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Apparatus for Thermal Analytic Microscopy and Its Application to Cholesteryl Nonanoate*

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The phase transitions of cholesteryl nonanoate were studied by means of a newly constructed apparatus for thermal analytic microscopy incorporating a thermoelement. It was possible to determine the heat of mesophase transitions more accurately. The transition values of cholesteryl nonanoate on cooling were obtained with the same accuracy as those on heating. The heat of the smectic-solid transition was obtained by slow cooling, which suppressed the supercooling. The vortices caused by cholesteric spherulites, which are similar to those of cholesteryl octanoate, change to circular arrangements of batonnets in the smectic phase. Color changes were observed at the cholesteric-smectic transition in extremely thick specimens. The relations between texture, color, and heat at the cholesteric-smectic transition of cholesteryl nonanoate are discussed.

On the basis of the results of previous works,1,2) a new apparatus was constructed for thermal analytic microscopy and applied to a study of the phase transitions of cholesteryl nonanoate.

Apparatus

The apparatus is shown in Fig. 1.

Sample Cells. The sample cells are similar to those used previously (Fig. 2)2) the microscopic cell being equivalent to the thermal ones. The body is made of stainless steel (0.5 mm thick) except for the center part. The cover and the center of the body are made of Pyrex glass. Two kinds of covers have been prepared. One is slightly smaller than the inner diameter of the body easily drops onto the center glass part. It is employed for thin layer (10—20 μ) samples. The other exactly fits the edge inside the top of the body (Fig. 2). It produces an extremely thick layer (300—350 μ) for the study of the characteristic colors at transformation, independent of the retardation color caused by the thin anisotropic layer.

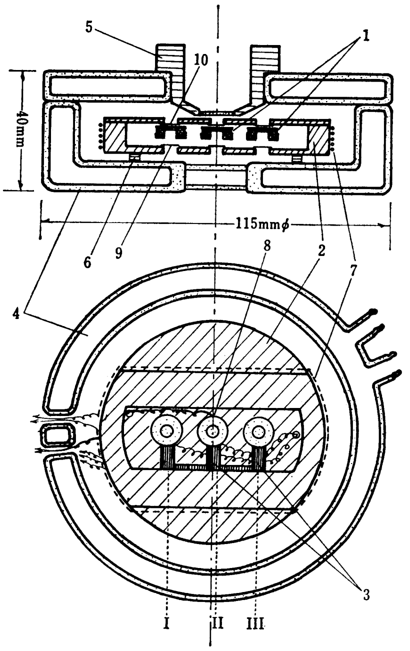

Fig. 1. Apparatus. 1: Sample cell; 2: copper block; 3: thermoelement; 4: jacket; 5: Teflon adapter for microscope; 6: Teflon support; 7: heater; 8: thermocouple for temperature measurement; 9 and 10: glass plates.

Copper Block. A disk-like copper block with a high heat capacity, part of which had been cut off, is used. It has a cover (1.5 mm thick) for maintaining a homogeneous temperature distribution in the interior of the block. A part of the block is cut off to make the thermal conditions as close as possible to those of the three cells.

Connection between Thermoelements. Since a thermoelement is a synthetic material made of several kinds of semiconductor compounds, the thermoelectric power is to some extent dependent on the properties of each thermoelement, and the contact potential differences arisen from the junctions between thermoelements and the block are not identical. Therefore, when the copper block has an inhomogeneous temperature distribution, the differences between each contact potential difference increase. In order to eliminate the potential disturbances, all the thermoelements are connected with the same thermoelement material (Fig. 1).

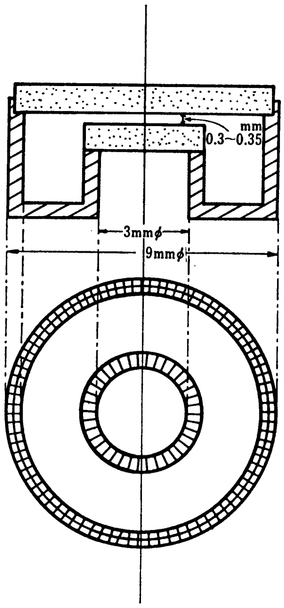

Fig. 2. Sample cells made of glass and stainless steel.

