Chelation of Chitosan Derivatives with Zinc Ions. I. *O,N*-Carboxymethyl Chitosan

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ABSTRACT: The chelation between *O,N*-carboxymethyl chitosan (ONCMCh) and zinc sulfate in aqueous solution was studied by kinetic experiments and characterized by inductively coupled plasma (ICP) and UV spectrophotometry. The experimental data indicated that the chelating processes were greatly controlled by the reaction conditions (i.e., reaction time, temperature, and Zn^{2+} ionic and ligand concentrations). The consequence of chelating Zn^{2+} onto ONCMCh was the formation of complexes with different solubilities. The favorable complexes for ONCMCh-Zn^{2+} chelate were at the low zinc ionic and ligand concentrations, as well as at the appropriate temperature. The evidence provided by the kinetic parameters and the changes in zinc concentration by ICP analysis further confirmed the plausible complexing mechanisms. While the formation of water-soluble products was occasioned by the electrostatic attraction mechanism, the water-insoluble products were predominantly formed by chelation of Zn^{2+} with *O,N*-carboxymethyl chitosan. © 2000 John Wiley & Sons, Inc. J Appl Polym Sci 77: 2246–2253, 2000

Key words: chelation; chitosan derivatives; zinc ions; *O,N*-carboxymethyl chitosan

INTRODUCTION

Chitin and its deacetylated product, chitosan, are the world's second most abundant natural polymers following cellulose. Their utilization represents a great challenge to both the scientific and chemical communities. The significance of chitin and chitosan in a material world is due to the fact that they are renewable and biodegradable. In addition, the recent increased understanding of their functions in biology and their potential in technological and biotechnological applications,[1–3] particularly metal chelation of chitosan and its derivatives, have attracted a great deal of attention and have been brought into vogue in research in recent years.[4–8]

The complexing ability of chitin and chitosan with polyvalent metals in hazardous waste water has recently identified.[9–12] However, researchers feel that the complexing ability has not been realized to a satisfying level. They believe the complexing properties can be enhanced by chemical modification of the functional groups, such as hydroxyl and amino groups, in chitin or chitosan.[13–15] For instance, carboxymethylation was prepared and regarded as a simple and effective process to facilitate the chelation ability of chitosan with transition metal ions.[16] Carboxymethyl chitosan was found to elicit a broad range of biological activities[17] and proved to be useful in cosmetic and pharmaceutical applications.[18]

Zinc ions possess nutritional features important to human health and health care.[19] Hence, the success of the preparation of chelated products using modified chitosan and zinc ions must have significant medicinal and nutritional values. This leaves us prone to prepare an *O,N*-carboxy-

methyl chitosan (ONCMCh) and to complex it with zinc ions. This article assesses the chelating processes and the kinetic factors that affect chelation reactions.

EXPERIMENTAL

Materials

High purity grade (99.99%) zinc sulfate and sodium monochloroacetate were obtained from Aldrich Chemical Company. They were used as received without further treatment. Chitosan was purchased from Eastman Chemical Company and was ground into 40-mesh size particles before using it to prepare ONCMCh.

Preparation of ONCMCh

The ONCMCh was prepared according to a modified Hayes' procedure described in a patent.[20] In brief, chitosan sodium salt reacted with monochloroacetic acid at 25 and 60°C for 1 and 3 h, respectively, to prepare O-substituted and N-substituted CMChs. In the present experiment sodium monochloroacetate was used to displace its acid form.

Synthesis of ONCMCh Chelates

Synthesis of ONCMCh chelates with Zn^{2+} was performed by the addition of 100 mL of zinc sulfate aqueous solution with a known concentration into the ONCMCh solution, which was prepared by dissolving it in deionized water. The reaction was allowed to proceed at various conditions for a designated period of time, after which the chelate was collected by centrifugation, washed with deionized water followed by ethyl alcohol, and air dried to a constant weight. The weight of the product is defined as the yield of the chelate.

Water-insoluble (A) and water-soluble (B) complexes were obtained from this procedure, that is schematically shown in Figure 1. Under certain conditions, stiff gels were obtained. In this case, they were dispersed in ethyl alcohol with stirring and then the precipitate was collected and dried.

