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A thermodynamic investigation on chitosan–divalent cation interactions

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Abstract

The α-form of the biopolymer chitin, with particle sizes in the 80–200 mesh range was deacetylated under hot alkaline conditions to yield chitosan. These biopolymers were characterized by nitrogen elemental analyses, infrared spectroscopy, X-ray diffractometry and carbon-13 NMR. The degree of deacetylation found was 80.5%, which was determined via infrared spectroscopy. Chitosan suspended in water adsorbs the divalent cations cobalt, nickel, copper and zinc, as represented by batch process isotherms, in which the number of moles adsorbed as a function of cation concentration gave a defined isotherm for each cation, indicating saturation of the available nitrogen centers on the organic polymeric chain by cations in an acid–base interaction. The cation–biopolymer interaction was quantitatively followed through calorimetric titration, whose values enabled the calculation of the thermodynamic values. The exothermic enthalpy, the negative Gibbs free energy and the positive entropic values are in agreement with a favorable thermodynamic condition for cation–chitosan amine group interaction. Based on negative enthalpic values, -26.66 ± 0.05, -27.58 ± 0.03, -39.05 ± 0.01 and $-27.71 \pm 0.04\,\mathrm{kJ\,mol^{-1}}$, the sequence expresses the Irving–Williams series, Co < Ni < Cu > Zn.

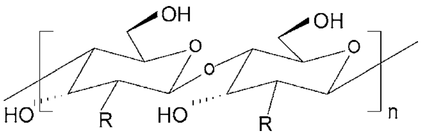
Keywords: Chitosan; Chitin; Coordination; Adsorption; Thermodynamics

1. Introduction

Native chitin, poly-β(1 → 4)2-acetamide-(2-deoxy-D-glucopyranose), and chitosan, poly-β(1 → 4)-2-amino-(2-deoxy-D-glucopyranose), biopolymers present interlayered β-glycosidic bonded structures, without any predominance of either monomer. Thus, the copolymer constitution does not include pure chitin or chitosan but always a mixture containing a certain amount of the deacetylated species [1,2]. Thus, chitin as extracted from natural sources contains around 10% of free amino groups, while chitin submitted to an alkaline chemical reaction yields chitosan through deacetylation processes, with the remaining acetylation not lower than 3% [3].

Investigations on such natural biopolymers concluded that chitin can be present in three distinct forms α, β and γ, nevertheless, the last is less studied, probably due to the fact that it appears in lower percentages than the α- and β-forms. In natural condition, chitin is normally found associated with proteins, forming glycoproteins. Chitin as well as chitosan is present in a percentage varying from 20 to 50% in arthropods [4], and forms the structural and protector material part of insets, marine animals and cellular microorganisms [5]. On the other hand, chitin can be artificially obtained [6] by opening the ring of the exazoline group, which is a sugar derivative or by biosynthesis from glucose. In this process glucose initially converted into an amino-sugar through an enzymatic operation, followed by acetylation and finally is polymerized by enzyme [5].

The free amino groups distributed on the chitosan polymeric chain obtained from chitin deacetylation are effectively active in a series of chemical modifications [7–12]. Moreover, this new biopolymer has applicability in many scientific and technological applications [2,7]. For comparison, a structural representation for chitin R = $-NHCOCH_3$ and chitosan R = $-NH_2$ is shown below.



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The original natural precursor chitin is the most important organic nitrogen containing polymer found in the oceans [13] and the second most abundant material of the planet, being superimposed only by cellulose [14], in which R = OH in the preceding scheme. It is worth remembering that chitin extraction does not cause any disturbance to the ecosystem, its embraces all advantages provided by polysaccharides, considering it as the source of chitosan, and both are biocompatible biopolymers for animal tissues with low toxicities and significant biomedical applications [15,16]. From the point of view of academic study its use has increased in many hybrid material with poly(ethylene glycol) [17], κ-carrageen [18], silylant agent [19], in enzyme immobilization [20] and some inorganic compounds [21].