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Table 1. Transition values for cholesteryl myristate

		Cryst.-Sm.		Sm.-Ch.		Ch.-Iso.	
		$\frac{T}{^\circ\mathrm{C}}$	$\frac{Q}{\mathrm{cal/g}}$	$\frac{T}{^\circ\mathrm{C}}$	$\frac{Q}{\mathrm{cal/g}}$	$\frac{T}{^\circ\mathrm{C}}$	$\frac{Q}{\mathrm{cal/g}}$
Heating	this work	70.6	18.9	78.4	0.55	83.5	0.37
	previous work[1)]	70.6	18.2	78.4	0.5	83.5	0.3
Cooling	this work	50—67	17.4	78.4	0.64	83.6	0.52
	previous work[1)]	52—67	16.3	78.5	0.6	83.5	0.5

Jacket. Water is circulated in the glass jacket. Teflon is utilized partly to buffer the heat effect caused by the aproach of the microscope. Three small Teflon pieces are used to place the copper block in the interior of the jacket.

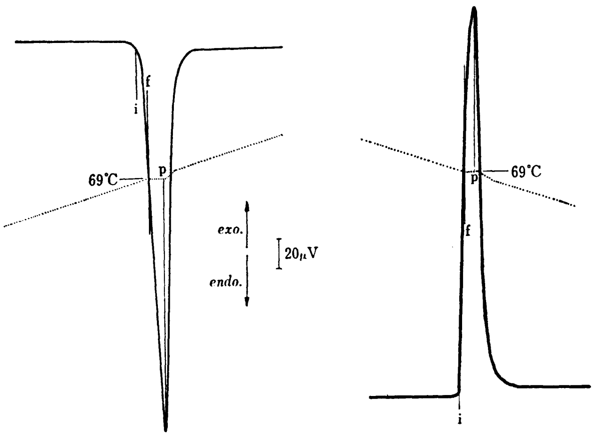
The block heater, the thermocouples for temperature measurement and monitoring, and the sample cell holders are of the same materials as in the previous apparatus.[1)]

Results

Calibration by Stearic Acid. By using the heat of the fusion of stearic acid[3)] the performance of the apparatus was examined. At the heating or cooling rate of 0.63 °C/min the following items were checked: (1) the sensitivities of the thermoelements, I, II, and III (Fig. 1); (2) the dependence of the heat effect on the sensitivity range of an amplifier, and (3) the dependence of the heat effect on the sample weight.

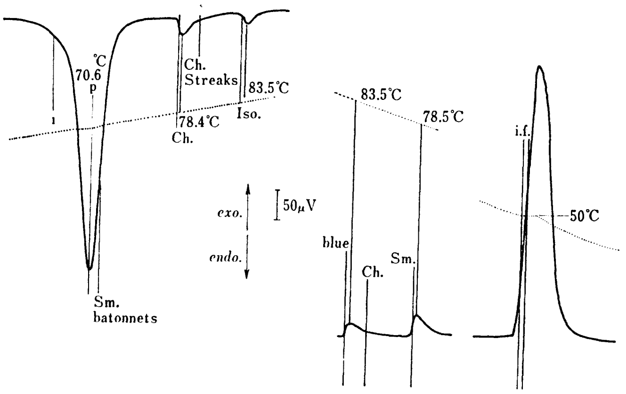
Comparison of the Apparatus with the Previous One. The peak area for the heat of fusion of stearic acid was found to be 40% greater than that of the previous work.[1)] This indicates that the connection of thermoelements is effective in improving the sensitivity of the apparatus. In addition, the base lines of the thermograms were greatly stabilized. The apparatus in the preceding work showed good base lines only with slow heating or cooling.

