OSJ | OPEN SCIENCE JOURNAL

RESEARCH ARTICLE

Gallium (III)-acetate speciation studies under physiological conditions.

Brahim Hacht*

Department of Chemistry, Faculty of Sciences, University Mohammed Ist, Oujda, Morocco

*Corresponding author: Brahim Hacht, email: b.hacht@ump.ma

Abstract:

This paper describes an experimental investigation in aqueous solution of the equilibria between gallium (III) metal ions and acetate ions as a model of low-molecular-weight ligand using potentiometric techniques under physiological conditions of temperature 37 °C and ionic strength 0.15 $mol.dm^{-3}$ NaCl. Potentiometric measurements were monitored with the help of a glass electrode calibrated daily in hydrogen ions concentrations. Several metal to ligand ratios were used and the respective titrations data obtained in the pH range of 2.3-5.3 were treated using the SUPERQUAD computer program to determine global stability constants. Different species were considered during the calculation procedure and the following hydroxides have been characterized: $Ga(OH)_3$, $Ga_6(OH)_{15}^{+3}$ and coexist with one mononuclear complex $Ga(CH_3COO)^{+2}$. Speciation calculations based on the determined constants were then used to simulate the species distribution as a function of pH

Key words: gallium (III), acetate, speciation, global stability constants, potentiometric titrations, physiological conditions.

OPEN ACCESS

Citation: Brahim H. (2016) Gallium (III)-acetate speciation studies under physiological conditions. Open Science Journal 1(1)

Received: 19th Febryary 2016

Accepted: 8th April 2016

Published: 27th April 2016

Funding: The author(s) received no specific funding for this work

Competing Interests: The author have declared that no competing interests exists.

Introduction

Gallium is a semimetal that occupies the third position of Group 13 of the Periodic Table. In aqueous solution gallium is more stable in its 3+ oxidation state and is classified according to Pearson (1) as a hard Lewis acid, giving rise to thermodynamically stable complexes with ligands that are hard Lewis bases. However, gallium in oxidation state 2+ is energetically unfavorable and, in physiological conditions, oxidation state 1+ is for gallium almost impossible. Redox chemistry is, therefore, not possible for Ga(III) in biological mediums (2). The coordination chemistry of gallium (III) ions has been largely studied in the last decades (3-9) where many ligands with oxygen and/or nitrogen donor atoms (such as carboxylate, phosphonate, phenolate, hydroxamate and amine groups) were used as chelating agents.

Due to the analogy with Fe(III) in terms of electronegativity and ionic radius (10), electron affinity (1), and coordination geometry, Ga(III) is considered as a good mimetic of Fe(III) (11). These properties suggest that Ga(III) ion follows biochemical routes similar to those found in iron metabolism. Recently, gallium ions were shown to exhibit antitumor effects in the form of simple salts such as gallium nitrate, and interferes with cellular iron metabolism (12, 13). Moreover, gallium (III) plays a very important role in the diagnosis of tumor and for possible treatment of malignancies. In the United States, this metal ion was approved by the FDA for i.v. administration and is used in its ^{67}Ga(III) salts or citrate complex to diagnose tumors (14).

Numerous studies and particularly potentiometric titration and ^{71}Ga NMR experiments (15, 16) have revealed that aluminium and gallium have a similar hydrolytic behavior. Indeed, among metals, aluminium, gallium and iron exhibit a particular hydrolytic properties as they give rise to polycationic structures in a given pH/concentration conditions (17).

Ga(III) metal ions are prone to hydrolyse within a large domain of pH including physiological pH to form mono and polynuclear hydroxide species (18). Although this phenomenon must be taken into account prior to any metal-ligand interactions.

Such studies have already been initiated concerning the hydrolysis of gallium, many of which were reviewed by Baes and Mesmer (19). More recently, several reviews were devoted to gallium and other rare elements hydrolysis in aqueous solution. Most of the available data refer to standard conditions of temperature and pressure and at different ionic strength.

However, in spite of the numerous studies previously carried out on Ga(III) hydrolysis, only few quantitative data are available regarding corresponding interactions in solution, and none of these refer to the physiological conditions of temperature and ionic strength. In a recent investigation we have reported the hydrolysis of Ga(III) with respect to these specific conditions (20).

With such a diverse array of reported works, it is useful to look for the interactions of the acetate anions as a low-molecular-weight ligand. Acetic acid (ACT) has been known for centuries and its biological role was studied very early in the biosynthesis of heme (21) and elucidated by the 1964 Nobel prize winner Konrad Emil Bloch (22). He found that acetic acid is the primary precursor for the production of cholesterol in the body as well as for fatty acids.

With a view to taking into account the leading role of metal-ligand interactions in biological systems (23, 24), it is extremely important that formation constants of complexes of metal ions with low molar mass bioligands be determined under experimental conditions as close as possible to those pertaining to *in vivo*.

