

Composition, Stability and Probable Structure of a Colourless Organometallic Complex (Gd(III)-Malic Acid)

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ABSTRACT

The formation of colourless gadolinium complexes (x,y,z), between x gadolinium ions, y ligands and z protons, of some organic acids, has been studied in aqueous solution. In this work we present the results of investigations on the interaction of the gadolinium ion (Gd^{3+}) with malic acid ($C_4H_6O_5$, α -hydroxyl dicarboxylic acid), in dilute aqueous solution for pH values between 5.5 and 7.5. Colourless gadolinium complexes of malate ions have no absorption band UV-visible, the indirect photometric detection (IPD) technique was used and studies have identified a major tri-nuclear complex of malate ion ($^-OOC-CH_2-CHOH-COO^-$). The formation of this new colourless complex is derived from three Gd(III) ions that react with two malate ions and two hydronium ions (H_3O^+), giving for this colourless complex, a (3,2,2) composition and apparent stability constant depends on the acidity of the medium, with $\log K'_{322} = 18.88 \pm 0.05$ at pH = 6.30. To complement previous results and to propose a probable structure for this new complex detected in solution, studies of IR spectroscopy have been conducted to identify the chelation sites for both ligands. The results were analysed and show that this organometallic gadolinium complex, contains two different sites, respectively, two lateral tetradentate mono-nuclear sites and a single central bidentate mono-nuclear site. From these results, the reaction of formation, the stability constant and the probable structure of this new colourless organometallic gadolinium complex are proposed.

Keywords: Gadolinium Complexes; Malate Ions; Indirect Photometric Detection (IPD); Apparent Stability Constant; Chelation Sites; Bidentate Mono-Nuclear; Tetradentate Mono-Nuclear

1. Introduction

In the field of analysis of very dilute solutions, we developed a new detection technique for determining the compositions and stabilities of some colourless organometallic complexes, which have no absorption band UV-visible. This technique is the indirect photometric detection (IPD), based on competitive reactions, by ligand-ligand exchange. The method is simple, reproducible, effective and applicable to very dilute solutions. Thus, the importance of IPD technique was also revealed by its adaptation to other techniques of separation and determination, such as liquid chromatography [1-3], capillary electrophoresis [4] and continuous flow analysis (FIA) [5-7]. Some studies [8,9] show that this technique is very effective in identifying some colourless tungstate complexes of sugars and organic acids.

The paramagnetic complexes of the trivalent gadolinium ion (Gd^{3+}), have emerged as very important agents of contrast, for many applications in Magnetic

Resonance Imaging (MRI), through interest electronic and magnetic properties of this ion [10,11]. Recent studies have shown that the lanthanide complexes of coumarin (1,2-benzopyrone), exhibit antiproliferative activity [12]. In this work, investigations by indirect photometric detection (IPD) were carried out to study the interaction of the trivalent gadolinium ions, with malate ions (conjugate base of malic acid), detecting the majority of colourless complex formed in solution and determine its composition and stability. To elucidate the structure and the chelation sites of this major malate complex of Gd^{3+} ions, the technique of IR spectroscopy has been used. Indeed, these two techniques (IPD and IR spectroscopy) are very useful for elucidating the formation reaction of the major complex for the system (Gd(III)-malic acid), completely, determine, the composition and stability, the nature of chelation sites for each of ligands ($^-OOC-CHOH-COO^-$), involved in the composition of detected complex and propose a probable structure for this major gadolinium complex of the malate ion.

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2. Experimental Methods

2.1. Chemicals

Malic acid, Chrome Azurol S (H_4Ch), Gd(III) nitrate and other chemicals were commercial products (Aldrich, Prolabo,...) of the purest available and analytical grade, used as received.

2.2. Indirect Photometric Detection

A standard Helios γ UV-visible spectrum-eter controlled by Vision 32 software was used for spectrometric measurements, using quartz cells of optical path length $l = 1$ cm. The absorption measurements have been performed at room temperature and at wavelength $\lambda_{max} = 545$ nm. Stock solutions of Gd(III) nitrate and Chrome Azurol S (H_4Ch), prepared with concentrations of respectively 10^{-2} M and 10^{-3} M. In a typical experiment, a solution ($v = 50$ mL) of the colored sacrificial complex (GdH_4Ch) was prepared

$$\left(\left[Gd^{3+} \right] / \left[H_4Ch \right] = 1.5 \right),$$

using as a buffer of MESH (0.1 M) [2-(N-morpholino) sulphonic ethane acid]. The initial solution also contained a calculated amount of 1 M NaOH in order to obtain the desired pH value $pK_{a(MESH)} = 6.20$, experimental pH range (5.5 - 7.5). pH values are measured with a Microprocessor pH Meter HANNA 210 equipped with a combined glass electrode and calibrated with commercial buffers (pH 4.00 and 7.00). Then aliquots ($v = 0.100 - 0.200$ mL) of an aqueous solution of the malic acid ($C_L = 2$ g/L) were added, using a Gilson micropipet of 0.200 mL. After each addition, the resulting solution was left at least 5 min in order to reach equilibrium (and thus a constant absorption value). Addition was repeated until a maximum volume of 2.00 mL of the organic acid solution was added. The change in the total volume was neglected. For fixed pH environments, the apparent formation constants $\log K'_{xyz}$ was calculated using a laboratory made computer program. The absorption values for the undissociated (A_f) and the totally dissociated (A_r), sacrificial complex (GdH_4Ch), are used as determined in experiments using pure Chrome Azurol S (H_4Ch), and an excess of Gadolinium(III) (performed at pH intervals of 0.20). Assuming various integers for the Gadolinium and organic acid stoichiometry, a formation constant is calculated for each added amount of ligand and corresponding absorption value. The results are rejected when a systematic variation of $\log K'_{xyz}$ occurs with increasing added amount of ligand or when individual values of $\log K'_{xyz}$ differed from the mean value by more than 2%.

