and $\log \beta_{1,1}$ are obtained from successive approximation procedure and independently by introducing experimental values and analytical concentrations of the reagents in a PC program.[24]

The refined values are collected in Table 1 and are used to calculate the curves drawn on the figures. The good agreement between the curves and experimental points supports the validity of the obtained results and the correctness of the procedure.

In all three columns of Table 1, two numerical values are collected for each species, the second of them is in parentheses. The difference between them is relative to the assumed form of arginine acting as the ligand. The bigger value is connected with eq 1, showing Pb^{2+}, H^+, and L as independent reagents. The other values are obtained by considering the ligand in the form H_2L (first line) and HL (second and third lines), respectively. In fact the difference between these values is equal to $\log k_1 = 12.35$ (second and third lines) or to $\log k_1 k_2 = 21.18$, where k_1 and k_2 are the protonation constants of arginine determined in 1.00 M NaCl.[11]

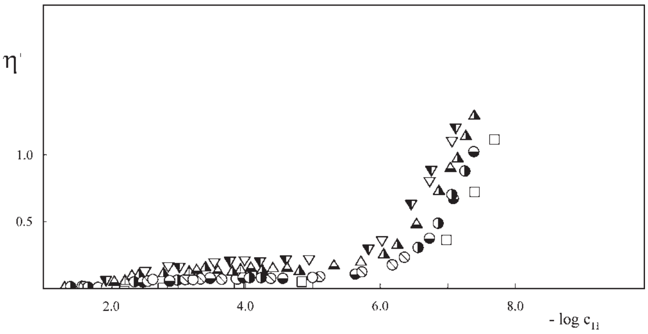

Figure 4. Dependence of η' on $-\log c_H$ in $NaClO_4$. Points are plotted at C_H = 0.150 M and C_{Pb} = (▽, ▼ = 0.25, 0.125) × 10^{-3} M; C_H = 0.100 M and C_{Pb} = (△, ◬, ◭, ▲ = 1.0, 0.5, 0.25, 0.125) × 10^{-3} M; C_H = 0.050 M and C_{Pb} = (○, ⦶, ◑, ◒ = 1.0, 0.5, 0.25, 0.125) × 10^{-3} M; and C_H = 0.025 M and C_{Pb} = (□) 0.125 × 10^{-3} M. At different C_{Pb} but at the same C_H points fall on the same curve.

The good agreement between results obtained by two different methods is evident.

3.2. Results Obtained in 1.00 M $NaClO_4$ and Discussion.

In 1.00 M $NaClO_4$ as ionic medium, experimental data are obtained for C_H = (0.050, 0.070, 0.100, and 0.150) mol dm^{-3} and C_{Pb} = (0.125, 0.25, 0.50, and 1.00) × 10^{-3} mol dm^{-3}.

The elaboration of the data to find the prevailing values of q, p, and r is similar to that applied above for the other ionic medium.

By taking into account eq 1 and the mass action law, from the lead(II) material balance, we can obtain the following function:

$$\begin{aligned}\eta' &= \log(C_{Pb}/c_{Pb}) \\ &= \log(1 + \sum\sum\sum q'\beta_{q',p',r'}c_{Pb}{}^{q'-1}c_H{}^{p'}c_L{}^{r'})\end{aligned} \tag{5bis}$$

In eq 5bis hydrolytic species of lead(II) can be neglected by considering the concentration range of the reagents in this work and the Carell and Olin investigation on lead(II) hydrolysis.[21,22]

The function η', directly obtained from experimental data, is plotted in Figure 4 versus $-\log c_H$.

In the plot η' versus $-\log c_H$, it can be observed that experimental points obtained at different total concentrations of lead(II), but at the same C_H fall on the same curve independent of the different C_{Pb}. As η' is an independent function of C_{Pb}, it is assumed that polynuclear species in lead(II) do not exist in appreciable concentrations and then $q = 1$.

To find the prevailing values of p and r and the constants $\beta_{1,p,r}$, the dependence of η' on the free concentration of the ligand, c_L is studied.

