Article

Chemical Equilibrium in the Complexation of First Transition Series Divalent Cations Cu²⁺, Mn²⁺ and Zn²⁺ with Chitosan

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O presente estudo forneceu dados para o cálculo das constantes de estabilidade formadas em um sistema aquoso e HCl contendo quitosana e cátions Cu²⁺, Mn²⁺ e Zn²⁺. Os diagramas de espécies foram gerados para demonstrar como o pH influencia esses equilíbrios estudados por titulações potenciométricas e espectrofotométricas. Os sólidos extraídos dessas soluções aquosas em HCl foram estudados por espectroscopia no infravermelho. A quitosana complexa-se com os cátions Cu²⁺, Mn²⁺ e Zn²⁺ a pHs >4 >5 e >2 respectivamente, na razão ligante:metal, 1:1. As constantes de estabilidade calculadas para ML apresentaram a seguinte ordem de estabilidade: Cu²⁺>Zn²⁺>Mn²⁺. Algumas estruturas para os possíveis complexos formados foram sugeridas envolvendo ambos os átomos de N e/ou O como sítios quelantes de uma ou duas unidades monoméricas do biopolímero estudado.

This study provided the data for calculating the stability constants formed in the aqueous HCl equilibrium of chitosan and Cu²⁺, Mn²⁺ and Zn²⁺, and determined the species distribution diagrams to show the influence of pH in the complexation systems based on data obtained from potentiometric and spectrophotometric titrations. The solid complexes further obtained from these aqueous HCl systems, were investigated by infrared spectroscopy. It was verified that chitosan coordinated with the cations Cu²⁺, Mn²⁺ and Zn²⁺ at pHs >4 >5 and >2 respectively, in ligand to metal ratio of 1:1. The logarithms of the binding constants for ML species presented the following stability order: Cu²⁺>Zn²⁺>Mn²⁺. Some possible complexed structures were suggested having both N and/or O atoms as binding sites with one or two monomers of the biopolymer studied.

Keywords: chitosan, divalent cations, pH, stability constants, biodegradable and biocompatible polymers

Introduction

Chitosan (Chit) is a linear polysaccharide composed of β (1 → 4) linked 2-amino-2-deoxy-D-glucose and 2-acetamido-2-deoxy-D-glucose. It presents biodegradable and biocompatible properties, gelling ability, and can be shaped into several derivatives by further modifications of its amino and hydroxyl groups.[1]

Chit is obtained on an industrial scale by the alkaline deacetylation of chitin, one of the most abundant biopolymers in nature which can be extracted from crabs and shrimp shells, fungal biomass, insect cuticle or squid pen.[2]

The main parameters that are used for chitosan characterization are deacetylation fraction, molecular weight and cristallinity of the biopolymer. When the degree of deacetylation of chitin is higher than 50%, the so called chitosan becomes soluble in aqueous acidic media. Higher deacetylation degree, more than 95%, are high cost and are generally reserved for biomedical applications.[3,4]

Chit is of commercial interest due to its high percentage of nitrogen, amino and hydroxyl groups on their chemical structure which act as chelation sites for

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metal ions. So, in the literature, there are many reported works taking advantage of the complexing ability of chitosan and some of its derivatives with polyvalent metal ion to remediate various hazardous waste waters.[4-7]

Although these many works in the literature showing the complexing ability of chitosan[8-10] and stating the dependence of pH in the complexation, virtually none so far, have studied the chemical equilibrium measuring the binding constants and the pH dependence in the formation and destruction of the complexed species.[11,12]

In the complexation studies of chitosan and metal ions, different coordination mechanisms have been proposed. Some examples[6-14] are structures found, called "bridge model", where the metallic ion is bound to four nitrogen atoms of either the same or to different chains. Another model, "pendant model", considers the metallic ion linked to the amino group like a pendulum. This last model was confirmed by Domard[8] through theoretical calculations of functional density (DFT) and it was suggested that chitosan coordinates with divalent ions, like Cu^{2+} and Ni^{2+}, through the nitrogen of the amino group and indistinctly through the hydroxyl groups of C-3 and C-6 of some sugar unit of the biopolymer.[9]

Much is yet to be said on chitosan complexation ability since the two type basic sites, $-NH_2$ and $-OH$ are capable of binding metal ions. In a recent review[3] in the literature, chitosan is presented as mainly a $-NH_2$ chelating agent although the literature showed complexation with metal ions through $-OH$ in the sugar unit of this and other biopolymers.[11,12,15]

It is important to state that it is not possible to determine the exact pKa value for each amino and C-6 $-OH$ groups in chitosan since its complex structure, therefore a mean value can be obtained as a function of the degree of dissociation which in itself, is dependent on several values amongst which are the degree of deacetylation (DD), molecular mass (MM) and experimental conditions such as ionic strength (I), pH, temperature and solvent.[1] It is important then, to state a mathematical model for the use of microcomputer programs to calculate the binding and protonation constants of the biopolymer studied. According to the literature,[11,12,16-18] considering small parts of the biopolymer repeating units proved to be useful among other set conditions.

It is well known that at trace levels metal ions are essential nutrients for living organisms, but their lack or excess leads to adverse effects.[19]

Studies in the literature involved absorption of metal ions in chitosan,[20-22] but complete equilibrium investigations with determination of binding constants, as well as pH speciation, have only recently been addressed in the literature.[23-26] Also, the reported studies presented different experimental conditions from this present work.

