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# Columnar Liquid Crystals in Polypeptides. 1. A Columnar Hexagonal Liquid Crystal Observed in Poly($\gamma$-octadecyl L-glutamate)

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ABSTRACT: Poly($\gamma$-octadecyl L-glutamate) can form a new type of thermotropic phase in addition to a cholesteric phase. This new phase appears in a higher temperature region than the cholesteric phase, and the transition between them can be determined by microscopic observation of the mesophase texture. This new phase can be distinguished from the cholesteric phase as it exhibits a fan-shaped texture and extremely high viscosity. Furthermore, an X-ray examination indicates that in this phase the rigid $\alpha$-helical molecules are packed into a regular two-dimensional hexagonal lattice. The new phase was thus assigned to a columnar hexagonal phase, which is first observed in a thermotropic polypeptide system.

## Introduction

Rigid polymers in solution form liquid-crystalline phases above a critical concentration depending on their axial ratio. Nematic or cholesteric (chiral nematic) liquid-crystalline phases have been experimentally recognized and theoretically predicted by Onsager,[1] Flory,[2] and others.[3,4]

Much less was known about the other types of liquid crystals that may be formed by these rigid polymers, but recently experimental data indicate that rigid polymers may form liquid crystals that are neither nematic or cholesteric. Livolant and Bouligand[5] and Lee and Meyer[6] have suggested from the analysis of microscopic textures that the synthetic polypeptide, poly($\gamma$-benzyl glutamate), may form a hexagonal columnar liquid crystal in its lyotropic state although a systematic study of this phenomenon has not been performed. Strzelecka et al.[7] have reported that a smectic-like phase in addition to a cholesteric phase can be seen in aqueous solutions of DNA, and subsequently, Livolant et al.[8] observed a columnar hexagonal phase in a similar DNA lyotropic system. Ballauff and Schimidt,[9] Wegner et al.,[10] and Ebert et al.[11] have shown that aromatic polyesters with long aliphatic side chains can form smectic-like liquid crystals with a characteristic layered structure, as revealed by X-ray analysis. These liquid crystals can be easily differentiated from the nematic or cholesteric liquid crystal formed by this kind of rigid polymers, but the classification of these mesophases has not clearly been made. On the theoretical side, Kimura et al.[12,13] and Frenkel[14] have predicted that, in rigid polymeric systems, smectic and columnar liquid crystals may arise as the result of an excluded-volume effect of the hard rods. The search for new liquid crystals of rigid polymers and their identification are proceeding now in both experimental and theoretical aspects.

This report describes the first observation of a thermotropic columnar liquid-crystalline phase formed in a polypeptide system. The polymer examined is poly($\gamma$-octadecyl L-glutamate), which has been already found to form a thermotropic cholesteric liquid-crystalline phase.[15] A columnar hexagonal phase appears at a higher temperature region than the cholesteric phase and was examined by microscopic observations and X-ray diffractions. The ability of this polymer to form such a phase will be discussed with respect to the structure of component molecules.

## Experimental Section

Poly($\gamma$-octadecyl L-glutamate) (PG-18) was synthesized by an ester-exchange reaction between poly($\gamma$-methyl L-glutamate) (PMLG) and *n*-octadecyl alcohol.[15] The reaction was performed in 1,2-dichloroethane with *p*-toluenesulfonic acid as a catalyst at 60 °C. The complete replacement of the methyl groups by octadecyl groups was confirmed by the NMR spectra. Five samples of PG-18 with degrees of polymerization (DP) of 70, 110, 220, 400, and 700 were prepared from PMLG with the corresponding values of DP. These specimens were designated as PG-18-$X$ where $X$ indicates the degree of polymerization.

Optical microscopic observation of the mesophase textures was made with an Olympus BH-2 polarizing microscope equipped with a Mettler FP82 hot stage. Wide-angle X-ray patterns were recorded with a flat-plate camera using a Rigaku-Denki X-ray generator with Ni-filtered Cu K$\alpha$ radiation. The sample was kept in a glass capillary tube and heated to the desired temperature by a heater that was maintained to within 1 °C of the desired temperature. The film-to-specimen distance was determined by calibration with silicon powder. The optical rotational dispersion (ORD) curves were measured with a JASCO automatic recording spectrometer.

