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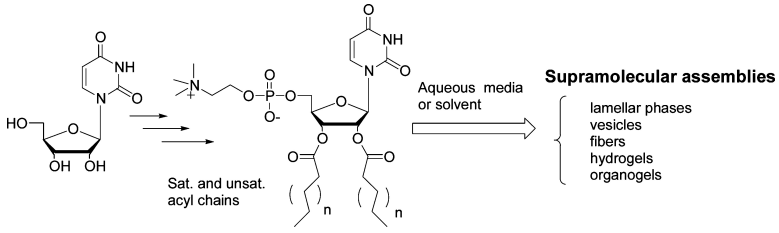

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Supramolecular Assemblies of Nucleoside Phosphocholine Amphiphiles

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Abstract: A family of new uridine phosphocholine amphiphiles that were prepared using a convenient four-step synthetic route is described. Physicochemical studies (differential scanning calorimetry, small-angle X-ray scattering, UV−vis and circular dichroism spectroscopies, light microscopy, transmission electronic microscopy, and scanning electron microscopy) show that these amphiphiles spontaneously assemble into supramolecular structures including vesicles, fibers, hydrogels, and organogels. In aqueous solution, the amphiphiles possessing saturated alkyl chains self-assemble into DNA-like helical fibers in the crystalline state below T_m and compact bilayers above the melting temperature (T_m). The transition from bilayers to fibers is thermally reversible. Above a threshold concentration (>6% w/w), a hydrogel is formed due to an entangled network of the fibers. A therapeutic agent such as DNA can be entrapped within the hydrogel structure. In addition to forming bilayer vesicles and hydrogels in aqueous solution, these nucleoside amphiphiles also form organogels in cyclohexane above T_m. Scanning electron microscopy shows a continuous multilamellar phase in the organogels.

Introduction

The design of supramolecular assemblies that mimic the molecular organization and compartmentalization observed in biological systems is of widespread interest. The most familiar examples of these assemblies are the phosphocholine lipid bilayers, which constitute prokaryotic and eukaryotic cell membranes.[1] These diacyl phosphocholines spontaneously organize to form spherically self-closed liposomes in solution. To better understand the chemical principles and structural characteristics that govern self-assembly, the molecular structures of conventional lipids and surfactants are being modified through incorporation of molecular recognition features (electrostatic interactions, H-bonding, chirality). It is well documented that molecular recognition principles can be used to create highly ordered molecular assemblies from small molecules.[2−9] For example, the molecular recognition features present in DNA have been incorporated in amphiphiles, including nucleobase bolaamphiphiles and cholesterol derivatives.[10−19] Nucleoside phosphocholine amphiphiles are new amphiphiles possessing both the molecular recognition information of nucleic acids and the compartmentalization characteristics of lipids. The introduction of the nucleoside structure extends the supramolecular organization capabilities of natural glycerol based phosphocholine lipids.

In this study, we present the synthesis and physicochemical properties of uridine based phosphocholine amphiphiles, hydrogels, and organogels. The experimental results from differential scanning calorimetry, small-angle X-ray scattering, UV−vis and circular dichroism spectroscopies, light microscopy, transmission electronic microscopy, and scanning electron microscopy support the presence of DNA-like helical fibers and a "fluid" lamellar state, below and above the phase transition temperature, respectively. At concentrations above 5% w/w, the entangled fibers form a hydrogel in aqueous solution. To evaluate if these hydrogels are of potential use for the delivery of therapeutics, we report the incorporation of calf thymus DNA in the hydrogel network. Furthermore, in cyclohexane the dimyristoyl and dipalmitoyl nucleoside amphiphiles also afford organogels as a result of the formation of multilamellar phases.

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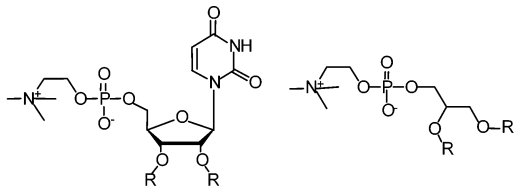

R= acyl chain length	uridine phosphocholines	glycerol phosphocholines
14, myristoyl	DMUPC	DMPC
16, palmitoyl	DPUPC	DPPC
18, stearyl	DSUPC	DSPC
20, arachidoyl	DAUPC	DAPC
18 (9-olefin), oleyl	DOUPC	DOPC

Figure 1. Uridine phosphocholine amphiphiles under investigation and glycerol phosphocholine analogues.