An expressive chitosan application is associated with its ability to extract cations from aqueous solution, due to the availability of the free amine groups. Thus, this biopolymer can be potentially used as an extractor support to remove undesirable cations from an ecosystem [22].

The extracting equilibrium condition for any cation is also related to a thermal effect, which can be calorimetrically followed through the titration method. For this heterogeneous system it is important to follow the thermal effect caused by solute increment, which arises from active acidic centers on the adsorbent polysaccharide, to give the integral thermal effect of adsorption that permits to calculate the more representative thermodynamic data for such an interaction [23]. Thus, the adsorption effect can be caused when a known number of moles of the adsorbent interact with the adsorbate center dispersed on a surface at a solid/liquid interface [24].

The present investigation deals with the determination of the integral thermal effect due to the adsorption of the divalent cations copper, nickel, zinc and cobalt on available amine nitrogen centers attached to the chitosan structure. The main purpose is to report these most precise calorimetric obtained data, based on integral thermal effects and compare the distinct behavior of these cations when they are coordinated.

2. Experimental

2.1. Chitin and chitosan

Chitin from shrimp shells (Fine Chemical Kito) was cut in a knife mill to yield particles which was sieved to the 80–200 mesh range. Chitosan was prepared by deacetylating 3.0 g of this chitin in 600.0 cm^3 of alkaline solution, 50% sodium hydroxide, for 2 h at 380 K. The solid was filtered and washed thoroughly with doubly distilled water until neutral pH and dried under vacuum at room temperature [19,23].

2.2. Characterization

The products obtained were characterized by nitrogen elemental analyses in a Perkin Elmer model PE 2400 instrument, infrared spectroscopy by using 2% samples in KBr pellets with 40 scans and 4 cm^{-1} resolution in a Bomem spectrometer series MB, ^{13}C NMR spectra were obtained on a AC 300/p Bruker spectrometer with magic angle spinning, operating in CP/MAS mode at 75.47 MHz with a pulse delay 4 s and a contact time of 1 ms. The amount of cation was analyzed in an ICP OES Perkin-Elmer 3000 DV apparatus and the X-ray patterns in a Cu Kα source with a Shimadzu model XD3A diffractometer.

2.3. Adsorption

The adsorption assays were performed at constant thermosted temperature of 298 ± 1 K in an orbitally stirrer instrument. For this adsorption process a series of divalent cations, copper, cobalt, nickel and zinc, dissolved in doubly distilled water, were chosen to establish the chitosan isotherm and their stock concentrations were determined through EDTA titration [25]. For a complete isotherm, a series of polyethylene flasks containing 5.0, 10.0, 15.0, 20.0, 30.0, 35.0, 40.0 and 50.0 cm^3 of any desired aqueous cation solution, was added to doubly distilled water to reach the maximum volume of 50.0 cm^3, in order that the cation concentration remained near $1.0 \times 10^{-3}\ \mathrm{mol\,dm}^{-3}$. To each final solution 30.0 mg of chitosan was added and all flask suspensions were stirred for 2 h. After this time at least two aliquots of the supernatant of each flask were separated to have the cation analyzed by ICP OES technique. The time to reach saturation of the isotherm was previously determined using similar stock cation solutions and varying the time and, in such conditions, the plateau was defined, being chosen 2 h as long enough for equilibrium for all cations. Taking into account the isotherm of concentration, the amount of cation adsorbed was analyzed from the supernatant for each sample and determined by the expression: $N_\mathrm{f} = (N_i - N_\mathrm{s})/m$, where N_f is the number of moles adsorbed per gram of the biopolymer, N_i the initial number of moles of cation added into the solution, N_s the number of moles of cation that remains in the supernatant and m the mass of the polymer used in each flask.