The chelating ability of chitosan makes it a powerful metal ion remediating agent as well as having other applications in the chemical industrial processes. The study of the interaction between this biopolymer and metallic cations in different experimental condition is a relevant topic, though. In this sense, the main objectives of this work were to study the influence of the H^+ concentration in the solubility of chitosan, calculation of the binding constants and the speciation according to pHs in different ligand to metal ratios in HCl aqueous media systems. In that sense, the analytical tools of potentiometric and spectrophotometric titrations, and FTIR spectroscopy were also employed to help unravel the possible complexed structures formed.

Experimental

Reagents

Chitosan (75% degree of deacetylation, medium molecular weight, 200000D, Aldrich, Germany) was dissolved in a standard HCl solution (Merck, Brazil), ≈ 0.1 mol dm^{-3}. Metal ion solutions were prepared from reagent grade salts, $CuCl_2\cdot2H_2O$, $MnCl_2\cdot4H_2O$, $ZnCl_2$ (Merck, Germany) and were used without further purifications. Standardized (≈ 0.1 mol dm^{-3}) KOH aqueous solution (Merck, Brazil) was the titrant, 0.100 mol dm^{-3} KCl (Merck, Germany) was the ionic strength providing electrolyte. A 0.01 mol dm^{-3} solution of standardized Na_2EDTA was employed to standardize the metal ion solutions, and bi-distilled water free of CO_2 was used in all solutions.

Potentiometry

The potentiometric titrations were carried out in duplicate, according to the methodology developed in the Chemical Equilibrium Laboratory-LEQ-UFPR[24-26] for biopolymers, in a Micronal pH meter (Brazil, model B-374) with a glass electrode H_3O^+ sensitive (Analyser-Brazil, model SM01), and a saturated calomel reference electrode (Analyser-Brazil, model SR 02). The potentiometric apparatus was calibrated with standard HCl and KOH (Merck, Brazil) solutions in order to read -log $[H^+]$ directly. The pK_w used for water at I = 0.100 mol dm^{-3} was 13.78. The air in the reaction cell was purged by an inert N_2 atmosphere (White-Martins, Brazil), previously purified in two cleaning flasks containing solutions of

KOH 1.0 and 0.1 mol dm^{-3}. A thermostated bath maintained the temperature (25.0 ± 0.1 ºC, MQBTC 99-20, Microquímica, Brazil) and a piston burette (Metrohm AG Herisäu / E 274, Switzerland) was used to deliver the titrant solution (KOH, 0.1 mol dm^{-3}, 0.05 cm^{-3} increments of volume, standardized by potassium hydrogen phthalate (Carlo Erba, Italy). Solutions of 0.10 mmol of Chitosan in 0.485 mmol HCl were titrated in the absence and in the presence of previously standardized[27] metal solutions in ligand to metal ratios of 1:1 and 2:1.

The Hyperquad[28] microcomputer program was used to calculate the protonation and the binding constants for the complexed species found in percentage higher than 10% of total metal concentration in the equilibria. The mathematical model took into account the experimental average pH data from each potentiometric titration employed in triplicate, the hydrolysis constants for the metal ions, Cu^{2+}, Zn^{2+} and Mn^{2+},[29] the millimols of chitosan, calculated taking into account the dimeric repeating unit of the biopolymer (MM=344.3 g mol^{-1}). The dimeric unit was chosen to minimize any bias introduced in the calculations by the use of a low deacetylation degree chitosan. The calculations proceeded until the simulated curve matched the experimental one, thus minimizing the propagation errors in the experimental conditions employed. HySS (Hyperquad simulation and speciation)[30] program was used to calculate the species distribution diagrams using as input Hyperquad[28] data.

Ultraviolet-Visible spectroscopy

The measurements were carried out using an HP (8452 Diode Array Spectrophotometer), UV-Visible spectrophotometer, using quartz cells (1 cm), from 190 to 820 nm.

Previously acidified (HCl) solutions of chitosan and metal ions (both at 10^{-3} mol dm^{-3}) were obtained by mixing proper quantities of the reagents to obtain different ligand to metal ratios in pH values from 1.8 to 8.0 depending on the metal ion employed.

Fourier Transform Infrared Spectroscopy

The IR spectra were obtained in an FTIR spectrophotometer, BioRad (USA), with KBr (Merck, Brazil) pellets from 400 to 4000 cm^{-1}. The solid complexes were obtained from aqueous solutions of ligand to metal ratio of 2:1 freshly prepared and the pH set at 1.8 to 8.0 with KOH solution. The resulting solutions were placed in an oven (40 ºC, for 48 h). The solid samples thus obtained were washed repeatedly with absolute ethanol,
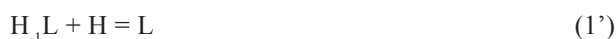
and placed once again in the oven at the same temperature for another 24 h. The resulting solid was ground and used in 1% m/m in the KBr pellets (total mass of 0.1000 g) made at 8 tons for 3 min.