Results and Discussion

Synthesis of Nucleoside Based Phosphocholine Amphiphiles (1,2-Dimyristoyluridinophosphocholine, DMUPC; 1,2-Dipalmitoyluridinophosphocholine, DPUPC; 1,2-Distearyluridinophosphocholine, DSUPC; 1,2-Diarachidoyluridinophosphocholine, DAUPC; 1,2-Dioleyluridinophosphocholine, DOUPC). Uridine phosphocholine amphiphiles possessing saturated acyl chain lengths from C14 to C20 as well as an unsaturated analogue were investigated (Figure 1). This series of amphiphiles will assess the effects of amphiphile structure—nucleobase, chain length, and chain unsaturation—on physicochemical properties and supramolecular organization. The synthesis of these nucleoside phosphocholine amphiphiles derived from uridine is shown in Scheme 1. Briefly stated, the uridine acetonide derivative **1** was reacted with an excess of chlorooxodioxaphospholane in tetrahydrofuran in the presence of triethylamine at 0 °C to afford the phosphate nucleoside derivative **2**. Compound **2** was transferred to a pressure tube and heated for 24 h with trimethylamine in acetonitrile to give **3**. The choline intermediate **4** was obtained after acetonide cleavage using HCl and formic acid. Next, the two fatty acid chains (myristic/palmitic/stearic/arachidic/oleic acid) were DCC coupled to the secondary hydroxyl groups to afford the uridine amphiphiles **5a**−**e**. All amphiphiles were isolated after purification by size exclusion chromatography (LH 20, DCM/MeOH 50/50). This synthetic strategy used readily available starting materials and is an improvement over the phosphoramidite route to phosphocholine;[20] also, it is amenable to the preparation of a large number of derivatives.

Thermal Analysis. The phase transition temperature (T_m) and phase transition enthalpies of the nucleoside phosphocholine amphiphiles were determined by modulated differential scanning calorimetry (MDSC). It is known that with natural glycerol phosphocholines the physical properties of the bilayers, such as permeability and overall stability, depend on the T_m. Phase transition temperatures or melting temperatures of 20.5, 41.8, 53.0, and 65.0 °C were measured for DMUPC, DPUPC, DSUPC, and DAUPC, respectively. The value of T_m increases with increasing alkyl chain length (Figure 2a). The melting temperatures observed for nucleoside amphiphiles are similar to that for the natural glycerol (Figure 2a) as well as ribose based phosphocholines, which lack the nucleobase.[20,21] The melting temperature of the unsaturated derivative DOUPC was not observed between 80 and −40 °C (DOPC, $T_m = -20$ °C).

As observed with natural glycerol phosphocholines, the enthalpies of the nucleoside amphiphiles also increase with chain length (Figure 2b). However, the enthalpies of the nucleoside amphiphiles are dramatically lower than that reported for the glycerol phosphocholines, as shown in Figure 2b.[20] For example, DAUPC exhibits a phase transition enthalpy of 1.84 kcal/mol whereas DAPC, the glycerol phosphocholine analogue, possesses a phase transition enthalpy of 11.9 kcal/mol. A similar correlation is observed with all chain lengths. Such a large difference in enthalpy indicates that the nucleoside moieties strongly affect the transition between the solid and the "fluid" liquid crystal state of the chains, likely reflecting amphiphile organization below and/or above the T_m. This conclusion is further supported by results obtained with the ribose phosphocholine analogues. Ribose phosphocholine amphiphiles, which lack a nucleobase, do not possess a small enthalpy and have values similar to the glycerol phosphocholines.[20] The role of the nucleobase is significant in this transition from the solid to the liquid crystalline state, even though the chain length is the primary determinant of the melting temperature.

