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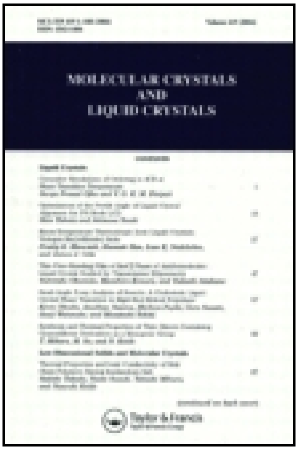

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Liquid Crystallinity in Lipid-Water Systems

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Liquid Crystallinity in Lipid-Water Systems

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Following a short review of the three most common liquid crystalline structures occurring in lipid-water systems, viz. the lamellar and the two hexagonal structures, attention is focused on the occurrence of cubic structures in such systems and on the various proposals for the arrangement on the molecular level. It is especially stressed that the tacitly assumed structure of globular aggregates, either of the 'oil in water" or "water in oil" type, is often difficult to reconcile with the interpretations of the experimentally obtained X-ray diffraction findings. The different structure proposals are discussed, and the connection between cubic phases of model systems and of systems of biological origin proper are stressed. Due to their optical anisotropy the occurrence of cubic phases in lipid-water systems has often been overlooked.

INTRODUCTION

Liquid crystallinity in systems containing amphiphilic lipids and water is a common feature and has been the subject of intensive research for a considerable time. At the Second International Liquid Crystal Conference in 1968, A. S. C. Lawrence[1] gave a lecture with almost the same title as above, and it is up to the present writer to give the topic a slightly different twist.

There is general agreement on the molecular arrangement of the three most common varieties of liquid crystalline structures in lipid-water systems, viz. those of the *lamellar*, the *normal hexagonal* and the *reversed hexagonal* phases, and on the location and sequence of the respective phases in a phase diagram. In addition, several other liquid crystalline phases may be found, which are interspersed between those three phases, or between one of them and the aqueous or oil-continuous solution phases, but the existence of these phases

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† Invited lecture, presented at Eighth International Liquid Crystal Conference, Kyoto (Japan), June 30–July 4, 1980.

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as independent ones and/or the arrangement on the molecular level has been debated.

The nature of amphiphilic liquid crystalline phases can be considered in two different ways. The liquid crystalline structure can be regarded as an ordered array of large micelles. In this sense, the structure would arise from interactions between micelles occurring at high surfactant concentration or between micelles whose capacity to solubilize another lipid and still remain in isotropic solution has been exhausted. There are also amphiphilic lipids whose solubility in water is so low that it is questionable if a micellar solution is formed, but the lipid transforms under the influence of water into a liquid crystalline state. The alternative approach considers the liquid crystalline phases as an intermediate stage between the solid phases and a melt/liquid phase. In this case, the formation of liquid crystalline phases would be due to the residual strong short-range intermolecular forces similar to those in solids. Both approaches have their merits, the first is applicable to aqueous amphiphilic systems, while the second is more applicable to thermotropic phases.

This review will start with a short recapitulation of the occurrence and the structures of the unequivocal liquid crystalline phases, and then proceed to the problems of the more questionable phases. Attention will to a large extent be focused on the structures of the cubic, "viscous isotropic", phases. Lyotropic amphiphilic liquid crystalline phases and their structures are dealt with in a large number of reviews, some of which are listed in references 2–11.

NOMENCLATURE

The nomenclature found in the literature can only be described as a mess. The terminology used for thermotropic liquid crystalline structures is not applicable. The only common ground is the lamellar structure, which is smectic. In lipophilic systems the symbols used to represent different phase structures vary from one research group to another. Attempts to compare the different systems may be found in the reviews of Tiddy[2], Skoulios[6] Ekwall[8] and Winsor.[10,11] The symbols used in this review will mostly be those of Ekwall.[8]

EXPERIMENTAL

"A trivial but nonetheless very important aspect in all work in lipid-water systems is that meaningful investigations presuppose a certain amount of information on the phase diagram."[12] One has to separate the different

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phases and to analyse their composition. In the separation, centrifugation techniques may be used to advantage.[13,14]

The principal experimental methods for studying the different phase structures are polarizing microscopy H, X-ray diffraction and NMR spectroscopy.

Most of the liquid crystalline phases are birefringent and the various phases exhibit different characteristic textures in the microscope, but identification of a particular phase is often something of an art. Hartshorne has given reviews of the microscopic technique applied to liquid crystalline systems.[15,16] Readers interested in comparison with texture observations of thermotropic liquid crystals may consult Demus and Richter.[17] Rosevear has made an attempt to classify the liquid crystalline phases occurring in soap-water systems.[18,19]

Electron microscopy has sometimes been used to reveal the morphology of a particular structure, but the results may be influenced by the technique of sample preparation.[20,21]

The final structure assignment must be confirmed by X-ray diffraction. X-ray work may also provide the dimensions of the aggregate units. The techniques employed and the interpretation go back to the work of the Luzzati-school.[9,22,23] The interpretation is based upon the fact that whereas in the "wide-angle" region a broad diffusion reffection is obtained around the 4.5 Å position indicative of a "liquid" state of the hydrocarbon chains, in the "low-angle" region one obtains a series of sharp reflections corresponding to "crystalline" spacings ranging from ten to several hundred Å and giving structure parameters in one, two or three dimensions. One difficulty with the X-ray diffraction technique is the common paucity of reflections and the fact that their intensity often is low. Neutron diffraction, which has been used rather seldom,[24] has the advantage in addition to the wavelength that by exchanging ordinary water for heavy water one may obtain an impression of the intensity variations of the individual reflections.

Recent developments in the use of several spectroscopic techniques have enriched our insight into aggregate structures on the molecular level.[3,4] Besides IR and Raman techniques, measurements of the lipid and water diffusion coefficients, and quadrupole splittings by NMR are of interest. ESR methods and measurements of polarized absorption and emission have also been used. Especially promising seem to be the recently developed techniques for measurements of time-resolved fluorescence.[25]

THE AMPHIPHILE PHASE DIAGRAM

Lipids which are amphiphilic have a dualistic nature, one part of the molecule being hydrophilic and the other lipophilic. This feature endows the amphiphiles with their capability to form micellar and liquid crystalline structures in combination with water. With respect to the interaction between lipid and water one may divide the amphiphilic lipids into two groups:

i) "insoluble swelling amphiphiles"

which are virtually insoluble but swell under the influence of water to form liquid crystalline phases;
and

ii) "soluble micelle-forming amphiphiles"

which separate in liquid crystalline form when the capacity to be dissolved as clear isotropic (micellar) solution is exhausted.

There is no sharp boundary between these two classes of amphiphiles, a rise in temperature may transform an insoluble swelling amphiphile to a soluble micelle-forming amphiphile. In addition there are other amphiphilic lipids which obtain the capability to form liquid crystalline phases in combination with some other amphiphile. Some amphiphilic lipids may be liquid crystalline at ordinary temperatures in the pure state, others may be liquid, but on the addition of water give liquid crystalline phases. However, most are solids at room temperature.