This study thus reports a potentiometric determination of the stability constants of Ga(III)-ACT under physiological conditions. Although made difficult by the occurrence of precipitates in the case of higher metal ion concentrations, this study led to definite conclusions as to the number and stoichiometry of hydroxides formed under physiological conditions (0.15 mol·dm^{-3} NaCl, 37 °C). In particular, mononuclear ML species could be characterized beside neutral $M(OH)_3$ and polynuclear $M_6(OH)_{15}{}^{3+}$ hydroxides (M represents metal ion and L stands for ACT).

Experimental

Reagents. — gallium (III) chloride solution was prepared from Sigma pure anhydrous crystals and was slightly acidified to prevent metal hydrolysis. It's metal title was checked using complexometric titrations (25) whereas its exact final acid content was deduced from potentiometric measurements using Gran titrations (26).

Sodium hydroxide solutions were prepared by diluting the contents of BDH concentrated volumetric solutions vials with freshly boiled doubly deionized water, which was saturated with purified nitrogen before use. These solutions, standardized against potassium hydrogenophtalate (Prolabo, Normapur) and proved to be carbonate-free from the features of the corresponding Gran titration plots, were stored under nitrogen atmosphere.

Solutions of acetic acid were freshly prepared and titrated against sodium hydroxide. They were first made sufficiently acidic to ensure that the dissociating group was significantly protonated at the outset of each experiment.

Experimental Conditions. — Sodium chloride (0.15 mol.dm^{-3}) was chosen as a background electrolyte to maintain constant activity coefficients and to ensure isotonicity with blood plasma.

Potentiometric titrations were performed using a Metrohm E105 combined microelectrode formed by a glass electrode (g.e.) and a saturated NaCl calomel electrode (s.c.e.), fitted in a thermostated Metrohm 4 cm^3 vessel according to the scheme cell unit:

$$\mathrm{g.e.} \,|\, \mathrm{Ga^{3+}, ACT, NaCl\ (0.15\ mol.dm^{-3})} \,|\, \mathrm{NaCl\ (s.c.e.)} \,|\, \mathrm{Hg_2Cl_2 - Hg} \qquad (1)$$

E.m.f. variations were monitored by a Beckman model 4500 digital mV-meter. The temperature of the reaction vessel was maintained at 37 ± 0.02 °C by circulating water and all titrations were carried out under a constant bubbling of purified and thermostated nitrogen. The above electrode system was calibrated in the concentration scale (27), the ionic product of water being found equal to $10^{-13.24}$ under the present conditions. The value of K_w was estimated using acid-base titration curves and the refined value was obtained using the SUPERQUAD (28) computer program. Accordingly, the symbol pH used throughout actually represents –log[H^+]. The slope s from the Nernst equation, $E = E_0 - s.pH$ was determined using acid-base titration (27). The electrode slope was found equivalent to the expected theoretical one (61.54 mV.pH^{-1}). In these conditions, the value of the standard electromotive force, E_0 takes into account the liquid junction potential and activity coefficient terms. With a view to avoid the day-to-day drift of E_0, the E of 0.01 mol.dm^{-3} HCl was measured at the beginning of each titration.

Successive aliquots of sodium hydroxide 0.02 mol.dm^{-3} were delivered from a Radiometer ABU 12 Autoburette equipped with a 2.5 cm^3 glass cylinder, both titrate and titrant containing 0.15 mol.dm^{-3} NaCl.

As may be noted in Table I, where titration data are summarized and whose results are given in the supplementary data (obtainable from the author), metal ions concentration were varied significantly over the set of these experiments in view to allow the formation of different metal-ligand stoichiometries.

Table I

System	**C_M**	**C_L**	**C_H**	**pH range**	**Exp.**	**n**	**R**
Ga(III)-acetate	1.945	19.788	1.581	2.76 – 4.93	+	31	0.1
	1.945	9.894	1.581	2.73 – 4.41	x	36	0.2
	5.187	19.788	4.216	2.43 – 4.08	□	32	0.26
	3.891	9.894	3.162	2.47 – 3.25	Δ	29	0.4
	4.863	9.894	3.953	2.49 – 3.08	▽	25	0.5
	4.863	4.947	3.953	2.49 – 2.85	◃	22	1
	9.726	4.947	7.905	2.27 – 5.29	▹	20	2

Calculation Procedures. — Potentiometric data were treated following our usual approach (29, 30) involving optimization and simulation in successive steps. For a general complex formula corresponding to $M_pL_qH_r$, M = Ga(III) metal ion, L = ACT and H = proton, the reaction for the complexation can be written :

$$pM^{3+} + qL^{-} + rH^{+} \leftrightarrow M_pL_qH_r^{3p-q+r} \qquad (2)$$

The numerical treatment of the data was performed by the program SUPERQUAD. To be in agreement with the notations of this program, the cumulative stability constant of the equilibrium (2) is given by the equation (3) where charges and molecules of water have been omitted for the sake of simplicity:

$$\beta_{pqr} = \frac{[M_pL_qH_r]}{[M]^p[L]^q[H]^r} \qquad (3)$$

According to SUPERQUAD notations, the hydroxo-complexes are referred to as (p, q, -r) and the hydroxide ion is thus (0, 0, -1).
The experimental protonation curves were plotted as a function of pH using equation (4). These curves represent the average number of protons bound to each ligand:

$$\bar{r} = \frac{C_L + C_H - C_{OH} + [OH^-] - [H^+]}{C_L} \qquad (4)$$

with C_L, C_H and C_{OH} are the total concentrations of acetate, strong acid and hydroxide.