Calibration by Cholesteryl Myristate. The transition values of cholesteryl myristate were estimated by a comparison with those of the previous work [1)] The results on heating or cooling at the rate of 0.63 °C/min are given in Table 1. The apparatus can determine the heat of mesophase transition to one more significant figure than that of the previous results.[1)] The heats of two mesophase transitions on cooling were found to be considerably larger than those on heating. The heat of the crystallization of stearic acid is 2% larger than that of fusion. Thus, the thermoelectric effect of the thermoelements shows a hysteresis. A small peak area, such as the mesophase-mesophase transition, depends greatly on the way of drawing a base line under the peak. The difference between the values on cooling and heating, however, amounts to more than 20% of the heat of the mesophase transition. It is much larger than 2% in the case of stearic acid.



(a) (b)

Fig. 3. DTA and temperature curves of stearic acid on heating(a) and cooling(b), with notations denoting microscopic change. Sample weight, 11.91 mg; scanning rate, 0.63 °C/min; amp. range, ±250 μV; chart speed, 240 mm/h. Full line: DTA; dotted line: temperature of sample. i: Initiation of melting or crystallization in the microscopic view. f: Finish of the change in the same view as 'i.' p: Peak point of the DTA curve.



(a) (b)

Fig. 4. DTA and temperature curves of cholesteryl myristate with notations denoting the microscopic change on heating(a) and cooling(b). Sample weight, 66.22 mg; rate, 0.63 °C/min; amp. range, ±500 μV; chart speed, 240 mm/h.

Simultaneous Observation in Microscopy and DTA. Figure 3 shows the thermograms and the temperature curves*** of stearic acid with notations of the microscopic changes. Point 'i' indicates the initiation of melting or crystallization, and 'f', the finish of the microscopic change. The temperature 'f' agrees with the starting point of the plateau (69 °C) on the temperature curve. The peak of fusion, 'p', agrees with the end point of the plateau. The temperature for both the complete view change in the microscopy and

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*** The temperature was measured by a thermocouple soldered to the copper cell holder of the center cell.

Table 2. Transition values for cholesteryl nonanoate.

Scanning rate and reference	Heating				Cooling					
	Solid-Ch.		Ch.-Iso.		Iso.-Ch.		Ch.-Sm.		Sm.-Solid	
°C/min	T °C	Q cal/g	T °C	Q cal/g	T °C	Q cal/g	T °C	Q cal/g	T °C	Q cal/g
0.6—2.5	77.8	10.6	92.0	0.24	92.0	0.27	72.5	0.19	43—30	10.3
Ref. 5	80.8	10.2	93.0	0.22	86	0.23	66	0.11	super-cooling	
Ref. 6	77.8	11.4	91.7	0.25	—	—	74.6	0.14	—	—

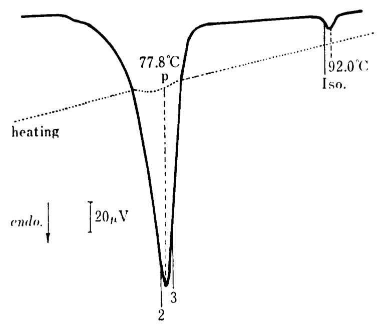


Fig. 5. DTA and temperature curves of cholesteryl nonanoate on heating, with the number of Photos. 2 and 3. Sample weight, 88.53 mg; rate, 0.63 °C/min; samp. range, ±250 μV; chart speed, 240 mm/h.

the peak on the thermogram is equal to 69 °C, the temperature of the fusion of stearic acid.

Figure 4 shows the curves of cholesteryl myristate, with notations denoting the appearance of typical textures. The isotropic-cholesteric transition peak on cooling agrees with the appearance of the homeotropic blue color, but not with that of the cholesteric focal conic texture. According to Gray, both the homeotropic and the focal conic textures belong to the same phase.[4]

The agreement between microscopic and DTA observations has thus been established.