Characterization

UV absorption of the ONCMCh-Zn chelates was measured using a Perkin–Elmer Lambda 3A UV-vis spectrophotometer in the range of 190–700 nm. Deionized water was used as the reference.
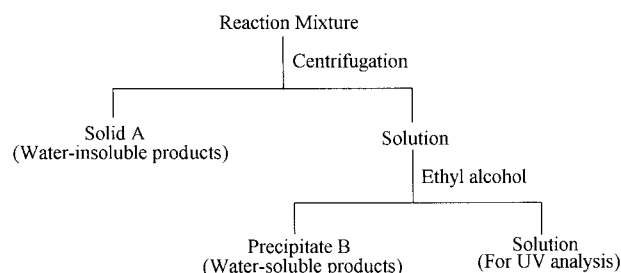
Figure 1 A schematic diagram for separation of ONCMCH-ZN^{2+} chelates.

Zinc contents (%) in the chelated specimens were determined by an inductively coupled plasma (ICP) spectrometer (Thermo Jarrell Ash emission spectrometer, ICP 610E) according to a wet ashing procedure.[21]

RESULTS AND DISCUSSION

Physical Characteristics of Chelates

The ONCMCh prepared from the Hayes' method is a light yellow powder with irregular shapes. When 2.0 g of ONCMCh powder was dissolved in 100 mL of water, the particles transformed into a very viscose, pale yellow solution. Within 30 min of reaction with zinc ions, ONCMCh formed complexes with Zn^{2+} ions and yielded gel-like precipitates. Under certain reaction conditions, ONCMCh reacted rapidly with Zn^{2+} ions to form stiff-gel chelates in the reactor. This phenomenon implied that ONCMCh is a very powerful chelating agent to establish dative bonds with Zn^{2+} in the aqueous solution. This is probably due to the presence of carboxyl, amino, and hydroxyl groups in ONCMCh. All the chelates obtained were colorless in the wet state and pale yellow in the anhydrous state.

Factors Influencing Chelating Yields

Important physicochemical parameters, which help in evaluating the basic qualities of a good chelate, are the chelating kinetics and equilibria.

Effect of Reaction Time

The chelating yield from ONCMCh as a function of time at a designated concentration of Zn^{2+} and temperature is presented in Figure 2.

ONCMCh readily reacted with Zn^{2+} to form a chelated compound. Within an hour at 50°C, 1.8 g
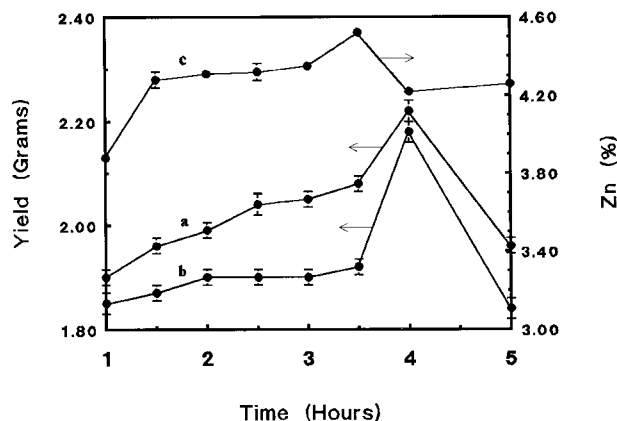
Figure 2 Effect of reaction time on chelation yield. Reaction conditions: ONCMCh, 2.0g; ZnSO$_4$, 0.005M; temperature, 50°C. (a) total complex; (b) water-insoluble complex.

of water-insoluble complex were generated. This suggested that chelation took place rapidly between zinc ions and ONCMCh at the early stage of the reaction. A small amount of water-soluble complex was also generated. Between 1 and 3.5 of reaction, only a slight increase in the yield of the water-soluble complex was noted whereas almost no change in the yield of the water-insoluble complex was observed. Absorption of zinc ions into ONCMCh took place slowly between 1 and 3.5 h of reaction. Obviously, the tendency for chelation of ONCMCh is less dependent on reaction time and is quite different from the chitosan chelating mechanism in which the complexes of chitosan with transition metal ions were conducted in a heterogeneous medium, and kinetics of the process is controlled by the diffusion of the solute. As a result, a gradual change in the yield curve was encountered in chitosan complexation.[22]

Interestingly, however, as the reaction approached 4 h, the yield of water-insoluble chelates increased rapidly whereas the water-soluble portion decreased. This phenomenon further suggested that the zinc absorption sites were situated in a proper configuration to convert into chelated products. Hence, water-insoluble products were increased. In other words, more ligand ONCMCh to chelate with Zn^{2+} caused a rapid increase in chelating yields and a reduction in the percentage of Zn in the chelated polymer. However, because the Zn^{2+} ions remain more or less constant during the course of the reaction, it is plausible to believe that Zn^{2+} ions were rapidly absorbed during the initial 90 min of reaction. The interaction of absorbed Zn^{2+} ions within the

polymer thereafter generated water-insoluble chelate.