Formation curves are obtained from equation (5) and express the average number of ligands bound per metal ion:

$$\bar{p} = \frac{C_L - [L].\sum_0^r \beta_{LH_r}.[H^+]^r}{C_M} \qquad (5)$$

Finally, the amount of ligand not complexed to metal is obtained from the free hydrogen-ion concentration and the ligand-protonation constants. The calculation is independent of the metal complex species existing in solution (6):

$$[L] = \frac{C_H - [H^+] + [OH^-]}{\sum_0^r r.\beta_{LH_r}.[H^+]^r} \qquad (6)$$

The equations (5) and (6) assume that all species are mononuclear and there is no hydroxo- nor protonated complexes. Thus, when these conditions are not valid, ($\bar{p}$,-$\log_{10}$[L]) becomes in fact a pseudo $\bar{p}$. These functions being ideal for showing the degree of variation from mononuclearity, and for comparing experimental with simulated titrations (31). In general, the shape of the formation curves ($\bar{p}$,-$\log_{10}$[L]) can be used to deduce the possible stoichiometries of the complex species present in each system as well as to obtain rough estimates of their formation constants.

In general, when metal-hydroxo/hydroxides complexes are predominant, estimates of formation constants cannot be made easily from the shape of these curves. Chemical considerations and used as an input data to be refined with the help of the SUPERQUAD computer program. The final discrimination between sets displaying similar SUPERQUAD analyses is based on the two parameters: σ (connected with the objective function) and χ^2 statistics. The critical values are assumed as $\sigma < 3$ and $\chi^2 \leq 12.60$ at the 0.95 confidence level. Theoretical protonation and formation curves were simulated with the help of our SIMDIS computer program (32) on the basis of the 'best' set of constants. This procedure confirms how accurately these constants describe the experimental data. Graphical comparisons between experimental curves and corresponding simulated data were used as final tests of coincidence. In addition, the same program was used to generate species distribution as a function of pH in a given analytical conditions.

Results and discussion

The constant that controls the free and protonated form of ACT has been already determined in a previous study and in the same conditions (33). It was then confirmed and used before studying gallium (III) equilibrium. In addition, as mentioned in the literature (34), in the case of Ga(III), it is recommended to determine the constants relative to metal-ions hydroxides in the same conditions as for metal –ligand interactions. Because of its very high polarizing power, the Ga(III) ion has such a high tendency to hydrolyse that, even in acidic pH, its hydrolysis equilibria must absolutely and accurately be taken into account in the calculations especially as the acetate is not a strong ligand to render hydroxides negligible.

Figure 1 shows the experimental protonation curves of acetate obtained in the presence of different concentrations of Ga(III) shown in Table I, to be compared with the dashed line calculated from the ligand protonation constant. The important shifts observed for increased gallium ions concentrations, which reflect the release of protons from complexing ligand and/or water molecules, suggest the formation of stable gallium acetate complexes and/or metal ion hydroxides. Indeed, both phenomenon (complexation and hydrolysis) leads to the release of protons into the solution. The shift between these curves is pH and metal-to-ligand ratio dependent. At pH ca 2, these curves remain practically identical but the gap becomes very important around pH 3.