Results and Discussion

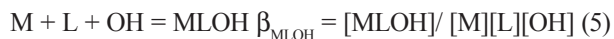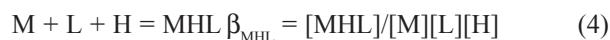*Potentiometric titrations and metal speciation*

The formation of the complexes can be described according to the Lewis acid-base theory, where an acid is the acceptor of electron pairs from a base. Chitosan (generally referred as L or Chit) is the Lewis base presenting –OH and –NH$_2$ basic sites, and Cu^{2+}, Zn^{2+} and Mn^{2+} are the acids (generally referred as M).

Equations 1 and 2 represent the protonation equilibria for the two potentially basic sites of the dimeric repeating unit of chitosan, the amino group –NH$_2$ and the hydroxyl of C-6, –OH. Equations 1' and 2' represent generically, the equations 1 and 2. The acidity of this C-6–OH group in other sugar-derived biopolymers was studied and determined previously:[23-26]

$$^-O\text{-Chit-NH}_2 + H^+ = HO\text{–Chit–NH}_2 \quad \log K_1 = 12.37 \pm 0.08 \quad (1)$$

$$H_{-1}L + H = L \quad (1')$$

$$HO\text{-Chit-} NH_2 + H^+ = HO\text{–Chit–NH}_3^+ \quad \log K_2 = 6.35 \pm 0.08 \quad (2)$$

$$L + H = HL \quad (2')$$

where H$_{-1}$L is the completely deprotonated sugar unit, L the chitosan dimeric unit with C-6 hydroxyl group protonated, and HL, the fully protonated species.

The complexed species with the overall stability constants associated with equilibrium in this work can be represented by the generic equations 3 to 5, charges omitted for simplicity.

$$L + M = ML \quad \beta_{ML} = [ML]/[L][M] \quad (3)$$

$$M + L + H = MHL \quad \beta_{MHL} = [MHL]/[M][L][H] \quad (4)$$

$$M + L + OH = MLOH \quad \beta_{MLOH} = [MLOH]/[M][L][OH] \quad (5)$$

The potentiometric equilibrium profiles are shown in Figure 1. Each curve represents a separate experiment and the shape gives qualitative information of equilibrium stoichiometry and gives suggestion of possible species present to start the binding constants calculation. The negative portion of the x axis represents the titration of the
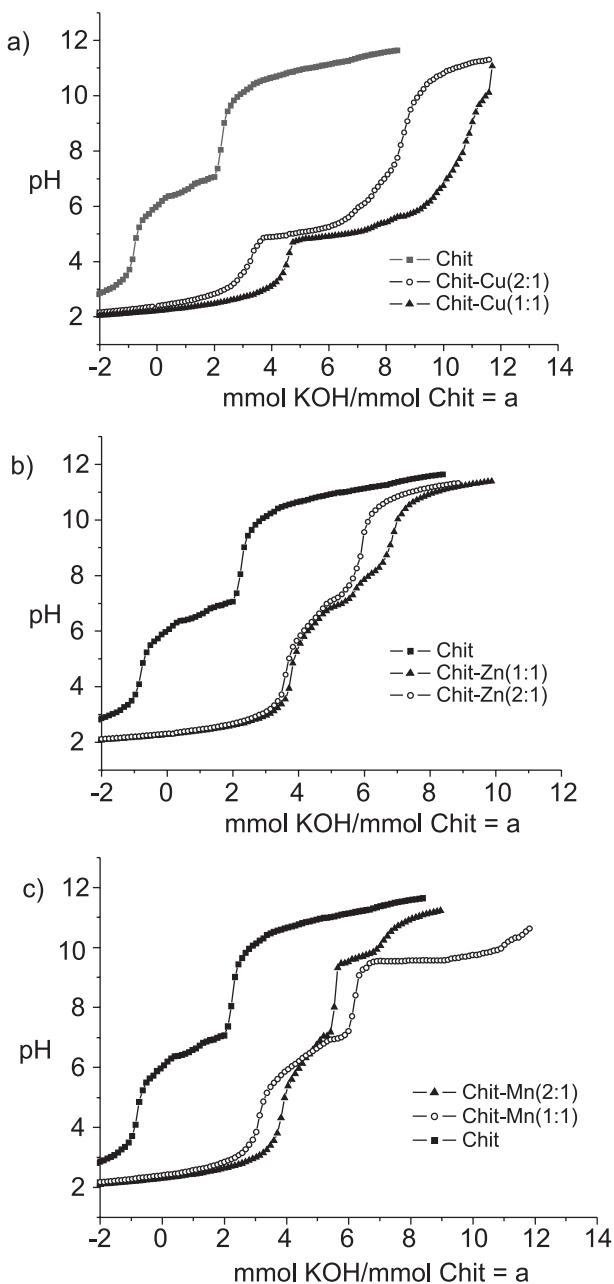
Figure 1. Potentiometric equilibrium curves for solutions containing 0.1 mmol of aqueous HCl chitosan and 0.1 and 0.05 mmol of the metal ion: a) Cu^{2+}; b) Zn^{2+}; c) Mn^{2+}; T = 25.0 ºC; I = 0.100 mol dm^{-3} (KCl).

mineral acid added to each experiment in order to prevent metal hydrolysis and to fully solubilize chitosan. The curve of the ligand in the positive portion of the x axis showed

the neutralization of chitosan from pH 6 to 11. Protonation constants for the dimeric sugar unit of chitosan were calculated from potentiometric data and were compared with the values reported in the literature for amino and primary alcohol groups in a generic sugar monomeric unit.