Supramolecular Assemblies. A variety of supramolecular structures are formed by uridine phosphocholine amphiphiles, and these structures have been studied by light and electronic microscopies (Table 1). The type of supramolecular structure formed was dependent on the composition of the amphiphile, temperature, concentration, and if the amphiphile was in water or cyclohexane. All five amphiphiles assemble into liposome-like structures in aqueous solution at temperatures above the T_m of the amphiphile. Importantly, the presence of the nucleobase on the amphiphilic structure does not prohibit the formation of lamellar phases, even though the incorporation of the nucleobase in the amphiphile increases the effective size of the headgroup. Depending on the procedure used, liposomes ranging from 40 nm to 15 μm in diameter can be prepared. For example, amphiphile **5e** (1,2-dioleyluridinophosphocholine (DOUPC)) forms vesicles in water as evident by light and electron transmission microscopy (TEM) (Figure 3). Small unilamellar vesicles (SUV) and "worm"-like structures can also be obtained depending on the preparation conditions.

These supramolecular structures are formed using two methods. In the first approach, the solid amphiphile is hydrated directly on the slide (method A). Hydration of the amphiphile (e.g., DOUPC) is observed under a microscope with formation of vesicular structures including multilamellar systems (Figure 3a). A more typical procedure to afford liposomes (method B) involves extrusion of an aqueous solution of the amphiphile. For example, in this method, 3 mg of DOUPC is first dissolved in 1 mL of buffer (Tris 100 mM, NaCl 20 mM, EDTA 10 μM, pH 7.4). After 20 min of agitation at room temperature, large liposomes are extruded 10 times through a polycarbonate filter (50 nm) at 21 °C to obtain small unilamellar vesicles. Particle sizing using quasi-elastic light scattering (QELS; Wyatt) shows particles with a hydrodynamic radius (R_h) of 20.5 nm. The presence of this SUV population was also confirmed by TEM experiments (Figure 3b).

In addition to forming vesicles in solution, some of these nucleoside phosphocholine amphiphiles form hydrogels at room temperature. Gels, either covalently or noncovalently cross-linked, continue to attract attention as materials for use in the

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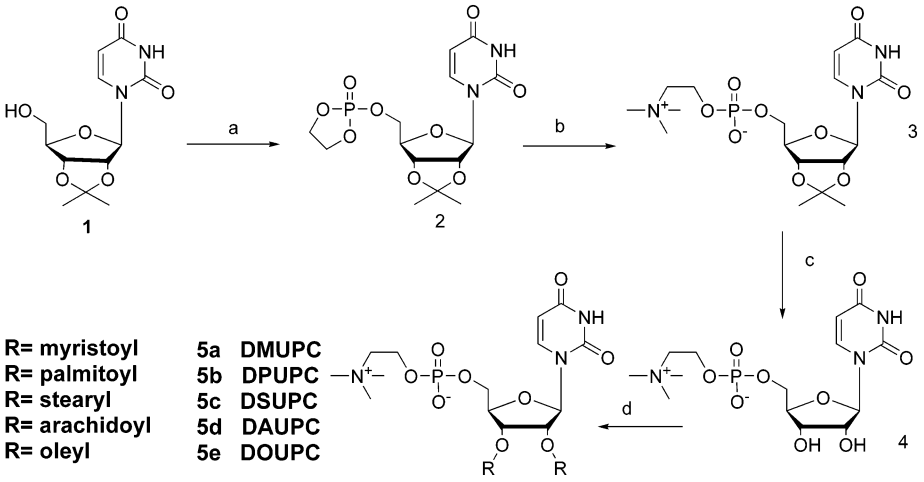
Scheme 1. Synthetic Scheme for Nucleoside Based Amphiphiles[a]



[a] Conditions: (a) chlorooxodioxaphospholane, TEA, THF, 0 °C, 15 h; (b) trimethylamine, AcCN, THF, 60 °C, 24 h; (c) HCl in HCO_2H/H_2O; (d) myristic/palmitic/stearic/arachidic/oleic acids, DCC, DMAP, DMF, RT.

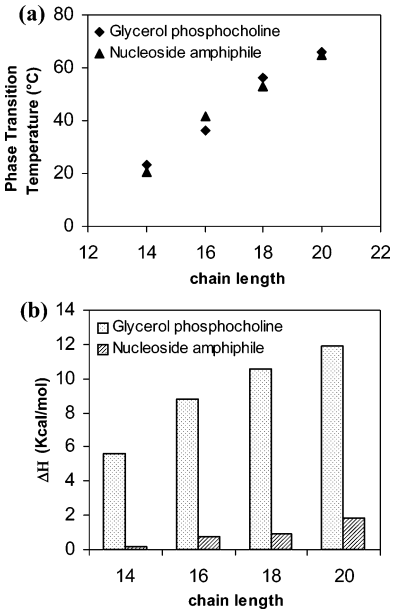


Figure 2. Phase transition temperature (a) and enthalpies (b) versus chain length of nucleoside amphiphiles compared to natural glycerol phosphocholines.