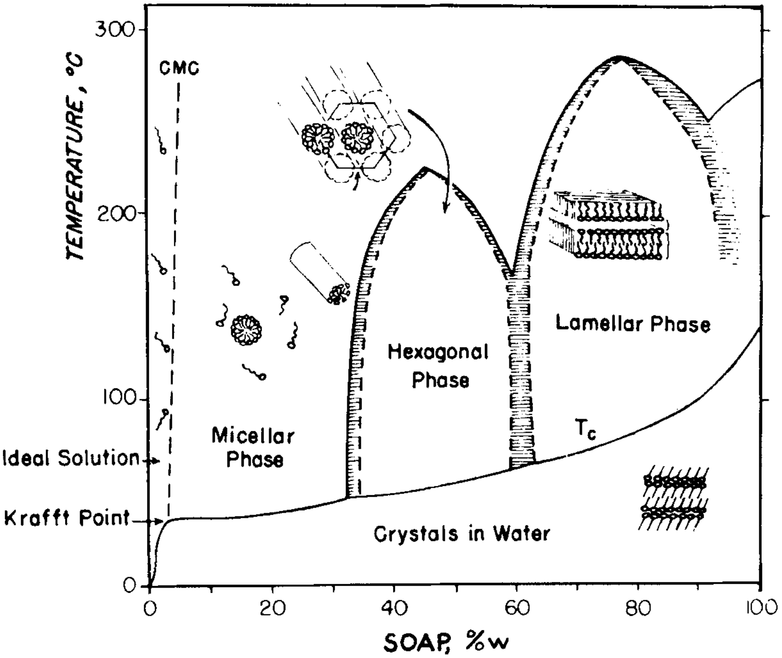

FIGURE 1 Type of phase diagram obtainable for soap-water systems. Modified after Small-Luzzati-McBain.[26]

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The amphiphilic lipids have anionic, cationic or non-ionic character. However, the overall features of their binary phase diagrams with water are remarkably similar, irrespective of character. Figure 1 shows the type of phase diagrams obtained for soap-water systems. Of importance is the course of the T_c-curve and especially the knee termed the Krafft point. Below the T_c-curve there are no liquid crystalline homogeneous phases. The phases sometimes called "gel" or "coagel" are just suspensions of crystals in water, or lamellar crystals that possess the capability to incorporate water in the structure, while the original rigid hydrocarbon arrangement is retained. The Krafft point is often called the critical micelle temperature. It is very sensitive to impurities and additives. A pure specimen has a higher Krafft point than an impure one, and the Krafft point may be elevated by the addition of salt. Together with hysteresis effects, this may be the explanation for the differences in experimental results of different investigators.

Peculiarly enough, very few recent temperature-concentration diagrams of amphiphile-water systems seem to have been published. One rare exception is the recent work by Perron and co-workers.[27, 28] As an example there is no published phase diagram for such a commonly used amphiphile as sodium dodecylsulphate ($C_{12}SO_4Na$). Figure 2 gives a rough phase diagram of its binary system with water. The Krafft point is reported to lie at 9–10°C,[29] but already moderately concentrated micellar solutions require much higher temperatures (at about 24°) in order to stay clear. Goodman and Walker have published a diagram for the C_{10} homologue,[30] but in the opinion of the present writer it is questionable if it is correct in all details. One drawback in the determination of the phase diagrams of alkyl sulphates is their thermal instability.

The phase diagram for the aqueous system of the cationic hexadecyltrimethyl-ammonium bromide (C_{16}TABr) is also not worked out in detail. The phase diagram contains in addition to the hexagonal phase, a cubic and a lamellar phase at elevated temperature (75°C) according to work by the Luzzati-school.[9,22,23] The Krafft point of this soap is usually regarded to be about 24°, but for very pure specimens it lies around 27°.

UNEQUIVOCAL PHASES

As mentioned above several different liquid crystalline phases may be found in one and the same lipid-water system. At constant temperature, as the lipid concentration is increased, one encounters the sequence isotropic micellar solution—hexagonal phase—lamellar phase—reversed hexagonal phase—low-water phases (crystalline solid, isotropic solution or melt) but interspersed between these phases other liquid crystalline phases may occur.

Several factors affect the occurrence of a particular phase. One factor is

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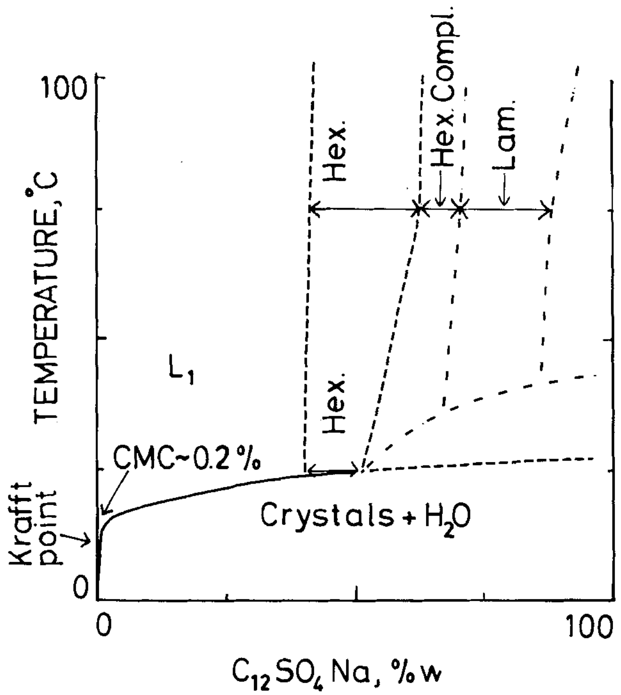

FIGURE 2 Rough phase diagram for the system sodium dodecylsulphate and water. The Krafft point lies at about 9°C[29] and the cmc is about 8 mM.[31] The phase behaviour at 75° is sketched according to Luzzati *et al.*[22,23]

purely geometrical, the proportions between the polar and non-polar parts of the amphiphile molecules. Other conditions being equal, a large polar head group favours the formation of a normal hexagonal phase, while the reversed hexagonal phase is obtained when the non-polar part is bulky. The chemical nature of the polar group also has some influence. The addition of another amphiphilic component modifies the proportions between the polar and non-polar parts of the amphiphilic "molecules", as well as the interactions between the polar groups. These interactions may also be altered by the addition of a common electrolyte like sodium chloride. All this gives possibilities for the occurrence of a large number of different liquid crystalline phases. In his recent review Tiddy gives a condensed summary of current theories for the formation of liquid crystalline structures.[2]

The basic reason for micelle formation in an aqueous solution of an amphiphilic lipid is the tendency of the alkyl-chains to avoid contact with water, in combination with the affinity of the water molecules for the polar head groups. In the absence of any directional forces, the amphiphilic molecules would prefer to pack as globular aggregates in order to minimize

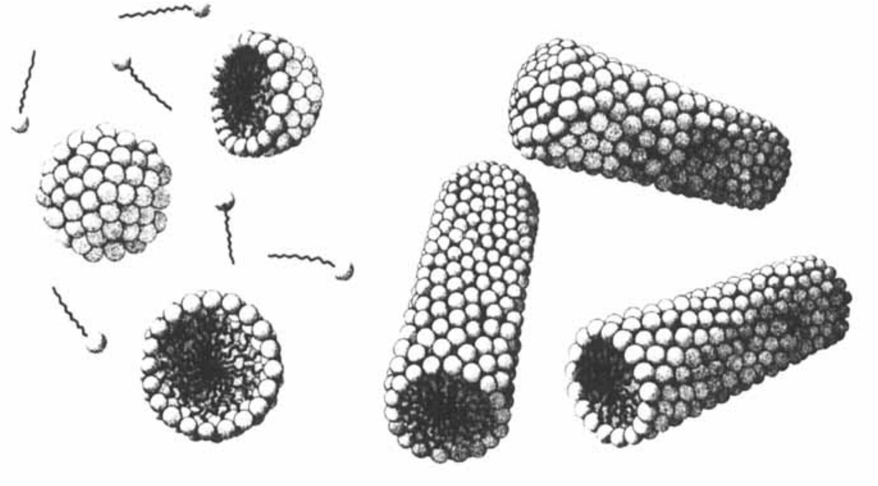

FIGURE 3a A common visualization of the structures of the normal micelles in aqueous amphiphile systems.[19] See text for criticism.