The potentiometric equilibrium profiles of chitosan in the presence of the metal ions studied (Figure 1) presented x axis displacement when compared to the profile of chitosan alone. The calculated equilibrium constants are given in Table 1. The species distribution curves for Cu^{2+}, Zn^{2+} and Mn^{2+} are presented in Figure 2, a, b and c, respectively. It can be seen that ML complexes of all metal ions studied appeared at pH values above 4. MHL complexes of Zn^{2+} were formed below pH 2 and the monohydroxo 1:1 ligand to metal complexes of all metals, appeared after pH 6. The late pH formation of all ML suggested that the formation of ML species probably happened through both possible binding sites of chitosan. Even knowing that C-6 hydroxyl group is to be deprotonated at high pHs, it may occur at lower values depending on the metal ions. On the other hand, the presence of MHL with Zn^{2+} showed that this metal ion was not capable to deprotonate the C-6–OH group. It was not possible to detect further species in the equilibrium with Zn^{2+}. The profiles were almost coincidental whatever the ligand to metal ratio employed.

In general, the ML order of stability found was: $Cu^{2+}>Zn^{2+}>Mn^{2+}$. This sequence maintained the trend in the well-known Irvin-Williams series.

Spectroscopy

The spectroscopic studies were carried out in the same ligand to metal ratios as employed in the potentiometric studies.

Ultraviolet-Visible spectroscopy

Previous work in the literature has showed that chitosan absorbs in the UV region, near 214 nm.[13]

Figure 3 shows the behavior of Cu^{2+}–chitosan system, where two bands appear in the ultraviolet region, one around ~ 206 nm and the other, between 249-270 nm. The first one was attributed to the free ligand and the second, to Cu^{2+}–chitosan charge transfer band (TCLM).[12,13,31] Although some authors suggest a 2:1 amino-copper ratio complex,

Table 1. Logarithms of calculated binding constants for chitosan (L) and Cu^{2+}, Zn^{2+} and Mn^{2+} (M) in HCl aqueous equilibria. T=25.0 ºC, I=0.100 mol dm^{-3} (KCl)

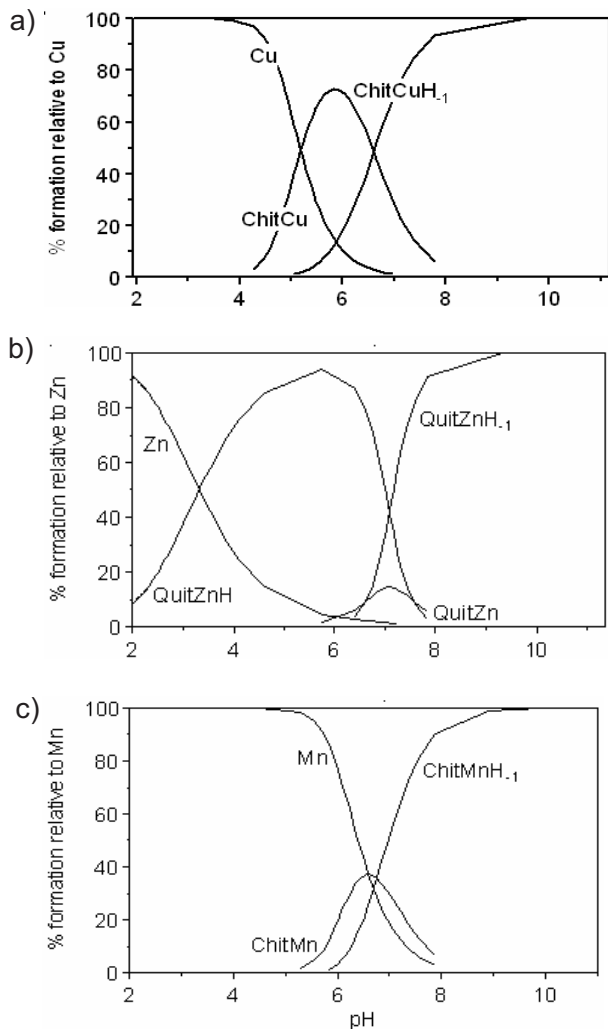| species	log K ± s.d	species	log K ± s.d.	species	log K ± s.d.
ChitH–Zn	7.5 ± 0.1	Chit–Cu	11.35 ± 0.06	Chit–CuH$_{-1}$	4.75 ± 0.06
		Chit–Zn	11.1 ± 0.3	Chit–ZnH$_{-1}$	4.5 ± 0.3
		Chit–Mn	9.3 ± 0.3	Chit–MnH$_{-1}$	2.6 ± 0.1

s.d.: standard deviation.

Figure 2. Species distribution curves of Cu^{2+}, Zn^{2+} and Mn^{2+}, aqueous HCl chitosan solutions, ligand to metal 1:1, with total metal concentration set at 100%. H_{-1} stands for OH^-.

Figure 3. UV-Vis spectra at various pHs of aqueous HCl chitosan solution and Cu^{2+}, ligand to metal ratio of 1:1.

based on the appearance of a second wide band in this region,[12,13] in this work it was associated to overlapping of bands of the $-O \rightarrow Cu^{2+}$ group in the hydroxyl groups (–OH) or due to water molecules in the metal coordination sphere.[32] A third much less intense d-d band ($^2T_{2g} \rightarrow {}^2Eg$) at 586 nm was identified. However, it was not possible to clearly identify the influence of N donor atom in the complexation of chitosan in this d-d transition band due to gelling of the solutions as concentration was increased.