Table 1. Supramolecular Structures Formed by Uridine Phosphocholine Amphiphiles and Melting Temperature, T_m, Data[a]

amphiphile	bilayers	fibers/hydrogels	organogels	T_m, °C
DMUPC	+	n.o.	+	20.5
DPUPC	+	+	+	41.8
DSUPC	+	+	−	53.0
DAUPC	+	+	−	65.0
DOUPC	+	n.o.	−	n.o.

[a] n.o., not observed.

medical, analytical, and material science areas.[9,22] In particular, hydrogels are of interest for medical applications (e.g., wound repair, tissue engineering, etc.) since these materials mimic the properties of soft tissue, and can be used as a delivery carrier for bioactive molecules and macromolecules.[23−25] Although a

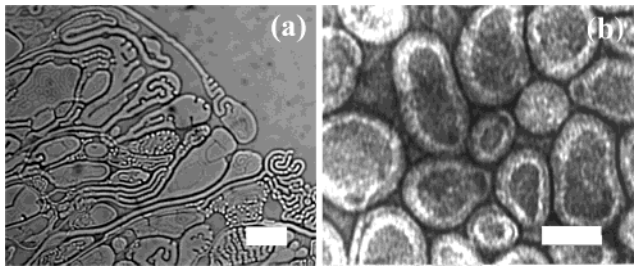


Figure 3. (a) Light micrograph of DOUPC supramolecular organizations upon hydration (bar = 30 μm, method A). (b) TEM image of small unilamellar vesicles obtained after extrusion of DOUPC through a 50 nm filter (bar = 50 nm, method B).

wide variety of polymeric and nonpolymeric hydrogelators have been described,[26−29] spontaneous gel formation from low molecular weight gelators, including phosphocholines, is less known. As shown in Figure 4a, DPUPC (**5b**) forms stable opaque hydrogels in aqueous solution at temperatures below the T_m. An amphiphile concentration higher than 6% w/w, confirmed by thermogravimetric analysis (TGA) experiments, is required to form the hydrogel. Scanning electron microscopy (SEM) shows an entangled fiber network throughout the DPUPC hydrogel (Figure 4b,c). Microcavities are present in the hydrogel of 1−10 μm in size (Figure 4b). Importantly, conventional phospholipids (e.g., DPPC) do not form hydrogels in aqueous media, again emphasizing the unique role the nucleoside plays in the formation of these supramolecular structures. At higher magnification, TEM images show aligned nanofibers that are helical for DPUPC in water at room temperature (Figure 4e,f). As shown in Figure 4g, the fiber diameter is approximately 4.2 nm, with an apparent helical pitch of 7.0 nm. Under these experimental conditions at 20 °C, DPUPC is in the solid crystalline state and not in the "fluid" liquid crystal state where vesicle structures would assemble. Similar hydrogels are observed for DSUPC and DAUPC. Fibers are not observed with