2.3. IR Spectroscopy

Samples were prepared by weighing the appropriate amount of malic acid (ligand) and $Gd(NO_3)_3$, adding H_2O , mixing and finally adjusting the pH with concentrated HCl or NaOH and concentrations of analyzed samples are 10^{-3} M. Analyses were performed using an infrared spectrometer, Fourier transform (FT-IR), Perkin Elmer BX, equipped with a DTGS detector, a splitter and a cesium iodide window. In this configuration, the interval of analysis is that the middle infrared, 6000 cm^{-1} to 250 cm^{-1} and analysis are conducted on small samples, whose size is less than 1 mm^3 . Liquid samples are placed between two plates of very pure salt (KBr), these plates are transparent to infrared light and the spectra relative to free ligand (malate ion) and complexed, have been plotted for frequencies from 4400 cm^{-1} to 400 cm^{-1} .

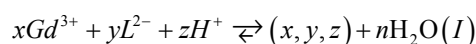
2.4. Raman Spectroscopy

The complex is precepts quickly at room temperature, at a concentration of 10^{-2} M and fixed pH value (5.86) (Higher than the acidity constant of malic acid). The precipitate was filtered and dried in drying oven and the complex formed is insoluble in water, methanol and ethanol. The Raman spectra of the ligand (Malic acid) and its new Gd(III) complex was performed using an Fourier transformation Raman spectrometer (FT-Raman) VERTEX 70 with a range of measurement ($4000 - 50$) cm^{-1} , laser source NdYag (1.064 μm), a nominal power of 500 MW, detecting Ge with high sensitivity and a resolution of 4 cm^{-1} (64 scan). The spectra relative to free malic acid and Gd(III)-carboxylate complex, have been plotted for frequencies from 3600 cm^{-1} to 200 cm^{-1} .

3. Results and Discussion

3.1. Indirect Photometric Detection Study

The complexation reaction of x gadolinium ions (Gd^{3+}) with y malate ions and z protons (H^+), is given by expression (I):



L^{2-} represents the ligand (malate ion).

The formation constant K_{xyz} (or stability constant (β_{xyz})) of the complex is defined as the equilibrium constant:

$$K_{xyz} = \left[(x, y, z) \right] \cdot \left[Gd^{3+} \right]^{-x} \cdot \left[L^{2-} \right]^{-y} \cdot \left[H^+ \right]^{-z} \quad (1)$$

Additionally, a conditional equilibrium constant K'_{xyz} is defined in case of constant pH value (buffered solution):

$$K'_{xyz} = \left[(x, y, z) \right] \cdot \left[Gd^{3+} \right]^{-x} \cdot (C_L)^{-y} \quad (2)$$

C_L represents the analytical concentration of the un-

complexed ligand, all experiments are performed for pH values higher than pK_{a1} and pK_{a2} of malic acid, therefore, we have $C_L = [L^{2-}]$. Using this equality, Equation 1 can be written as:

$$K_{xyz} = [(x, y, z)] \cdot [Gd^{3+}]^x \cdot (C_L)^{-y} \cdot [H^+]^{-z} \quad (3)$$

$$= K'_{xyz} [H^+]^{-z}$$

$$\text{Thus } \log K_{xyz} = \log K'_{xyz} + z \cdot pH \quad (4)$$

The complexation of the ligand can be studied using a spectrophotometric method. Since the reagent (malic acid) and its detected gadolinium complex do not possess a characteristic UV-visible absorption spectrum, a second ligand (called the sacrificial ligand) is introduced. This second ligand must absorb in the UV-visible spectrum and form a colored complex with Gd^{3+} ions. Based on ligand-ligand displacement, the photometric method is said to be in the indirect mode. The dissociation of this colored complex has to cause large variations in the UV-visible spectrum which allow the calculation of the concentration of the sacrificial complex. Using the formation constant of this colored complex, the concentration of the unknown complex can be obtained. Therefore, the sacrificial ligand must form a single colored complex of lower stability than the gadolinium complex under study. In this work, 3''-sulfo-2'',6''-dichloro-3,3'-dimethyl-4'-hydroxy-fuchsone-5,5'-dicarboxylic acid, often called **Chrome Azurol S** and noted H_4Ch , has been used as sacrificial ligand. Chrome Azurol S is a tetraprotic acid with pKa values of 2.25 (H_3Ch^-/H_2Ch^{2-}), 4.71 (H_2Ch^{2-}/HCh^{3-}) and 11.82 (HCh^{3-}/Ch^{4-}) [13]. In the experimental pH range (5.5 - 7.5), the formation of H_4Ch , H_3Ch^- and H_2Ch^{2-} has been neglected. It is an indicator which is generally used for the photometric proportioning of the metal ions in solution [14,15]. The interaction of the H_4Ch with gadolinium ions (Gd^{3+}), gives a coloured reagent ($\lambda_{max} = 545$ nm) of average stability for values of pH ranging between 5.50 and 7.50. The buffer "MESH". [2-(N-morpholino) sulphonic ethane acid] was adopted to fix pH in the study of the sacrificial complex $Gd-H_4Ch$ and the detected gadolinium-malate complex. We chose this buffer because it does not present any interaction with Gd^{3+} ions and so that we would be able to work in range of pH where the stability of the sacrificial complex is maximal.