The Zn^{2+}–chitosan system (Figure 4) presented a spectrum similar to that of the free polymer with only a band around 204 nm. The electronic configuration of Zn^{2+} presenting completely full "d" orbitals, does not allow the occurrence of d-d transitions and the configuration is extremely stable. Under the experimental conditions employed, the energy difference between the orbitals that give or receive electrons would be high in such a way that
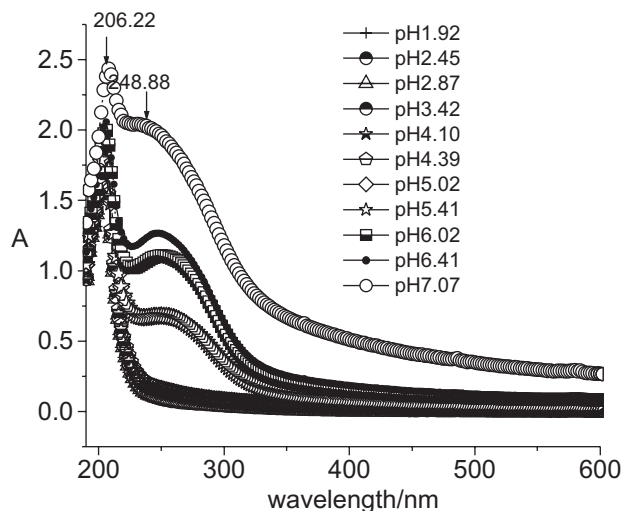
the absorption of the type ($TCLM–NH_2 \rightarrow Zn^{2+}$) would be displaced toward smaller wavelength, being able to overlap with the observed band.

The Mn^{2+}–chitosan system in the ultraviolet region (Figure 5) presented a spectral behavior similar to the one with copper, although the charge transfer band ($TCLM–NH_2 \rightarrow Mn^{2+}$) appeared as a shoulder around 306 nm at pH>3. As the shoulder extends up to 400 nm, associated bands like those arisen from $O \rightarrow Mn^{2+}$ group, were probably overlapped.[32] No bands were observed in the visible region as a consequence of semi-filled "d" orbital configuration of Mn^{2+}, implying prohibited electronic spin transitions. Isosbestic points were observed mainly at pH around 4.5 till 6.1, confirming the presence of the new species with the variation of pH.
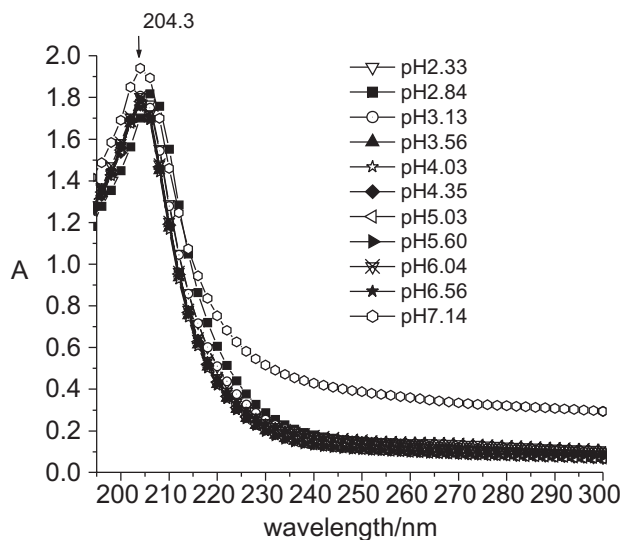

Figure 4. UV spectra at various pHs of aqueous HCl chitosan solution and Zn^{2+}, ligand to metal ratio of 1:1.
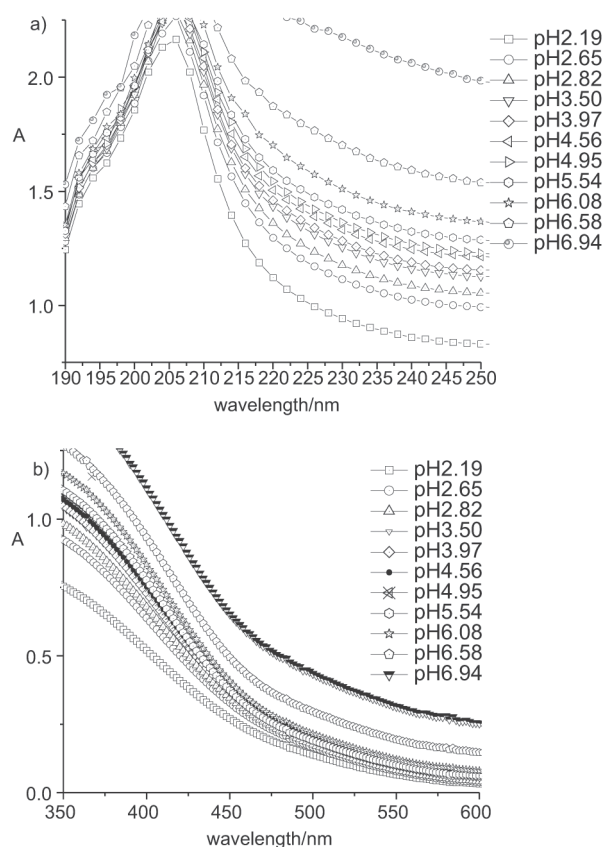
Figure 5. UV-Vis spectra at various pHs of aqueous HCl chitosan solution and Mn^{2+}, ligand to metal ratio of 1:1. a) 190 to 250 nm; b) 350 to 600 nm.