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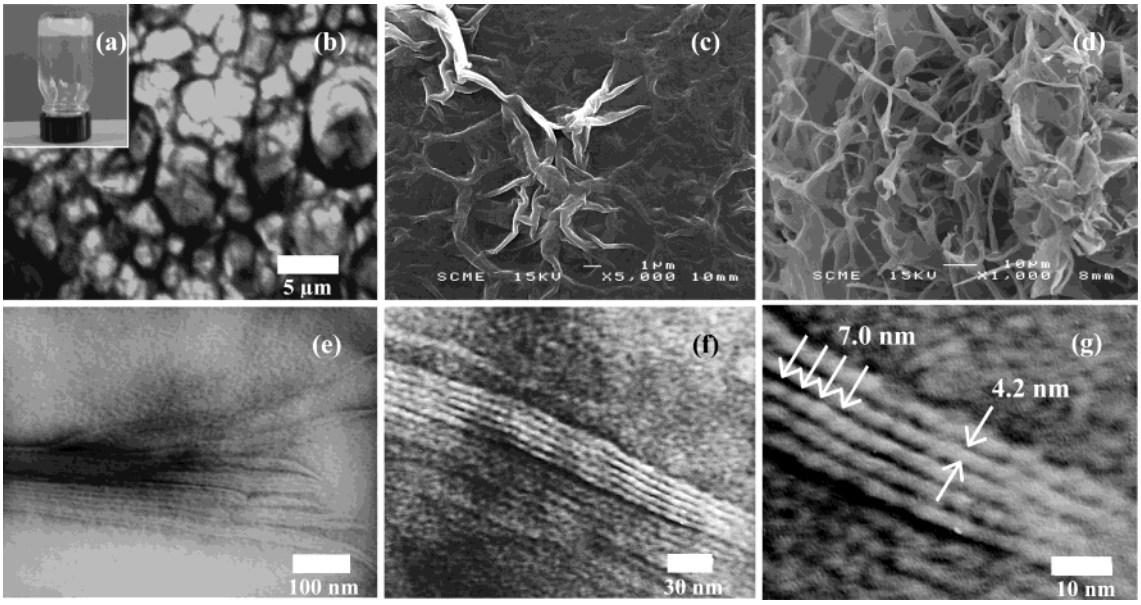

Figure 4. (a) Photograph of DPUPC hydrogel (6% w/w). (b) TEM image of DPUPC hydrogel at 6% w/w showing a microscopic network made of nanofibers. (c) SEM micrograph of the DPUPC hydrogel at 6% w/w. (d) SEM image of a freeze-dried DPUPC hydrogel. (e−g) High-resolution TEM images of DNA-like nanofibers obtained from DPUPC hydrogel at 6% w/w: (e) micrograph showing that the gel network is composed of nanofibers; (f) nanofibers aligned parallel; (g) arrows point to a fiber diameter of 4.2 ± 0.5 nm with an apparent helical pitch of 7.0 ± 0.5 nm.

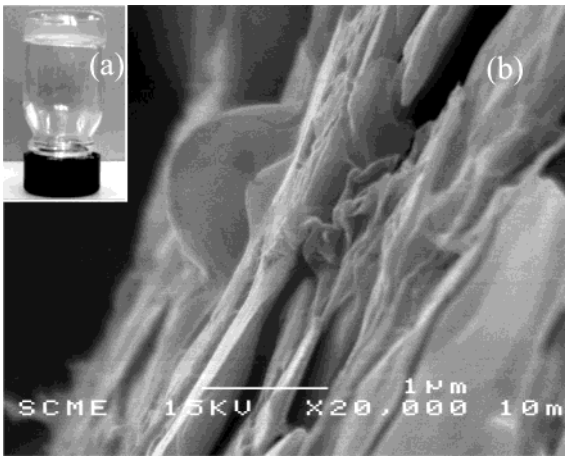

Figure 5. (a) Photograph of organogel. (b) SEM of DPUPC organogel.

DMUPC, **5a**, and DOUPC, **5e**, at room temperature since the "fluid" liquid crystal state is present at these temperatures (i.e., T_m < 25 °C). In a related but structurally different system, a helical structure was reported for an anionic phospholipid−nucleoside conjugate. The resulting supramolecular assemblies were attributed to the formation of larger bilayer helical ribbons of 10−150 nm in diameter.[30−32] However, the hydrogel structure created by these uridine amphiphiles, **5b**−**e**, are a consequence of the entangled network formed by the smaller diameter nonlamellar DNA-like fibers.

These uridine amphiphiles also form gels in nonaqueous solution (Figure 5). Specifically, DMUPC and DPUPC form clear organogels in cyclohexane. The longer chain derivatives (DSUPC, DAUPC, and DOUPC) did not form organogels, and this is a likely consequence of their increased solubility in cyclohexane compared to DMUPC and DPUPC. Simple glycerol based phospholipids (e.g., DMPC) do not self-assemble into gels in cyclohexane; however, organogels are formed in cyclohexane/water mixtures.[33] No water is needed to stabilize the organogels with these nucleoside phosphocholine amphiphiles. Organogels are observed at concentrations similar to those for the hydrogels and are prepared only above the T_m where increased chain fluidity exists. For example, DPUPC forms organogels in cyclohexane at low concentrations (6% w/w) and at temperatures above 42 °C. A SEM image shows the formation of a multilamellar system that stabilizes the organogel (Figure 5).

