Chiral nematic suspensions of cellulose crystallites; phase separation and magnetic field orientation

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Suspensions of rod-like cellulose crystallites of axial ratio ≈ 20–40, prepared by acid hydrolysis of natural cellulose fibres with sulphuric acid, give stable ordered fluids that display well-formed textures and disclinations characteristic of chiral nematic liquid crystalline phases. The critical volume fraction for phase separation of salt-free suspensions is typically 0·03, with a relatively narrow biphasic region. Because of the negative diamagnetic susceptibility of cellulose, the ordered phase becomes oriented in a magnetic field with its chiral nematic axis parallel to the applied field.

1. Introduction

The phase separation of suspensions of anisotropic rod-like species such as tobacco mosaic virus [1], DNA fragments [2], and collagen [3] into fluids displaying nematic or chiral nematic order is a widely observed phenomenon. Recently, it has been demonstrated that suspensions of rod-like cellulose crystallites form a chiral nematic phase [4]. The rodlets were prepared by acid hydrolysis of natural cellulose fibres from several sources. It is surprising that well-formed textures and disclinations characteristic of chiral nematic liquid crystals are readily formed by dilute suspensions of particles with relatively simple chemistry and geometry. This spontaneous ordering may be related [4–7] to the helicoidal structures often observed in naturally occurring composites containing cellulose, chitin and other chiral components.

In this preliminary report, we describe qualitatively the concentration-driven phase separation, and some of the changes in liquid crystalline textures that result from the application of a magnetic field.

2. Experimental

The samples were prepared by sulphuric acid hydrolysis of cellulose fibres [4]. Typically, samples of filter paper (Whatman Grade 1) or of a bleached softwood kraft

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pulp were disintegrated in a Wiley mill to pass a 20 mesh screen, and then treated with 64 wt% sulphuric acid at times ranging from 10 min at 70°C to 3 h at 45°C. The suspension was diluted to 2 l with distilled water, concentrated by centrifugation and decantation, and then dialysed until the water outside the dialysis membrane was neutral. The suspension was treated with a mixed-bed ion exchange resin and dispersed by an ultrasonic treatment (Branson Sonifier Model 350) for 1 min. The preparation and properties of several suspensions are summarized in the table.

On standing, the suspensions separated into an upper isotropic and a lower birefringent phase. The concentrations were measured gravimetrically before and after evaporation of the water. The sulphur content of the rodlets, measured by elemental analysis, was in the range 0·5–1·0 per cent sulphur by dry weight, depending on the hydrolysis conditions. Samples of these suspensions for optical microscopy were sealed in flat-sided glass capillaries, 4 mm wide with optical path length 0·4 mm (Microslides, Vitrodynamics Inc., Rockaway, New Jersey 07866, U.S.A.). One capillary was filled with the anisotropic phase of sample C. Another, prepared from the isotropic layer of suspension B was partially sealed with Parafilm©, to allow the water to evaporate very slowly (over weeks), in order to follow the formation of the anisotropic phase. Photomicrographs were taken between crossed polars with a Zeiss Universal polarizing microscope. Unless otherwise indicated, the sample tubes were oriented such that their long axis runs from top to bottom of the photomicrographs and the crossed polars are parallel to the edges of the photomicrographs. A homogeneous magnetic field of 7 T was applied parallel to the long axis of the sample tubes with a solenoid (Thor Cryogenics) having a horizontal 50 mm diameter room temperature bore. The sample tubes were removed from the still-energized magnet for observation and photomicrography; their relaxation time was slow enough to permit leisurely observation.

3. Results and discussion

As reported previously, the cellulose rodlets making up the suspension are of the order of 200 nm long by 5 nm wide [4]. The precise dimensions of the rodlets and the critical concentration necessary to form an ordered suspension are sensitive to the source of the cellulose, the hydrolysis conditions and the ionic strength. When a suspension at a concentration just below the critical concentration is allowed to evaporate, small ordered regions form, develop and coalesce to give a lower ordered region and an upper isotropic suspension.

(i) Initially, small, uniformly birefringent objects can be detected by optical microscopy with crossed polars [4]. They appear as spherical or oval droplets against the dark isotropic background; observation with a first order red plate and crossed polars shows that at this stage the cellulose rodlets are oriented

Properties of cellulose suspensions.