Figure 6. IR spectra of solid complexes of chitosan and Mn^{2+} extracted from aqueous HCl solutions at various pHs in ligand to metal ratio of 1:1.

Charge transfer bands from the ligand to the metal "TCLM" presented by complexes in this work, have similarities to those of certain dinuclear compounds of the studied metallic cations with chlorine atoms bridging two metallic centers. The bands for Cu^{2+} complex appeared at 265-420 nm,[33] for zinc at 260 and 316 nm[33] and for manganese, at 265-415 nm.[33,34] It can be reasonably supposed that these matching bands are due to the presence of structural units similar to the compounds obtained in this present work.

Fourier Transform Infrared Spectroscopy

The infrared spectra were obtained from pH 4 to 8 where the highest interaction was detected with same mass of samples.

All infrared bands presented different intensities and thus showed that the binding groups when the chit-metal complexes were formed have undergone different bonding as pHs changed. It has also been demonstrated that the spectrum of chit for the same pH interval showed little variation.[35]

The IR spectrum of chitosan presents ν_{OH} bands at 3453 cm^{-1}, ν_{NH} at 3345 cm^{-1}, ν_{CH} at 2923 cm^{-1}, ν_{C-O} at 1089 cm^{-1} [36] and when chitosan is not completely deacetylated, the

appearance of a ν_{CO} band at 1670 cm^{-1} and β_{NH2} at 1590 cm^{-1} has also been reported.[36,37]

The Cu^{2+}–chitosan system is presented in Figure 7. The region above 2700 cm^{-1} shows characteristic bands of the aliphatic groups, specifically CH_2 and CH, between 2880 and 2960 cm^{-1}, as well as that of OH of alcohol at 3445 cm^{-1} and of amine NH_2 at 3365 cm^{-1}. These last two groups showed a decrease of 8 and 20 units, respectively, toward higher wave numbers if compared to chitosan alone. In the case of Zn^{2+} (Figure 8) and Mn^{2+} (Figure 6) complexes, the shifts were 67 and 75 units, respectively. The three complexes also presented a sharp difference in the band of δ_{NH2} around 1600 cm^{-1}, the bending vibrations of NH groups, as the pH was steadily increased.

The region between 1020 and 1080 cm^{-1} presented alterations in the intensity and displacement. This region accounts for ν_{CO} stretching vibrations of primary and
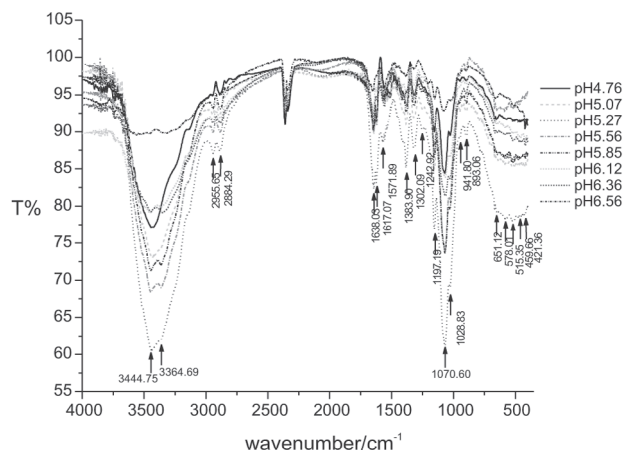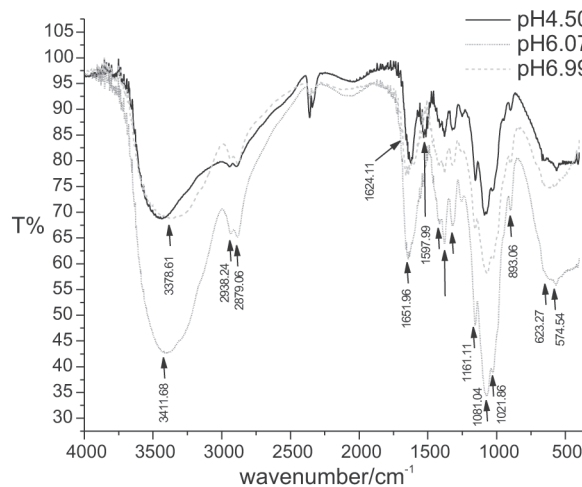

Figure 7. IR spectra of solid complexes of chitosan and Cu^{2+} extracted from aqueous HCl solutions at various pHs in ligand to metal ratio of 1:1.

secondary alcohols respectively.[38] The region around 1150 cm^{-1} (ν_{CO}) also showed changes in intensity and displacement which also indicates changes in the secondary carbon hydroxyl groups. In the IR spectra of chitosan alone, no significant bands are seen in the region below 1000 cm^{-1}.[35] However, there were sharp changes in the spectra when chitosan is complexed to the metal ions studied. Such were the cases of M–L vibrations, ν_{NH2} and ρ_{NH2} (amino complexes),[6,35,38] this last one possibly coinciding with the vibrations of ρ_{HOH} (aquo complexes),[38] ν_{OH} (hydroxyl complexes) and ν_{OH} (aquo complexes)[38] (refer to Table 2).