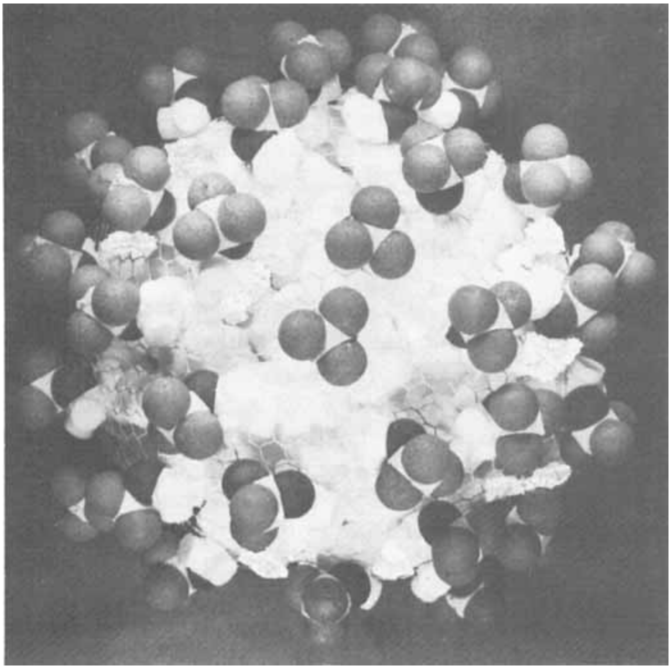

FIGURE 3b A visualization of a globular micelle where account has been taken of the space requirements of the different parts of the dualistic amphiphile molecules. The model is supposed to depict a sodium dodecylsulphate micelle containing 60 entities.[3]

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the repulsion between the polar head groups. When the concentration of a micellar solution is increased, the crowdedness of the system deforms the globular micelles into rod-shaped ones, and when their length reaches a critical value, a hexagonal liquid crystalline phase separates. If a dilute micellar solution becomes unstable due to the addition of a weakly polar amphiphile, a lamellar liquid crystalline phase will separate instead.[32]

In the visualizations of the molecular arrangement of the entities in micellar phases one often sees pictures such as those shown in Figure 3(a). However the raspberry-like appearance is misleading since the polar groups are shown as too tightly packed. The presentation takes no account of the space requirements of the different parts of the dualistic amphiphile molecules. It is hoped that Figure 3(b) gives a more realistic impression.

The hexagonal liquid crystalline phases have a very stiff consistency. For the naked eye they are transparent, but they are birefringent and display characteristic textures in the polarizing microscope.[18,19] The basic information obtained by X-ray diffraction is the distance between the hexagonal axes in a rod structure, and with knowledge of the composition one can estimate the aggregate parameters, such as the diameter of the rod-aggregates and the area per polar head group at the interfaces between the polar and non-polar regions.[9,33]

The structure of the normal hexagonal phase is visualized in Figure 4. As for the micellar structures the common presentation is misleading; for purely geometrical reasons the polar head groups cannot be as tightly packed as shown in the figure. The values for the polar head group area depend on where one locates the interface between the polar and non-polar regions, but the area values obtained are about 50 Å^2 (33–36). The reversed variant gives values below 20 Å^2 (33–36).

The lamellar phases are not as transparent as the hexagonal ones and are often more fluid. The microscopical texture is also different.[18,19] X-ray diffraction reveals one-dimensional periodicity. The structure is considered to consist of lipid bilayers alternating with water layers. The common conceptual presentations almost always imply that the water layers are thinner than the bilayers (Figure 5), but under suitable conditions the lamellar phase may take up large amounts of water and still be thermodynamically stable.

For instance, this behaviour occurs in the lamellar phases of the binary systems of sodium diethylhexylsulphosuccinate (Aerosol OT) and water, and of several ternary systems of sodium or potassium carboxylates, or hexadecyltrimethylammonium bromide and long-chain alcohols and water ($C_nNa - C_nOH - H_2O$, $KC_n + C_nOH - H_2O$, $C_{16}TABr - C_nOH - H_2O$).[37–41] The same behaviour may be found in systems of uncharged lipids and water (monoglycerides, lecithins), although it is often enhanced by small additions of a charged compound such as sodium chloride.[35,42] The general

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FIGURE 4 The structure of the two-dimensional hexagonal phases.

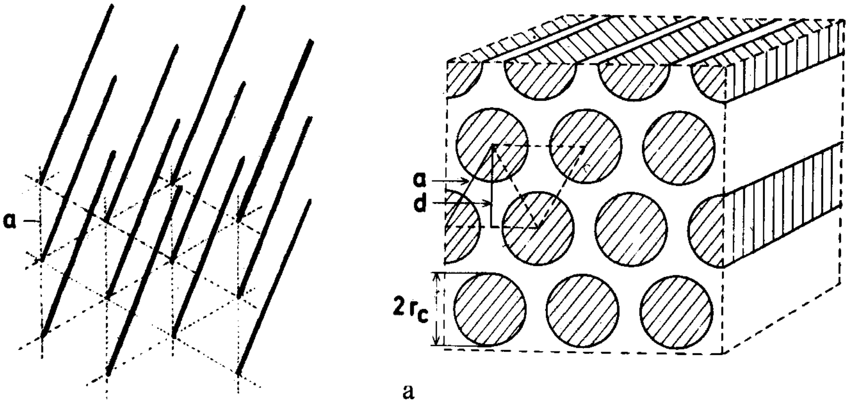

FIGURE 4a The two-dimensional array of indefinitely extended long parallel rods, *a* is the hexagonal lattice parameter, *d* the spacing with the indicees $(10\bar{1}0)$, $2r_c$ the diameter of the rod-aggregates, "normal" or "reversed".

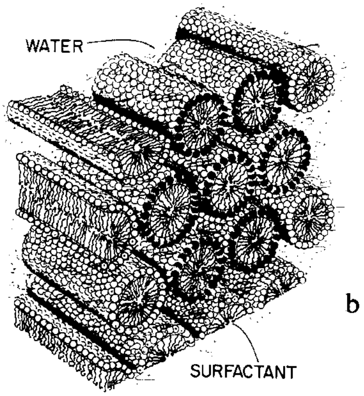

FIGURE 4b A visualization of the rod-structure of the normal hexagonal liquid crystalline phase.[19] See text for criticism.

impression of the studies of these systems with "ideal swelling" is that the average molecular packing of straight alkyl chains in the lamellar bilayer is about 25 Å^2 and not especially influenced by the size of the polar head group or its counter ion.[37–41] Figure 6 shows the experimental results for one typical system.

The lamellar phase of the binary system of the diethyl amine salt of the fully fluorinated nonanoic acid and water also exhibits the same type of swelling but in this case the molecular packing is around 40 Å^2.[41]

A practical consequence of the phenomenon of "ideal swelling" which is

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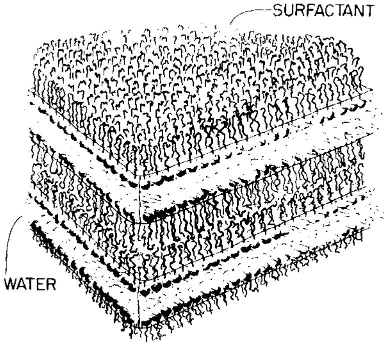

FIGURE 5 A common schematic visualization of the structure of the lamellar liquid crystalline phase of lipid/water systems.[19]

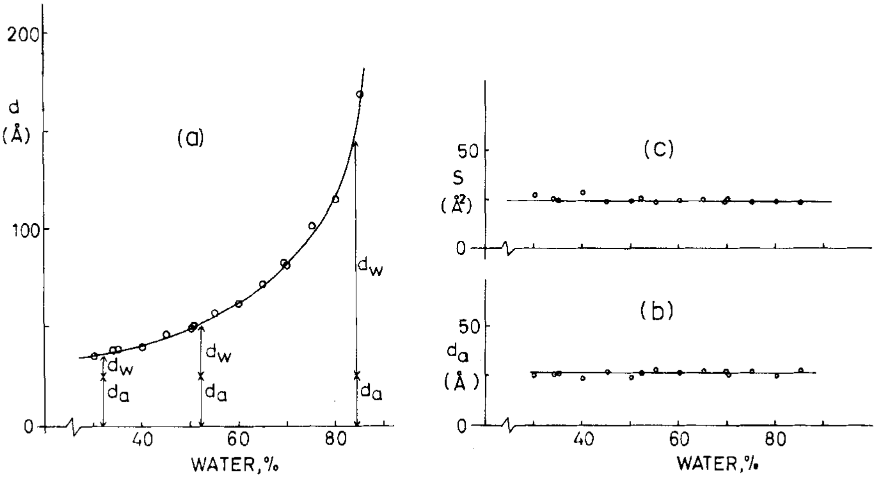

FIGURE 6 One example of the "ideal swelling" with the water content in a lamellar liquid crystalline phase. (a) Plot of the interplanar spacing, d, *vs* concentration. The relative proportions between the thicknesses of the lipid, d_a, and water layers, d_w, in different concentration regions are indicated. (b) and (c) The constancy of the lipid bilayer thickness and the molecular packing inside the lipid bilayers. The example is taken from the system hexadecyltrimethylammonium bromide-hexan-1-ol-water, the weight ratio of alcohol to soap is 40/60 and the experimental temperature 25°C.[40,41]

not always realized is that a simple plot of interplanar spacings *vs* concentration cannot be rectilinear (Figure 6a).