## Results

As described in previous papers,[15–17] poly($\gamma$-octadecyl L-glutamate) has an $\alpha$-helical main chain surrounded by octadecyl side chains. The side chains are long enough to form a crystalline phase composed of paraffin-like crystallites. These crystallites force the $\alpha$-helical main chains to pack into a characteristic layered structure with the crystallites located between the layers.[15]

After melting of the side-chain crystallites at a temperature of 60 °C, these polymers form a thermotropic liquid-crystalline phase. The liquid crystal is fluid and its cholesteric nature can be well identified by the presence of its fingerprint texture with the fine retardation lines in the microscopic observation.[18–20] The typical texture of a cholesteric phase is shown in Figure 1a. The pitch can be determined from the spacings between the lines[19] and is plotted against temperature for PG-18-220 in Figure 2. As shown in Figure 2, the pitches are observed to be in the range of 1–2 $\mu$m for all specimens employed here. It is also apparent that there is a relatively weak temperature dependence in accordance with results obtained for thermotropic cholesteric phases of polypeptides with the long alkyl side chains.[20] From the negative sign of optical rotation as measured in the wavelength region of 400–700 nm, it was found that the cholesteric phases of PG-18

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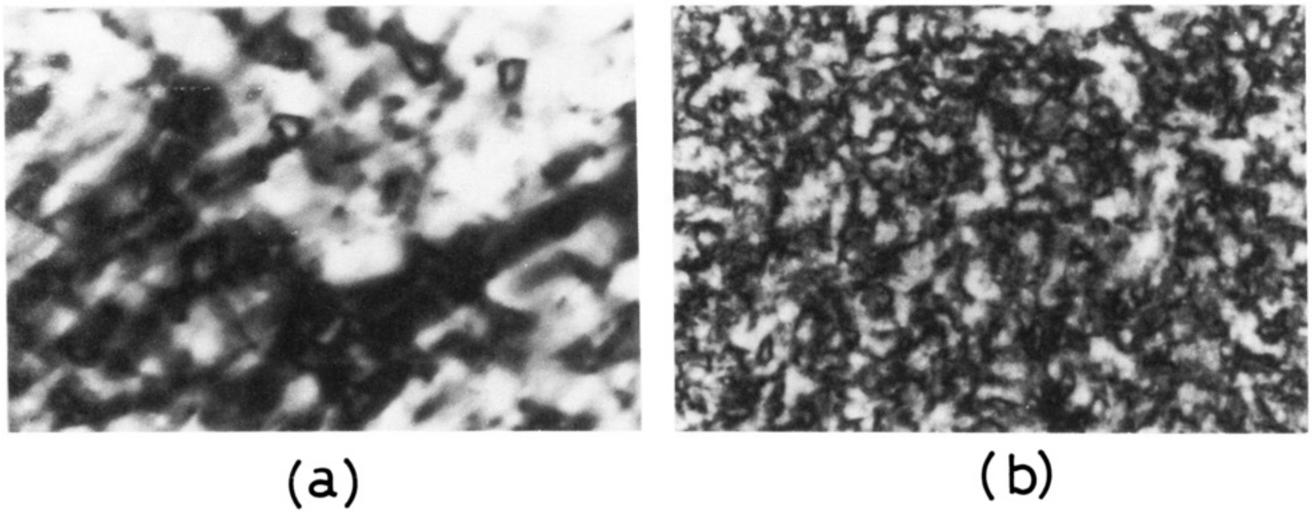

**Figure 1.** Optical microphotographs observed for the mesophases of PG-18-220: (a) a cholesteric mesophase at 150 °C and (b) the high-temperature mesophase at 210 °C.

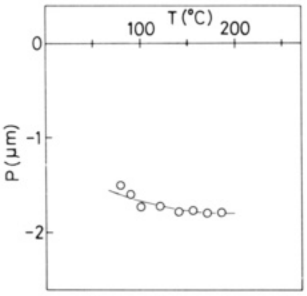

**Figure 2.** Temperature dependence of the cholesteric pitch (P) observed for PG-18-220. Here, the negative value of pitch indicates that the specimen forms a left-handed cholesteric structure.