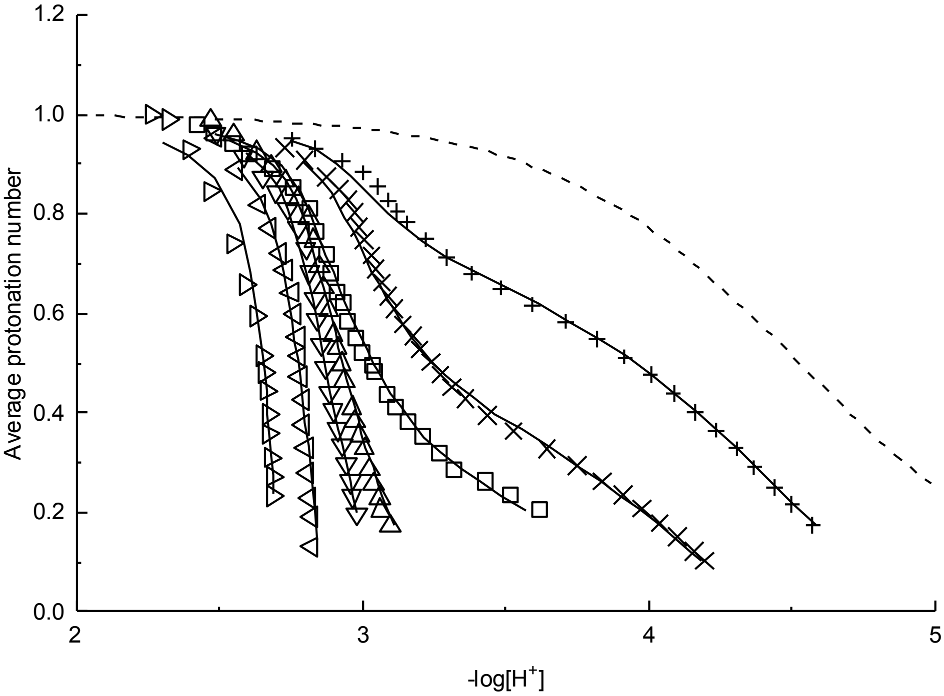

Figure 1: Protonation curves relative to Ga(III) –ACT mixture. Symbols as in Table I. Dashed line represents the calculated protonation curve of ACT in the absence of Ga(III). Solid lines are theoretical curves calculated with the help of SIMDIS program using results of Table II.

The plot of the formation curves ($\bar{p}$, -$\log_{10}$[L]) was necessary to distinguish the effect of the complexation and the metal hydrolysis. Figure 2 shows the experimental formation curves calculated according to equations (5) and (6). As can be seen, none of the metal-to-ligand ratio has superimposable formation curves and the overall spread in these curves, is metal-to-ligand ratio dependent. Thus, the presence of either deprotonated or hydroxo-complexes (or both) is indicated. In addition, these curves do not present marked plateau around $\bar{p} = 1$ or 2 which are usually observed respectively for mononuclear species ML and ML_2 in the case of strong ligands (35). In addition, the ‘‘tails’’ displayed by the experimental curves are regarded as specific to the formation of hydroxo species. Accordingly, monomeric with several mono and polymeric hydroxo species – beside metal hydroxides themselves taken from our previous investigations on gallium hydroxides (20) – were used for the refinement procedure.

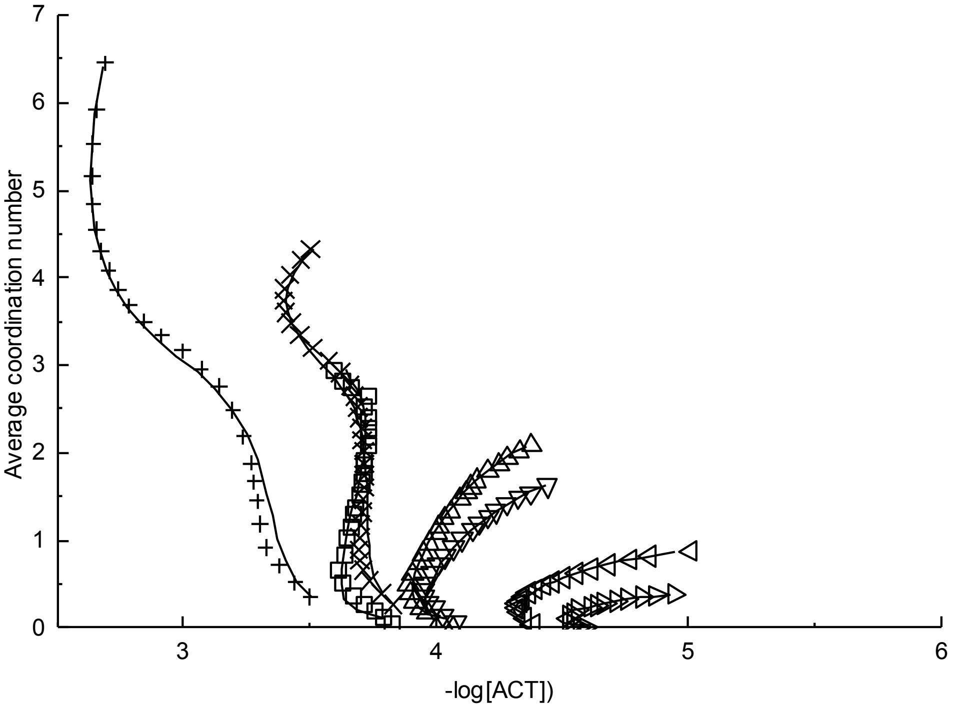

Figure 2: Formation curves relative to Ga(III) –ACT mixture. Symbols as in Table I. Solid lines represent the corresponding simulated protonation curves as obtained by means of SIMDIS using data in Table II.

Given that ACT is a very weak ligand and as recommended in the literature (36), the set of constants relative to Ga(III) hydroxides determined in the same conditions of temperature and ionic strength (20) were in the first step maintained constant and only Ga(III)-acetato complexes were refined. However, when the entire set of hydroxides constants is refined beside Ga(III) acetate complexes, this leads to the existence of only $M(OH)_3$ and $M_6(OH)_{15}^{3+}$ with a better result for the SUPERQUAD statistical values σ and χ^2. In addition, when they are allowed to vary, the resulting refined constants benefit from a good accuracy and were very close to the recommended values (20). This confirms the validity of the entire procedure.