The free concentration of arginine, c_L, can be calculated from the material balance of the analytical excess of hydrogen, C_H:

$$C_H = c_H + \sum nk_n c_H{}^n c_L + \sum\sum p'\beta'_{1,p',r'}c_H{}^{p'}c_L{}^{r'} \tag{6bis}$$

In eq 6bis, the k_n values are the protonation constants of arginine, determined in the same experimental conditions.[11] To calculate c_L, the last term of eq 6bis, relative to complexes formed with the participation of protons can be neglected without loss of accuracy, because in all the investigated solutions $C_{Pb} \le 0.01C_H$. From the same eq 6bis also $c_{HL} = k_1c_Hc_L$ and $c_{H2L} = k_1k_2c_H{}^2c_L$ can be calculated.

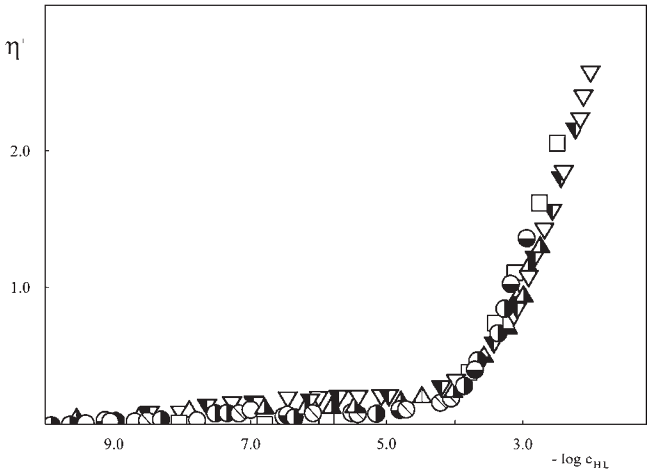

Figure 5. In the range $4 \leq -\log c_{HL} \geq 3$, points obtained in $NaClO_4$ at different C_H and C_{Pb} fall on the same curve: $C_H = 0.150$ M and C_{Pb} = (▽, ▼ = 0.25, 0.125) × 10^{-3} M; $C_H = 0.100$ M and C_{Pb} = (△, ◭, ▲, ▲ = 1.0, 0.5, 0.25, 0.125) × 10^{-3} M; $C_H = 0.050$ M and C_{Pb} = (○, ⦶, ◑, ◒ = 1.0, 0.5, 0.25, 0.125) × 10^{-3} M; $C_H = 0.025$ M and C_{Pb} = (□) 0.125 × 10^{-3} M.

Applying a procedure similar to that applied in the case of NaCl as ionic medium, the function η' is plotted as a function of $-\log c_L$. It can be observed that points obtained at different C_H fall on different curves and in particular η' is an increasing function of C_H.

Also in this case the formation of complexes with the assumption of protons takes place, i.e., $p \geq 0$.

By taking into account considerations similar to those above formulated in the case of the 1.00 mol dm^{-3} NaCl ionic medium, we performed a trial to plot η' as a function of $-\log c_{HL}$. The approach had partial success, because in the range $4 \geq -\log c_{HL} \geq 3$, corresponding approximately to the range $4 \leq -\log c_H \leq 6.5$, points obtained at different C_H fall on the same curve, as can be seen in Figure 5.

It is observed that at $4 \leq -\log c_H$, η' is still an increasing function of C_H, while, at $6.5 \leq -\log c_H$, the opposite trend takes place, because η' becomes a decreasing function of C_H.

As the three ranges are next to each other, it seems reasonable to treat the data by a successive approximation procedure.

As from the examination of the points in Figure 5, it can be deduced that the slope is less than 2, and the hypothesis can be formulated that the two species PbHL and PbH_2L_2 are formed, because the abscissa of Figure 5 is $-\log c_{HL}$. In this case and for this range ($4 \geq -\log c_{HL} \geq 3$), it can be written

$$\eta' = \log(1 + \beta'_{1,1}c_{HL} + \beta'_{1,2}{c_{HL}}^2) \qquad (7\text{ bis})$$

By the application of the graphical method proposed by Sillèn,[23] eq 7 bis can be normalized in the following form:

$$Y' = \log(1 + \alpha' u' + u'^2)$$

where

$$\log u' = \log c_{HL} + \frac{1}{2}\log \beta'_{1,2} \qquad \text{and}$$

$$\log \beta'_{1,1} = \log \alpha' + \frac{1}{2}\log \beta'_{1,2}$$

Superimposing the normalized curve family to the points and moving the two plots parallel to abscissa gave the best fit. In this point, on the basis of the mathematical positions, the first approximation values of $\log \beta'_{1,1}$ and $\log \beta'_{1,2}$ can be obtained.