2.4. Calorimetric titration

The thermal effects were obtained by the titration method using a Thermometric 2277 (Thermal Activity Monitor) precision calorimetric system [23,24]. In this procedure, a glass calorimetric vessel with 15.0 cm^3 of capacity was used and, for each titration, nearly 50.0 mg of the biopolymer was suspended in 14.0 cm^3 of doubly distilled water. The suspension was maintained under mechanical turbine stirring (90 rpm) and thermostated at 298.15 ± 0.02 K. The titration was followed by adding successively 10 μL increments of the titrand solutions, for example, a 0.1085 $\mathrm{mol\,dm}^{-3}$ copper nitrate solution, and the thermal effect was recorded after each increment of titrand. To complete the titration 11 increments were done in a maximum time of 1.5 days for all cations. Outside the calorimeter, the same titration was simulated in order to follow the possible variation in pH, which could be due to the cation hydrolysis, mainly in case of copper, and from the beginning to the end of the operation the pH value was in the interval 6.4 ± 0.4.

The thermal effects obtained for each interaction of any divalent cation (M^{2+}) with chitosan (Ch) were determined from a series of experiments and the complete thermodynamic cycle for each cation can be summarized as follows:

$$\mathrm{Ch_{(aq)}} + \mathrm{M^{2+}_{(aq)}} = \mathrm{Ch} \cdot \mathrm{M^{2+}_{(aq)}}, \quad Q_\mathrm{r} \qquad (1)$$

$$\mathrm{Ch_{(aq)}} + n\mathrm{H_2O_{(aq)}} = \mathrm{Ch} \cdot n\mathrm{H_2O_{(aq)}}, \quad Q_\mathrm{h} \qquad (2)$$

$$\mathrm{M^{2+}_{(aq)}} + n\mathrm{H_2O_{(aq)}} = \mathrm{M^{2+}} \cdot n\mathrm{H_2O_{(aq)}}, \quad Q_\mathrm{d} \qquad (3)$$

The sequence of reactions (1)–(3) for each individual calorimetric titration experiment was carried out in duplicate for each cation employed. Based on these individual thermal effects, the net value for reaction (1), was obtained by subtracting the dilution Q_h and Q_d from the obtained Q_r value determined.

The net thermal effect of these interactions between chitosan and cations gave a calorimetric thermal effect Q_int by applying the expression: $Q_\mathrm{int} = Q_\mathrm{r} - Q_\mathrm{h} - Q_\mathrm{d}$. However, the hydration thermal effect of the biopolymer was determined as null, that is, $Q_\mathrm{h} = 0$, then the expression reduced to $Q_\mathrm{int} = Q_\mathrm{r} - Q_\mathrm{d}$.

3. Results and discussion

The attached pendant N-acetyl groups of natural chitin were partially removed using alkaline conditions to yield the more reactive derivative chitosan. The infrared spectra for both biopolymers are shown in Fig. 1. The effectiveness of the deacetylation process was successfully followed by a well-established infrared spectroscopy technique. Thus, the degree of deacetylation (DD) can be calculated [26] from Eq. (4), and for this determination the absorbance ratio from the infrared spectrum of chitosan was obtained for the bands at 3400 and 1655 cm^{-1}, respectively, as shown in Fig. 1b. By applying these values in the corresponding equation a DD = 80.5% was obtained, which corresponds to an acceptable degree for this biopolymer, and compares with other technique applied for this same determination, such as proton NMR methodology [27]:

$$\mathrm{DD} = 97.67 - \left[26.486\left(\frac{A_{1655}}{A_{3400}}\right)\right] \qquad (4)$$

From the chitin and chitosan infrared spectra shown in Fig. 1, the common large and intense bands located at 3700 and 3000 cm^{-1} can be attributed to axial OH and NH group deformations, which are more evident in the chitin spectrum. On the other hand, the absorption band at 3480 cm^{-1} can be assigned to the hydrogen bond between OH on carbon 5 of the biopolymer structure with the carbonyl acetamide group, attributed to amide II in the 1700 cm^{-1} region [28]. The other absorptions at 3260 and 3100 cm^{-1} are associated with the intermolecular hydrogen bond to acetamide vibration. The axial carbon–hydrogen bond that appears in the 3000–2800 cm^{-1} range is more intense for the chitin biopolymer. The symmetrical and asymmetrical carbon–oxygen–carbon ring [29] gives rise to the bands located at 1051 and 1161 cm^{-1}, respectively. Finally, it is important to mention the absorptions at 1420 and 1380 cm^{-1}, that are assigned as CH_2 and CH bending bands, respectively, but with low intensity, as shown in Fig. 1a.