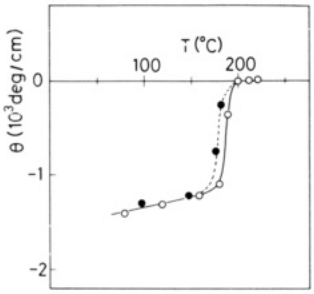

**Figure 3.** Variation of optical rotation ($\theta$) with temperature as observed for a specimen of PG-18-220 with a thickness of 10 $\mu$m at a wavelength of 600 nm. The open and closed circles were obtained on heating and cooling, respectively, after annealing for 20 min at respective temperatures.

samples assume a left-handed helicoidal structure over the observed temperature region.[19]

For PG-18-220, the cholesteric mesophase is observed over the temperature region of 60–190 °C. On heating beyond this region, the texture gradually changes, and around 200 °C, the fingerprint texture is replaced by a fan-shaped or broken fan-shaped texture as shown in Figure 1b. Simultaneously, the viscosity of the mesophase increases substantially. Furthermore, the negative optical rotation (OR) attributable to the cholesteric helix gradually decreases and finally approaches a value of zero at temperatures above 200 °C (Figure 3) although the weak OR of the individual $\alpha$-helical molecules is still observed. These observations indicate that there is a transition from a cholesteric mesophase to another kind of mesophase at temperatures above 200 °C. The transition is completely reversible, with the cholesteric mesophase reappearing on cooling below 190 °C, which can be realized in Figure 3. The corresponding transition at around 200 °C could not be detected by DSC, suggesting that it is a weakly first-order or a second-order transition.

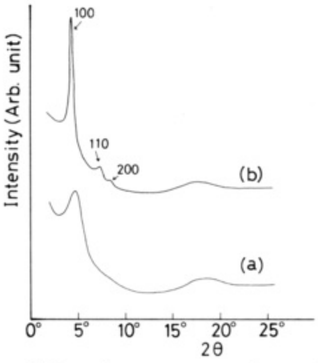

**Figure 4.** X-ray diffraction patterns observed for the mesophases of PG-18-220: (a) a cholesteric mesophase at 150 °C and (b) the high-temperature mesophase at 210 °C.

The structural features of the two mesophases were analyzed by X-ray diffraction. Curve a of Figure 4 shows the X-ray diffraction profile for a cholesteric mesophase of PG-18-220 taken at 150 °C. The profile includes two inner and outer broad bands with the reciprocal spacings of $^1/_{20}$ and $^1/_{4.7}$ Å$^{-1}$. These are expected for a cholesteric mesophase; the former band reflects a somewhat disordered lateral packing of the $\alpha$-helical rods, and the latter may result from the fifth-layer line of an $\alpha$-helical molecule and/or the alkyl side chains with a high degree of orientational disorder. On the other hand, the high-temperature mesophase exhibits a different X-ray profile as shown in curve b of Figure 4. This diffraction profile shows three sharp inner reflections in addition to the outer broad reflection of around 4.7 Å that remains unchanged. The lattice spacings of the sharp reflections are 21.1, 12.1, and 10.6 Å. These are in a ratio of 1, $1/\sqrt{3}$, and 1/2, indicating that the $\alpha$-helical rods are laterally packed into a regular two-dimensional hexagonal lattice. The density of 0.83 g/mL is calculated on the assumption that each $\alpha$-helical molecule passes through each unit cell. This value is in agreement with the actual bulk density of around 1 g/mL[15] when considering the thermal expansion of a molar volume.

It is noteworthy that the phase transition of the cholesteric mesophase to the higher temperature mesophase depends on the DP of polymers. This can be seen in Figure 5 where the transition temperatures are plotted against

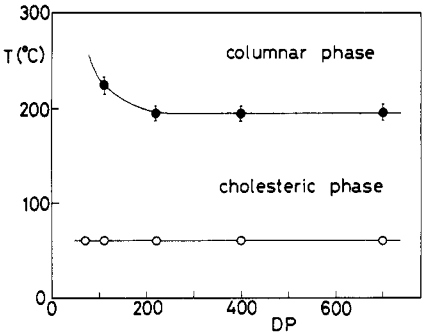

**Figure 5.** Variation in the transition temperatures with the degree of polymerization (DP) of the polymer. The open circles indicate the melting temperature of side-chain crystallites, and the closed circles represent the transition temperature from the cholesteric phase to the columnar hexagonal phase.