3.2. Formation of the Colored Sacrificial Complex $Gd_x(HCh)_y$

A series of experiments was conducted to determine, the composition, the stability of the colored sacrificial complex ($Gd_x(HCh)_y$) and its formation reaction. Indeed, in 50 cm^{-3} of the auxiliary ligand solution (HCh^{3-}) of a concentration $C_{HCh} = 10^{-4} \text{ M}$ and $pH = 5.91$, we in-

troduced increasing quantities of a Gd^{3+} ions solution of initial concentration 10^{-2} M . The evolution of UV-visible spectra for the formation of this sacrificial complex is represented by the diagram in following **Figure 1**.

The UV-visible spectra show clearly that the maximum absorbance of the sacrificial complex is located at $\lambda_{max} = 545 \text{ nm}$. So, we have studied the formation of this sacrificial complex at this maximum wavelength, depending on the ratio

$$q = (\text{metal/ligand}) = [Gd^{3+}] / [HCh^{3-}]_{total}$$

as shown in the **Figure 2**.

The curve in **Figure 2** indicates that the value of the ratio q , relative to the complete formation of this sacrificial complex is the intersection of the linear (positive slope), with the final segment (slope = 0) of the curve, this value of q is close to 1.50. This indicates that the

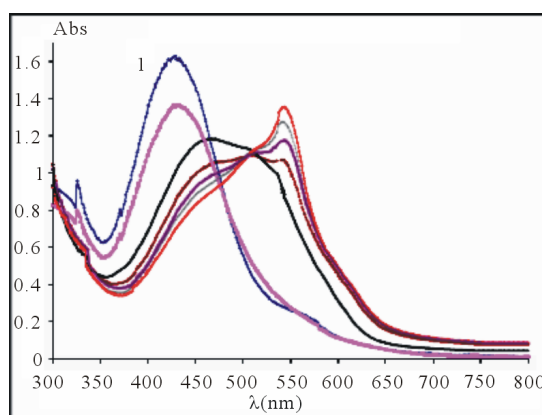


Figure 1. UV-visible spectra for the formation of sacrificial complex $Gd_x(HCh)_y$, $pH = 5.91$, $[HCh^{3-}]_{total} = 10^{-4} \text{ M}$ and $[Gd^{3+}]_0 = 10^{-2} \text{ M}$, $l = 1 \text{ cm}$, Curve (1) free CAS.

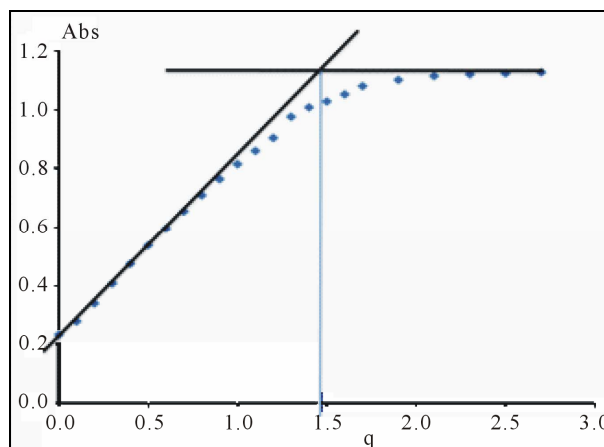
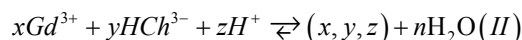


Figure 2. Formation of the sacrificial complex, depending on the ratio $q = [Gd^{3+}] / [HCh^{3-}]_{total}$, buffer $MESH^-/MESH$, $pH = 5.70$, $\lambda_{max} = 545 \text{ nm}$, $l = 1 \text{ cm}$, $[HCh^{3-}]_{total} = 10^{-4} \text{ M}$.

formation of the sacrificial complex is according to the following balanced reaction:



$$1.5C_{HCh} \quad C_{HCh} \text{ fixed } 0$$

$$C_{HCh}(1.5 - x\alpha) \quad C_{HCh}(1 - x\alpha) \quad \alpha C_{HCh}$$

C_{HCh} represents the analytical concentration of ligand HCh^{3-} in all its forms, z the number of protons involved during the reaction, while C_{HCh} is the coefficient of complex formation, calculated from the experimental values of absorbance by the relation

$\alpha = (A - A_f) / (A_f - A_i)$, with A_i and A_f which respectively represent the initial absorbance (free ligand) and final (100% of the complex).

Therefore, the expression of the apparent constant on the formation reaction according to the balanced reaction (II) is given by the following equation :

$$K'_{x,y,z} = [(x, y, z)] \cdot [Gd^{3+}]^x \cdot (C_{HCh})^y \quad (5)$$

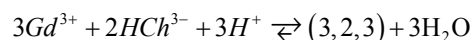
$$= \alpha (C_{HCh})^{1-x-y} / (1.5 - x\alpha)^x \cdot (1 - y\alpha)^y$$

$K'_{x,y,z}$ is calculated using a computer program written from the balanced reaction (II), we obtained constant values of K'_{32z} for the pair ($x=3, y=2$) for all experimental points, with a value of $\log K'_{32z} = 7.40$ at $pH = 5.91$. Under the same conditions of temperature and concentrations, several experiments were performed for pH values between 5.50 and 7.50. The same calculation program was used and the results clearly indicate that only the detected sacrificial complex (3,2,z), is formed in solution. For all studied solutions at known values of pH, the apparent constants K'_{32z} were calculated and the obtained results are summarized in the **Table 1**. These results show that the stability of the sacrificial complex depends on the acidity of the medium.