On the other hand, after 4 h the reaction was mainly controlled by the desorption and dechelation processes and thus reduced yields of both water-insoluble and water-soluble products. Apparently, the ONCMch-Zn^{2+} chelates were unstable after 4 h of reaction.

Effect of Reaction Temperature

The influence of the reaction temperature on chelating of ONCMCh with Zn^{2+} is shown in Figure 3. The absorption of Zn^{2+} ions appeared to increase as the temperature was raised from 30 to 50°C. The yield of water-soluble products increased slightly from 30 to 40°C and slowed down at 50°C. On the contrary, the yield of water-insoluble products increased insignificantly from 30 to 40°C but significantly from 40 to 50°C. Beyond this temperature, the water-soluble and water-insoluble products both decreased because of desorption and dechelation. It is apparent that at the lower temperature (<50°C) the formation of ONCMCh-Zn complexes is favored. A high temperature (>50°C) favors the desorption and dechelation. This result is very similar to the previous work in which the chelation capacity of N-carboxymethyl chitosan was found to decrease significantly at a temperature above 55°C.[23] From the above observations it is clear that the chelation between ONCMCh and Zn^{2+} is markedly dependent on the reaction temperature. This result is also distinct from the work of McKay et al.[9] who pointed out that the sorption capacities of
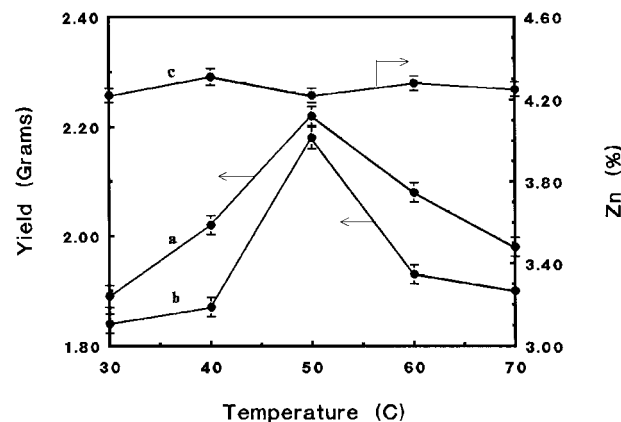
Figure 3 Effect of reaction temperature on chelation yield. Reaction conditions: ONCMCh, 2.0g; ZnSO$_4$, 0.005M; time, 4 h. (a) total complex; (b) water-insoluble complex.

zinc and nickel ions for chitosan were not influenced by temperature.

Even though the chelating yield is dependent on temperature, the ease of formation and stability of water-insoluble products were quite different. At low temperature (30–40°C) there seemed to be no gain in yield, and between 40 and 50°C the yield improved rapidly. However, between 50 and 60°C the yield decreased rapidly because of dechelation and beyond this temperature the yield remained constant again. These results suggested that two types of different chelates were formed: one is thermally stable and the other one is unstable.

A small amount of water-soluble products increased slightly between 30 and 40°C, but they also lost their yield rapidly after 60°C. This finding implied that a higher temperature the desorption took place preferentially in the water-soluble products.

The concentration of Zn contents in the water-insoluble products is also shown in Figure 3. Apparently, the Zn content is almost constant throughout the range of reaction temperatures. And it is likely that most of these Zn^{2+} ions were chelated with ONCMCh between 30 and 40°C.

Effect of Zn^{2+} Ionic Concentration

Generally, the increase in metal ionic concentration resulted in a decrease in complex capacities. For instance, the influence of ionic concentration resulted in the decrease in yields of Cr^{6+}-chitosan,[24] Cu^{2+}- and Ni^{2+}-chitosan bead,[25] and Nr^{2+}-N-carboxymethyl chitosan.[23] For the preparation of ONCMCh-Zn^{2+} chelates, a similar result was observed when 0.005–0.03 M of $ZnSO_4$ was used. The results are shown in Figure 4.