The addition of salt to systems of a charged amphiphile, a long-chain alcohol and water may sometimes cause the formation of low-viscous liquid crystalline phases that are orientable in a magnetic field, so-called Lawson and Flautt phases.[43] They are also often termed "nematic" lyomesophases, and they occur as two variants with positive or negative diamagnetic susceptibilities.[44–46] Their structures are considered to be hexagonal or lamellar,

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respectively, but whether or not these low-viscosity phases are continuous with the adjoining high-viscosity regions in salt-free systems is not yet established. Their inner molecular arrangements have been debated, but it now seems definite that the phase with positive magnetic susceptibility has a rod-structure, while the structure proposal for that with negative magnetic susceptibility is not as certain. A structure of lamellar bilayer discs has been proposed for the latter phase. The proof of this conclusion rests on NMR work as well as on studies of polarized absorption and X-ray diffraction.[47–50] However, it is uncertain whether the obtained X-ray experimental results warrant any definite conclusions. Aqueous systems of fluorocarbon surfactants also possess this property of being oriented in a magnetic field even without the addition of any salt.[2,51]

EQUIVOCAL PHASES

As already mentioned several other liquid crystalline phases may occur in amphiphile system. They are interspersed between the three established ones or between one of them and the aqueous micellar or the oil-continous solution. In many instances the existence of these phases as real ones and their inner structure has been questioned. The B, C, K and M phases in Ekwall's nomenclature, and the hexagonal complex and rectangular phases and the anhydrous thermotropic phases studied by the Luzzati-school will not be considered in the present context. Attention will instead be focused on the structures of the cubic, viscous isotropic, phases.

THE CUBIC PHASES

The cubic phases are optically isotropic phases of very stiff consistency. They are much stiffer than the hexagonal phases. They give X-ray diffraction patterns which give a three-dimensional crystalline cubic pattern in the low-angle region in addition of the diffuse reflection in the 4.5 Å region.

In the first instances when a cubic phase was observed and recognized as an independent phase, it was tacitly assumed that the aggregate entities were globular and in cubic close packing, either of the type "oil in water" or "water in oil" (Figure 7). This opinion still prevails, in spite of much contradictory experimental evidence. An assumption of globular aggregates was for a long time thought to be the only way to interpret the high-resolution NMR-spectra obtained from these phases. However, the work by Charvolin and Rigny had demonstrated that the nature of the spectra may be reconciled with other cubic aggregate alternatives.[52]

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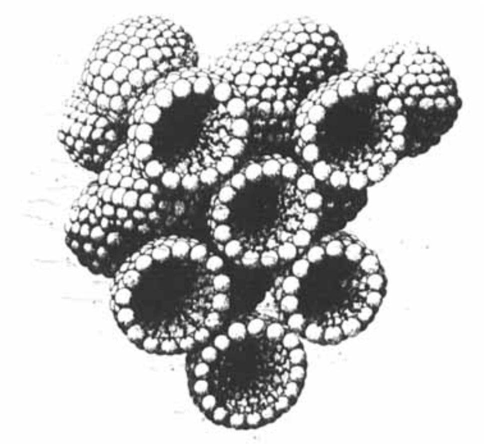

FIGURE 7 A common conceptual visualization of the structure of a viscous isotropic, cubic, phase. The "oil-in-water" type, that is globular micelles in close-packed face-centered arrangement.[19] As for globular aggregates of aqueous micellar solutions the raspberry-like appearance of the aggregates is unrealistic.

In a binary phase diagram of an amphiphilic lipid and water one may in principle find cubic phases in the following locations:

i) between the micellar aqueous solution and the normal hexagonal phase

ii) between the normal hexagonal and the lamellar phases

iii) between the lamellar and the reversed hexagonal phases and

iv) between the reversed hexagonal and the reversed micellar phase (or isotropic melt).

(In the phase diagrams of multicomponent systems there are still more possible locations.)

OBSOLETE STRUCTURE PROPOSALS

In one of the first attempts to elucidate the structures of cubic phases by X-ray diffraction the Luzzati-school studied some phases at location (ii). Cubic phases of potassium soaps (KC_{12}, KC_{14} and KC_{16}) and of alkyltrimethylammonium bromides (C_{12}TABr and C_{16}TABr) and water were being investigated.[22,23] They first assumed that the structure was face-centered and of the type "oil in water" but as a consequence of criticism by Clunie *et al.*,[53] as well as of inconsistencies in the values obtained for the area per polar group in the sequence hexagonal → cubic → lamellar phase they revised their model in favour of the "water in oil" type.[54] However, further studies more and more indicated that the cubic phases at this location in fact had body-centered structures, and that the structures were similar to that found for the anhydrous strontium soaps (SrC_{14}–SrC_{22}).[55]

THE Ia3d STRUCTURE

For the strontium soaps at elevated temperature (around 235°C) Luzzati and Spegt found a cubic phase which gives 17 reflections,[55] and they could unambiguously show that the structure belongs to the body-centered space group Ia3d (no. 230 in the International Tables 56). They proposed a structure in which the primary units are short rod-like aggregates, which are joined three and three at each end and form two interwoven but otherwise independent three-dimensional networks. The unit cell showing the positions of the axes of the rods, as well as a more easily understood visualization of the structure proposal, is given in Figure 8. The short rod-aggregates form screws which penetrate the structure in three directions at right angles. The large number of reflections, as well as the good agreement between obtained and calculated intensities, provide the proposal a high degree of crystallographic credibility. The structure of the strontium soaps will be of the "water in oil" variant, viz. the polar groups of the fatty acid molecules together with the strontium ions form the rod aggregates.

The same X-ray pattern has been found for some other cubic low water

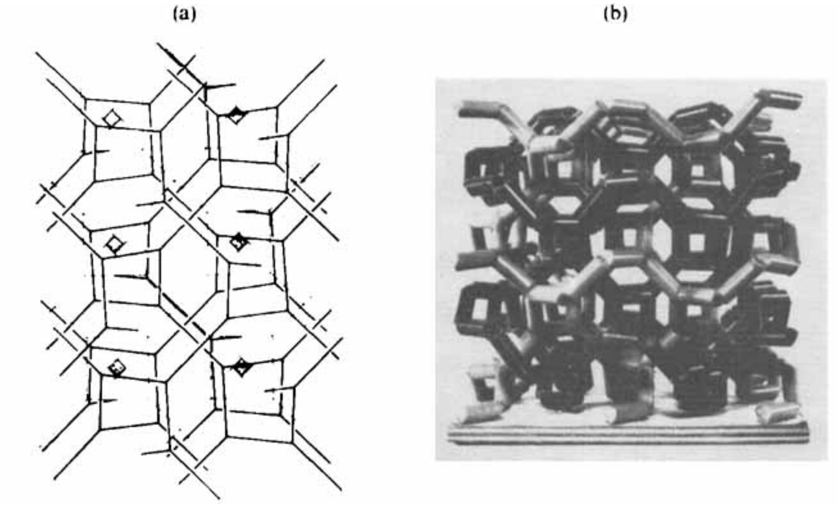

FIGURE 8 Schematic representation of the cubic structure Ia3d. (a) Perspective picture of the axes of the rod-aggregates. The dotted lines are the projections of the axes on the basal plane. (b) An attempt to visualize the two three-dimensional networks.