For all these solutions, the formation constant K'_{32z} on the sacrificial complex is given by the relationship $K'_{xyz} = K'_{32z} / [H^+]^z$. Hence $\log K_{32z} = \log K'_{32z} + z \cdot pH$, (K'_{32z} conditional constant for a given pH), we plot the curve $\log K'_{32z}$ depending on the medium pH (**Figure 3**), and the slope value is the equivalent number of H^+

ions that react, with 1.5 equivalents of Gd^{3+} ions and 1 equivalent of HCh^{3-} ions (composition adopted to make simple calculations). The linear function shown in **Figure 3**, clearly verifies the accuracy of the studied relationship, and its slope $p = 1.50$, calculates the equivalent number of hydronium ions, involved in the formation reaction of the detected gadolinium complex, and hence determines its exact composition (3,2,3), whereas the intercept expresses the stability constant of this sacrificial complex, with $\log K_{xyz} = 16.27$ at V.

Thus, the complexation of Gd^{3+} ions by HCh^{3-} ions in the pH range 5.50 to 7.50, led to the formation of the colored tri-nuclear complex, by following balanced reaction:



The formation constant of this (3,2,3) complex is defined as:

$$K_{323} = [(3, 2, 3)] \cdot [Gd^{3+}]^{-3} \cdot [HCh^{3-}]^{-2} \cdot [H^+]^{-3} \quad (6)$$

The conditional equilibrium constant for a fixed value of pH is given by:

$$K_{323} = [(3, 2, 3)] \cdot [Gd^{3+}]^{-3} \cdot (C_{HCh})^{-2} \quad (7)$$

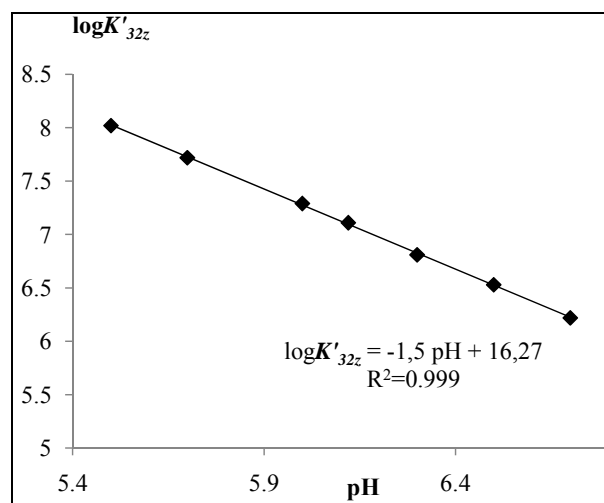


Figure 3. The stability of the tri-nuclear complex Gd_3HCh_2 , depending on the medium acidity.

Table 1. Evolution of the sacrificial complex stability, depending on the acidity of the medium.

pH	5.50	5.70	5.90	6.00	6.12	6.30	6.50	6.70
$\log K'_{32z} \pm 0.05$	8.02	7.72	7.40	7.29	7.11	6.81	6.53	6.22
A_i	0.358	0.253	0.301	0.388	0.225	0.295	0.285	0.265
A_f	1.0605	1.179	1.316	1.466	1.616	1.666	2.704	2.826
$\varepsilon = (M^{-1} \cdot cm^{-1})$	11000	12000	13000	15000	16000	17000	27000	28000

3.3. The Composition and Stability Study of the System: Gd(III)-Malic Acid

If the studied ligand (malate ions), is added to a colored solution of the sacrificial complex (3,2,3), this chrome azurol S (HCh^{3-}) complex will dissociate. To calculate the complexed concentration of the HCh^{3-} ions, the absorption values of the totally complexed A_F , and completely dissociated HCh^{3-} ions have to be determined A_I . Then the following equations can be used:

$$[(3,2,3)] = (C_{HCh})_0 \cdot (A - A_I) / (A_F - A_I) \quad (8)$$

$$(C_{HCh})_{free} = (C_{HCh})_0 \cdot (A - A_I) / (A_F - A_I) \quad (9)$$

When the conditional equilibrium constant K'_{323} of the sacrificial complex is known, the concentration of free gadolinium ion ($[Gd^{3+}]$), can be calculated using Equations (7)-(9).

Knowing $[(3,2,3)]$ and $[Gd^{3+}]$, the concentration of the gadolinium complex under study $[(x,y,z)]$ (balanced reaction I) can be determined using the gadolinium mass balance equation :

$$x \cdot [(3,2,3)] = C_{Gd} - [(3,2,3)] - [Gd^{3+}] \quad (10)$$

C_{Gd} being the initial gadolinium concentration. In a similar way, the concentration of the free ligand is obtained by

$$(C_{L^{2-}})_{free} = (C_{L^{2-}})_0 - y[(x,y,z)] \quad (11)$$

It should be also noted that a perfect knowledge of the characteristics of the sacrificial colored complex Gd_3HCh_2 is necessary. The determination of the composition and the stability of the sacrificial complex and the precision of the conditions of its formation are paramount stages to apply the indirect photometry technique. In each experiment, the ligand (malate ions), is added stepwise in order to measure the absorption at different values (at least 12) of the overall initial concentration of this studied ligand. The correct K'_{xyz} is looked for by varying x and y in order to obtain a constant value for all values of $C_{L^{2-}}$. If K'_{xyz} is determined at different pH values, the slope of the $\log K'_{xyz} \cdot pH$ plot reveals the number z of protons, necessary for the formation of the studied gadolinium complex by use of equation 4, since the value of K'_{xyz} is independent of pH. Now the stability and the total composition of the gadoliniummalate complex have been determined.