For water-insoluble products the yield of ONCMCh-Zn^{2+} complex with the highest value (2.18 g) was obtained at a low concentration of 0.005M of $ZnSO_4$. Beyond this point a sharp decrease in yield was observed. Between 0.01 and 0.02M the yield became more or less constant, and beyond 0.02M the yield decreased slowly. These changes suggested that the favorable concentration for formation of ONCMCh-Zn^{2+} chelate is at the low zinc ionic concentration because the increase in the ionic strength not only decreased the ion activity but also increased the concentration of competitive anions in the metal ligand used.[24] Hence, the increase in Zn^{2+} concentration would probably benefit the reverse reaction to form zinc sulfate and simultaneously interfere with the chelation reaction.
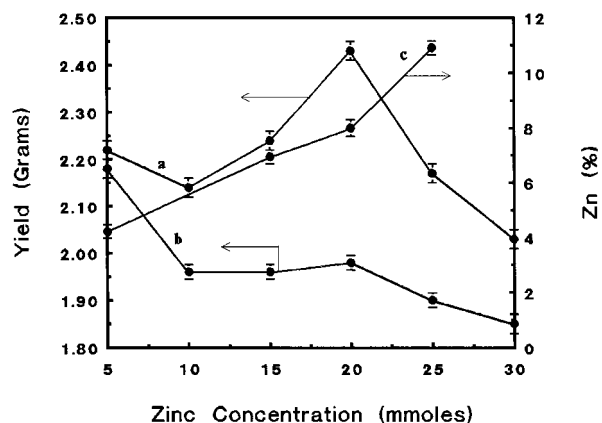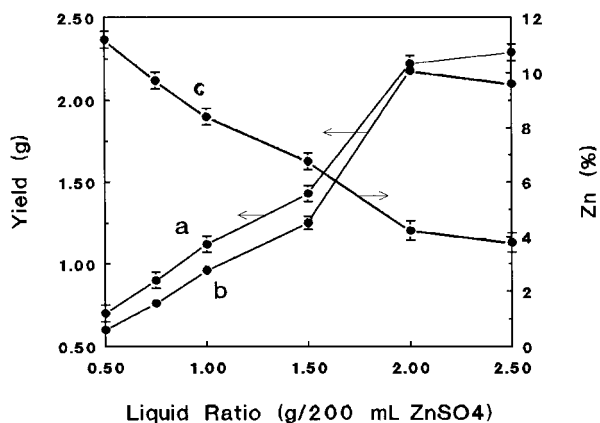

Figure 4 Effect of zinc concentration on chelation yield. Reaction conditions: ONCMCh, 2.0 g; temperature, 50°C; time, 4 h. (a) total complex; (b) water-insoluble complex.

It is also of interest to recognize that when Zn^{2+} ion concentration increased from 0.005 to 0.03M, which is a sixfold increase in concentration, it resulted in only a 15% decrease in the yield of water-insoluble products. This result demonstrated that the formation of water-insoluble products was not dominated by the electrostatic attraction (adsorption). If the electrostatic attraction did play a primary role in the complex formation between Zn^{2+} and ONCMCh, the adsorption would be sensitive to the change in the electrolyte concentrations.[12] On the contrary, ONCMCh displayed an impressive ability to coordinate with Zn^{2+} ions and thus to yield the insoluble crosslinked ONCMCh-Zn Chelates. Muzzarelli et al.[23] recently proved the chelations between Cu^{2+}, Ni^{2+}, Co^{2+}, or Fe^{2+} metal ions and N-carboxymethyl chitosan.

For the water-soluble products it was found that with the increase in Zn^{2+} concentration from 0.005 to 0.01, 0.015, and 0.02M, a drastic increase in yields from 0.04 to 0.18 and 0.28 to 0.45 g, respectively was observed (Fig. 4). These changes further showed that the variation in ionic concentration had a pronounced affect on the interactions between ONCMCh and Zn^{2+}, and the formation of water-soluble products was governed by the electrostatic attraction mechanism. Beyond 0.02M the desorption rate was greater than the complexing rate; hence, the yield decreased.

The monotonous increment in the zinc content in the chelates with increasing zinc ionic concentration in aqueous solution is understandable because of the decrement in chelating yields.

Figure 5 Effect of ligand concentration on chelation yield. Reaction conditions: ZnSO$_4$, 0.005M; temperature, 50°C; time, 4 h. (a) total complex; (b) water-insoluble complex.