As far as the monomeric hydroxide species such as $M(OH)^{+2}$, $M(OH)_2^{+1}$ and $M(OH)_4^{-1}$ are concerned, any attempt to refine their constants alone or in presence of the ML species and in various combinations was unsuccessful. Our results are in good agreement with those of Baes and Mesmer (19) suggesting that Ga^{3+} can generate easily polynuclear hydroxide rather than monomeric species. A summary of the best set of stability constant describing the interaction Ga(III)-ACT is presented in Table II.

Table II

System	**p**	**q**	**r**	**$\log_{10}\beta pqr$**	**σ**	**χ2**	**n**
Proton-acetate	0	1	1	4.53	From reference [33]		
Ga(III)-hydroxides	1	0	-3	-9.53	From reference [20]		
	4	0	-11	-25.17			
	6	0	-15	-30.46			
Ga(III)-acetate	This work						
	1	1	0	2.74 ± 0.05	0.93	16.95	195
	4	0	-11	-9.29 ± 0.01			
	6	0	-15	-30.93 ± 0.04			

The pseudo-experimental curves are shown in Figure 2 and represented using solid lines. They were computed with the help of our SIMDIS program (32) on the basis of constants from Table II and analytical conditions from Table I. A quite satisfactory coincidence may be observed between this figure and the experimental one, which confirms the validity of the approach used.

It is worth noting that for some pH values depending on Ga^{3+}-to-ligand ratio, a slight opacity was observed in the titration cell, indicative of the initiation of a precipitation process which can be assigned to the neutral hydroxide $Ga(OH)_3$. Titrations were then stopped in view to limit data to the true equilibrium conditions.

The solubility product for $Ga(OH)_3$ is not well documented especially in moderate media. Table III summarizes the product $[Ga^{3+}][OH^-]^3$ calculated for each point, particularly from start pH to the titration endpoint using the formation constants of Table II and data of experiment 7, where the metal-to-ligand ratio = 2 is the most favorable among the present work, for the formation of this neutral hydroxide which could precipitate at acidic pH. In the present conditions, the solubility limit can be estimated near 1.3E-34.

Table III

pH	%M	%M(OH)$_3$	%M$_6$(OH)$_{15}$	%ML	[M].[OH]^3
2.32	98.12	0.46	0.00	1.42	1.46E-35
2.395	97.64	0.76	0.00	1.60	2.33E-35
2.469	96.91	1.27	0.02	1.80	3.72E-35
2.546	95.68	2.13	0.18	2.02	5.97E-35
2.593	94.24	2.90	0.72	2.15	7.92E-35
2.627	92.30	3.59	1.87	2.23	9.64E-35
2.65	90.34	4.12	3.29	2.26	1.08E-34
2.656	89.67	4.27	3.80	2.26	1.12E-34
2.663	88.94	4.43	4.37	2.26	1.15E-34
2.669	88.18	4.60	4.97	2.25	1.18E-34
2.673	87.83	4.68	5.25	2.24	1.20E-34
2.679	86.99	4.85	5.93	2.23	1.23E-34
2.684	86.33	4.98	6.47	2.23	1.25E-34
2.687	85.92	5.07	6.80	2.22	1.27E-34

Figure 3 shows the variation with the pH of the computed composition of a solution containing equimolar 2 mmol·dm^{-3} of ACT and Ga(III) as a percentage of the total gallium. As was mentioned above, acetic acid is very weakly complexed by Ga(III) cations under the present conditions. Indeed, it remains predominant (see figure 4) exclusively in its protonated and free forms LH. The ML species appears at pH 2.0 and reaches only 1.8% at pH 2.9 while gallium exists in this interval mainly in its free form Ga^{+3}. The neutral hydroxide M(OH)$_3$ becomes significant at pH 2.7 (6%) and dominates almost entirely (95%) the distribution at pH = 4.4. The polynuclear $M_6(OH)_{15}^{+3}$ hydroxide reaches ca. 8% near pH 3 and dominates the repartition with 50% at pH = 3.3.