Phase Transitions of Cholesteryl Nonanoate. *Thermal Data of Cholesteryl Nonanoate*: The sample was purified as in the previous work.[1] Table 2 gives the transition values of cholesteryl nonanoate. The heat of crystallization was measured by slow cooling, which helped to suppress the supercooling.[5] The heat of the isotropic-cholesteric transition on cooling is somewhat larger than that on heating, as was found for cholesteryl myristate.

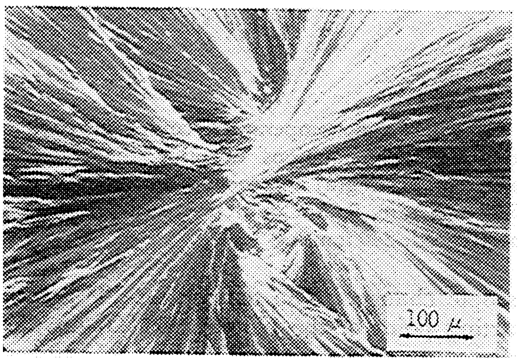


Photo. 1. Spherulitic center of a solid of cholesteryl nonanoate crystallized from the melt.

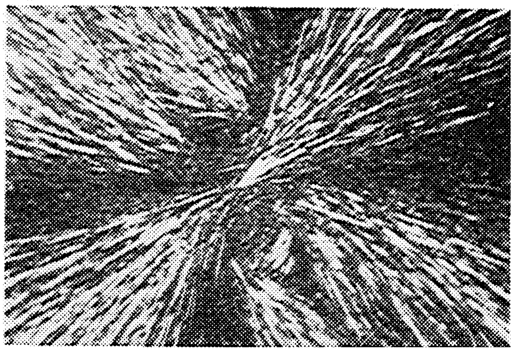


Photo. 2. Melting state around the peak point of the transition to the cholesteric phase.

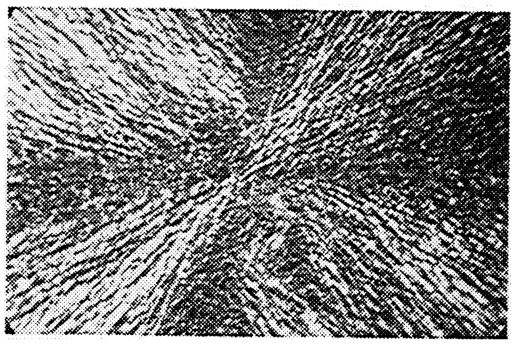


Photo. 3. Cholesteric texture just after the peak.

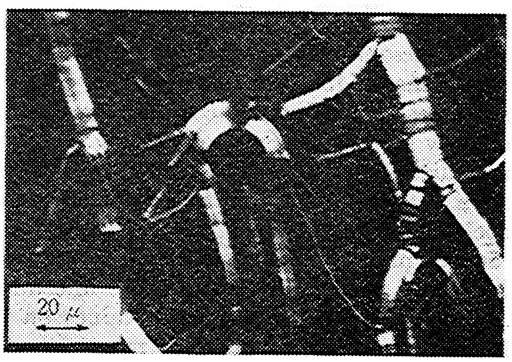
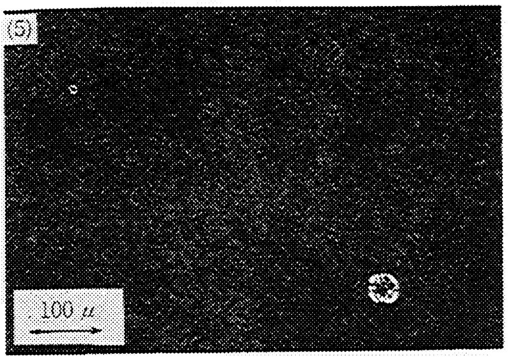
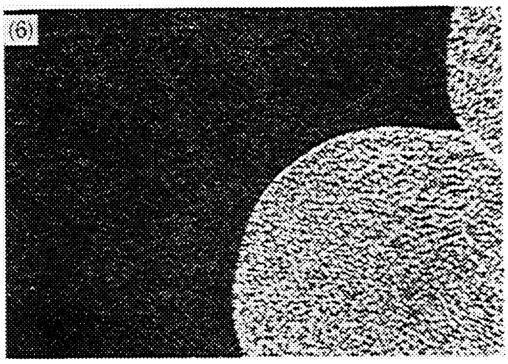
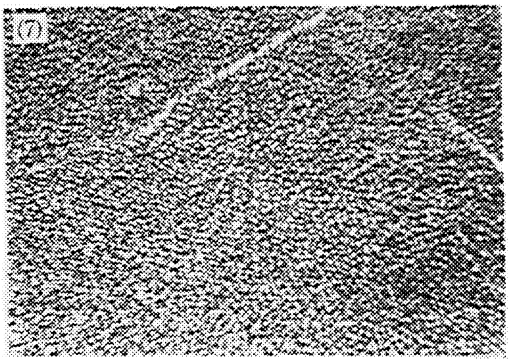