Effect of Ligand Concentration

The liqand concentration is defined as the ONC-MCh content in the zinc sulfate aqueous system (i.e., the amount of ONCMCh in grams in 200 mL ZnSO$_4$ solution).

The fascinating phenomenon shown in Figure 5 is the notable similarity in the yield curves between water-insoluble and water-soluble products in the range of 0.50 to 1.50 concentrations. Beyond the 1.50 concentration, the yield of water-insoluble products increased steeply and then leveled off whereas the water-soluble products slowly decreased and then reversed to increase slightly at a 2.5 ligand concentration. In short, an enhancement of chelating yield at higher ligand concentration seemed more striking. However, if we take into account the change in yield against the ligand concentration, different results are shown in Table I.

It is obvious that at a lower ligand concentration it is more promising for the ONCMCh to chelate with Zn^{2+} ions. This situation (i.e., the ligand concentration effects the chelating yield) does not occur in chitosan with metal ions. It is known that the chelating capacity is not dependent on the amounts of adsorbent added.[26] This result is particularly important because changing the ligand concentration allows chelating polymers with various zinc contents to be obtained.

UV Spectra and ICP Analysis

In order to check the zinc content in the chelated specimens, UV analysis was conducted to qualitatively determine the amount of Zn^{2+} ions remaining in the solution. However, it is very difficult to directly measure the zinc content in the aqueous solution because Zn^{2+} ions are not a UV absorber. It is known that Zn^{2+} can form a blue water-soluble complex with Zincon in alkaline solution; hence Zincon can be added to the solution after the chelation reaction to determine the residual Zn^{2+} ions by UV spectroscopy.

The amount of zinc ions existing in the solution can be characterized by the occurrence of absorption peaks at about 500 nm (due to zinc-Zincon complex) and at about 620 nm (free Zn^{2+}) in the UV spectra. The change in zinc ion contents varied with the various experimental conditions presented in Figures 6–9.

Analysis of the UV spectra revealed that the variation in zinc content in each chelated product is also controlled by the kinetic experimental conditions, such as reaction time, temperature, ion concentration, and ligand concentration.

On the other hand, when comparing the UV spectra with the results of water-insoluble products by ICP, opposite trends in the curve changes between the zinc content in the solid (in the top of the figures) and the residual zinc in the solution (in the UV spectra) showed experimental conformity. The alteration in zinc content is certainly associated with different chelating mechanisms that will be discussed in detail in a subsequent publication.

Table I Changes in Percentage Yields with Ligand Concentrations

	Ligand Concn. (g/200 mL ZnSO$_4$)					
	0.50	0.75	1.0	1.50	2.00	2.50
A (%)	+20	+14	−4	−16	+9	−16
B (%)	+20	+6	+16	+12	+2	+8