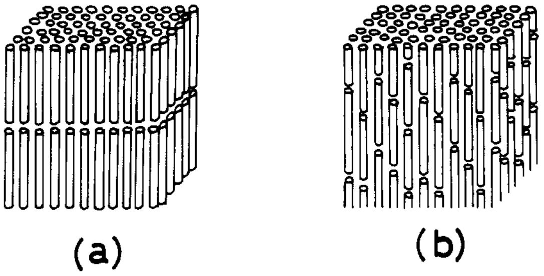

**Figure 6.** Model of two liquid-crystalline phases that satisfy the present observations. In the smectic B phase, (a) a layer with a thickness close to the molecular length is constructed. Within a layer, the polymers are packed into a two-dimensional hexagonal lattice. In the columnar hexagonal phase, (b) the polymers are packed into a similar two-dimensional lattice but with translational disorder along the polymer-chain axis.

DP. For the materials with a DP of more than 200, the transition temperature remains constant at around 200 °C, but it increases to 230 °C for PG-18-110, and finally it may move to a temperature beyond the decomposition temperature of 250 °C for PG-18-70, the lowest DP sample used in this study. Although the variation in the DP is limited, the results show that the transition temperature decreases with an increase in the DP; in other words, the high-temperature mesophase is more stable at higher degrees of polymerization.

## Discussion

The observation of a new type of mesophase, other than cholesteric, for a liquid-crystalline polypeptide is a significant discovery that may help us better understand the liquid-crystalline behavior of the rigid polymeric systems. The structural properties of this new mesophase are summarized as follows.

(1) The mesophase is highly viscous and exhibits a fan-shaped texture.

(2) The mesophase possesses no supramolecular helicoidal structure.

(3) The $\alpha$-helical molecules are packed into two-dimensional hexagonal lattice.

Smectic and columnar hexagonal phases can both be considered as liquid-crystalline phases that exhibit such structural properties. In a smectic phase, the layer consists of a parallel side-by-side arrangement of polymer molecules; hence, the thickness of each layer should be close to the average length of the molecules, as illustrated in Figure 6a. Since the molecules are laterally packed into a two-dimensional hexagonal lattice and no helicoidal structure exists, the mesophase may be assigned to the smectic B phase.[21] On the other hand, the columnar hexagonal phase can be distinguished from the smectic B phase only on the basis that it possesses translational disorder in a direction along the polymer-chain axis (see Figure 6b). At the present time, we have no direct evidence to rule out the smectic B phase as the high-temperature phase, but it would be difficult to accept the assignment of a smectic layered structure when considering the wide distribution of molecular lengths generally observed in the polymeric systems and the low number density of polymer ends. It would therefore seem more reasonable to assign this new phase to a columnar hexagonal phase.

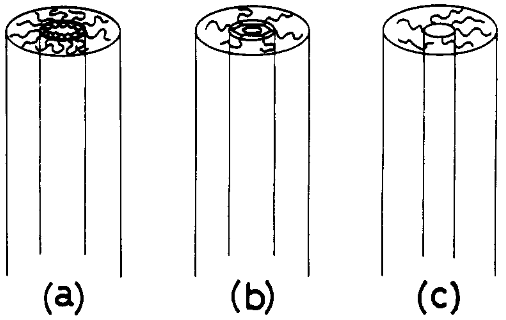

**Figure 7.** Models of columns formed by three different types of molecules. Columns a and b are formed by amphiphilic molecules and disk-shaped molecules, respectively. Both molecules have two groups which are different with respect to the polarity and/or geometric shape, and their columns may be viewed as a kind of micellar structure derived from such differences. Similar structural peculiarities can also be seen in the polypeptide molecule that may behave as a column (c) by itself.

Two classes of molecules are known to form a columnar hexagonal phase: amphiphilic molecules[22] and disk-shaped molecules with long aliphatic side groups.[23] In both cases, the molecules have two groups that are different with respect to their polarity and/or geometric shape, and their columnar phases may be viewed as a kind of micellar structure derived from such differences (see Figure 7a,b). A similar structural peculiarity can be seen in the present material since the molecule is composed of a polar $\alpha$-helical main-chain core surrounded by aliphatic side-chain groups as illustrated in Figure 7c. For this reason, it is also plausible that a columnar mesophase is formed.