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lipid systems and also for cubic phases located petween the lamellar and reversed hexagonal phases (location (iii)).[33] This structure is of the "water in oil" type.

The supplementary variant of the "oil in water" type is found for phases at location (ii) (KC_8–KC_{16}, $C_{12}TABr$, $C_{12}TACl$, $C_{16}TABr$).[33] In this case the derived parameters for the rod-aggregates show that they are almost isodimensional, i.e. the length is nearly equal to the diameter.

A CUBIC STRUCTURE PROBABLY BUILT OF GLOBULAR AGGREGATES

A cubic system of globular aggregates is most likely to be found in location (i) viz. between the aqueous micellar solution and the hexagonal phase. It is easy to envisage that a system of globular micelles will separate from an aqueous solution when the concentration is increased (or when the micelles have swollen due to the incorporation of a hydrocarbon). As the separated aggregates lack directional forces one would expect them to form a close-packed, face-centered structure.

The X-ray diffraction patterns for these cubic phases often show very few reflections, and thus it is not possible to determine whether the cubic structure is primitive, body-centered or face-centered. However, in some cases the obtained patterns are indicative of primitive or body-centered structures.[57] (Note that it is always possible to "force" a cubic X-ray diffraction pattern into the face-centered variant, but the consequence for the lipid/water systems

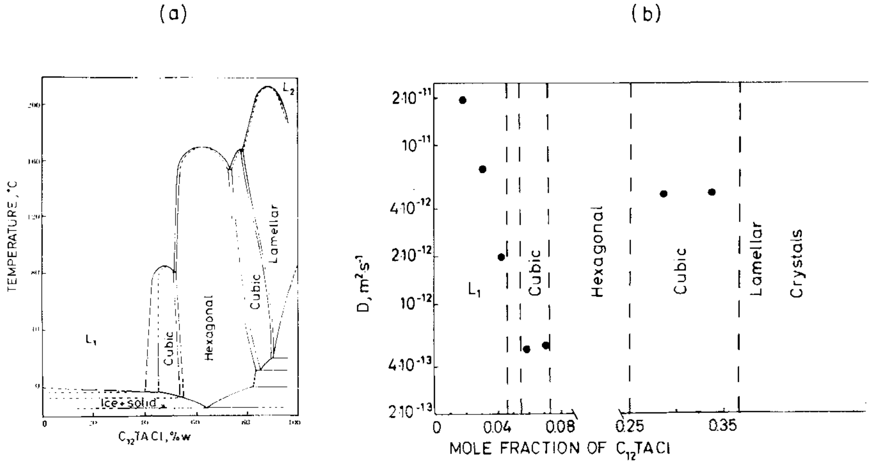

FIGURE 9 (a) The binary phase diagram of the system dodecyl trimethyl ammonium chloride/water.[59] (b) The amphiphile diffusion coefficient in the aqueous solution phase and in the two cubic phases.[60]

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will be unreasonably large unit cells; consequently the idea of non-polar "lipid" aggregates in an aqueous environment must be abandoned and structures based upon multilayered aggregates considered instead.)

It has been possible to assign this cubic phase (location (i)) to a definite space group, Pm3n (no. 223 in the International Tables 56). The space group seems to be rather firmly established as two different research groups have obtained it with different experimental techniques for this phase in the system dodecyltrimethylammonium chloride-water[58,59] (Figure 9(a)). Luzzati and Tardieu have proposed a structure composed of a three-dimensional network of short "amphiphilic" rods of equal length connected to three others at one end and to four at the other, thus forming a chlathrate structure in which each basket in addition holds a globular aggregate.[58] The globular aggregates would be in a body-centered cubic array but the network degenerates the structure as whole into a primitive one. However, the structure proposal is open to criticism because the calculation of the structure factors of the individual reflections includes several independently adjustable parameters, and furthermore the basic assumption of equal area values for the polar head groups at the interfaces between polar and non-polar regions leads to improbable size relations between the globular aggregates and the rod-aggregate units. (The size of the globular aggregates would be four times the size of the nearly isodimensional rod-aggregate units.)

On the other hand, support for the idea of globular aggregates as the only building block for the cubic structure in this phase location is given by the NMR diffusion studies in the above mentioned C_{12}TACl system[59]. The lipid diffusion coefficient decreases monotonically inside the micellar solution region with decrease in water content, and shows no discontinuity on passing into the neighbouring cubic phase (Figure 9(b)). (The fact that the coefficient obtains a tenfold higher value for the second cubic phase of this system, on the other side of the hexagonal phase (location (ii)), and also has a considerably higher value than that obtained for rather concentrated micellar solutions, implies that the latter cubic phase cannot have the reversed structure of "water in oil" type, but must have a structure based upon another principle, for instance the rod-structure Ia3d discussed above. It is also possible to distinguish between cubic phases with rod and lamellar structures by NMR diffusion methods; see below.)

The existence of cubic phases which with certainty are in the location (iv) and have the structure of globular aggregates of the "water in oil" type has not been demonstrated.

As mentioned above the occurrence of cubic structures based upon globular aggregates would be most likely at locations (i) and (iv), but the existence of such structures in these phases has not yet been satisfactorily demonstrated. The contradiction between the X-ray diffraction and NMR diffusion results

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must be eliminated. The X-ray favours for phases at location (i) primitive or body-centred structures and consequently contradicts the existence of globular aggregates while the NMR indicates the opposite.[57–61]

One occasionally encounters the argument that globular micellar aggregates should be capable of forming a body-centered structure since cubic plastic crystals with this structure are known to exist. This argument disregards the fact that the globular molecules forming cubic plastic crystals are not strictly symmetrical.[62,63]

THE Im3m STRUCTURE

So far we have only been discussing cubic structures based upon globular or rod-like aggregate units. Proposals of cubic structures based upon lamellar disc aggregates have also been made. Cubic phases are found in systems of monoglycerides and water at chain lengths starting from C_{14}, and these phases are located between the lamellar phase and the reversed hexagonal phase, which in the monoglyceride systems occurs on the water-rich side of the lamellar phase[64] (Figure 10). In addition, the phase diagrams may be of a such type that by increasing the temperature of a particular specimen it is possible to transform an originally lamellar structure into a cubic structure and further into a hexagonal one (Figure 11).[65] The X-ray diffractograms of the cubic phase can be indexed as belonging to a body-centered structure, and about ten years ago Larsson gave some general arguments for a lamellar organization of the amphiphile molecules in the structural units.[66] A structure consisting of closed polyhedra, the tetrakaidekahedra of Lord Kelvin, was proposed (Figure 12(a)). This polyhedron has six quadrilateral and eight hexagonal faces, and if it is assumed that the lipid molecules form the faces, the maximum amount of water may be enclosed in a body-centered structure.

However, subsequent studies of water and lipid diffusion by NMR techniques gave information which excluded a structure composed of closed aggregates, and instead required a structure with continuous water and lipid regions.[67] Table I shows that in a monoglyceride/water system the directly measured lateral diffusion coefficient of the lipid in the lamellar phase agrees well with the calculated lateral diffusion coefficient for the cubic phase when it is assumed that its structural units are lamellar entities. As a comparison, the results from the lamellar phase and the cubic Ia3d phase in the Aerosol OT system show that the measured values for the lateral diffusion coefficient of the lamellar phase are similar to those obtained for the cubic phase, assuming the existence of a rod-structure.