A, water-insoluble product; B, water-soluble product. The percentage change against the ligand concentration: (−) decrease in percent; (+) increase in percent.
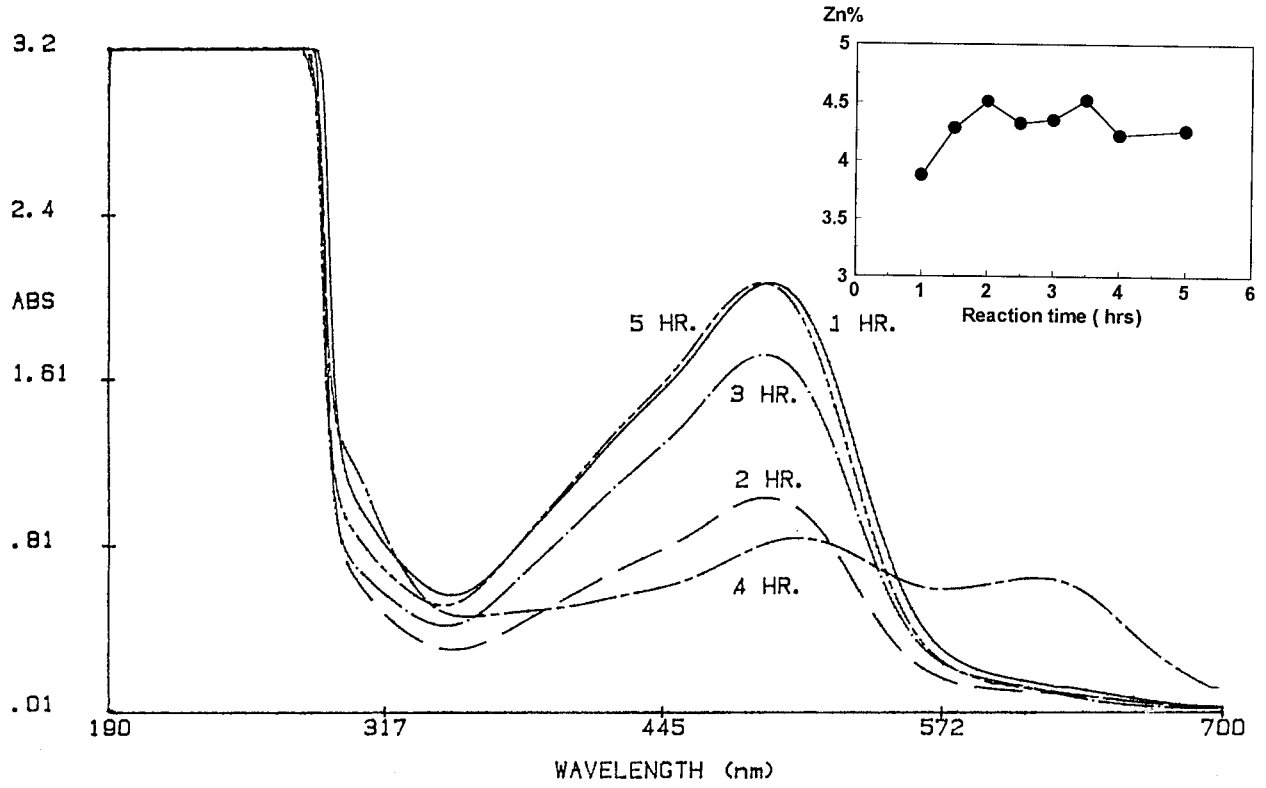
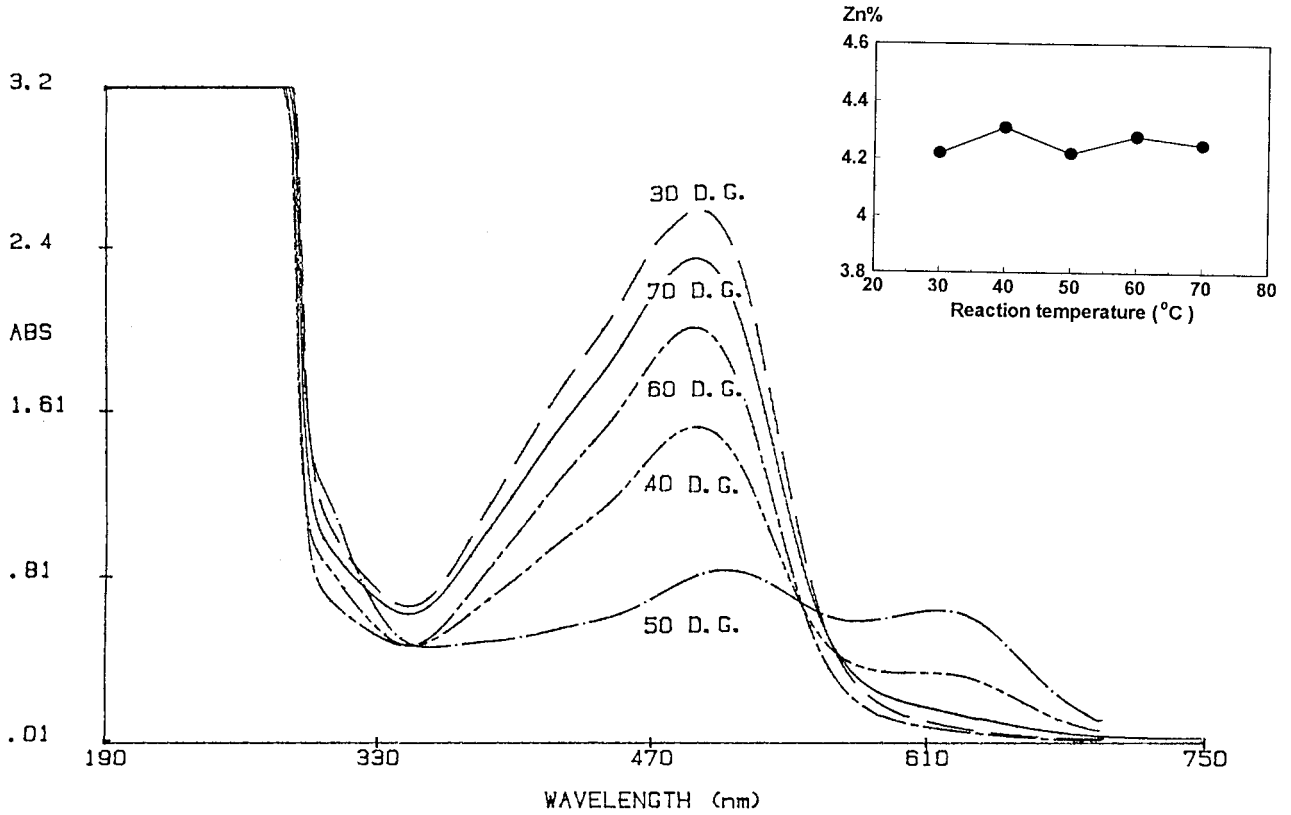**Figure 6**   Change in zinc content as a function of reaction time.