The amine chitosan group reactivity can be demonstrated through chemical surface modifications by reacting easily, for example, with an aldehyde function [30]. However, the degree of such active centers attached to the structural backbone reflects in nitrogen amount determined through elemental analysis, found as 4.36 and 6.36% for chitin and

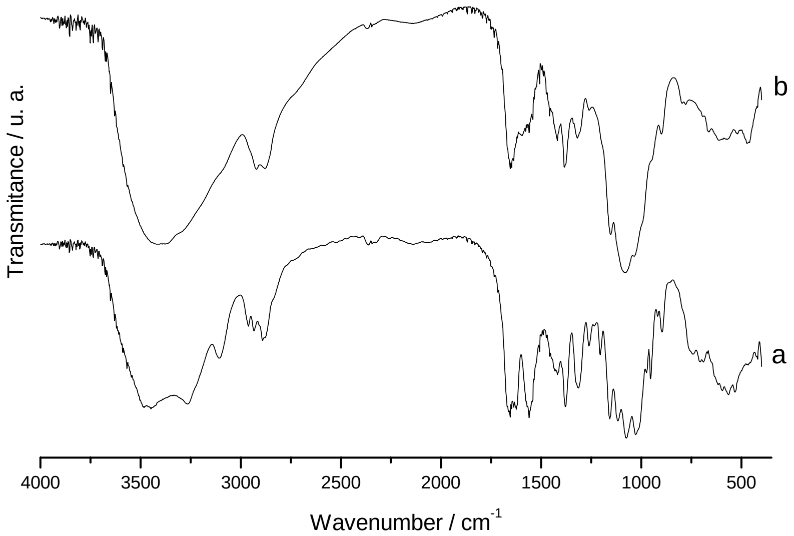

Fig. 1. Infrared spectra for chitin (a) and chitosan (b).