Another argument against a closed structure for the cubic phase in monoglyceride/water systems is that the values for the X-ray diffraction spacings decreased when the temperature was increased.[67]

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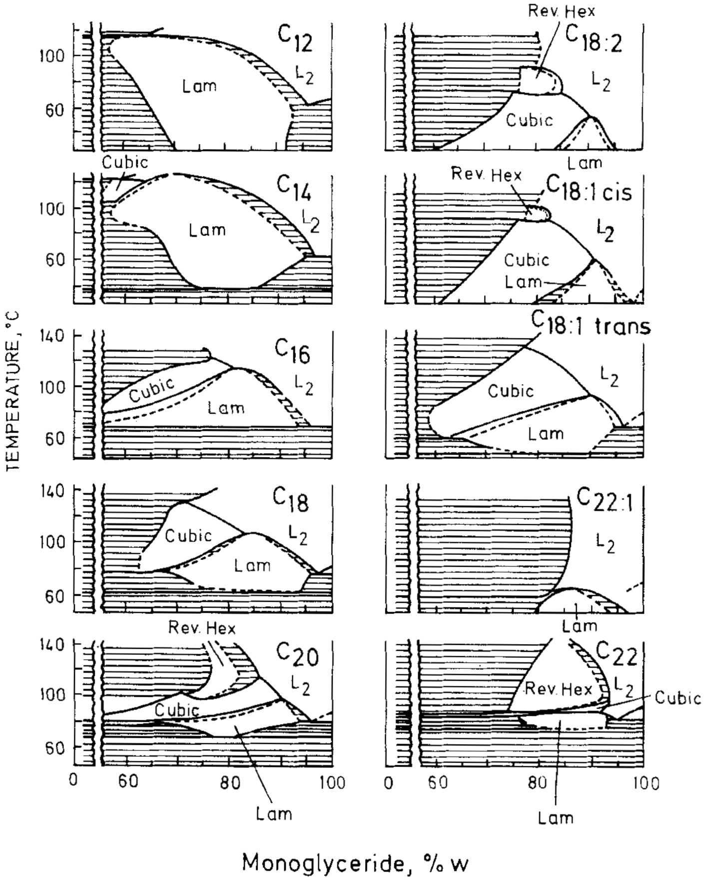

FIGURE 10 The binary phase diagram for some aqueous monoglyceride systems. Modified after Lutton.[64]

The structure for this phase belongs to space group Im3m (no. 223 in the International Tables[56]). If the closed tetrakaidekahedra structure is opened by removing the quadrilateral faces a water- and lipid-continuous system is formed. The visualization given in Figure 12(b) shows the location of the methyl end group planes in the lipid bilayers and is very schematic, since there will in addition be a smooth continuous change in the curvature (see below).

A closer inspection of the structure proposal shows furthermore that the structure is bicontinuous with respect to water.[68] The lipid bilayer membranes separate two independent three-dimensional systems of water channels.

The proposal resembles, and is in fact identical with, one of the structures which is created by "intersection free infinite periodic minimal surfaces". Such structures have been reviewed by Schoen.[69] Scriven has recently dis-

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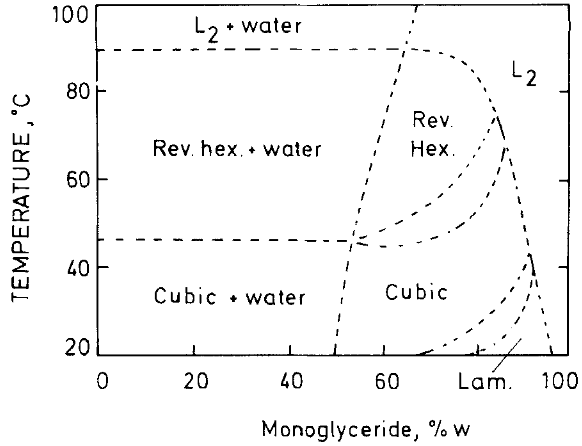

FIGURE 11 The binary phase diagram for the system "sunflower monoglyceride"-water modified after Krog *et al.*[65] The diagram illustrates how, by simply raising the temperature, one obtains the transition lamellar → cubic → hexagonal phase.

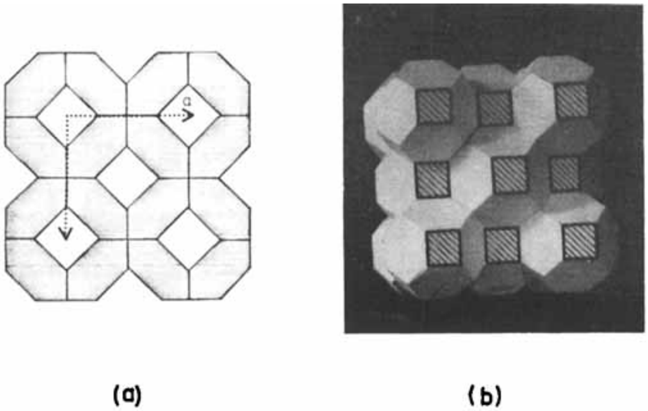

FIGURE 12 The tetrakaidekahedral polyhedron proposed as the building stone of the body-centered cubic structure in monoglyceride/water systems. The cubic unit cell axes are dotted. The structure unit consists of hexagon-shaped and quadratic lipid monolayers. In the original proposal the unit was closed[66] but in a later version the quadratic faces were removed and thus giving an open structure.[67] (a) Schematic illustration of the unit cell. The planes have the positions $\pm$ [(111), $(\bar{1}11)$, $(1\bar{1}1)$, $(11\bar{1})$]. (b) An attempt to visualize the structure, the shaded areas represent the two sets of water channels separated by lipid bilayers.

cussed the possibilities for "equilibrium bicontinuous structures" to exist in amphiphilic systems.[70,71] One of the structures with "periodic minimum interfaces" is the "Schwarz′ primitive minimal surface" (Figure 13). This structure has a primitive cubic lattice and is the same as that obtained when

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TABLE I

Experimental and calculated values for the amphiphile diffusion coefficient as function of temperature[a] (67).

Composition (w/w)	Temp. °C	Phase structure	$D \times 10^{11}$, m^2/s	$D_L \times 10^{11}$, m^2/s	$D_L^{cub} \times 10^{11}$, m^2/s	$D_{\parallel}^{cub} \times 10^{11}$, m^2/s
Monoolein-heavy water 88/12	22	lamellar	0.73	1.1		
	29	lamellar	0.85	1.4		
	35	lamellar	1.1	1.6		
	43	cubic	1.5		2.3	4.5
	49	cubic	2.0		3.0	6.0
	57	cubic	2.6		3.9	7.8
Aerosol OT-heavy water 70.9/29.1	24	lamellar	0.67	1.0		
	29	lamellar	0.93	1.4		
	35	lamellar	1.1	1.7		
72.4/27.6	24	cubic	0.36		0.54	1.1
	29	cubic	0.50		0.75	1.5
	35	cubic	0.59		0.89	1.8

[a] D is the experimentally determined effective diffusion coefficient. D_L represents the lateral diffusion coefficient in a lipid bilayer, obtained by correcting D for the "magic angle", D_L^{cub} and $D_{\parallel}^{cub}$ are estimated diffusion coefficients for translational motion along the amphiphilic surface at assumed different aggregate structures for the cubic phase (lamellar and rod-like aggregate units, respectively).

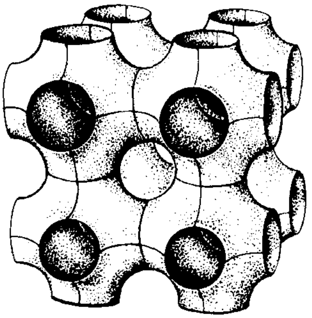

FIGURE 13 Schwarz' primitive periodic minimal surface, with simple cubic symmetry.[71] The surface partitions space into two equal, infinitely connected interpenetrating subvolumes.

the hexagon-shaped planes of Figure 12(b) are given a curvature and the bilayers degenerate into methyl contact planes. The primitive cubic lattice will be transformed into a body-centered one if we consider lipid bilayers instead of methyl end group planes and take into account the symmetry of the lipid bilayers. The space group will be the same, Im3m, as that arrived at from the X-ray findings.