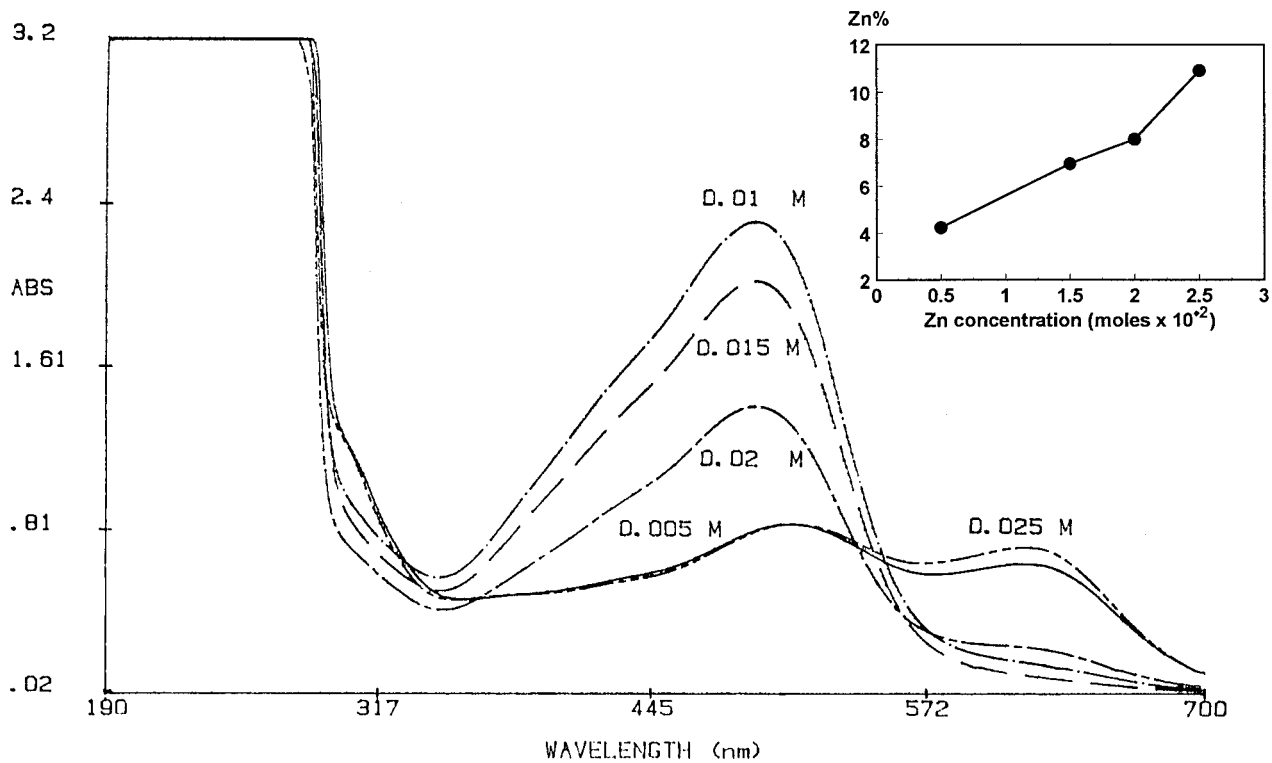**Figure 7**   Change in zinc content as a function of reaction temperature.