Suspension	A	B	C
Cellulose source	Bleached kraft	Filter paper	Bleached kraft
Hydrolysis time	10 min	2 h	50 min
Hydrolysis temperature/°C	70	45	<70†
Suspension pH	2·8	2·2	2·3
Pitch of anisotropic phase/μm	23	60	12
Total concentration/wt%	5·5	5·7	4·0

† Hydrolysis with 60 wt% sulphuric acid.

parallel to the long axis of the oval droplets. These initial ordered domains are similar to the 'tactoids' observed by Bernal and Fankuchen in their pioneering study of suspensions of tobacco mosaic virus [8].

(ii) As the size of these ordered regions grows, their birefringence becomes less uniform and dark patterns form, often becoming resolved into lines running parallel to a diameter [4]. Further domain growth occurs normal to the lines, resulting in the rather irregularly shaped banded regions shown in figure 1.

(iii) As the droplets sediment, hydrodynamic forces sometimes generate thin birefringent tails that trail vertically. The sedimenting 'tactoids' coalesce on reaching the bottom of the sample tube, to give the bulk anisotropic phase.

(iv) The anisotropic phase initially shows a texture that reflects the dimensions and orientation of the droplets from which it formed. In this polydomain texture, the parallel fingerprint patterns characteristic of long-pitch chiral nematic liquid crystalline materials are visible in some regions.

(v) For sealed samples, prepared at concentrations where both ordered and isotropic phases are in equilibrium, the interface between the phases is sharp, and the polydomain texture of the ordered phase rapidly coalesces and reorients into a planar texture, with the chiral nematic axis normal to the walls of the sample tube. An example is shown in figure 2.

Cholesteric droplets: the 'tactoids' observed on phase separation normally correspond to a class of cholesteric spherulites described by Bouligand and Livolant [9], in which the cholesteric layers show a uniform orientation in the droplet. Systems forming these droplets include concentrated aqueous solutions of DNA, and MBBA containing a small amount of chiral additive [9], and aqueous dispersions of collagen [10] and chitin [11]. This droplet morphology contrasts with the case where the chiral nematic layers are parallel to the droplet surface that is observed in many other systems, including the synthetic polypeptides first studied by Robinson [12]. In the droplets of cellulose suspensions, the uniform orientation of the layers means that the direction of orientation intersects the isotropic–anisotropic interface at a variable angle, often close to 90°. This suggests that the orientation of the cellulose rodlets at the interface between the ordered and isotropic phases is not a critical factor in determining the stability of the droplet morphology (the rodlets are not constrained to lie completely parallel or normal to the interface), but the tendency to elongate along the
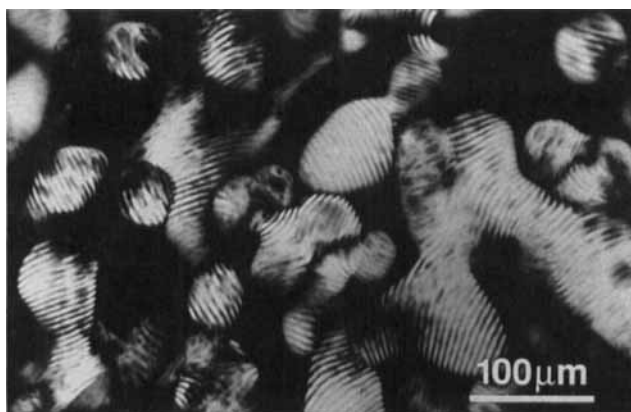

Figure 1.   Growth of anisotropic domains, 'tactoids', in a suspension of cellulose crystallites as water evaporates from isotropic layer of sample A.
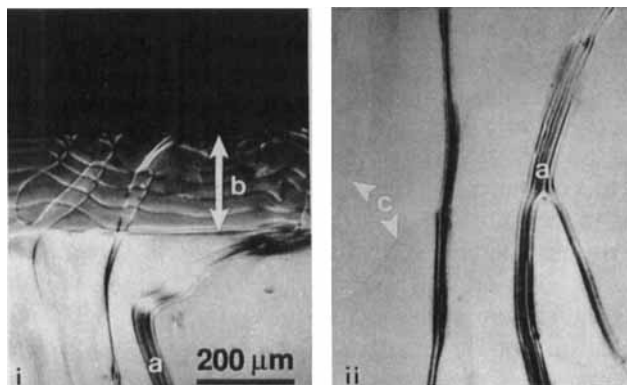
Figure 2.   (i) Isotropic–anisotropic interface of suspension A. The upper dark region is the isotropic phase. The vertical walls separating planar regions (*a*) show a chiral nematic pitch of about 20 mm. The interface (*b*) is distorted by the flow of the anisotropic phase when the sample tube is laid horizontally on the microscope stage. (ii) Anisotropic phase of sample A. The vertical walls separating planar regions (*a*) and Grandjean planes at the surface of the sample tube (*c*) are visible.