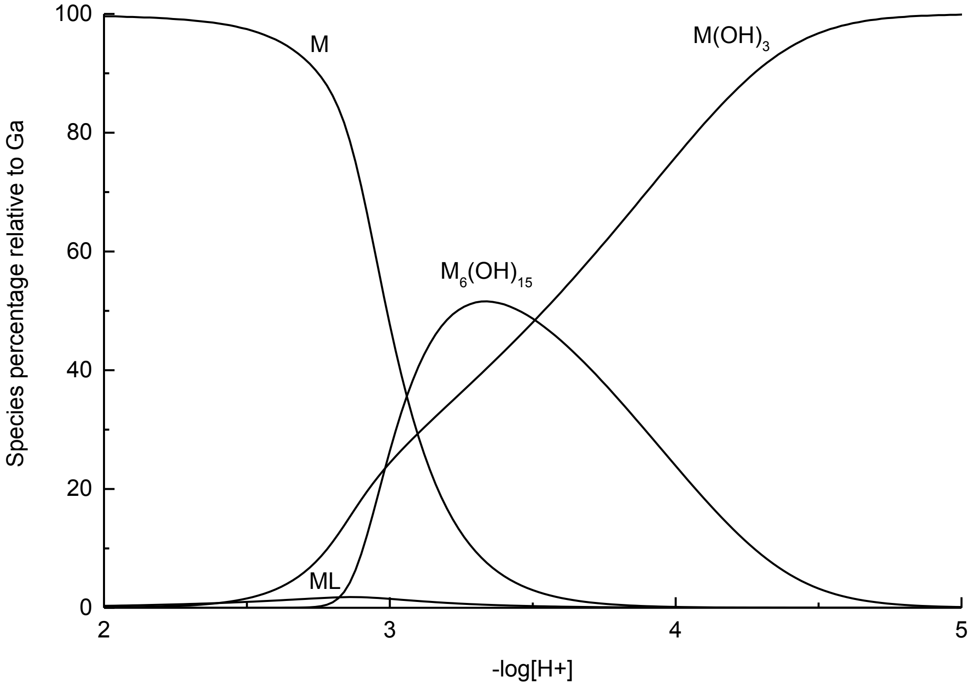

Figure 3: Simulated distribution of the Ga(III) into its free, complexed and hydroxides species as a function of pH, with Ga(III) = 2 mmol.dm^{-3} and ACT = 2 mmol.dm^{-3}.

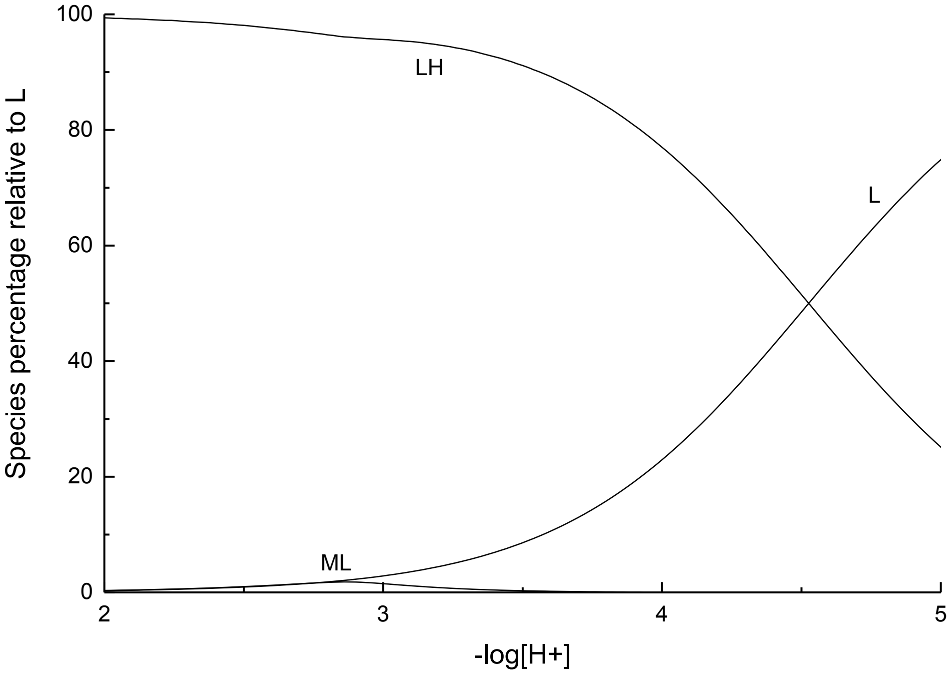

Figure 4: Simulated distribution of the ACT into its free, complexed and protonated species as a function of pH, with Ga(III) = 2 mmol.dm^{-3} and ACT = 2 mmol.dm^{-3}

Moreover, interaction between Ga^{3+} and Cl^- has been neglected in our analysis. Indeed, as pointed out in the literature, this complexation becomes significant only at very high chloride ion concentrations (37). This was not the case for our study, since 0.15 mol.dm^{-3} NaCl can be considered as relatively a low ionic strength medium.

In order to make suitable comparisons with literature data, it is useful to consider experiments made under identical conditions because the respective values of the stability of these complexes and the differences between these them are quite small. As noticed above, coordination chemistry of gallium (III) with several ligands is widely described in the literature, but unfortunately, no studies were devoted to using physiological conditions except those realized by our group in the same medium with calcium and magnesium as divalent cations (20) and trivalent aluminium (30). Calcium and magnesium forms separately very weak complex with acetic acid with a $\log\beta_{110}$ ca 1 but both metal ions generate ML_2 species with $\log\beta_{120} = 2.77$ and 2.95 respectively. Al(III) forms only dimeric species ($\log\beta_{120} = 4.12$) besides two hydroxo species. $M_2L(OH)_2$ and $M_2L(OH)_3^-$ and hydroxides. In the same conditions, Ga-ACT system do not generate dimeric species nor hydroxo ones and seems then to behave differently from its so called homologue Al(III).