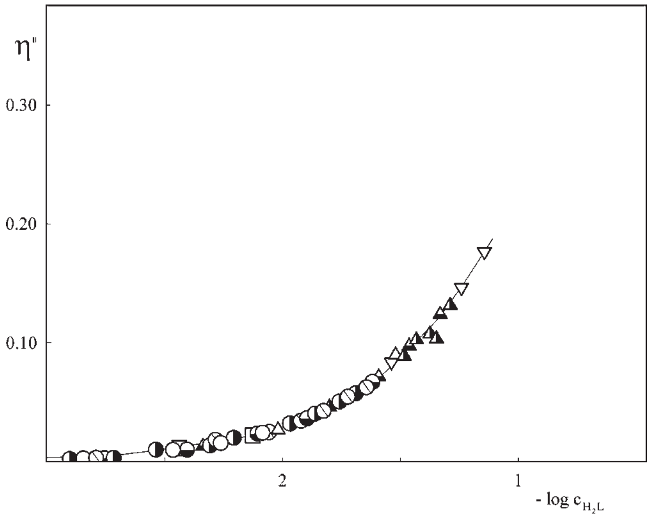

Figure 6. Experimental points obtained in $NaClO_4$ and in the range $-\log c_H \leq 5$ fall on the same curve independently of C_H and C_{Pb}. $C_H = 0.150$ M and C_{Pb} = (▽, ⧨ = 0.25, 0.125) × 10^{-3} M; $C_H = 0.100$ M and C_{Pb} = (△, ◭, ▲, ▲ = 1.0, 0.5, 0.25, 0.125) × 10^{-3} M; $C_H = 0.050$ M and C_{Pb} = (○, ⦶, ◑ ; ◒ = 1.0, 0.5, 0.25, 0.125) × 10^{-3} M; and $C_H = 0.025$ M and C_{Pb} = (□) 0.125 × 10^{-3} M. The curve is the normalized one in the position of best fit.

Points obtained at low $-\log c_H$ (range $4 \geq -\log c_{HL}$) are elaborated by taking into account that the lone form of arginine in this acid range is H_2L.

For each point, the quantity C''_{Pb}, is calculated, as follows:

$$C''_{Pb} = C_{Pb} - c_{Pb} - \beta'_{1,1}c_{Pb}c_{HL} - \beta'_{1,2}c_{Pb}{c_{HL}}^2$$

From C''_{Pb} the corresponding η'' (=$\log C''_{Pb}/c_{Pb}$) is plotted versus $-\log c_{H2L}$ in Figure 6. As can be seen, all points fall on the same curve and the following equation can be written: $\eta'' = \log(1 + \beta'_{1,2,1}c_{H2L})$, which can be normalized with the following $y' = \log(1 + u'')$, where $\log u'' = \log \beta'_{1,2,1} + \log c_{H2L}$. In the position of best fit, obtained by superimposing the normalized curve to experimental points $\{\eta'' = \log(1 + \beta'_{1,2,1}c_{H2L})\}$ and moving the two plots parallel to the abscissa, the value of the constant $\log \beta'_{1,2,1} = 0.80 \pm 0.15$ is obtained. Also the value of $\log \beta'_{1,2,1}$ can be considered a first approximation.

At higher $-\log c_H$ ($-\log c_H \geq 6.5$), the formation of arginine is in a completely deprotonated form, i.e., L, exists. By applying a similar procedure to elaborate the data at high $-\log c_H$, in this case, from $C'''_{Pb} = C_{Pb} - c_{Pb} - \beta'_{1,2,1}c_{Pb}{c_H}^2c_L - \beta'_{1,1}c_{Pb}c_{HL} - \beta'_{1,2}c_{Pb}{c_{HL}}^2$, we can calculate the function η''', and it is plotted in Figure 7 versus $-\log c_L$.

As is evident, all points are independent of C_H and C_{Pb} and fall on the same curve, it is therefore reasonable to assume the existence of the species PbL alone. By applying the graphic method proposed by Sillén,[23] we obtain $\log \beta'_{1,0,1} = 8.7 \pm 0.15$.