3.4. Determination of the Composition and the Stability Constant of the Gd-Malate

With an aim of determining the composition and the stability constant of the gadolinium-malate complex, we

monitored the evolution of the absorbance during disappearance of the sacrificial complex by addition of increasing quantities of malate ions. For a given volume (50 ml) of a solution containing sacrificial complex (3,2,3) 10^{-2} M, we added increasing quantities of malate ions solution, with known concentration. The spectrophotometric study carried out with fixed wavelength ($\lambda_{max} = 545$ nm), showed a reduction in the absorbance of the solution progressively with the addition of the malate ions (Figure 4). The dissociation of the sacrificial complex, relating to the reduction in the absorbance by the addition of malate ions solution, is done in favour of the formation of the colourless complex between Gd^{3+} and malate ion species.

The curve C.2 in Figure 4, clearly shows that the absorbance decreases and stabilizes. This stability indicates that all Gd^{3+} ions, initially present in the solution have reacted with added malate ions. Knowing the concentration of gadolinium ions and the quantity of ligand from the added volume of the malic acid solution, necessary to reach this stage of absorption, we could determine the molar ratio q ($Gd^{3+}/malate$), involved in the complexation reaction. The preceding experiment was carried out for different values of pH between 5.50 and 7.50, the way in which the absorption decreases, depends on the pH of the medium and on the formation constant of the detected complex (Gd-malate), as well as on the absorption values (A_I) and (A_F) of the free and totally complexed Chrome Azurol S (HCh^{3-}). Analyzing the experimental data with the computer program written from the balanced reaction (I), the results confirm the reproducibility of the molar ratio q and to determine the composition and apparent stability constant K'_{xyz} (Table 2). The data-processing treatment of the preceding experimental results, shows that this tri-nuclear detected complex

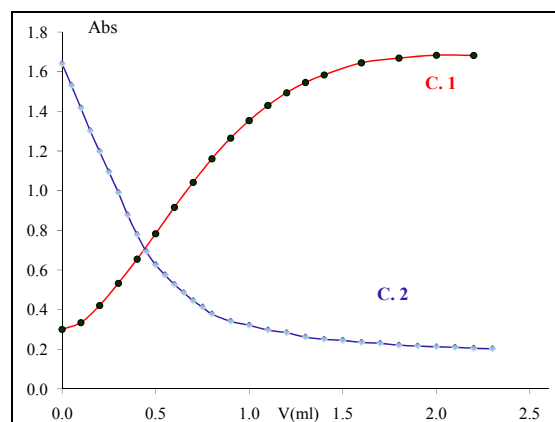


Figure 4. C.1 : Formation of colored sacrificial complex $Gd_3(HCh)_2$; C.2 : Decomposition of this complex by addition of a malic acid solution (2g/L), $pH = 6.50$, $[HCh^{3-}]_{total} = 10^{-4} M$, $q = [Gd^{3+}] / [HCh^{3-}] = 3/2$.

Table 2. The apparent stability of the tri-nuclear detected complex, depending on the acidity of the medium.

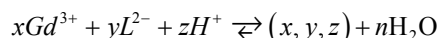
pH	5.63	5.80	6.11	6.30	6.54
$\log K'_{32z}$	20.21	19.86	19.26	18.88	18.38

formed between the gadolinium ions and malate ions, resulting from the interaction of **three** Gd^{3+} ions equivalents with **two** equivalents of malate species, so a molar ratio $q = x(Gd)/(malte) = 3/2$.

Buffer $MES^-/MESH$, $\lambda_{max} = 545 \text{ nm}$, $l = 1 \text{ cm}$,

$$[HCh^{3-}]_{total} = 10^{-4} \text{ M}, [Gd^{3+}]_0 = 10^{-2} \text{ M}$$

For all pH range we obtained ($x = 3, y = 2$), therefore, for the complexation reaction of Gd^{3+} ions with malate ions at this pH range, these results and the Equations (1)-(3) allow to write the following expressions:



The value of z can be positive, negative or null. The stability constant of this formed complex is defined by:

$$K_{32z} = [(3, 2, z)] \cdot [Gd^{3+}]^{-3} \cdot [L^{2-}]^{-2} \cdot [H^+]^{-z}$$

Conditional stability constant K'_{32z} is defined in case of constant pH value (buffered solution):

$$K'_{32z} = [(3, 2, z)] \cdot [Gd^{3+}]^{-3} \cdot (C_L)^{-2}$$

For these experimental pH values higher than the pK_{a1} and pK_{a2} of malic acid, $C_L = [L^{2-}]$ thus:

$$\begin{aligned} K_{32z} &= [(3, 2, z)] \cdot [Gd^{3+}]^{-3} \cdot (C_L)^{-2} \cdot [H^+]^{-z} \\ &= K'_{32z} \cdot [H^+]^{-z} \end{aligned}$$

And $\log K'_{32z} = \log K_{xyz} - z \cdot pH$

The evolution of $\log K'_{32z}$ at different pH values is represented on **Figure 5**, this evolution is linear and the slope of the straight line is equal to -2 . The value ($z = 2$) represents the number of protons involved in the formation reaction of this new tri-nuclear complex (3,2,3)

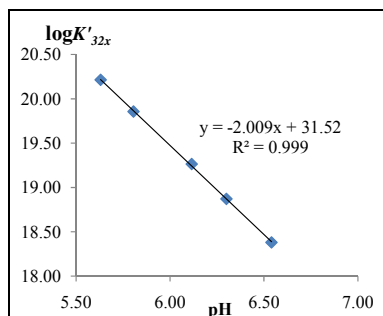


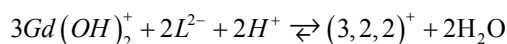
Figure 5. Evolution of $\log K'_{32z}$ for trinuclear Gd(III)-malate complex, at different pH values.