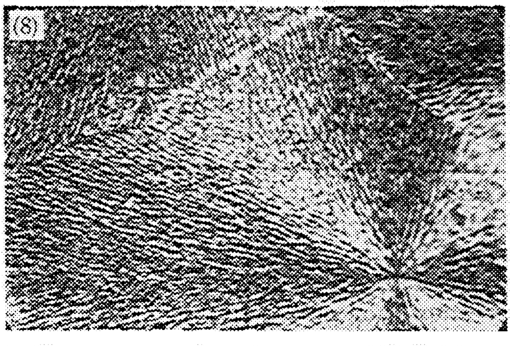
Photo. 4. Oily streaks of the cholesteric phase of cholesteryl nonanoate.

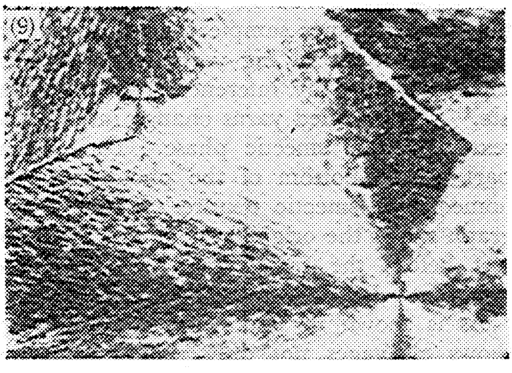
Microscopic observation and DTA: Figure 5 shows the curves on heating (0.63 °C/min), along with the numbers of the photographs. Photograph 1 shows the center of a solid phase radially grown from the smectic phase, taken at room temperature. Photo. 2 shows a transient