To obtain refined values for the constants, a second approximation is carried out and the results are collected in Table 2. Independently, the data C_{Pb}, C_H, C_L, and c_{Pb} and c_H together with the protonation constants of arginine, are introduced in the PC program.[24]

The refined values of the constants, relative to 1.00 mol dm^{-3} $NaClO_4$ as the ionic medium, are collected in Table 2 and are used

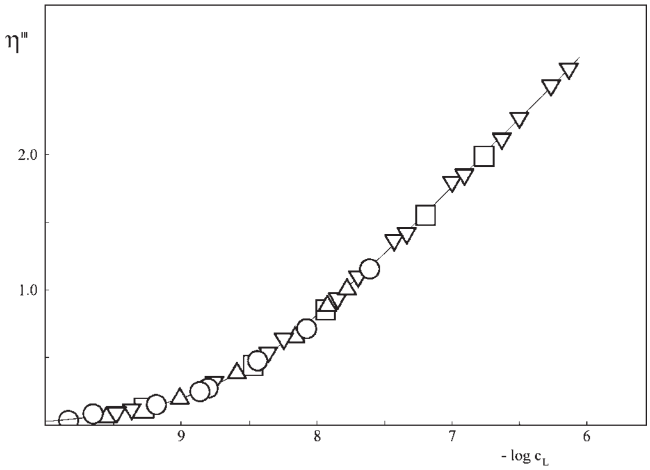

Figure 7. Dependence of η''' on $-\log c_L$ in $NaClO_4$. Experimental points obtained in the range $6.5 \leq -\log c_H \leq 8$ fall on the same curve independently of C_H and C_{Pb}: C_H = (□, ○, △, ▽ = 0.025, 0.050, 0.100, 0.150) M. The curve is the normalized one in the position of best fit.

Table 2. Species Assumed To Explain the Experimental Data for the System Lead(II)−Arginine and Values of Relative Constants at 25 °C and 1.00 M $NaClO_4$

species	graphic method	PC elaboration	proposed value
PbH_2L	0.80 (22.16) ± 0.15	0.85 (22.21) ± 0.10	0.85 (21.21) ± 0.10
PbHL	3.71 (16.24) ± 0.10	3.72 (16.25) ± 0.10	3.72 (16.25) ± 0.10
PbH_2L_2	6.09 (31.15) ± 0.10	6.04 (31.10) ± 0.10	6.04 (31.10) ± 0.10
PbL	8.7 ± 0.15	8.72 ± 0.10	8.72 ± 0.10

to calculate the curves drawn on the relative figures. The good agreement between the curves and experimental points supports the validity of the obtained results and the correctness of the procedure.

In the first three lines of Table 2 for each species, two numerical values appear, the second of them is in parentheses. The difference between them is due to the assumed form of arginine acting as the ligand. The bigger value is relative to the assumption that Pb^{2+}, H^+, and L are independent reagents, as eq 1 shows. The other values are obtained by considering the ligand in the form H_2L (first line) and HL (second and third lines), respectively. In fact, the difference between these values corresponds to the values of the protonation constants k_1 and k_2. The protonation constants of arginine determined in 1.00 M $NaClO_4$[7] are $\log k_1 = 12.53$ and $\log k_1k_2 = 21.36$.

The good agreement between the results obtained by the two different procedures is evident.

■ CONCLUSION

The complex formation between lead(II) and arginine was studied over a large range of reagent concentration in two ionic media 1.00 M NaCl and 1.00 M $NaClO_4$.

The behavior of arginine as a ligand is quite different in the two cases.

To explain the data obtained in NaCl the presence of three species PbH_2L, PbHL, and PbH_2L_2 was assumed with the relative stability constants. By using the values of the constants of Table 1, we calculate the distribution curves as a function of $-\log c_H$, and they are plotted in Figure 8A, for $C_L = 0.020$ M. The presence of PbL (65%) and PbH_2L_2 (11%) at $-\log c_H = 8$, can be explained because the concentrations of the different forms of arginine depend on both the hydrogen ion and arginine concentrations. Therefore, one can calculate $c_L = 7.6 \times 10^{-8}$ mol dm^{-3}, while $k_1^2 c_H^2 c_L^2 = 6.64 \times 10^{-6}$ mol dm^{-3}. That means the second is about 100 times the concentration of deprotonated arginine.