(Gadolinium-Malate) species.

Consequently

In the light of the spectrophotometric results relating to the interaction of Gd^{3+} ions with the malate ions, namely that the complexation reaction uses three hydrolyzed Gd^{3+} ions for two malate species and requires fixation of two protons.

According to the literature [16], the Gd^{3+} ion can be presented in various hydrolyzed forms in aqueous solution and this **new** tri-nuclear gadolinium complex detected in solution at experimental pH range, is probably formed from the hydrolyzed form $Gd(OH)_2^+$, according to the following reaction:



In order to confirm our results, to have more information on the nature of this **new** gadolinium complex and likely to propose a probable structure for this tri-nuclear specie, we carried out IR spectroscopy investigations. These studies by IR spectroscopy carried out in the same pH range but with more concentrated solutions, help to identify the nature of the chelation sites and the probable structure of this **new** gadolinium complex formed by interaction of malate ions with this hydrolyzed form of Gd^{3+} ions.

3.5. IR Spectroscopy Investigations

The IR spectroscopic studies can identify different groups of malic acid (ligand), which participate in chelation sites for the formation of the detected gadolinium complex. Three dilute solutions (10^{-1} cm), respectively, the free malate ion ($pH = 5.60$), free malic acid ($pH = 2.02$) and detected gadolinium complex of malate ion ($pH = 5.60$), were prepared and their spectra recorded (**Figure 6**) and analyzed. **Table 3** contains bibliographic data [17, 18], relative to vibrations intervals of various groups: OH, C=O, COO^- and C-C=O, and the intervals of vibration on the analyzed spectra (1, 2 and 3).

The experimental spectra obtained for the three analyzed samples, clearly indicate that the frequency for vibration of the groups: OH, C=O and COO^- , have

Table 3. The vibration frequencies, for the studied spectra and bibliographic data.

Groups	Experimental spectra	Ref. [17,18]
	$\nu \text{ (cm}^{-1}\text{)}$	
O-H	3300 - 3600	3650 - 3500
C=O	1620 - 1680	1800 - 1700
COO^-	400 - 900	590 - 700
C-C=O		465 - 550

seriously reduced 75%, 80% and 66% passing the free to complexed ligand (**Figure 6**). So, for each of the two malate ions involved in the formation of this detected tri-nuclear complex, the four oxygen atoms of the ionized carboxylic groups, participate in chelation sites, and the OH group in α position of the ionized group COO^- .

3.6. FT-Raman Spectroscopy Investigations

FT-Raman spectra of free malic acid and its complex with Gadolinium ions Gd^{3+} , are shown in **Figure 7**. A detailed analysis of vibrations in Raman spectroscopy was performed on the basis of comparison of experimental vibrational spectra obtained of malic acid and its gadolinium complex. The Raman spectra obtained show that the OH vibrations $3350 \pm 104 \text{ cm}^{-1}$ [12,19] and the free malic acid present two vibrations of C=O at 1677 cm^{-1} and 1635 cm^{-1} because the malic acid present two carbonyl functions aren't the same environment [19,20], these vibrations of OH and C=O groups, were not detected in the complex spectra and the vibration of C-C=O group is reduced from free malic acid to its gadolinium complex. So, all oxygen atoms involved in the chelation sites of this new tri-nuclear complex.

The vibration spectra (weak broad) of the complex in 1600 cm^{-1} , indicate the existence of water molecules [21], the intense vibration observed in the complex near 1069 cm^{-1} indicate the existence of the nitrate ions [12]. The new band vibrations related to Gd(III)-Oxygen carboxylate functions, are located at 600 and 547 cm^{-1} with a

weak bands [21,22], and the strong vibration of Gd(III)-oxygen nitrate appeared at 187 cm^{-1} [23]. The different vibrations (stretching and deformation) of C-C (aliphatic chains) appeared in a long field ($1400 - 400 \text{ cm}^{-1}$) with intense and medium intensities [21,20,24]. Bands in the $2590 \pm 80 \text{ cm}^{-1}$ region were assigned to the vibrations of CH and CH_2 [25,26], these bands are moderate intensities in complex spectra

These results clearly indicate that, this new tri-nuclear gadolinium complex contains two types of sites, a single central bidentate mono-nuclear site with participation of only OH groups in α position and two lateral tetradentate mono-nuclear sites, each consisting of four oxygen atoms of two ionized carboxylic functions, belonging to the two malate ions ($^-\text{OOC-CHOH-COO}^-$), involved in the formation of this new detected gadolinium organometallic complex. Indeed, all these results can offer for this tri-nuclear *Gd-Malic acid*, the structure presented in **Figure 8**.