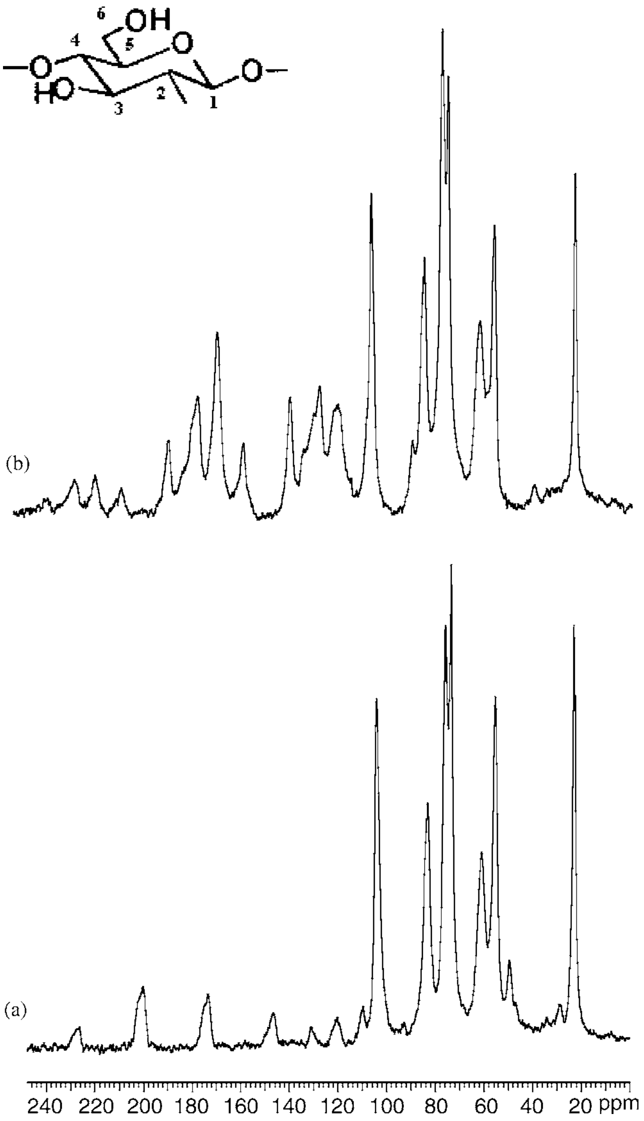

Fig. 2. ^{13}C NMR for chitin (a) and chitosan (b).

chitosan, respectively. A quantitative indication of the effectiveness of these basic centers comes from the amounts of cation adsorbed. For chitosan the calculated value gave 3.95 mmol/g of chitosan, based on a monomer of this biopolymer of 161 g and the present value is very close to that observed in other determinations [31].

The carbon-13 NMR technique is an important tool to characterize such kinds of biopolymers. The spectra of the pristine chitin and its derivative deacetylated form chitosan are shown in Fig. 2. The set of observed signals is in agreement with the starting chitin being in the alpha form, whose deacetylated compound gave signals near 75 ppm, an intensity which is much lower in carbon 5 than 3 [32], instead of an inversion observed for chitin in Fig. 2b. For β-chitosan the signals shown in Fig. 2a around of 75 ppm are not well defined with an identical chemical shift. On the other hand, chitin the in α-form presents satellite bands related to methyl groups and the intensity of the carbonyl group is higher than for chitin found in the β-form. The peaks at 175, 103, 85, 60, 57 and 22 ppm refer to $C{=}O$, C_1, C_4, C_6, C_2 and CH_3 [33], respectively, as represented by inserted numbered structure. These series of peaks remain practically with the same chemical shift in the chitosan spectrum, as shown in Fig. 2b. Based on the spectra presented in Fig. 2, it is worth mentioning that the original chitin biopolymer is not completely acetylated, due to the presence of signals in the 170–240 ppm range, which are similar to those found on the chitosan spectrum. The chitosan preparation conditions use hot concentrated alkaline solution, which could cause some polysaccharide dissociation to give some glycan α-chitin component. As the starting material contains glycans [34], the chitosan spectrum interpretation raises difficulties, which reflect mainly in a decrease of carbonyl from the original signal around 175 ppm.

The degree of crystallinity for chitin and chitosan biopolymers was evaluated through X-ray diffractometry, in a pro-

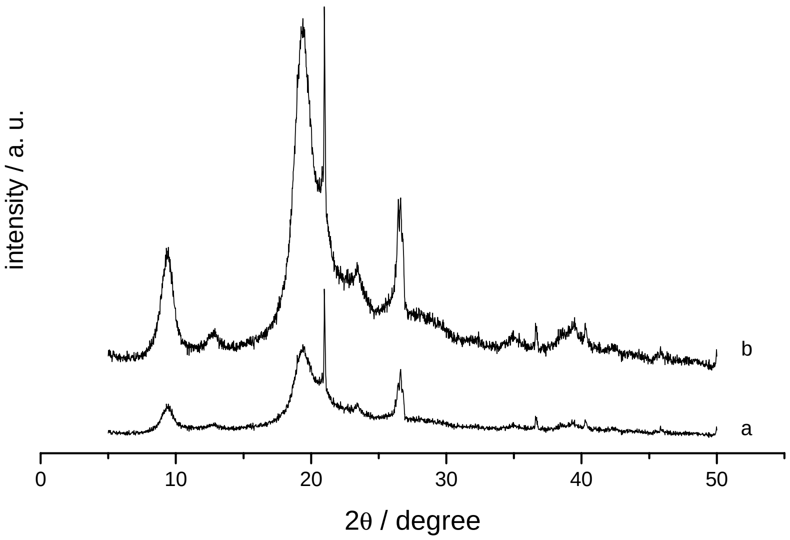

Fig. 3. X-ray diffractometry for chitin (a) and chitosan (b).