Moreover, another signal was detected for the complexes, around 1250 cm^{-1} (Figures 6 to 8) associated with δ_{CO} in the sugar ring which was not present in the spectrum of the ligand alone. This could be associated with structural rearrangement induced by the coordination which causes a higher polarization of this group and then leads to the appearance of bands in the infrared region.[39]

Although it is more likely that chitosan-metal cation complexation occurs primarily through the amino group (as ligand), hydroxyl groups in the sugar unit can play an important role when the stereochemistry favors the complexation through these groups or when either the pH or the metal ion strength deprotonate C-6 OH. It is not possible to rule out the participation of –OH groups (either from C-6 or C-3) in chitosan complexation since competition of the different metal ion Lewis acidity can provide the proper conditions for this complexation and
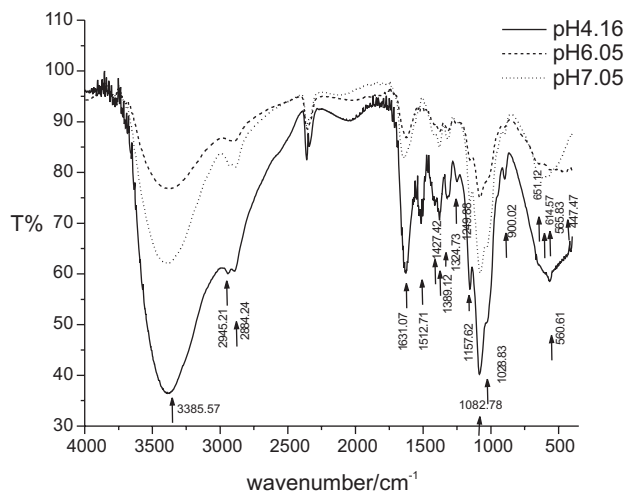

Figure 8. IR spectra of solid complexes of chitosan and Zn^{2+} extracted from aqueous HCl solutions at various pHs in ligand to metal ratio of 1:1.

also that the deprotonation constant of C-6 OH is within the pH range of aqueous solution (refer to equation 1). Above pH 8, all studied systems formed insoluble hydrolytic products.

Proposed coordination mechanism and possible structures

Data obtained so far allowed proposition of some structures for the complexed species formed in the chemical equilibria studied. The proposed structures may bear Cl$^-$ as the counter anion in the complex structure, to give the

Table 2. UV-Vis and IR bands for the solid complexes of chitosan (L) and Cu^{2+}, Zn^{2+} and Mn^{2+} extracted from HCl aqueous equilibria at different pHs

| assignment	UV-Vis observed bands / nm			
	Chitosan	Cu–Chitosan	Mn–Chitosan	Zn–Chitosan
π-π^*	214	206	208	204
TCLM		249-270	306-400	
d-d		580.85		
	IR observed bands / cm^{-1}			
ν (OH)	3453	3445	3386	3411-3378
ν (NH)	3345	3365	n.d.	3411-3378
ν (CH)	2923	2955, 2885	2945, 2884	2938, 2879
ν (CO) amide	1650	1638	1631	1652-1624
ν (HOH)	-	1618	1631	1652-1624
δ (NH)	1590	1572	1513	1598-1523
δ (OH) ring	1421	n.d.	1427	1415
δ (CH$_2$)	1370	1384	1389	1377
δ (CH) ring	1318	1302	1325	1318
δ (CO) ring		1243	1250	1264
ν (-COH-)$^{sec\ alcohol}$	1152	1198	1158	1161
	1089	1071	1083	1081
ν (-COH-)$^{primary\ alcohol}$	1029	1029	1029	1022
ρ (NH$_2$) complexes		942, 893	900	893
ρ (HOH)$_{aquo\ complexes}$		578	615	623
ν (M-NH$_2$)$_{amine}$		515	566	574
ν (M-OH)$_{alcohol}$		460	448	
ν (M-OH)$_{aquo}$		422		