4. Conclusion

In this work, we used some techniques to study the interactions of the trivalent Gd(III) ions with different ionic forms of malic acid and identify the composition, stability and structure of the major colourless complex, formed in solution for pH values between 5.50 and 7.50. The photometry in indirect mode (IPD) was used successfully to determine the composition and the stability of this major gadolinium complex. Only the tri-nuclear

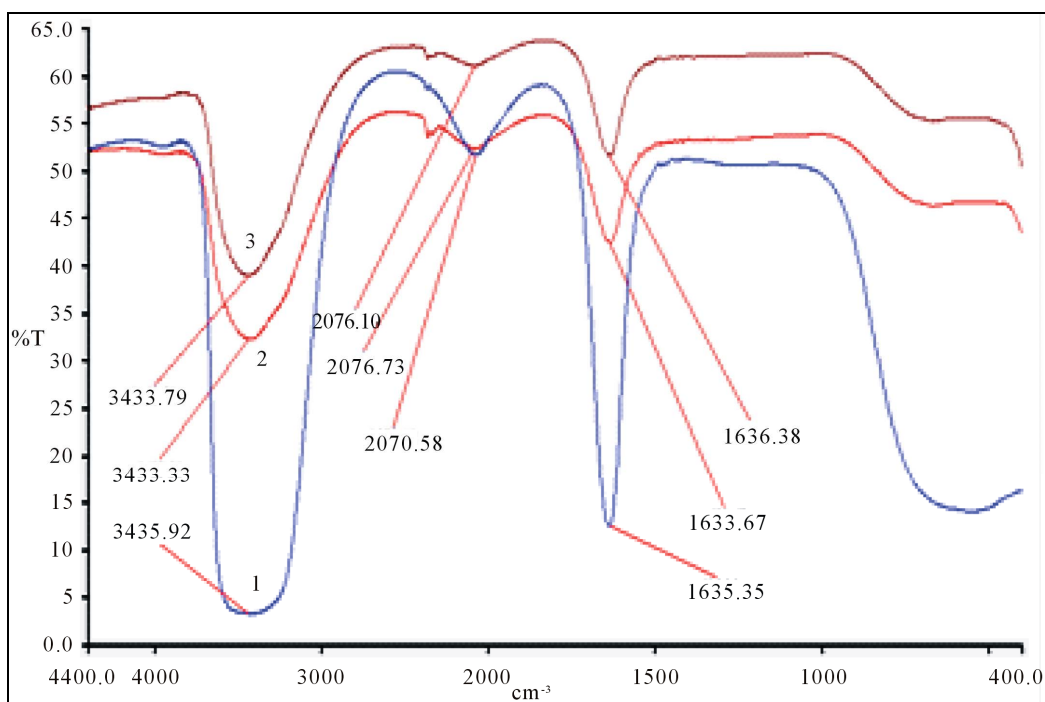
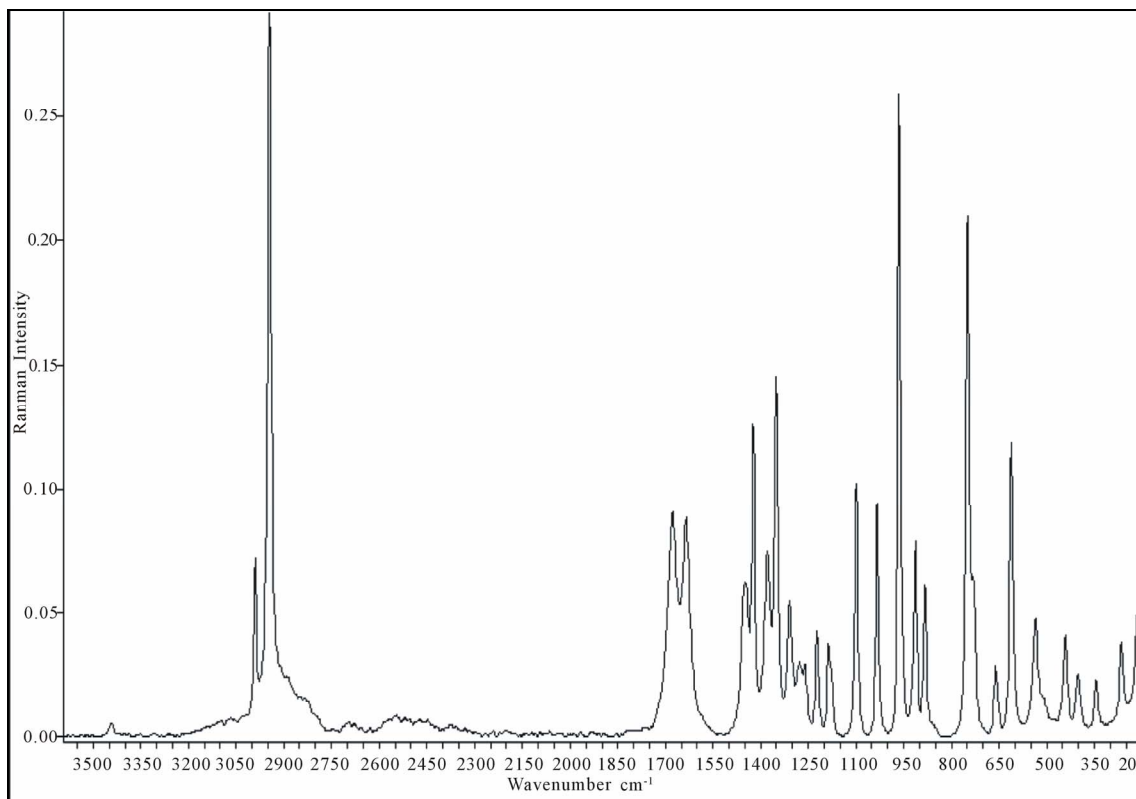
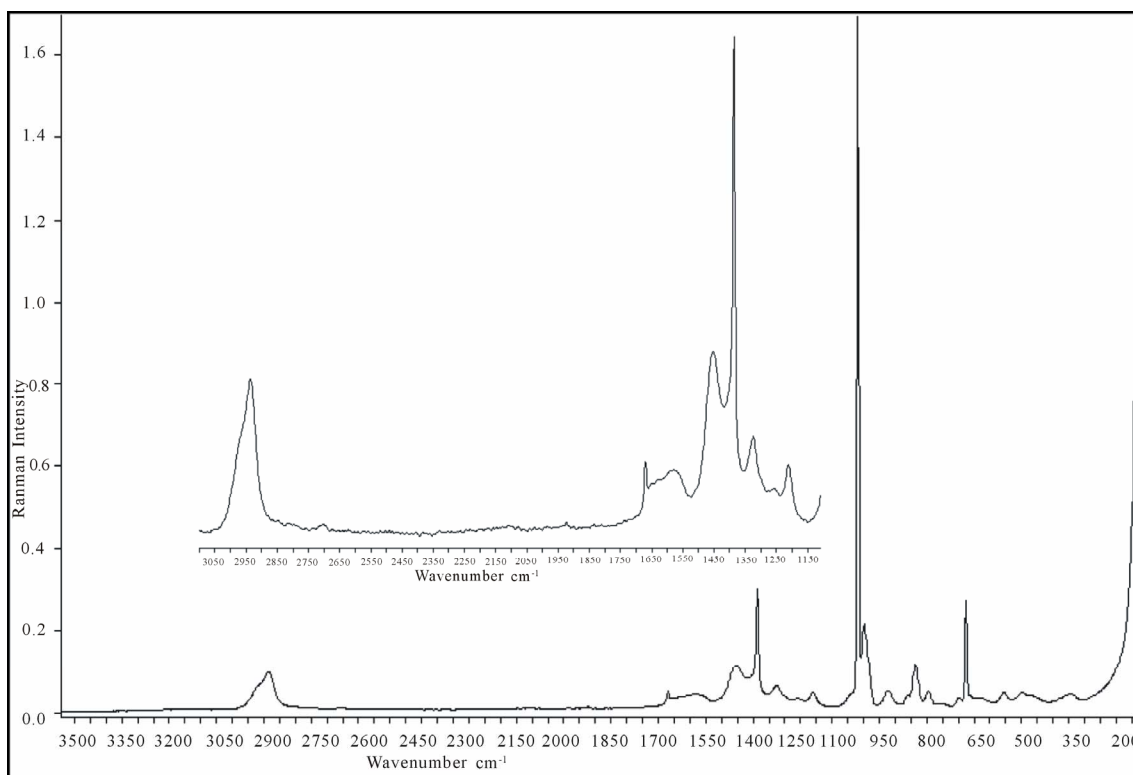