Conclusion

The aim objective of this study was to bring a contribution towards the solution equilibrium studies of gallium (III) in aqueous medium with a weak ligand and near physiological conditions. This was reported for the first time using potentiometric techniques assisted by computer calculations.

Investigations at the higher basic pH range was not possible due to the strong tendency of the Ga(III) ion to hydrolyze.

From the overall presented study, it is concluded that in physiological conditions (37°C, 0.15 mol·dm^{-3} NaCl) and for the range of concentrations used, gallium(III) ions gives rise to one monomeric complex ML species coexisting with a monomeric $M(OH)_3$ along with polycationic hydroxide species $M_6(OH)_{15}^{+3}$. Our results indicate the tendency of Ga(III) to engender mono and polycationic hydroxides and also the difficulty to determine dimeric acetato complex. Moreover, our data show the difference between Ga(III) and Al(III) regarding aqueous coordination of acetate ligand, since the latter produces dimeric complexes and polymeric hydroxo species under the same conditions of temperature and ionic strength. The lack of literature data obtained in the same conditions does not permit us any comparison. Therefore, new determinations of the complex formation constants of these systems under physiological conditions will thus be necessary in the near future.

Acknowledgements

The author thanks Professor G. Berthon (Retired, Univesity Paul Sabatier, Toulouse, France) for his support of the present work.

References

1. Pearson R. Hard and soft acids and bases. Chem Brit. 1967;3:103-7.
2. Jakupec MA, Galanski M, Arion VB, Hartinger CG, Keppler BK. Antitumour metal compounds: more than theme and variations. Dalton transactions. 2008;2:183-94. DOI: 10.1039/b712656p.
3. Sun Y, Anderson CJ, Pajeau TS, Reichert DE, Hancock RD, Motekaitis RJ, et al. Indium (III) and gallium (III) complexes of bis(aminoethanethiol) ligands with different denticities: stabilities, molecular modeling, and in vivo behavior. Journal of medicinal chemistry. 1996;39(2):458-70. DOI: 10.1021/jm9505977.
4. Green MA, Welch MJ. Gallium radiopharmaceutical chemistry. International journal of radiation applications and instrumentation Part B, Nuclear medicine and biology. 1989;16(5):435-48. DOI: 10.1016/0883-2897(89)90053-6.
5. Chaniotakis NA, Alifragis Y, Konstantinidis G, Georgakilas A. Gallium nitride-based potentiometric anion sensor. Analytical chemistry. 2004;76(18):5552-6. DOI: 10.1021/ac049476h.
6. Kubicek V, Havlickova J, Kotek J, Tircso G, Hermann P, Toth E, et al. Gallium(III) complexes of DOTA and DOTA-monoamide: kinetic and thermodynamic studies. Inorganic chemistry. 2010;49(23):10960-9. DOI: 10.1021/ic101378s.
7. Aldridge S, J. Downs A. The Group 13 Metals Aluminium, Gallium, Indium and Thallium: Chemical Patterns and Peculiarities. John Wiley & Sons, Ltd. 2011:726.
8. Enyedy EA, Domotor O, Varga E, Kiss T, Trondl R, Hartinger CG, et al. Comparative solution equilibrium studies of anticancer gallium(III) complexes of 8-hydroxyquinoline and hydroxy(thio)pyrone ligands. Journal of inorganic biochemistry. 2012;117:189-97. DOI: 10.1016/j.jinorgbio.2012.08.005.
9. Ghosh S, Gowda RR, Jagan R, Chakraborty D. Gallium and indium complexes containing the bis(imino)phenoxide ligand: synthesis, structural characterization and polymerization studies. Dalton transactions. 2015;44(22):10410-22. DOI: 10.1039/c5dt00811e.
10. Shannon R. Revised effective ionic radii and systematic studies of interatomic distances in halides and chalcogenides. Acta Crystallographica Section A. 1976;32(5):751-67. DOI: 10.1107/S0567739476001551.
11. Frangipani E, Bonchi C, Minandri F, Imperi F, Visca P. Pyochelin Potentiates the Inhibitory Activity of Gallium on Pseudomonas aeruginosa. Antimicrobial Agents and Chemotherapy. 2014;58(9):5572-5. DOI: 10.1128/AAC.03154-14.
12. Bernstein LR. Mechanisms of therapeutic activity for gallium. Pharmacological reviews. 1998;50(4):665-82.
13. Arion VB, Jakupec MA, Galanski M, Unfried P, Keppler BK. Synthesis, structure, spectroscopic and in vitro antitumour studies of a novel gallium(III) complex with 2-acetylpyridine (4)N-dimethylthiosemicarbazone. Journal of inorganic biochemistry. 2002;91(1):298-305. DOI:10.1016/S0162-34(02)00419-1.
14. Anderson CJ, Welch MJ. Radiometal-labeled agents (non-technetium) for diagnostic imaging. Chemical reviews. 1999;99(9):2219-34.
15. Bradley SM, Kydd RA, Yamdagni R. Comparison of the hydrolyses of gallium(III) and aluminium(III) solutions by nuclear magnetic resonance spectroscopy. Journal of the Chemical Society, Dalton Transactions. 1990;9:2653-6. DOI: 10.1039/DT9900002653.
16. Bradley SM, Kydd RA. Comparison of the species formed upon base hydrolyses of gallium(III) and iron(III) aqueous solutions: the possibility of existence of an [FeO4Fe12(OH)24(H2O)12]7+ Polyoxocation. Journal of the Chemical Society, Dalton Transactions. 1993;15:2407-13. DOI: 10.1039/DT9930002407.
17. Michot LJ, Montargès-Pelletier E, Lartiges BS, d'Espinose de la Caillerie J-B, Briois V. Formation Mechanism of the Ga13 Keggin Ion: A Combined EXAFS and NMR Study. Journal of the American Chemical Society. 2000;122(25):6048-56. DOI: 10.1021/ja9941429.
18. Wood SA, Samson IM. The aqueous geochemistry of gallium, germanium, indium and scandium. Ore Geology Reviews. 2006;28(1):57-102. DOI: 10.1016/j.oregeorev.2003.06.002.
19. Baes J, C.F., Mesmer RE. The Hydrolysis of Cations. Krieger Publishing Company, Malabar, Florida. 1986:489.
20. Hacht B. Gallium(III) Ion Hydrolysis under Physiological Conditions. Bulletin of the Korean Chemical Society 2008;29(2).
21. Radin NS, Rittenberg D, Shemin D. The role of acetic acid in the biosynthesis of heme. Journal of Biological Chemistry. 1950;184(2):755-68.
22. Lectures N. Physiology or Medicine 1963–1970. Elsevier, Amsterdam. 1972.