This structure proposal for the cubic phase occurring in monoglyceride-water systems is of biological interest in that the structure greatly resembles that proposed for plastids in etiolated Avena leaves.[72] The plastids contain the precursors of the chloroplast membranes, and these prolamellar bodies give electron micrographs (Figure 14) which indicate a framework of tubules lying in the direction of the three major axes of a cube type of lattice. Figure 15 shows a three-dimensional representation of the membrane system given by Gunning. There is a close resemblance with the structure proposal given in Figure 12.

The cubic phase of monoglyceride systems also has a physiological connection. *In vitro* studies of the lipolysis of triolein performed in an environment resembling the intestine have shown that a cubic phase exists as an intermediate state between fat emulsion droplets and micelle-like aggregates of monoglyceride, fatty acid and bile.[73]

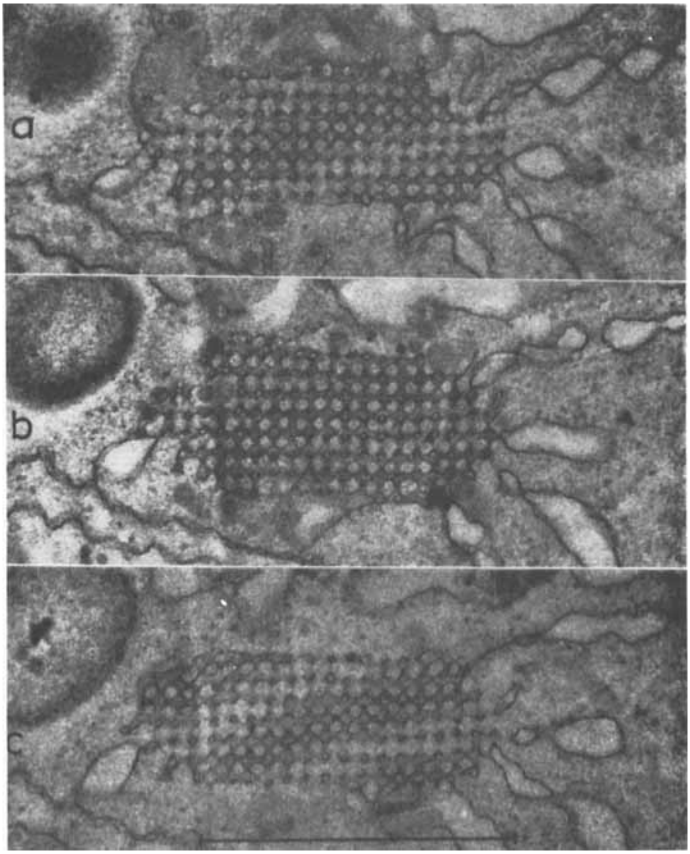

FIGURE 14 Electron micrographs of serial sections of a prolamellar body in a guard cell plastid obtained from an etiolated *Avena* leaf. The sectioning of the prolamellar body is close to a major lattice plane.[72]

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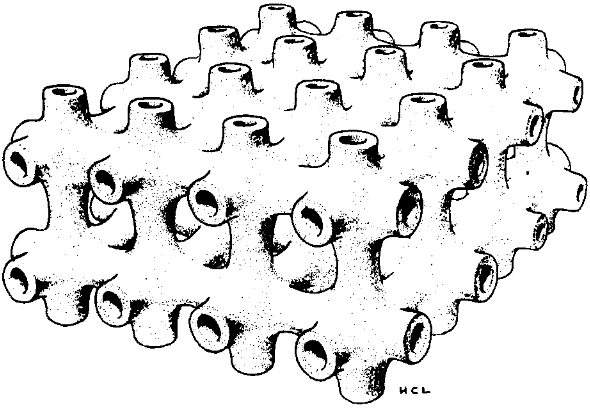

FIGURE 15 The three-dimensional visualization by Gunning of the membrane system occurring in plastids from etiolated *Avena* leaves.[72] (Note that the electron micrographs cannot give the curvature.)

A cubic phase has also been found *in vitro* in mixtures of monoglucosyl- and diglucosyl-diglycerides obtained from Acholeplasma Laidlawii.[74]

CONCLUSION

Liquid crystallinity is a common feature in lipid-water systems. The phases that occur in these systems have very varying structures. Common for all variants is that on the short range level (<5 Å) the state is liquid, while the state on the long range level (>15 Å) gives a crystalline superstructure of molecular aggregates ordered in one, two or three dimensions. This is a consequence of the dual nature of the amphiphilic molecules, which have a polar group in addition to a non-polar part of aliphatic and/or aromatic character. The balance between these two parts of the molecule determines its behaviour towards water and is the reason for the liquid crystallinity of lipid-water systems. There are three common structural variants, viz. the lamellar, the hexagonal and the reversed hexagonal ones, and the structure of these three is commonly agreed upon among workers in the field of lyotropic liquid crystals. But several other liquid crystalline phases also exist. One important group of these phases consists of the cubic, viscous isotropic, phases. The original tacitly assumed structure principle for this group was globular aggregates either of the "oil in water" or "water in oil" type, but this structure principle seems often to be contradicted by experimental facts. Consequently, structure proposals based upon the principles of bicontinuous systems of rod- or disc-like aggregates have been made. Since there are several different locations in the phase diagram where cubic phases may be found, the existence of only one building principle is excluded and one is forced to consider the existence of several different structure variants. The internal structure of these variants must be in logical relation to the structures of neighbouring phases.

One interesting aspect regarding the cubic phases is that they have been found in biological systems proper and it should be stressed that a cubic phase with a high lipid content may be in equilibrium with an aqueous system where the lipid content is low. Also it has to be stressed that the cubic phases may be more abundant than previously realized; due to their optical anisotropy one cannot see them in the polarizing microscope.

It has also been demonstrated that the cubic phase of a monoglyceride system possess the capability to incorporate in its structure other lipid substances such as diglycerides, cholesterol, bile acid salts and also proteins.

This review has to a large extent dealt with the conditions in liquid crystalline systems of simple amphiphilic lipids, but since the fundamentals are the same, reasonable conclusions may also be drawn for more complicated lipid systems.

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References

1. A. S. C. Lawrence, *Mol. Cryst. Liq. Cryst.*, **7**, 1–57 (1969).
2. G. J. T. Tiddy, *Physics Report*, **57**, 1 1–46 (1980).
3. B. Lindman and H. Wennerström, *Topics in Current Chemistry*, **87**, 1–83 (1980).
4. H. Wennerström and B. Lindman, *Physics Report*, **52**, 1 1–86 (1979).
5. J. Charvolin and A. Tardieu, in *Liquid Crystals, Solid State Physics* (eds. H. Ehrenreich *et al.*), Academic Press 1978, 201–257.
6. A. Skoulios, *Ann. Phys.*, 421–450 (1978)
7. R. D. Laughlin, *Adv. Liq. Cryst.* (ed. G. H. Brown), **3**, 41–98, 99–147 (1978).
8. P. Ekwall, *Adv. Liq. Cryst.* (ed. G. H. Brown), **1**, 1–142 (1975).
9. V. Luzzati, in *Biological Membranes* (ed. D. Chapman), **1**, 71–123 (1968).
10. P. A. Winsor, *Chem. Rev.*, **68** 1–40 (1968).
11. P. A. Winsor, in *Liq. Crystals and Plastic Crystals* (eds. G. W. Gray and P. A. Winsor), Ellis Horwood 1974, Vol. I chaps. 1; 2.2; 2.3 and 5, Vol. 2 chap. 6.
12. Ref. 4 p. 50.
13. L. Mandell and P. Ekwall, *Acta Polytechn. Scand. Chem. Met. Series*, **74**, 1 (1968).
14. L. Mandell, K. Fontell, and P. Ekwall, in *Ordered Fluids and Liquid Crystals* (eds. R. S. Porter and J. F. Johnson), Advances in Chemistry Series 63, Am. Chem. Soc. 1967, 89–124.
15. N. H. Hartshorne, in *Liquid Crystals and Plastic Crystals* (eds. G. W. Gray and P. A. Winsor), Ellis Horwood 1974, Vol. 2, chap. 2.
16. N. H. Hartshorne, *Microscopy of Liquid Crystals*, Microscope Publications Ltd., London, 1974.
17. D. Demus and L. Richter, *Textures of Liquid Crystals*, Verlag Chemie, Weinheim, 1978.
18. F. B. Rosevear, *J. Am. Oil Chem. Soc.*, **31**, 628–639 (1954).
19. F. B. Rosevear, *J. Soc. Cosmetic Chem.*, **19**, 581–594 (1968).

