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SHORT COMMUNICATIONS

Thermotropic Polypeptides VIII. Anomalous Phase Behavior in Low Molecular-Weight Poly(γ-octadecyl L-glutamate)

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Rigid rod polymers in solution were found to 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 is known about other types of liquid crystals that may be formed by the rigid polymers, but recently experimental data indicate that rigid polymers may form liquid crystals that are neither nematic nor cholesteric. Livolant and Bouligand[5] and Lee and Meyer[6] have suggested from the analysis of microscopic textures that the synthetic polypeptide, poly(γ-benzyl glutamate), may form hexagonal columnar liquid crystals in its lyotropic solution. The authors[7] also found the thermotropic hexagonal columnar phase in addition to the cholesteric phase in poly(γ-octadecyl L-glutamate). Strzelecka *et al.*[8] 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.*[9] observed a columnar hexagonal phase in a similar DNA lyotropic system. The smectic A phase was also observed in the lyotropic solution of tobacco mosaic virus by Meyer *et al.*[10] On the other hand, several interesting liquid crystalline phases with a characteristic layered structure have been found in the aromatic polyesters with long aliphatic side chains initially by Ballauff and Schmidt,[11] and succeedingly by Wegner *et al.*,[12] Ebert *et al.*,[13] and Harkness and Watanabe.[14] These liquid crystals can be easily distinguished from the nematic or cholesteric liquid crystals, but the structural details of these mesophases have not clearly been given. On the theoretical side, Kimura *et al.*[15] and Frenkel[16] have predicted that, in hard-rod particles, 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 short report describes the anomalous phase behavior of a thermotropic polypeptide, which suggests two types of liquid crystals other than the well-known cholesteric. The polymer examined here is poly(γ-octadecyl L-glutamate), which was prepared by ester-exchange reaction of poly(γ-methyl L-glutamate) with a degree of polymerization of 70 and octadecyl alcohol. It is designated PG-18-70 as in a previous paper.[7]

Poly(γ-octadecyl L-glutamate) (PG-18) has an α-helical main chain surrounded by octadecyl side chains. The side chains are long enough to form a crystalline phase composed of paraffin-like crystallites.[17] These crystallites force the α-helical main chains to pack into a characteristic layered structure with the crystallites located between the layers. After melting of side-chain crystallites at a temperature around 60°C, the polymer forms a thermotropic liquid crystalline phase which is fluid under the shearing.[17]

The liquid crystalline phase had been considered to be only a cholesteric, but the low molecular-weight PG-18-70 was found to exhibit several interesting optical textures which suggest the existence of a few other types of liquid crystals. The typical microscopic textures are shown in Figures 1a to 1e. Here, it should be noted that mesophases were annealed at least for a day to assure phase equilibrium at respective temperature since they are fairly viscous. The first typical texture of Figure 1a was observed for the mesophase at 70°C. In this case, the optically isotropic phase coexists with the anisotropic phase so that the entire morphology appears with the worm-like texture composed of an alternative arrangement of black and white stripes. The second one as observed at 110°C is shown in Figure 1c, where the almost entire phase is anisotropic and exhibits the pseudo-focal conic texture. The last one observed at 180°C is typical of the cholesteric phase as shown in Figure 1e. These observations of textures indicate that there are at least three different phases as the thermotropic mesophases of the PG-18-70.

At first, we refer to the worm-like texture. This kind of texture is observed in a temperature region from 60°C to 90°C. The worm-like texture is well developed after annealing for 1 day at respective temperature.