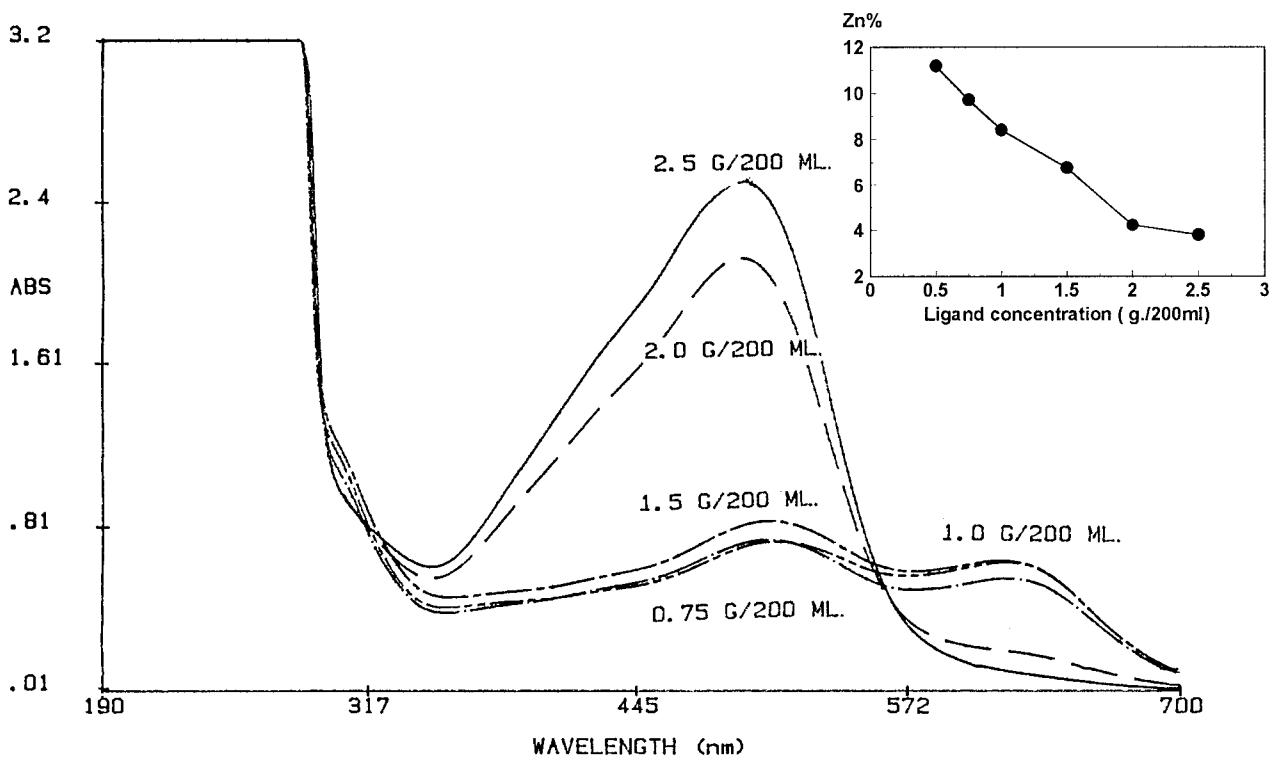**Figure 8**   Change in zinc content as a function of zinc concentration.

Figure 9   Change in zinc content as a function of ligand concentration.

CONCLUSIONS

The chelation between ONCMCh and zinc sulfate in aqueous solution was successfully demonstrated. It was found that the chelation processes were greatly controlled by the kinetic parameters; which are the reaction temperature, zinc ionic strength, and ligand concentration. The chelation processes were less dependent on the reaction time. Favorable conditions for ONCMCh-Zn^{2+} chelate formation were at the lower zinc ionic concentration ($0.005M$), temperature ($50°C$), and ligand concentration (0.5 g/200 mL $ZnSO_4$). Based on the studies by kinetic parameters and by ICP analysis of the changes in zinc concentrations, different complexing mechanisms were elucidated. It is important to recognize that the formation of water-insoluble products was mainly due to chelation between ONCMCh and zinc ions, and the formation of water-soluble products was mainly due to electrostatic attraction between ONCMCh and zinc ions. The understanding of the mechanisms of the chelation formation process would provide valuable information on the utilization of chelation products of ONCMCh-Zn^{2+} for specific applications.

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