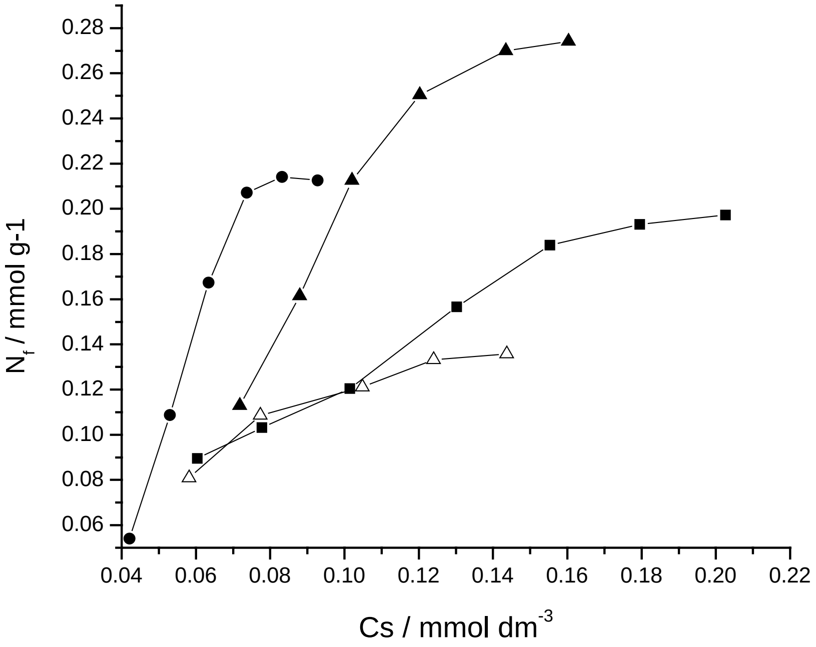

Fig. 4. Isotherms of adsorption of chitosan with $Cu(NO_3)_2$ (▲), $Co(NO_3)_2$ (●), $Ni(NO_3)_2$ (■) and $Zn(NO_3)_2$ (△).

cess which refers to cellulose as standard [35], due to the fact that the studied materials did not present the crystallographic planes that could be characterized as crystalline. As known, cellulose displays an absence of crystallinity, but its use is related to natural structural features, which are very close to both biopolymers chitin and chitosan. In this process expression (5) was employed [36] to determine the crystallinity index (CrI) for chitin and chitosan, and two important peaks due to the maximum intensity in the diffraction at 21 and 26° were considered as shown in Fig. 3a and b, respectively. The first one corresponds to the maximum intensity ($I_{\Theta M}$) and the second to the amorphous diffraction ($I_{\Theta A}$), gave the values 28.3 and 20.8% for α-chitin and chitosan, respectively. These values suggested that when chitin is chemically treated to produce chitosan, a decrease in crystallinity is observed:

$$\mathrm{CrI} = \frac{I_{\Theta M} - I_{\Theta A}}{I_{\Theta A}} \times 100\% \qquad (5)$$

The X-ray diffraction patterns obtained were shown to be as an important procedure to distinguish these two kinds of α- and β-chitin forms by means of the use of the crystallinity index, however, the same operation is unsuccessful in identifying the missing γ-form, due to the fact that it is always found in natural sources in a mixture of the preceding types.