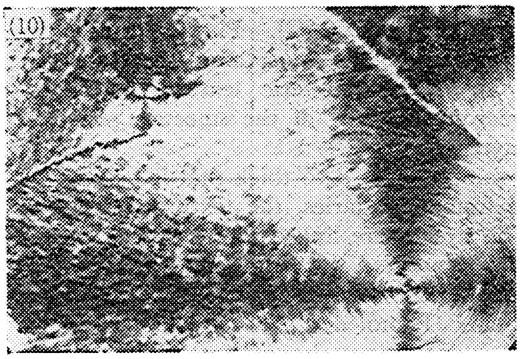


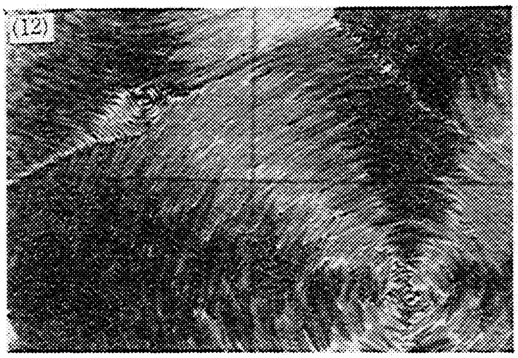


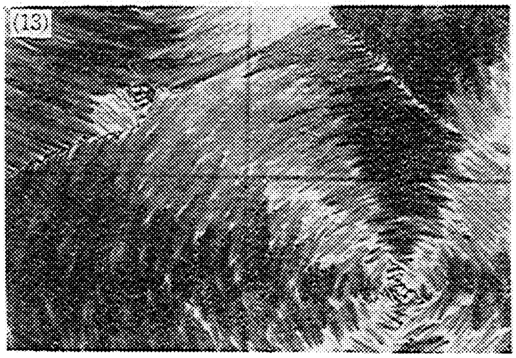


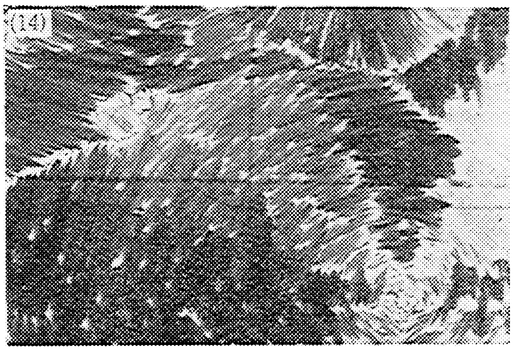












Photos. 5—14. The microscopic views from the occurrence of the spherulite of cholesteric texture to the appearance of the solid phase in cholesteryl nonanoate on cooling. 5: 84.3 °C; 6: 82.3 °C; 7: 79.5 °C; 8: 73.8 °C; 9: 73.5 °C, 10: 73.2 °C; 11: 72.7 °C; 12: 72.6 °C; 13: 72.2 °C; 14: 43 °C.

state to the mesophase (77.6 °C). Photo. 3 shows the cholesteric phase at 78.3 °C, just after the transition peak (77.8 °C). Th cholesteric texture closely resembles a microscopic view of the smectric phase in cholesteryl myristate.

When the sample layer is so thick that it behaves as a liquid free from boundaries, the texture shows oily streaks.[7] The streaks are linked to each other or are branched and separated by isotropic parts, which become clear as the cholesteric-isotropic transition point approaches (Photo. 4). The streak marked by the light and dark portions changes into small spherulites arranged linearly near the transition point. The spherulite is optically negative.

Cholesteryl nonanoate also forms large spherulites like spherical assemblies of the focal conic spherules in the cholesteric phase, much like cholesteryl octanoate. The difference from octanoate is in the formation of the basket-like texture, consisting of smectic batonnets, after the vortex formation. The smectic texture is extremely stable until the solid phase appears. Figure 6 shows the thermogram on cooling (0.63 °C/min), along with the numbers of Photos. 5—14. These photographs show transient states from the spherulitic growth of the cholesteric phase to the appearance of the solid phase, passing through the basket-like texture of the smectic phase. Photos. 5—7 show the spherulitic growth of the cholesteric focal conic texture, much as in the case of octanoate. No heat evolution accompanies the growth, much as in the case of the cholesteric phase of myristate. Photo. 8 shows the occurrance of a radial ordering at 73.8 °C, which corresponds to the starting point of the peak of the cholesteric-smectic transition. Photo. 9 indicates an ordering in the direction vertical to the radial one. The spherulitic center at the right bottom in the photograph looks like a fylfot or an Airy's spiral. Photo. 10 shows a very speedy vortex around the spherulitic center, which clearly indicates the occurrence of some kind of association on spherical surfaces from the center to the boundary. Photos. 11—13 show the growth of the basket-like texture of the smectic batonnets. The peak of the cholesteric-smectic transition is 72.5 °C ('p' in Fig. 6). The microscopic phenomena are almost finished by 'p'. Photo. 14 shows a fan-like solid phase, which appears in the right top portion in the photograph. The 'f' in Fig. 6 corresponds to the point where the solid covers the smectic texture.

