

# Studies on the Critical Phase-Transition Behavior of Cholesteric *N*-Phthaloyl Chitosan/Dimethyl Sulfoxide Solutions by Five Techniques

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**ABSTRACT:** A selectively side-modified chitosan derivative, *N*-phthaloyl chitosan (PhthCs), was prepared. Five techniques, including circular dichroism (CD) spectropolarimetry, ultraviolet-visible (UV-vis) spectroscopy, small-angle light scattering (SALS), polarized optical microscopy (POM), and differential scanning calorimetry (DSC), were simultaneously applied to investigate the order-disorder phase transition for the lyotropic PhthCs/dimethyl sulfoxide system. POM observations suggested that the critical concentration was 48 wt %, which agreed well with the DSC measurements. A four-leaf SALS picture was recorded in biphasic solutions. The four-leaf type became a circular symmetric scattering picture with a further increase in the concentration. A sharp peak at 330 nm was recorded in dilute

isotropic solutions with CD measurements. The sharp peak turned into a broad tailed peak from 330 to 800 nm after the phase transition occurred. The sharp peak was induced by a chromophore (phthoyl group). The broad peak was attributed to the formation of short-range-ordered organization in the concentrated solutions. In UV-vis studies, when the system varied from isotropic to anisotropic, the absorption band turned into platform. These five techniques were proven to be feasible methods for measuring the phase transition of cholesteric liquid crystals. © 2003 Wiley Periodicals, Inc. *J Appl Polym Sci* 90: 583–586, 2003

**Key words:** UV-vis spectroscopy; differential scanning calorimetry (DSC); solution properties

## INTRODUCTION

A great number of rigid or semirigid natural polymers (e.g., proteins,<sup>1</sup> polypeptides,<sup>2</sup> celluloses,<sup>3–6</sup> and chitins and chitosans<sup>7–10</sup>) have been found to form lyotropic liquid-crystalline phases (or mesophases) under suitable conditions. The phase-transition behavior of the lyotropic mesophases and the liquid-crystalline textures have attracted the interest of many researchers. Theoretical studies<sup>11–13</sup> have enabled us to predict the value of the critical concentration ( $C^*$ ), the point at which the order-disorder or anisotropic-isotropic phase transition occurs, for many rigid or semirigid polymer solutions. However, an experimental phase diagram is still necessary for the comprehension of these systems. Therefore, several techniques have been developed to detect the formation of ordered phases. These approaches include classic polarized optical microscopy (POM), NMR spectroscopy,<sup>14</sup> IR spectroscopy,<sup>15,16</sup> refractometry,<sup>17–19</sup> and differential

scanning calorimetry (DSC).<sup>20</sup> Nevertheless, a comparison of the different methods has only rarely been reported.

Like other natural polymers, chitosan and its derivatives mostly exhibit cholesteric phases characterized by a twisted lamellar structure, which is known to have several important optical properties, such as optical rotatory dispersion, selective reflection, and circular dichroism (CD). In the past 2 decades, hundreds of scientific publications have detailed the optical characteristics of a large number of concentrated cholesteric solutions, using CD spectropolarimetry and/or ultraviolet-visible (UV-vis) spectroscopy. However, most of the attention has been focused on conformation studies (e.g., handedness, twisting sense, and pitch) of cholesteric mesophase solutions. In addition, the polymers used in these studies have nearly been limited to cellulose and its derivatives, as reviewed by Zugenmaier.<sup>21</sup>

Previously, we have reported the critical phase transition as a function of the side groups for a variety of liquid-crystalline chitin and chitosan derivatives in some proton solvents, mainly using POM.<sup>9,10,22–24</sup> In this study, we inspected the critical phase transition of the cholesteric phase for another chitosan derivative, *N*-phthaloyl chitosan (PhthCs), with three other techniques—CD, UV-vis, and small-angle light scattering (SALS)—in addition to traditional approaches with POM and DSC. We used all of these techniques for one

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polymer system to compare the feasibilities of these methods.

## EXPERIMENTAL

### Synthesis of PhthCs

A commercial reagent-grade chitosan with a weight-average molecular weight of  $7.4 \times 10^5$  and a degree of deacetylation of 84%, as supplied by Xiamen Second Pharmaceutical Factory (Xiamen, China), was used in this study after a full deacetylation treatment with the method proposed by Shigehiro et al.<sup>25</sup> The degree of deacetylation was almost 100% after this process.