The biopolymer chitosan was assayed to remove cations from aqueous solution at the solid/liquid interface in a heterogeneous condition by using the batch process. After equilibrium was established, the isotherm of adsorption for each cation was obtained as outlined in Fig. 4. Basically, the isotherms have the same form, showing a tendency to reach a plateau, with copper the most adsorbed, followed by cobalt and nickel nearly with the same order of ability in adsorption, and finally, the zinc cation. These results are listed in Table 1. The maximum adsorption capacity is one important parameter to express the ability in cation/basic center interaction, as given by the values 0.21 ± 0.02, 0.27 ± 0.02, 0.37 ± 0.04 and 0.38 ± 0.01 mmol g^{-1}, for zinc, cobalt, nickel and copper, respectively. For this sequence copper is the most adsorbed, as was observed for other chemically modified natural and other inorganic surfaces. For example, previously chitosan [37] gave 0.32 mmol g^{-1}, which is a little bit small than silica immobilized with dithiocarbamate [38] and 2,4-dichlorophenoxyacetic acid [39] with 0.57 and 0.44 mmol g^{-1}, respectively. However, Amberlite XAD-2 resin gave variable values, dependent on the immobilized agent, as observed with pyrocathecol [40], tiron [41] and chromotropic acid [42] with 92.5, 0.22 and 0.13 mmol g^{-1}, respectively. On the contrary, for another similar chitosan natural biopolymer cellulose, chemically modified with methyliminodiacetic acid [43], only 0.020 mmol g^{-1} of copper cation is adsorbed.

The data obtained for such isotherms can be linearized after adjusting to a modified Langmuir equation [23,24] and for this fit the degree of cation adsorbed to the chitosan

Table 1
Divalent cations (M^{2+}) chemisorbed on chitosan gave the number of moles adsorbed (N_f), the equilibrium constant (K) and the maximum adsorption capacity (N_s)

M^{2+}	N_f (mmol g^{-1})	K (dm^3 $mmol^{-1}$)	N_s (mmol g^{-1})
Co	0.21 ± 0.01	12000 ± 10	0.27 ± 0.02
Ni	0.20 ± 0.03	5920 ± 14	0.37 ± 0.04
Cu	0.28 ± 0.05	7920 ± 10	0.38 ± 0.01
Zn	0.14 ± 0.07	13600 ± 10	0.21 ± 0.02

Table 2
Thermodynamic data for chitosan–divalent cation interactions, indicating the thermal effect on a monolayer (Q), the variation in enthalpy (ΔH), Gibbs free energy (ΔG) and the calculated entropy of interactions (ΔS)

M^{2+}	Q ($J\,g^{-1}$)	$\ln K$	$-\Delta H$ ($kJ\,mol^{-1}$)	$-\Delta G$ ($kJ\,mol^{-1}$)	ΔS ($J\,mol^{-1}\,K^{-1}$)
Co	7.20 ± 0.01	9.39	26.66 ± 0.05	23.3 ± 0.4	11 ± 1
Ni	10.20 ± 0.02	8.69	27.58 ± 0.03	21.5 ± 0.2	20 ± 1
Cu	14.84 ± 0.01	8.98	39.05 ± 0.01	22.3 ± 0.1	56 ± 1
Zn	5.82 ± 0.04	9.52	27.71 ± 0.04	23.2 ± 0.3	15 ± 1

surface can be determined as represented by expression (6):

$$\frac{C_s}{N_f} = \frac{C_s}{N_s} + \frac{1}{N_s K} \tag{6}$$

where C_s is the supernatant concentration after the equilibrium of the system, K the equilibrium constant, N_f the number of moles adsorbed per gram of chitosan and N_s the maximum adsorption capacity of the biopolymer that is covered by a monolayer of cations. N_s and K are obtained after isotherm linearization process [23].