In transmitted light, the visible light intensities increase with the thickness of the sample layer. A thick sample shows, at first, a light blue color with the peak of isotropic-cholesteric transition before the generation of the cloudy cholesteric phase. The color variation during the cholesteric-smectic transition is: first light yellow, then strong yellow, orange, red and finally deep blue. The colors become mixed with the variation Red and other colors remain in the microscopic view when the final blue appears. The arrows in Fig. 6 show the ranges of color appearance. The 'n' in the figure means "no color." The view at 'n' is only a dim light as the result of mixing all the colors. The color variation is similar to that in the case of taking an analyser off. The vortex in Photo. 10 is accompanied by so speedy a color variation that observation cannot follow it.

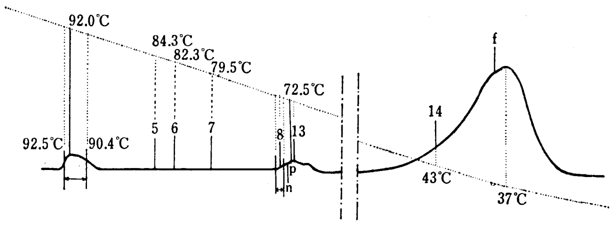


Fig. 6. DTA and temperature curves of cholesteryl nonanoate on cooling, with the number of Photos. 5—14. The experimental conditions are the same as in Fig. 5. The arrows show the ranges of color appearance.

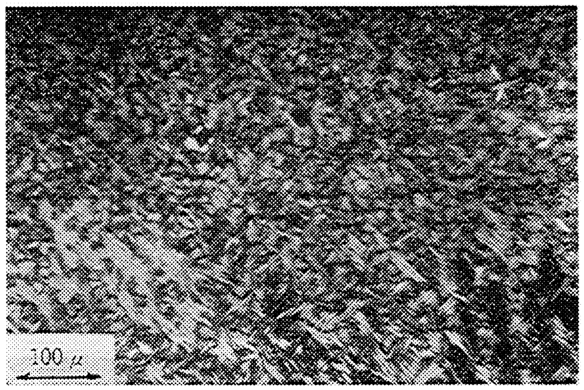


Photo. 15. Cholesteric-smectic transition state of cholesteryl nonanoate in a thin specimen.

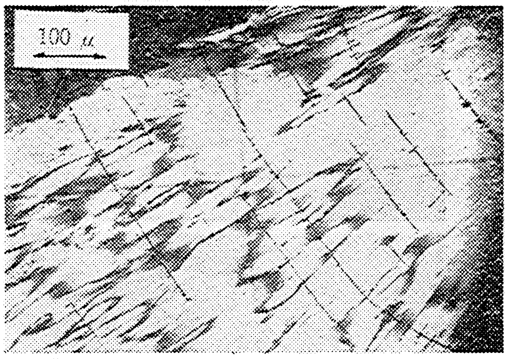


Photo. 16. Cleavages of thin solid film of cholesteryl nonanoate.

Photograph 15 shows the cholesteric-smectic transition for a thinner sample than that of Photo. 5—14. The coils made of big yarns in Photo. 15 suggest a smectic texture by inference from cholesteryl octanoate.[1] The coiled yarns change into the homeotropic texture of the smectic phase. The optically extinct part at the bottom of the photograph indicates the homeotropic smectic texture. The top of the photograph shows the original cholesteric focal conic texture just before the transformation. The selective color light scattering does not appear in this case.

As is shown in Photo. 16, a radially grown solid shows cleavages not only radially but also circumferencially. Solidification on a free surface gives dendritic crystals similar to cholesteryl octanoate.