23. Berthon G. Handbook of Metal-Ligand Interactions in Biological Fluids; Bioinorganic Chemistry. Dekker, New York. 1995;vols. 1-2.
24. Berthon G. Handbook of Metal-Ligand Interactions in Biological Fluids; Bioinorganic Chemistry. Dekker, New York. 1995;vol. 2., Part 5.
25. Schwarzenbach G. Complexometric Titrations. Metuhen and Co, London. 1957.
26. Rossotti H. The Study of Ionic Equilibria. Longman, London. 1978.
27. Linder PW, Torrington RG, Williams DR. Analysis Using Glass Electrodes. . Open University Press, Milton Keynes. 1984.
28. Gans P, Sabatini A, Vacca A. SUPERQUAD: an improved general program for computation of formation constants from potentiometric data. Journal of the Chemical Society, Dalton Transactions. 1985;6:1195-200. DOI: 10.039/DT9850001195.
29. Hacht B. Complex Formation of Acetic Acid with Ca(II) and Mg(II) Under Physiological Conditions. Journal of Solution Chemistry. 2007;37(2):155-63. DOI: 10.1007/s10953-007-9233-3.
30. Hacht B. Speciation studies of aluminium(III)-acetate complexes under physiological conditions. Chemical Speciation and Bioavailability. 2008;20(4):207-15. DOI: 10.3184/095422908X379282.
31. Corrie AM, Makar GKR, Touche MLD, Williams DR. Thermodynamic considerations in co-ordination. Part XX. A computerised approach as an alternative to graphical normalised curve fitting as a means of detecting oligonuclear complexes in metal ion-ligand solutions and its application to the zinc(II)-, lead (II)-, and proton-glycine peptide systems. Journal of the Chemical Society, Dalton Transactions. 1975;2:105-10. DOI: 10.1039/DT9750000105.
32. Hacht B. Simdis. unpublished programm. 1997.
33. Hacht B, Berthon G. Metal ion-FTS nonapeptide interactions. A quantitative study of zinc(II)-nonapeptide complexes (thymulin) under physiological conditions and assessment of their biological significance. Inorganica Chimica Acta. 1987;136(3):165-71.
34. Berthon G. Chemical speciation studies in relation to aluminium metabolism and toxicity. Coord Chem Rev. 1996;149:241-80. DOI:10.1016/S0010-8545(96)90030-2.
35. Berthon G, Hacht B, Blais M-J, May PM. Copper-histidine ternary complex equilibria with glutamine, asparagine and serine. The implications for computer-simulated distributions of copper(II) in blood plasma. Inorganica Chimica Acta. 1986;125(4):219-27. DOI: 10.1016/S0020-1693(00)81215-3.
36. Venturini M, Berthon G. Aluminium speciation studies in biological fluids. A new investigation of aluminium hydroxide equilibria under physiological conditions. Journal of the Chemical Society, Dalton Transactions. 1987;5:1145-8. DOI: 10.039/DT9870001145.
37. van Gaans PFM. Thermodynamics of aqueous gallium chloride: Activity coefficients in dilute and high chloride solutions with consideration of the effects of hydrolysis and chloride complex formation. Chemical Geology. 1993;104(1–4):139-57. DOI: 10.1016/0009-2541(93)90147-B.