The quantitative chitosan–cation interaction process can be determined through calorimetric titration by collecting the thermal effect obtained from the three required determinations (Eqs. (1)–(3)) in order to obtain the net thermal effect. These values were adjusted to a modified Langmuir equation (7) [23,24] to calculate the integral enthalpy involved in the formation of a monolayer per unit mass of the adsorbate $\Delta_{int}h$, as shown in Fig. 5, as an illustration for cobalt adsorption:

$$\frac{\sum X}{\sum Q} = \frac{1}{(K-1)\Delta_{int}h} + \frac{\sum X}{\Delta_{int}h} \tag{7}$$

In this equation $\sum X$ represents the sum of the mole fractions of the cation in solution after adsorption, $\sum Q_{int}$ the integral enthalpy of adsorption ($J\,g^{-1}$) obtained by dividing the thermal effect resulting from adsorption by the number of moles of the adsorbate and K a proportionality constant that also includes the equilibrium constant. By using the angular and linear values from $\sum X/Q_{int}$ versus $\sum X$ plot, it was possible to obtain the $\Delta_{int}h$ value. Then, the enthalpy of adsorption ΔH can be calculated by means of the expression $\Delta H = \Delta_{int}h/N_s$.

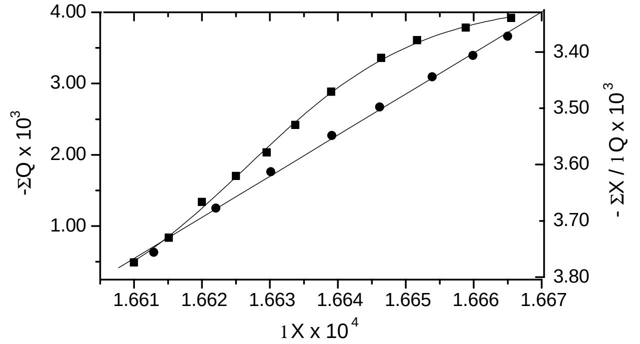

Fig. 5. Calorimetric titration isotherm of chitosan with cobalt at 298.15 ± 0.02 K; the straight line is the linearized form of the isotherm.

From K values the corresponding negative Gibbs free energy data were calculated ($\Delta G = -RT \ln K$) for cation–basic center interactions on the chitosan surface in spontaneous reactions. The missing thermodynamic entropic value was calculated through the expression $\Delta G = \Delta H - T\Delta S$, as previously reported [23,24], and the values are listed in Table 2. These positive values should be related to cation desolvation, an effect which causes an entropic enhancement. The transference of water molecules from the hydration sphere to the medium of the reaction promotes disorganization of the system, by increasing in entropy in all reactions, with the largest value obtained with copper [23,24,44].

These sets of favorable thermodynamic values imply an interaction between the hard nitrogen center available on organic backbone with the cations, with the resulting enthalpic values following the order Co $<$ Ni $<$ Cu $>$ Zn. This series of values is adjusted to a sequence proposed in the Irving–Williams series [45], which denotes a well-behaved cation–biopolymer interaction.

4. Conclusion

The biopolymer chitin in its alpha form with a degree of deacetylation of 80.5% develops a preferential ability in adsorbing divalent cations, as demonstrated by batch adsorption isotherms. However, the basic nitrogen atom center from the amine group on the chitosan surface interacts with cations at the solid/liquid interface as is manifested in corresponding thermal effect values. The change in chemical effects in any system can be calorimetrically followed by titration, whose curves demonstrate precise different saturation behavior to distinguish the cation sequences. Consequently, this sensitive technique enables establishing an order for the cation interactions using the thermodynamic data determinations.

For the set of cations investigated: cobalt, nickel copper and zinc, all the available basic centers were not saturated, which fact can be interpreted taking into account that the remaining sites on the polymeric surface may not be favorably placed for metal coordination. Therefore, the thermodynamic data for all cations reflect the main characteristic of each individual cation in relation to the same standard basic center. These quantitative data are favorable for such equilibrium, in this heterogeneous system, showing a spontaneous interaction as indicated by the negative Gibbs free energy, the exothermic enthalpic values and the positive entropic results.

This first set of thermodynamic data obtained for divalent cations gave exothermic enthalpic values in the order Co $<$ Ni $<$ Cu $>$ Zn, which fit to the Irving–Williams series. This well-behaved system suggested that the chemically modified natural product chitosan can be successfully employed for cation removal from wastewater or industrial effluents, with a preference for copper cations from this mixture.

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