chiral nematic axis, shown in figure 1, does indicate an anisotropy of surface energy, with lower values for layers normal to the interface. The process of development observed for these suspensions, from a small quasi-nematic germ with uniform orientation, of dimensions much smaller than the chiral nematic pitch, through progressive twisting of this structure, to the elongated layered droplets, is in accord with the structures described by Bouligand and Livolant for spherulites with a uniform orientation of the cholesteric layers [9].

On standing for several days, the boundaries between the domains in the anisotropic region that reflected the initial orientation of the spherulites disappear, and they reorient to give a planar texture with optical axis normal to the walls of the capillary. Furthermore, the axis also develops normal to the anisotropic–isotropic interface, indicating that the equilibrium orientation of rodlets is parallel to this interface. This indicates that other factors must be operative in the small droplets; possibly the energy penalty for bending along the chiral nematic axis is greater than that for oblique orientation of the rods in the interface.

Suspension stability and phase separation: the suspensions of cellulose, prepared as described, are clear and free from precipitant. Furthermore, biphasic suspensions, sealed in glass, show a constant proportion of isotropic and anisotropic phases after storage for several weeks at room temperature, but some desulphation and loss of stability does occur in time. The addition of NaCl to give a salt concentration of less than 0·1 M causes the immediate separation of a gel, so it is reasonable to infer that the suspensions are stabilized by the negatively charged sulphate groups on the surface of the cellulose rodlets.

Transmission electron microscopy of the cellulose crystallites shows that they are fairly uniform; typically, crystallites made from a bleached kraft wood pulp are straight, 100–200 nm long and about 5 nm wide [4].

Elemental analysis of a suspension made from bleached kraft pulp gave a sulphur content of 0·73 wt%, based on the dry weight of the cellulose in the suspension. This corresponds to a degree of substitution, DS, of 0·038 sulphate groups per anhydroglucose unit, assuming that the total mass of cellulose is available for sulphation. However, heterogeneous esterification of cellulose normally occurs initially at the

surface of crystalline substrates. The cross-section of the cellulose I unit cell, containing two chains, is about 0.6 nm^2. Taking the cross-section of the crystallites as 25 nm^2, then the rodlets contain about 81 chains. If the rodlets have a square cross-section, with 9 chains per side, then about 40 per cent of the cellulose chains are on the surface. Thus, of the order of 10 per cent of the anhydroglucose units on the surface contain sulphate groups. This corresponds to a surface charge density of around 0.2 sulphate groups per nm^2.

In the suspensions considered here, as much as possible of the free acid and salt have been removed, and residual sulphate groups are in the acid form. Typical values for the phase separation, in this case for suspension C, are as follows. Starting with a total concentration of 4.09 wt% of cellulose in water, two phases separated, giving measured concentrations of 3.85 wt% in the upper isotropic layer (which was about 40 per cent of the total volume of this sample) and 4.26 wt% in the lower anisotropic phase. Taking the density of microcrystalline cellulose as 1.52 [13], and that of water as 1, the estimated volume fractions for the isotropic and anisotropic phases, respectively, assuming no change of volume on mixing, are $\phi_i = 0.026$ and $\phi_a = 0.028$.

The phase separation of rod-like species into an ordered fluid phase was first considered by Onsager [14], who predicted that the critical concentrations for ordered phase formation would depend only on the axial ratio L/d of rods of length L and diameter d. For volume fractions of rods $\phi < \phi_i$, the solution is isotropic, for $\phi > \phi_a$ it is anisotropic, and for $\phi_i < \phi < \phi_a$, the solution separates into isotropic and anisotropic phases, where

$$\phi_i = 3.3d/L \quad \text{and} \quad \phi_a = 4.5d/L. \tag{1}$$

The critical concentrations predicted for axial ratios of 20–40 ($\phi_i = 0.17$–0.08) are significantly larger than those observed above for a sample prepared from kraft pulp. The calculated values are also outside the range where the second virial approximation is valid. The theory, based on the second virial approximation, considers only repulsive forces arising from the mutual impenetrability of the rods. Extensions of the Onsager theory have been reviewed by Semenov and Kokhlov [15].