PhthCs was then prepared according to the literature:<sup>26</sup> 18.0 g of phthaloyl anhydride and 320 mL of DMF were added to a flask, and they were followed by 4.0 g of pretreated chitosan ( $n_{\text{anhydride}}/n_{\text{NH}_2}$  molecular ratio = 5:1). The mixtures were stirred at 120°C under a nitrogen atmosphere for 7 h until a clear, homogeneous solution was achieved. Distilled and deionized water (400 mL) was then poured into the solution to make the polymer precipitate. The precipitate was collected through filtration and then was washed with ethanol and water several times. The wet outcome was dried *in vacuo* below 70°C to give a balanced product weight of about 7.1 g.

<sup>1</sup>H-NMR (Varian Unity 500-MHz NMR instrument with CD<sub>3</sub>SOCD<sub>3</sub>; ppm, USA) showed broad signals in region 2.7–5.3 and region 7.2–8.1; these were assigned to chitosan backbone protons and aromatic protons, respectively. From a comparison of the integrated aromatic proton signals and the backbone proton signals, the degree of the side-group substitution could easily be calculated to be approximately 1.0. IR (Nicolet Avator 360 with KBr; cm<sup>-1</sup>, USA) experiments showed  $\nu(\text{C}=\text{O}) = 1715$  (s) and 1770 (m) and  $\nu(\text{benzene ring}) = 722$  (m). The results of <sup>1</sup>H-NMR and IR experiments also indicated that the product was an imine instead of an amine. Therefore, a predication could be made that the phthaloyl group was almost introduced on the —NH<sub>2</sub> group at the C<sub>2</sub> position, and an imine ring was nearly formed after an amine chitosan intermediate was built. The ideal molecular structure of PhthCs is presented in Figure 1.

### Sample preparation

Solutions of PhthCs and dimethyl sulfoxide (DMSO) over a concentration range of 1 to 65 wt % were prepared in small glass vials separately. The vials were tightly sealed to prevent the solvent from vaporizing after the stirring. Most of the samples were aged at room temperature for 3 days and were made homogeneous in an oven at 100°C for 10 min before use.

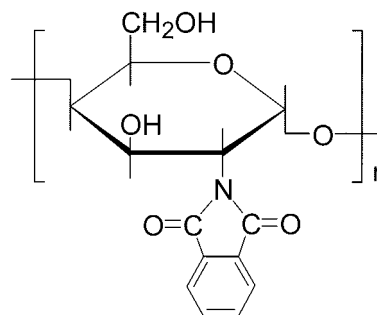


Figure 1 Ideal chemical structure of PhthCs.

### Measurements

For POM observations, the solutions were sandwiched between two glass slides to form sample cells. The cells were observed at 25°C under a polarized optical microscope (Olympus BH2, Japan). The incipient concentration at which birefringence appeared was defined as the critical concentration. SALS experiments were conducted on an LS-1 SALS apparatus (Yingkou, China) with the same sample cells.

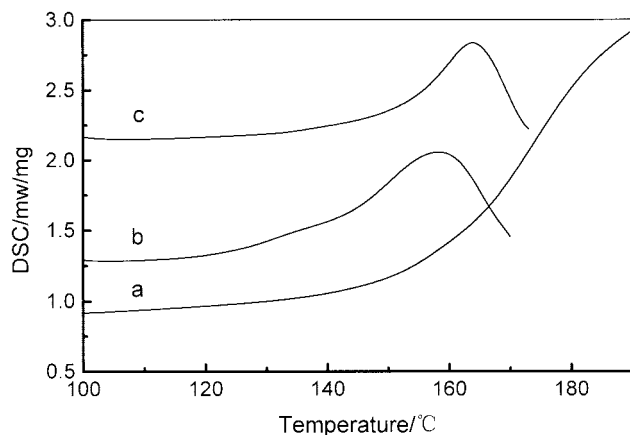
For the DSC measurements, 10.0-mg samples were sealed in an aluminum crucible. The crucible was then tested on a Netzsch DSC 204 thermal analyzer (Germany) at a heating rate of 20°C/min under a nitrogen atmosphere.