Figure 6. IR spectra, (1) free malate ion 10^{-3} M ; (2) free malic acid 10^{-3} M ; (3) Gd-malate, $[\text{Gd}^{3+}]/[\text{L}^{2-}] = 3/2$.



(a)



(b)

Figure 7. Experimental FT-Raman spectra in the range (3600 - 200) cm^{-1} : (a) free Malic acid; (b) Gd(III)-Malate , $[Gd^{3+}]/[L^{2-}] = 3/2$.

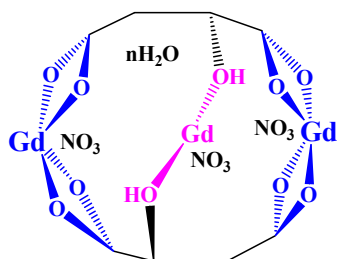


Figure 8. Probable structure for the new trinuclear gadolinic complex $Gd_3Malte_2(NO_3)_3 \cdot nH_2O$.

complex with a composition (3,2,3) and a high stability $\log_{322} = 31.52 \pm 0.05$, has been identified for the interval of studied pH. The results of studies on IR and FT-Raman spectroscopy, clearly show that in this trinuclear complex type of Gd^{3+} ions, all oxygens of the two ionized carboxylic functions COO^- of malate ion, and the oxygen atom of OH group in the α position, are involved in chelation sites, while, studies conducted by Hlaïbi *et al.* [8,9,27,28] show, that for the tungstic complexes of α -hydroxyl carboxylic acids, carbonyl function C=O is excluded. All these results also indicate, that the combination of these two techniques is very effective for identification and characterization of colourless organometallic complexes of Gd^{3+} ions, in fact, is the first time that this tri-nuclear complex was revealed, that its stability has been determined and a probable structure was proposed

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Abbreviations

IPD: Indirect Photometric Detection
MRI: Magnetic Resonance Imaging,
H₄Ch: Chrome Azurol S,
MESH: 2-(N-morpholino) sulphonic ethane acid.
A_I and **A_F**: The absorption values for the undissociated and the totally dissociated sacrificial complex.
K'_{xyz}: The apparent formation constant
K_{xyz}: The formation constant (or stability constant)

L²⁻: The ligand (malate ion)
C_L: The analytical concentration of the ligand
C_{HCh}: The analytical concentration of ligand **HCh³⁻** in all its forms
C_{Gd}: The initial gadolinium concentration
Gd₃HCh₂: The sacrificial colored complex
q: The molar ratio (gadolinium/malate ion).