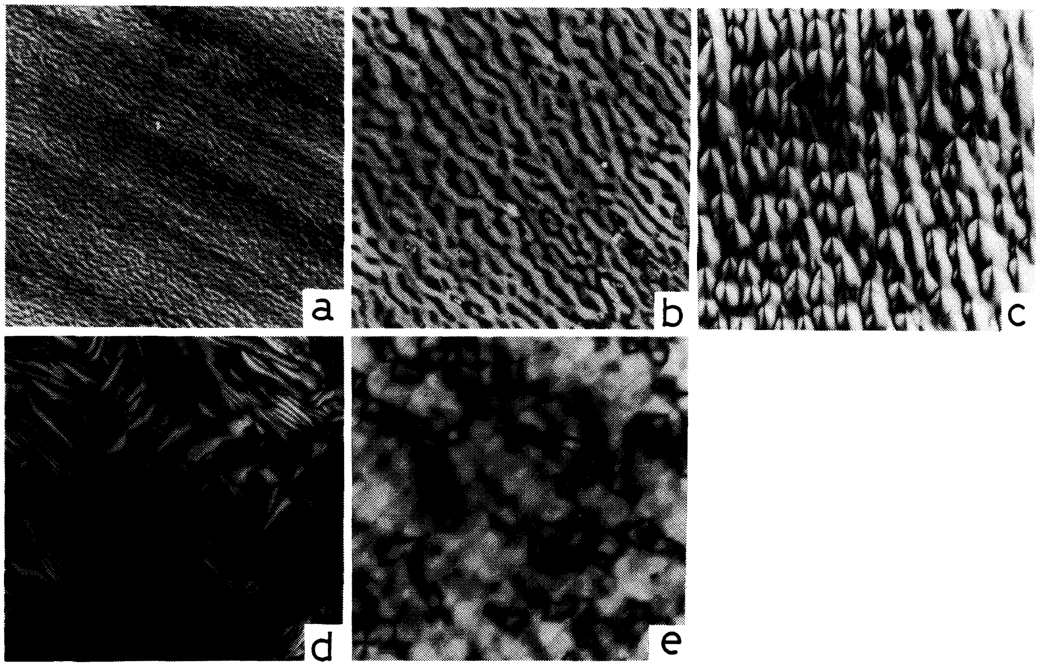

Figure 1. Liquid crystalline phases observed by polarized light microscopy. A sample of PG-18-70 with a thickness of 10 μm was placed between glasses, sealed with glue, and annealed for a day. Samples were observed through crossed polarizers with the Olympus BH-2 polarizing microscope (Magnification × 200). a and b, the worm-like textures observed at 70°C and 80°C, respectively; c, the pseudo-focal conic texture observed at 110°C; d, the texture including the transition bars observed at 130°C; e, the cholesteric finger-printed texture with equally spaced lines observed at 180°C.

The averaged width of the worms increases with annealing time and finally reaches a constant value, which increases with preparation temperature (refer to Figures 1a and 1b). Initially, we considered that the black and white stripes in this texture correspond to the periodic retardation lines due to a superhelical structure of some other phase than the cholesteric phase. However, this assignment can be ruled out from no observation of optical rotation. The worm-like texture is hence likely to arise from the phase separation of two different phases. One phase is optically isotropic and another is the anisotropic phase. On heating beyond 90°C, in fact, the domain structure exhibiting the phase separation of two phases becomes more distinct. The domain of anisotropic phase substantially grows leaving the optically isotropic phase as small droplets. The coexistence of the two phases is also clarified from the DSC measurements which were examined with respect to the crystallization of side chains. In Figure 2 are shown the DSC thermograms observed on cooling the mesophases. Two transition peaks well separated can be observed for the mesophases prepared at 60°C to 90°. One broad peak appears at 25°C and another sharp one at 45°C. It is also apparent that an amplitude of the broad peak decreases with a preparation temperature of mesophase while the sharp peak becomes predominant. This trend is reasonable if the crystallization occurs in a different manner between the two phases; that is, the broad and sharp peaks arise from the optically isotropic and anisotropic phases, respectively. The broad appearance of the transition peak at a lower temperature for the isotropic phase indicates the poor organization of side chains into a crystalline form which may be caused by a lack of the orientational order in a packing of α-helices.

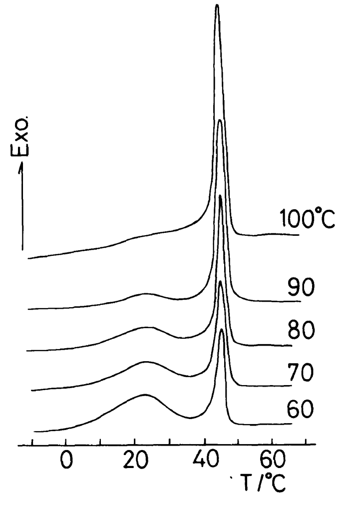

Figure 2. DSC thermograms exhibiting transitions due to crystallization of side chains. The measurements were performed with a Perkin–Elmer DSC-II calorimeter at a cooling rate of 10°C min^{-1}. The temperature given on each DSC curve indicates a preparation temperature of the mesophase.