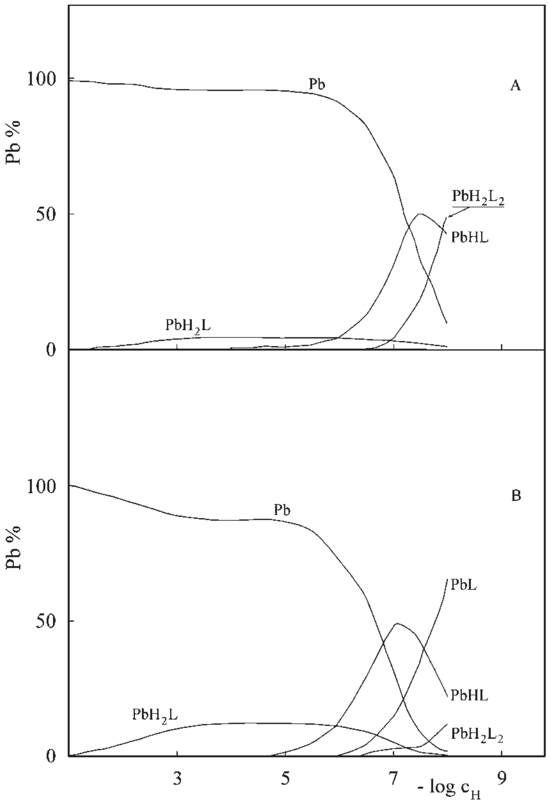

Figure 8. Distribution curves of lead(II) as a function of $-\log c_H$, at C_L = 0.020 M. Figure 8A is relative to the ionic medium 1.00 M NaCl, whereas Figure 8B is relative to the ionic medium 1.00 M $NaClO_4$.

From the figures and the values of the constants, it can be deduced that, as expected, the species formed in $NaClO_4$ are more stable than those formed in NaCl.

The presence of the ionic medium plays an important role. In fact, the difference between NaCl and $NaClO_4$ can be attributed to the property of Cl^- to form complexes with lead(II),[25] while ClO_4^- shows poor capacity as a ligand toward lead(II).

In both cases only PbH_2L is the complex formed in the range $-\log c_H \leq 5$, but in $NaClO_4$ as the ionic medium it reaches about 12%; its percentage in NaCl is less than 5%.

PbHL seems to be quite independent of the ionic medium because in both cases it is able to reach about 50%. However, in NaCl it reaches its highest percentage at $-\log c_H \approx 8.5$, whereas in $NaClO_4$ its percentage is 50% already at $-\log c_H \approx 8$.

A remarkable difference can be observed for PbH_2L_2. Its highest percentage is about 50% in NaCl, while in $NaClO_4$ it reaches only 12%. This can be explained because the presence of Cl^- prevents the formation of PbL, which is formed in $NaClO_4$ reaching a percentage of 50%.

From the comparison between Figure 8A,B, it can be observed that the formation of complexes begins at a higher $-\log c_H$ for NaCl than for $NaClO_4$. At $-\log c_H = 6.5$ the percentage of lead not bound to arginine is 83% in NaCl, while it is less than 60% in $NaClO_4$ as ionic medium.

It is known that emf data are not able to give structural information, but from the species assumed to explain the experimental data, some hypotheses can be formulated.

It seems reasonable to suppose that the complexes PbHL and PbH_2L_2 are chelates with a five-member ring, where arginine acts through carboxylate and amino groups. Also, PbL could be a chelate. In this case the chelate could be formed not only by means of carboxylic and amino groups but also with the participation of the guanido group. The species PbH_2L cannot be a chelate because, when the ligand acts in the form H_2L, the amino group of arginine is protonated and the carboxylate represents the lone possibility to bind the cation.

It is interesting to observe that the main formation of complexes takes place in the range of neutrality. From Figure 8A,B, it can be deduced that at $-\log c_H \approx 7.4$ (corresponding to the value of the blood) the free concentration of lead(II) is about 15% in solution of NaCl and less than 10% in solution of $NaClO_4$.

The results obtained here can be compared with literature data. Only the paper of Pelletier[13] reports the formation of PbHL and PbH_2L_2. It is really very hard to compare the values of the constants because the experimental conditions adopted in the work of Pelletier[13] are not defined. In any case the values proposed by Pelletier can be compared more with those obtained here in NaCl, than with the other obtained in $NaClO_4$.

The assumption of the species PbH_2L in both ionic media and of PbL in the case of $NaClO_4$, to explain the obtained data is a remarkable difference between this paper and the literature data.

It is also stressed that other researchers,[14] by studying the behavior of arginine as a ligand, explain their data by assuming that the ligand acts only as HL.

The narrow range of the concentration investigated by other authors explains why they have not found all the species found here.

The results of this paper suggest that extending the investigation in a future activity to the behavior of arginine toward other toxic cations will be worthwhile.

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