We next consider the structural factors responsible for the formation of columnar liquid crystals in this type of rigid polymeric system. As mentioned above, the peculiarity of chemical structure in the molecule is likely to facilitate the formation of a columnar liquid crystal in the present system. However, this may not be the only factor since columnar phases have been observed in two other systems of rigid polymers with dissimilar chemical structures. One of these is the lyotropic system of poly($\gamma$-benzyl glutamate)[6] and another the lyotropic DNA system;[8] in both systems, the columnar phase is observed at a higher concentration region than the cholesteric phase. Although complex interactions between polymer and solvent may be taking place in these systems, the results indicate that other factors that may be related to the anisotropy of molecule are included in the formation of a columnar liquid-crystalline phase. On this point, it is very interesting to note that the columnar hexagonal phase has been theoretically predicted to appear as the result of excluded-volume effects of the hard rods.[12-14]

Finally, we must comment on the exciting and significant phenomenon where the highly ordered columnar phase arises at a higher temperature region than the cholesteric mesophase. This phenomenon can be understood as an inverse sequence which has been rarely observed even in liquid-crystalline systems of low molecular weight materials.[21] At the present time, we cannot offer a reasonable explanation for this phenomenon, although it can be speculated that the anisotropic shape or large dipole moment of the present α-helical molecules are responsible for this behavior since the columnar phase is more stable at higher degrees of polymerization as shown in Figure 5.

To conclude, the discovery of a columnar phase in addition to the inverted cholesteric-columnar sequence in a thermotropic polypeptide system will lead to some new fields of research in the area of rigid-rod liquid-crystalline polymers.

## References and Notes

(1) Onsager, L. *Ann. N.Y. Acad. Sci.* **1949**, *51*, 627.
(2) Flory, P. J. *Proc. R. Soc. London* **1956**, *A234*, 60.
(3) Grosberg, A. Y.; Khokhlov, A. R. *Adv. Polym. Sci.* **1981**, *41*, 53.
(4) Odijk, T. *Macromolecules* **1986**, *19*, 2313.
(5) Livolant, F.; Bouligand, Y. *J. Phys.* **1986**, *47*, 1813.
(6) Lee, S.; Meyer, R. B. *Liq. Cryst.* **1990**, *7*, 451.
(7) Strzelecka, T. E.; Davidson, M. W.; Rill, R. L. *Nature* 1988, *331*, 457.
(8) Livolant, F.; Levelut, A. M.; Doucet, J.; Benoit, J. P. *Nature* **1989**, *339*, 724.
(9) Ballauff, M.; Schimidt, G. F. *Mol. Cryst. Liq. Cryst.* **1987**, *147*, 163.
(10) Rodriguez-Parada, J. M.; Duran, R.; Wegner, G. *Macromolecules* **1989**, *22*, 2507.
(11) Ebert, M.; Herrmann-Schonherr, O.; Wendorff, J. H.; Ringsdorf, H.; Tschirner, P. *Liq. Cryst.* **1990**, *7*, 63.
(12) Hoshino, M.; Nakano, H.; Kimura, H. *J. Phys. Soc. Jpn.* **1979**, *46*, 1709.
(13) Kimura, H. Private communication.
(14) Frenkel, D. *Liq. Cryst.* **1989**, *5*, 929.
(15) Watanabe, J.; Ono, H.; Uematsu, I.; Abe, A. *Macromolecules* **1985**, *18*, 2141.
(16) Ono, H.; Watanabe, J.; Abe, A. *Koubunshi Ronbunshu* **1988**, *45*, 69.
(17) Yamanobe, T.; Tsukahara, M.; Komoto, T.; Watanabe, J.; Ando, I.; Uematsu, I.; Deguchi, K.; Fujito, T.; Imanari, M. *Macromolecules* **1988**, *21*, 48.
(18) Watanabe, J.; Goto, M.; Nagase, T. *Macromolecules* **1987**, *20*, 298.
(19) Watanabe, J.; Nagase, T. *Macromolecules* **1988**, *21*, 171.
(20) Watanabe, J.; Nagase, T.; Ichizuka, T. *Polym. J.* **1990**, *22*, 1029.
(21) Gray, G. W.; Goodby, J. W. G. In *Smectic Liquid Crystals*; Leonard Hill: Glasgow and London, 1984; p 25.
(22) Luzzati, V.; Husson, F. *J. Cell. Biol.* **1962**, *12*, 207.
(23) Chandrasekhar, S. *Mol. Cryst. Liq. Cryst.* **1981**, *63*, 171.