Heating to a temperature of 110°C, the anisotropic phase occupies almost all the space. As mentioned above, this phase shows the pseudo-focal conic texture (see Figure 1c) and also no distinct optical rotation. The uniaxial orientation produced by shearing does not disappear even after annealing for a long time. This can be recognized from a well alignment of focal conic domains in Figure 1c and was also confirmed from the oriented X-ray pattern of Figure 3a taken for the same sample, in which the reflection attributable to a lateral packing of α-helices appears with the strong intensity on equatorial line. Such preservation of the uniaxial orientation can distinguish this phase from the cholesteric phase in which the uniaxial orientation immediately disappears because of the formation of helical twisted structure (refer to Figures 1e and 3b).

Further heating to the temperatures above 110°C results in the transition bars[18] (Figure 1d) and the anisotropic phase exhibiting the focal conic texture gradually disappears. Simultaneously, the well-known cholesteric phase appears, which can be assured from its characteristic finger printed texture with the equally spaced lines and also from its distinct optical rotation. The coexistence of two anisotropic phases is likely to expand in a temperature region from 120°C to 150°C, and

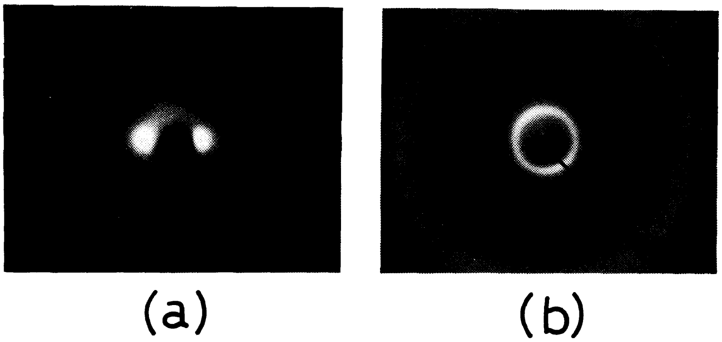

Figure 3. X-Ray pattern taken (a) for the mesophase at 110°C and (b) for the mesophase at 180°C by a Rigaku-Denki X-ray generator. The same samples giving the textures of Figures 1c and 1e were used for the measurement. The broad reflection with a spacing of 22 Å in the photographs was due to lateral packing of α-helices.

beyond this temperature region the uniform cholesteric domain can be attained as shown in Figure 1e. The uniformity of cholesteric phase is maintained up to a decomposition temperature of around 250°C.[19]

In summary, PG-18-70 forms three types of thermotropic phases after the side-chain crystallites melt. One of the phases, observed in the lowest temperature region, is optically isotropic, although the uniform domain of this phase cannot be attained even at a lowest mesophase temperature of 60°C. The second phase is optically anisotropic which is distinguished from the cholesteric phase finally appearing in the highest temperature region.[19]

With respect to the optically isotropic phase, we can consider two types of phases. One is a cubic isotropic mesophase and another is just an isotropic liquid. The cubic isotropic phases have been discovered in the certain lipids[20,21] and in the certain low molecular-weight mesogenic materials.[22] They are called the cubic amphiphilic phase and smectic D phase. Both may have a jointed-rod structure made up of cylinders. The component cylinder is the rod-like micelle in the cubic amphiphilic phase or the rod-like column in the smectic D phase. The latter is formed by the stacking of discs composed of aggregations of molecules.[23] Both phases exhibit striking resemblance in the X-ray patterns which contain several reflection spots in a small angle region and have been interpreted by the cubic lattice. In the present materials, the component molecule has a cylindrical shape and so the similar cubic structure can be proposed. However, the observed X-ray pattern does not offer evidence for this, showing only a diffuse reflection with a spacing of 25 Å in a small angle region. On the other hand, it is very hard to assign this isotropic phase to the isotropic liquid since it is a very exceptional case that the isotropic liquid is placed in a lower temperature region than the anisotropic mesophases. Thus, we have no decisive evidence as a basis for selecting one of the two possible phases.