In CD and UV-vis measurements, for a polymer concentration greater than 10%, the solutions were sandwiched between two quartz planes 0.2–0.5 mm thick. However, for a polymer concentration less than or equal to 10%, a quartz cell with a volume of 1 mL was used instead of the quartz planes. CD spectra of the specimens were recorded on a Jasco 810 CD spectropolarimeter (Japan) with a band width of 1 nm, a data pitch of 0.1 nm, a scanning speed of 200 nm/min, a time constant of 1 s, and a temperature of 25°C. UV spectra were recorded with a Beckman DU-7400 (USA) and a Shimadzu UV-2501 PC (Japan) with the same specimens used for CD measurement.

## RESULTS AND DISCUSSION

Solutions of PhthCs and DMSO were observed under an optical microscope between two crossed polarizers.  $C^*$  of these systems was determined to be 43%. The characteristic textures of the lyotropic systems were found to be a domain texture and a plane texture, both of which were characteristics of cholesteric liquid crystals.

The results of POM matched well with DSC experiments. For solutions with a polymer concentration below  $C^*$ , no obvious peak was recorded in DSC traces at the temperature used. However, for solutions that presented birefringence at room temperature under POM, a broad endotherm peak was observed (see Fig.

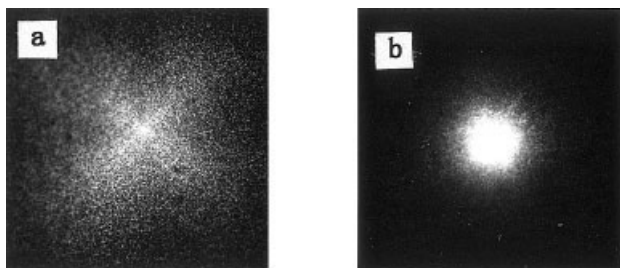


**Figure 2** DSC traces of PhthCs/DMSO solutions with concentrations of (a) 35, (b) 48, and (c) 55%.

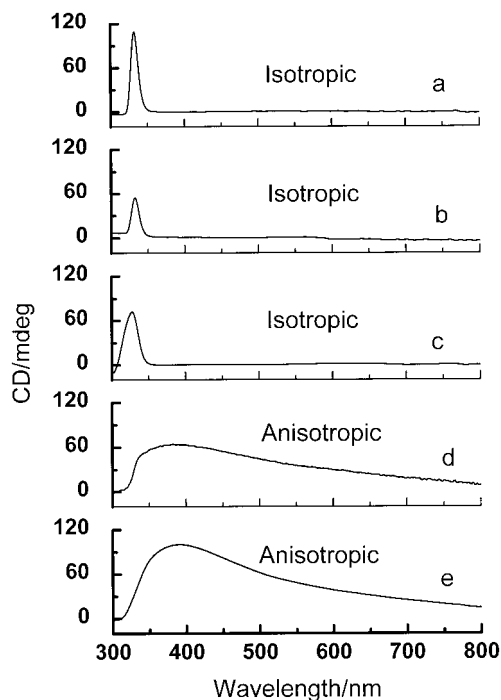
2). The peak was not related to the solvent, in that the boiling point of DMSO was 189°C at atmospheric pressure, but was associated with the order–disorder transition of the liquid-crystalline phases, that is, the so-called clear temperature.

SALS is a powerful tool for studying crystalline polymer morphology. Essentially, both liquid crystals and crystals are constructed with ordered molecular structures. Therefore, SALS experiments were also used to study the ordered textures forming in liquid-crystalline solutions. In our experiments, for solutions that did not exhibit any birefringence under POM, no scattering picture was observed by SALS. However, for biphasic solutions with concentrations of 43–55%, a four-leaf-type scattering picture was recorded [see Fig. 3(a)]. The four-leaf-type scattering picture turned into a circular symmetric type when the polymer concentrations were above 55%, at which point a homogeneous liquid-crystalline phase formed [see Fig. 3(b)]. These two kinds of scattering pictures were attributed to the formation of the domain texture and plane texture, respectively, that were observed under POM.<sup>27</sup> These results also agreed well with the POM experiments.