20. J. F. Goodman and J. S. Clunie, in *Liquid Crystals and Plastic Crystals* (eds. G. W. Gray and P. A. Winsor), Ellis Horwood 1974, vol. 2, chap. 1.
21. C. J. James and J. F. Heathcock, in *Surface Active Agents*, Society of Chem. Industry, London 1979, 219–232.
22. V. Luzzati, H. Mustacchi, A. Skoulios, and F. Husson, *Acta Cryst.*, **13**, 660–667 (1960).
23. F. Husson, H. Mustacchi, and V. Luzzati, *Acta Cryst.*, **13**, 668–677 (1960).
24. R. M. Hedge, R. K. Thomas, M. Mortimer, and J. W. White, *J. C. S. Faraday I*, **70**, 236–243 (1980).
25. L. B. Å. Johansson and B. Lindblom. *Q. Rev. Biophys.*, **13**, 63–118 (1980).
26. D. M. Small, *J. Am. Oil Chem. Soc.*, **45**, 108–117 (1968).
27. C. Madelmont and R. Perron, *Coll. Polymer Sci.*, **254**, 581–598 (1976).
28. R. Perron and J. L. Curat, *Chem. Phys. Lipids*, **19**, 301–311 (1977).
29. M. Hato and K. Shinoda, *J. Phys. Chem.*, **77**, 378–381 (1973).
30. J. F. Goodman and T. Walker, in *Chem. Soc. Spec. Period. Reports* (senior reporter D. H. Everett), *Colloid Sci.* **3** (1979) chap. 5.
31. P. Mukerjee and K. J. Mysels, *Critical Micelle Concentrations of Aqueous Surfactant Systems*, NSRDS—MBS 36, Washington D.C., US Government Printing Office 1971.
32. H. Wennerström, *J. Coll. Interface Sci.*, **68**, 589–590 (1979).
33. K. Fontell, in *Liquid Crystals and Plastic Crystals* (eds. G. W. Gray and P. A. Winsor), Ellis Horwood, Chichester 1974, Vol. 2, chap. 4.
34. Ref. 8, p. 26.
35. P. Ekwall, L. Mandell, and K. Fontell, *Acta Chem. Scand.*, **22**, 373–375 (1968).
36. K. Fontell, P. Ekwall, L. Mandell, and I. Danielsson, *Acta Chem. Scand.*, **16**, 2294–2298 (1962).
37. K. Fontell, *J. Coll. Interf. Sci.*, **44**, 318–329 (1973).
38. L. Mandell, K. Fontell, and P. Ekwall, *Acta Chem. Scand.*, **22**, 1543–1550 (1968).
39. L. Mandell, K. Fontell, and P. Ekwall, *J. Coll. Interf. Sci.*, **31**, 508–529 (1969).
40. L. Mandell, K. Fontell, and P. Ekwall, *J. Coll. Interf. Sci.*, **33**, 215–235 (1970).
41. K. Fontell, unpublished work.
42. T. Gulik-Krzywicki, A. Tardieu, and V. Luzzati, *Mol. Cryst.*, **8**, 285–291 (1969).
43. K. D. Lawson and T. J. Flautt, *J. Am. Chem. Soc.*, **89**, 5489–5491 (1967).
44. K. Radley, L. W. Reeves, and A. S. Tracey, *J. Phys. Chem.*, **80**, 174–182 (1976).
45. L. W. Reeves, A. S. Tracey, and M. M. Tracey, *Can. J. Chem.*, **57**, 747–753 (1979).
46. F. Fujiwara, L. W. Reeves, M. Suzuki, and J. A. Vanin, in *Solution Chemistry of Surfactants* vol. 1., (ed. K. L. Mittal), Plenum Press, New York 1979, p. 63–77.
47. B. J. Forrest, L. Hecker, and L. W. Reeves, *Mol. Cryst. Liq. Cryst.*, **58**, 223–232, 233–244 (1980).
48. J. Charvolin, D. M. Levelut, and E. T. Samulski, *J. Phys. Lettres*, **40**, p. L-587–L-592 (1979).
49. O. Söderman, G. Lindblom, L. B. Å. Johansson, and K. Fontell, *Mol. Cryst. Liq. Cryst.*, **59**, 121–136 (1980).
50. L. Q. De Amaral, C. Pimentel, and M. Travares, *J. Chem. Phys.*, **71**, 2940-2945 (1979).
51. N. Boden, P. H. Jackson, K. McMullen, and M. C. Holmes, *Chem. Phys. Lett.*, **65**, 476–479 (1979).
52. J. Charvolin and P. Rigny, *J. Magn. Reson.*, **4**, 40–46 (1971).
53. J. S. Clunie, J. M. Corkill, and J. F. Goodman, *Proc. Roy. Soc.*, **A 285**, 520–533, 534–541 (1965).
54. V. Luzzati and F. Reiss-Husson, *Nature*, **210**, 1351–1352 (1966).
55. V. Luzzati and P. A. Spegt, *Nature*, **215**, 701–704 (1967).
55. *International Tables for X-ray Crystallography*, The Kynoch Press, Birmingham 1952.
57. K. Fontell, K. Mandell, and P. Ekwall, *Acta Chem. Scand.*, **22**, 3209–3223 (1968).
58. A. Tardieu and V. Luzzati, *Biochim. Biophys. Acta*, **219**, 11–17 (1970).
59. R. R. Balmbra, J. S. Clunie, and J. F. Goodman, *Nature*, **222**, 1159–1160 (1969).
60. T. Bull and B. Lindman, *Mol. Cryst. Liq. Cryst.*, **28**, 155–160 (1974).
61. G. Lindblom and H. Wennerström, *Biophys. Biochem.*, **6**, 167–171 (1971).
62. G. W. Gray and P. A. Winsor, *Mol. Cryst. Liq. Cryst.*, **26**, 305–327 (1974).
63. Ref. 11, chap. 2.2 in vol. 1.

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64. E. S. Lutton, *J. Am. Oil Chem. Soc.*, **42**, 1068–1970 (1965).
65. J. V. Boyd, N. Krog, and P. Sherman, in *Theory and Practice of Emulsion Technology* (ed. A. L. Smith), Academic Press 1976, p. 61.
66. K. Larsson, *Z. Phys. Chem.*, (Frankfurt am Main), **56**, 173–198 (1967).
67. G. Lindblom, K. Larsson, L. B. Å. Johansson, K. Fontell, and S. Forsén, *J. Am. Chem. Soc.*, **101**, 5465–5470 (1979).
68. K. Larsson, K. Fontell, and N. Krog, *Chem. Phys. Lipids*, **27**, 321–328 (1980).
69. A. H. Schoen, Nasa Technical Notes (1970) NASA TN D–5541.
70. L. E. Scriven, *Nature*, **263**, 123–125 (1976).
71. L. E. Scriven, in *Micellization, Solubilization and Microemulsions* (ed. K. L. Mittal), vol. 2, Plenum Press, New York 1977, p. 877–893.
72. B. E. S. Gunning, *Protoplasma*, **60**, 111–130 (1965).
73. J. S. Patton and M. C. Carey, *Science*, **204**, 145–148 (1979).
74. Å. Wieslander, L. Rilfors, L. B. Å. Johansson, and G. Lindblom, *Biochemistry*, in press 1980.

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