We have also no direct means to identify the second anisotropic mesophase. From observations of focal conic texture, uniaxial orientation property and no optical rotation, only a suggestion is given that the mesophase may be smectic A in which a smetic layer is constructed by a parallel side-by-side packing of α-helical molecules. Such a smectic A structure, in fact, has been theoretically predicted for the hard-rod particles[15,16] and experimentally identified for the lyotropic solution of tobacco mosaic virus through the direct observation of layer structure due to the light scattering method.[10] It has also been reported that the smectic A phase may be formed in the lyotropic solution of DNA from the electric microscopic observations.[8]

Although this study, indicates the existence of three kinds of thermotropic phases, identification of mesophases except for the cholesteric phase was not possible. As one reason for this, it can be cited that mesophase transitions take place being accompanied by the wide coexistence of two phases and so each phase except for the cholesteric phase cannot be fully isolated. This may be caused by a wide distribution of molecular weight ($M_w/M_n = 1.65$) in the present polymer. For better understanding of the phase structure and phase transition behavior, it is necessary to prepare a polymer with a narrow distribution of molecular weight. This is now being done.

REFERENCES AND NOTES

1. L. Onsager, *Ann. M.Y. Acad. Sci.*, **51**, 627 (1949).
2. P. J. Flory, *Proc. R. Soc. London, Ser. A*, **234**, 60 (1956).
3. A. Y. Grosberg and A. R. Khokhlov, *Adv. Polym. Sci.*, **41**, 53 (1981).
4. T. Odijk, *Macromolecules*, **19**, 2313 (1986).
5. F. Livolant and Y. Bouligand, *J. Phys.*, **47**, 1813 (1986).
6. S. Lee and R. B. Meyer, *Liq. Cryst.*, **7**, 451 (1990).
7. J. Watanabe and Y. Takashina, *Macromolecules*, **24**, 3423 (1991).
8. T. E. Strzelecka, M. W. Davidson, and R. L. Rill, *Nature*, **331**, 457 (1988).
9. F. Livolant, A. M. Levelut, J. Doucet, and J. P. Benoit, *Nature*, **339**, 724 (1989).
10. X. Wen, R. B. Meyer, and D. L. Casper, *D. Phys. Rev. Lett.*, **63**, 2760 (1989).
11. M. Ballaff and G. F. Schimidt, *Mol. Cryst. Liq. Cryst.*, **147**, 163 (1987).
12. J. M. Rodriguez-Parada, R. Duran, and G. Wegner, *Macromolecules*, **22**, 2507 (1989).
13. M. Ebert, O. Herrmann-Schonherr, J. H. Wendorf, H. Ringsdorf, and O. Tschiner, *Liq. Cryst.*, **7**, 63 (1990).
14. B. R. Harkness and J. Watanabe, *Macromolecules*, **24**, 6759 (1991).
15. H. Kimura, Private communication; H. Kimura and M. J. Tsuchiya, *Phys. Soc. Jpn.*, **59**, 3563 (1990).
16. D. Frenkel, *Liq. Cryst.*, **5**, 929 (1989),
17. J. Watanabe, H. Ono, A. Abe, and I. Uematsu, *Macromolecules*, **18**, 2141 (1985).
18. D. Demus and L. Ritchiter, in "Textures of Liquid Crystals," Verlag Chemie, New York, N.Y., 1978.
19. It should be noted that in the higher molecular-weight PG-18 specimens we have observed the columnar hexagonal phase in the higher temperature region than the cholesteric phase (refer to ref 7).
20. V. Luzzati, A. Tardieu, T. Gurik-Krzywicki, E. Rivas, and F. Reiss-Husson, *Nature*, **220**, 485 (1968).
21. A. Tardieu and V. Luzzati, *Biochim. Biophys. Acta*, **219**, 11 (1970).
22. G. W. Gray and J. W. F. Goodby, in "Smectic Liquid Crystals," Leonard Hill, Glasgow and London, 1984, p 68.
23. A. Tardieu and J. Billard, *J. Phys. (Paris)*, **37**, C3-79 (1976).