The suspension may be stabilized by the charged sulphate ester groups on the surfaces of the crystallites. If it is assumed that the repulsion can be crudely modelled simply by increasing the effective diameter of the rods by a factor m, thus decreasing the axial ratio by m, but simultaneously increasing the apparent volume fraction by m^2, then

$$d_c = m.d \quad \text{and} \quad \phi_{i,c} = m^2 \phi_i \tag{2}$$

where d_c is the apparent diameter and $\phi_{i,c}$ is the apparent volume fraction of the charged rods. The phase separation of the charged rods is given by

$$\phi_{i,c} = 3.3/(L/d_c). \tag{3}$$

Combining (2) and (3) gives

$$m = 3.3d/(L.\phi_i). \tag{4}$$

For the experimental value for ϕ_i of 0.026, $m = 6.3$ for $L/d = 20$, 3.15 for $L/d = 40$. For rods of 5 nm diameter, this corresponds to an effective repulsive layer thickness of 5–12 nm.

The effect of charge on the ordered phase separation of rods was considered systematically by Stroobants *et al.* [16], who predicted that the effective diameter should increase by an amount approximately proportional to the double layer

thickness. They also predicted that the rods should exert a (nonchiral) twisting force on each other. Semenov and Kokhlov [15, 17] predicted a more complex phase behaviour depending on the relative importance of the electrostatic repulsion, which in turn is sensitive to charge density and electrolyte concentration. Other theories for the phase separation of spherocylinders are due to Lee [18] and Sato and Teramoto [19]. Unfortunately we are unable to test these theories quantitatively at present, because preliminary analyses have shown that a fractionation is occurring between the two phases, with the shorter rodlets preferentially migrating to the isotropic phase.

Effect of magnetic field: recently, Sugiyama et al. [20], showed that dilute suspensions of a highly crystalline tunicate cellulose, when allowed to evaporate in a magnetic field of 7 T, dried down to a film in which the crystallites were oriented with their long axes perpendicular to the field. The negative diamagnetic anisotropy of crystalline cellulose, indicated by this result, has interesting implications for the behaviour of chiral nematic suspensions of cellulose. Rather than untwisting the chiral nematic structure, the field should simply reorientate the cholesteric axis in the direction of the field. This behaviour has also been observed, for example, with liquid crystalline DNA [21]. Although the diamagnetic anisotropy of cellulose is relatively weak per molecular repeat unit, the crystalline rodlets are long and heavy, so that their total diamagnetic anisotropy is rather large, compared to TMV or DNA particles.

A few minutes exposure to a field of 7 T, applied along the axis of the sample tube, readily realigned the samples. For suspension B, initially in a predominantly planar texture, but with some boundaries running more or less parallel to the cell walls (see figure 3 (i)), application of the field for 10 min led to the formation of multiple lines, with
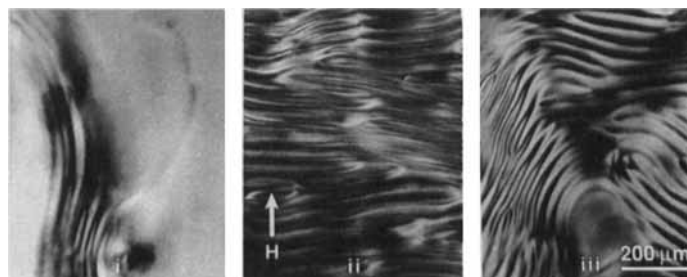
Figure 3.   Effect of magnetic field on texture of suspension B. Sample tube and magnetic field are oriented vertically. (i) Initial texture; (ii) after 10 mins in field; (iii) after 2 h relaxation.
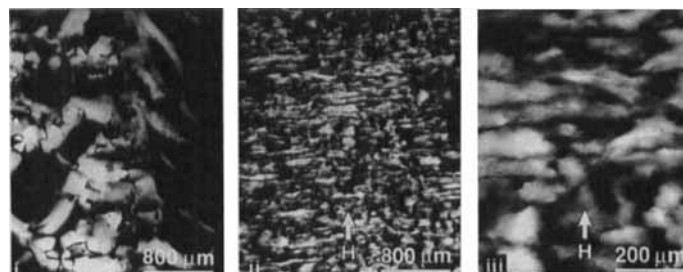