n.d.: not detected
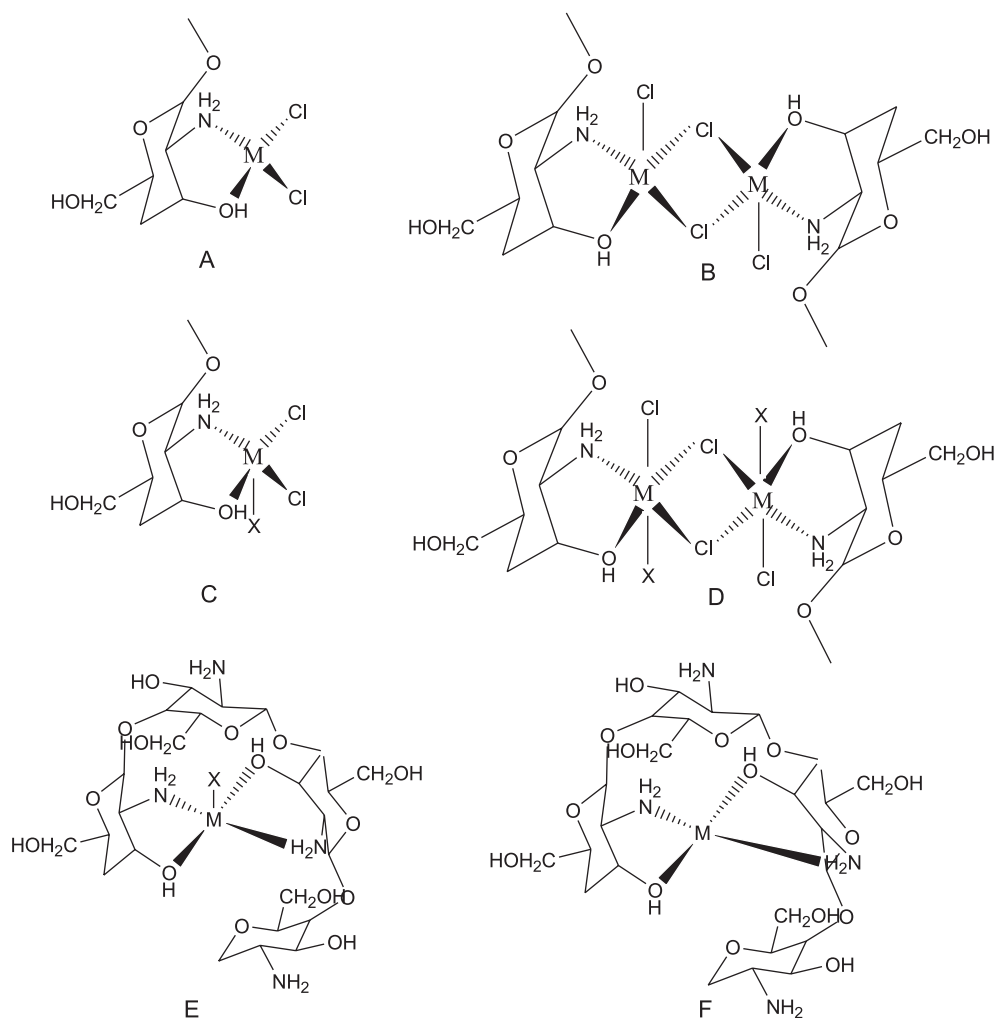
Figure 9. Proposed structures for chitosan–metal ion complexes in the experimental conditions of this work. A: MChitCl$_2$ or [MNOCl$_2$]; B: M$_2$(Chit)$_2$Cl$_4$ or [M$_2$N$_2$O$_2$Cl$_4$]; C: MChitCl$_2$X or [M$_2$N$_2$O$_2$Cl$_4$X$_2$]; D: M$_2$(Chit)$_2$Cl$_4$X or [M$_2$N$_2$O$_2$Cl$_4$] ; E: MChitX or [MN$_2$O$_2$X]; F: MChit or [MN$_2$O$_2$], where : M, metal ion (Copper(II), Manganese(II), Zinc(II)); Chit, Chitosan (A-D, Chit as monomer and, E and F, Chit as dimer); N, Nitrogen; O, Oxygen; Cl, Chloro; X, H$_2$O or OH.

species, zero final charge balance. Nitrate is not likely to be in the complex structures but can be present probably in the second coordination sphere of the complexes for the same reason stated as for Cl$^-$. Since the three dimension-structure of a biopolymer, chitosan can coordinate with the metal ions forming mono or dinuclear units with possible metallic bridges with chlorine atoms.[40-42] In this way, tetracoordinated [MNOCl$_2$], pentacoordinated [MNOCl$_2$X] or [μ–Cl–MNOCl$_2$] hexacoordinated [μ–Cl–MNOCl$_2$X] structures can be formed, where X are OH$^-$ or H$_2$O molecules (Figure 9).

Conclusions

This work provided the calculated binding constants for chitosan and the divalent cations Cu^{2+}, Zn^{2+} and Mn^{2+}.

The order of stability found was Cu^{2+}>Zn^{2+}>Mn^{2+}. The coordination happened through amino and hydroxyl groups (coordination through either C-6 or C-3 hydroxyl group of the dimeric sugar unit) in bridge-like structure with the metallic centers, originating tetracoordinated [MNOCl$_2$], pentacoordinated [MNOCl$_2$X] or [μ–Cl–MNOCl$_2$] hexacoordinated [–Cl–MNOCl$_2$X] compounds, with chloride anions completing the coordination sphere of the complexes. These species are in agreement with the literature[43] where the thermodynamic parameters for chit and copper(II) have shown that after the primary interaction with the N atom, a second interaction happens through –OH of C-3 involving one or more monomer of the same or different chain, with the same cation forming stable complexes and with the references 40 and 42, where there are the reports of structures of two metal ions bridged

by two Cl atoms bound to two monomers from different chains of chit or to a dimeric unit of the same chain of chit (refer to Figure 9).

It has to be emphasized that as the dimeric structure of chit was considered in all calculations as the minimum structure, ML can be the representation of the complexation through N or O binding sites of either a monomeric or dimeric sugar units of chit. In this way the equilibrium was conceived, ML_2 is hardly to be expected.

The present study helped clarify Cu^{2+}, Zn^{2+} and Mn^{2+} selectivity of the coordinating and remediating agent chitosan, in aqueous HCl chemical equilibrium.

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