Discussion

Relation between the Light Scattering in Transmission and in Reflection. A thin, anisotropic specimen generally looks colored under crossed polars because of its own retardation. The thickness most sensitive to retardation is in the range of less than 100 μ. In this work, color observation was carried out for extremely thick samples of more than 300 μ. Therefore, the color observed at the transformation is independent of the thickness. In transmitted light, the visible light intensity increases much more for a thick specimen than for a thin one, making a good contrast to the case of reflected light.[8] In a specimen thicker than 300 μ the molecules in a mesophase may be oriented randomly as a whole, since numbers of molecules restricted by boundary are negligible. Therefore, a theory based on the homeotropical model seems not to be applicable. If the mesophase, however, consists of a crystal structure with a higher order, such as the spherical symmetry of spherulite, the optical axis may naturally be normal to the glass boundary.

The color variation number reflected light was observed using a Mettler fusion-point apparatus. The colors from blue to red were observed in the range of 74—72 °C. Except for the red color, the selective reflection color and the color obtained by our apparatus are complementary. Ennulat[8] studied the selective light reflection of several cholesteryl esters with a monochromator; he found that the wavelengths from 470 nm to 590 nm showed, in the temperature range of 76.78—76.12 °C, the respective intensity maxima with regard to nonanoate. The characteristic colors of this work correspond to the complementary colors of those observed at Ennulat, but the temperature does not agree with his result, although the temperature range 0.7 °C found by us is almost the same as his result (0.66 °C).

Mechanism of the Occurrence of the Vortex. A large spherulite in the cholesteric phase consists of small spherulites with a focal conic texture. In other words, the spherulite has a double structure. An ordinary cholesteric focal conic texture changes into the randomly coiled yarns of the smectic phase, but not into the vortex (Photo. 15). Only an ordered structure of spherical symmetry makes a vortex at the cholesteric-smectic transition. Before being transformed into a vortex, a spherulite seems to be almost solidified by the development of the radial order. In Photo. 9 the isotropic part disappears except for the isogyres of the spherulites. If the same order develops further, the stress force may be accumulated in the system, which will thus be destroyed, and the substance will start to flow as a fluid. In the field of spherical symmetry, the center may be regarded as a singular point similar to a source in hydrodynamics. Consequently, a vortex will come from every spherulite. This inference is just one of many possibilities. A more essential mechanism of vortex formation may be related to the transformation from an optically negative phase to an optically positive phase. A color-light scattering accompanied by a vortex may also be related to some antisymmetric force, such as a twist force caused by the transformation.

Relation between Optical and Thermal Phenomena. It was observed that the microscopic changes in both the fusion and the crystallization of stearic acid agreed with the starting points of the peaks (Fig. 3), suggesting that an optical change precedes the corresponding thermal change. Similar relations were also observed for all the transitions of cholesteryl myristate and nonanoate except for the fusion. The peaks of their fusion show loose curves because of a premelting effect, which makes the starting points of the peaks less clear.

We wish to express our thanks to Dr. Yorihiro Satomi of National Chemical Laboratory for Industry for his guidance regarding the employment of the Mettler fusion-point apparatus.

References

1) K. S. Kunihisa and S. Hagiwara, *Bull. Chem. Soc. Jpn.*, **49**, 1204 (1976).

2) K. S. Kunihisa and T. Shinoda, *Bull. Chem. Soc. Jpn.*, **48**, 3506 (1975).

3) P. E. Slade, Jr. and L. T. Jenkins, "Thermal Characterization Techniques," Marcel Dekker, New York (1970); K. S. Markley, "Fats and Oils," 2nd ed. Part 4, Fatty Acids, Interscience Publishers, (1967).

4) G. W. Gray, "Molecular Structure and the Properties of Liquid Crystals," Academic Press, London and New York (1962).

5) E. M. Barrall II, R. S. Porter, and J. F. Johnson, *J. Phys. Chem.*, **71**, 1224 (1967).

6) G. J. Davis, R. S. Porter, and E. M. Barrall II, *Mol. Cryst. Liq. Cryst.*, **10**, 1 (1970); *ibid.*, **11**, 319 (1970).

7) G Friedel, *Ann. Phys., Paris*, (9) **18**, 273 (1922).

8) R. D. Ennulat, *Mol. Cryst. Liq. Cryst.*, **13**, 337 (1971).