The CD spectra of polysaccharides are often uninformative because there are no chromophores with strong absorption bands in the UV–vis region in many



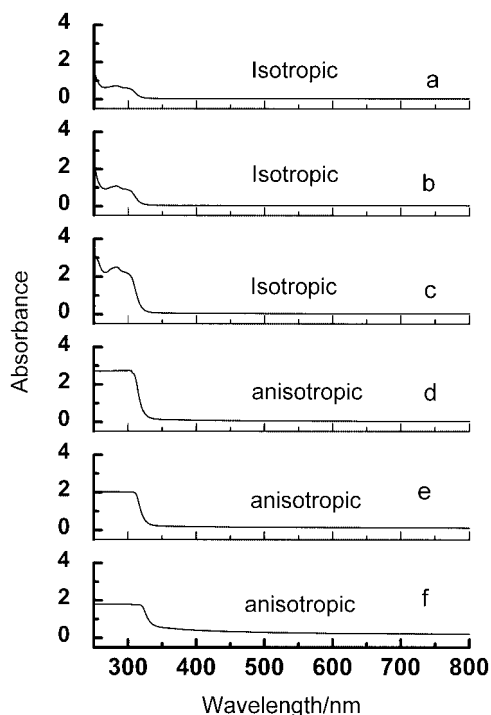
**Figure 3** SALS patterns of PhthCs/DMSO solutions (work distance = 10 cm) with concentrations of (a) 45 and (b) 55%.



**Figure 4** CD spectra of PhthCs/DMSO solutions with concentrations of (a) 1, (b) 25, (c) 40, (d) 45, and (e) 50%.

cases. However, for their derivatives (e.g., chromophore groups introduced into the polymer chains), CD spectra are easily achieved. In our studies, a phthaloyl group was selectively introduced into a chitosan molecule. This modification not only resulted in good solubility and extended liquid-crystalline properties in organic solvents but also benefited CD investigations. Figure 4 shows CD spectra of PhthCs/DMSO solutions with different concentrations. For polymer concentrations below  $C^*$ , a sharp absorption peak at about 330 nm was found to exist for all the samples. For concentrations above  $C^*$ , a broad peak tailing from the UV region to the visible region was found.

The peak at 330 nm existed regardless of the supermolecular structure of the mesophase because this peak still appeared when the solutions were isotropic. We also found that this peak was irrelevant to the solvent, in that the solvent DMSO was not chiral and had no chromophore with the absorption band at this region. Therefore, this peak was attributed to the absorption of the phthaloyl group affected by the inherent twisted conformation of macromolecules in solution. The tailed band was interpreted by Rout et al.<sup>26</sup> as the formation of mesophases consisting of only short-range-ordered domains not showing uniform cholesteric organizations. We noticed that CD signals of the biphasic systems differed distinctively from those of the monophasic isotropic solutions because of the peculiar structure of the cholesteric liquid-crystalline phases. Obviously, this kind of mutation for CD



**Figure 5** UV spectra of PhthCs/DMSO solutions with concentrations of (a) 15, (b) 25, (c) 35, (d) 45, (e) 50, and (f) 55%.

spectra provided us a new veracious approach for determining the critical phase transition for the liquid crystals, which were cholesteric in state.

The UV-vis absorption spectra (250–600 nm) of PhthCs/DMSO solutions (dilute to concentrated) are shown in Figure 5. The dilute solution spectrum shows two absorption bands at 289 and 306 nm induced by the  $\pi \rightarrow \pi^*$  transition of the conjugate phthaloyl groups. Interestingly, the absorption for the entire specimen turned into a platform when the solution concentrations were above a certain concentration ( $C^*$ ). The platform was not caused by the cutoff of the absorbance. Although the reason that this happened is not clear at present, we still believe that the phenomena were not accidental because we repeated our measurements several times and obtained the same results. We also tried using another solvent, DMF (dimethyl formamide), and the platform recurred under similar conditions. Furthermore, for the same specimen, nearly identical data were recorded on two instruments provided by different manufacturers (Beckman DU-7400 and Shimadzu UV-2501 PC). These facts made us believe that the conversion was undoubtedly related to the formation of the ordered phase in the solutions. Therefore, we believe UV-vis measurements are also a feasible way of determining the phase transitions of liquid-crystalline solutions for this chitosan derivative.

## CONCLUSIONS

Five techniques—CD, UV-vis, SALS, POM, and DSC—were simultaneously applied to investigate the order-disorder phase transition for the lyotropic PhthCs/DMSO system. Although CD, UV-vis, and SALS have seldom been used to study the critical phase transitions of liquid crystals, our work has proven that they are also feasible methods for determining this mutation in concentrated polymer solutions, and the results of these techniques can agree very well. To some degree, it is more sensitive and reliable to detect the critical phase transition with CD and UV-vis than with the traditional POM method, especially for cholesteric-type liquid crystals.

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