Figure 4.   Effect of magnetic field on texture of suspension C. Sample tube and magnetic field are oriented vertically. (i) Prior to application of field; (ii) after 1 h in field; (iii) higher magnification showing cholesteric layers.
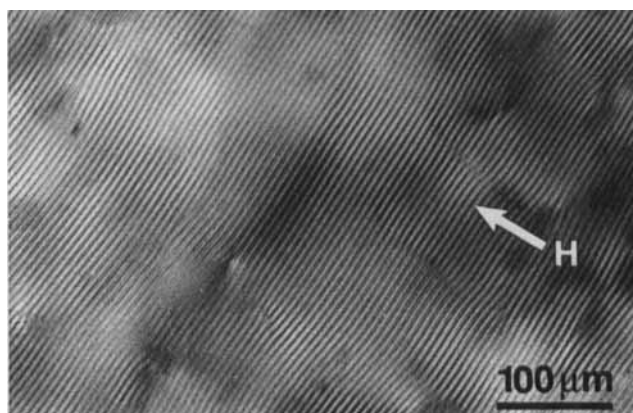
Figure 5. Orientation of cholesteric layers in suspension C after 12 h in a 7 T magnetic field. Sample tube orientation and field direction as indicated, crossed polars vertical and horizontal.
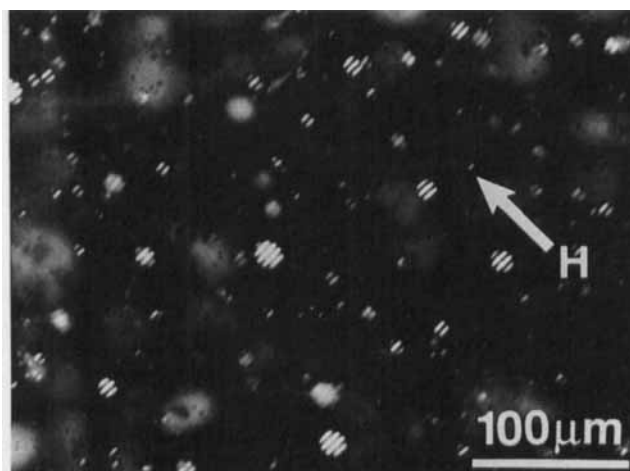

Figure 6. Orientation of tactoids in a magnetic field.

many disclinations, orthogonal to the field direction (see figure 3 (ii)). After a couple of hours relaxation, the texture reorients (see figure 3 (iii)), becoming essentially planar overnight. This sample has a very long 60–70 μm pitch, and the thickness of the sample is about four pitch-lengths, causing some distortions in the figures.

Suspension C is completely anisotropic, with a shorter pitch (12 μm). Prior to placing in the field, the sample showed a very jumbled texture at low magnification (see figure 4 (i)). After 1 h in a 7 T field, the texture changed, with many disclination lines running perpendicular to the field (see figure 4 (ii)). At higher magnification (see figure 4 (iii)), the fingerprint lines are also seen to be tending to line up perpendicular to the field. After about 12 h in the field, the orientation of the cholesteric layers is close to perfect (see figure 5) over the entire sample tube. The orientation was perpendicular to the field, even at the walls of the sample tube and at the surface of air bubbles in the sample, but the relaxation process on removal from the field was fastest at these walls, with the layers taking up their preferred parallel orientation.

The tactoids in the biphasic region are also readily oriented in the magnetic field (see figure 6). The dark areas in this figure are of course the isotropic phase. We are unable to say whether the rodlets in the isotropic phase also orient in the magnetic field; if so, they relax rapidly when removed from the magnet to give the observed isotropic phase.

4. Conclusions

The following observations concerning these readily accessible suspensions of cellulose seem significant.

(1) The dilute suspensions form a chiral nematic phase.

(2) The anisotropic phase separates at relatively low concentrations; typically, for a salt-free suspension containing $4 \, wt\%$ cellulose, the volume fractions of cellulose in the isotropic and anisotropic phases were estimated to be 0·026 and 0·028, respectively.

(3) The anisotropic phase separates as 'tactoids', with a uniform orientation of the cholesteric layers.

(4) The suspensions display a negative diamagnetic susceptibility; the chiral nematic axis aligns parallel to the magnetic field.

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