X-ray Diffraction Studies. To better understand the molecular organization within the supramolecular structures—fibers vs bilayers—formed by these amphiphiles, small-angle X-ray scattering (SAXS) experiments were performed below and above the T_m of the amphiphiles. For bilayers in aqueous solvent, the repeat distance of the amphiphile multilayer d is related to the membrane thickness d_1 and solvent thickness in the intermembrane space d_w via the expression $d = d_w + d_1$ (Figure 6). Data collected from several SAXS experiments above the T_m show that the nucleoside phosphocholine amphiphiles arrange into compact lamellar phases. For instance, aqueous solution of DPUPC in the "fluid" liquid crystal state at 50 °C (L_α, 20% w/w in water; DPUPC T_m = 41.8 °C) possesses a lamellar repeat period of 4.6 ± 0.2 nm. This distance is almost 2 nm shorter than that for the natural homologue DPPC.[34] Likewise, measurements of DOUPC above the T_m at 20 °C (L_α; 20% w/w in water) give a repeat period of 4.5 ± 0.2 nm, which again is 2 nm smaller than the value reported for natural DOPC (6.5 nm) in the L_α state.[35] The smaller repeat period and thus more compact bilayer can be explained by either a decrease in the

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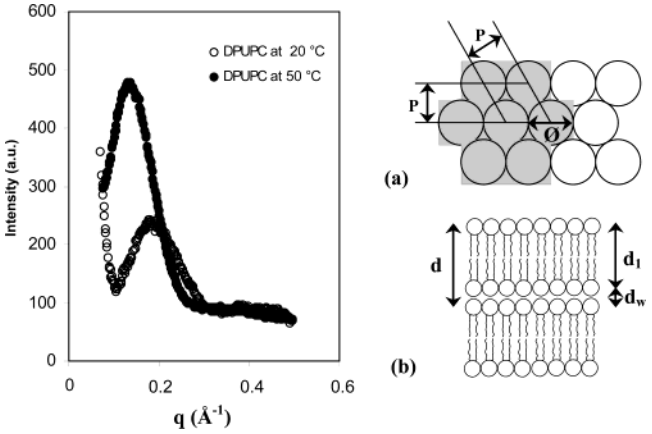

Figure 6. Left: SAXS patterns of DPUPC below (20 °C) and above T_m (50 °C). This plot shows two Bragg peaks at $q = 0.187$ and 0.137 Å^{-1} corresponding to the experiments at 20 and 50 °C, respectively. For the data at 20 °C, $p = 2\pi/q = 3.4 \pm 0.2$ nm, and for the data at 50 °C, $d = 2\pi/q = 4.6 \pm 0.2$ nm. Right: schematic views of the (a) compact hexagonal phase due to strand aggregation and fiber formation and (b) multilamellar arrangement. At 20 °C, the repeat period measured is p spacing $= 3.4 \pm 0.2$ nm, which correspond to a diameter $\varnothing = 2p/\sqrt{3} = 3.9 \pm 0.2$ nm ($\varnothing = 4.2 \pm 0.5$ nm was measured on high-resolution TEM images as shown in Figure 4g). At 50 °C, a lamellar repeat distance, d, is obtained of 4.6 ± 0.2 nm.

intermembrane space d_w due to nucleoside interactions or interpenetration of lipophilic chains resulting in low values for the total membrane thickness d.

In addition to the compact bilayers observed in the L_α state, the nucleoside phosphocholine amphiphiles exhibit small repeat periods below T_m. At 20 °C the lipid chains of DPUPC are in a gel state. At this temperature and a concentration of 6% w/w, DPUPC forms a fiber network and stable hydrogels are observed by TEM and SEM (Figure 4). The repeat period measured by SAXS (p spacing $= 3.4 \pm 0.2$ nm, Figure 6a) indicates the presence of strongly aggregated assemblies.

This result was confirmed by high-resolution TEM images, which show nanofibers of a diameter of about 4.2 ± 0.5 nm and an apparent helical pitch of 7.0 ± 0.5 nm (Figure 4). A cross section of the fiber illustrating the basic repeat unit as seen by SAXS is shown in Figure 6a. It is noteworthy that helical fiber formation is observed below the melting temperature and does not depend on the amphiphile concentration as evidenced by TEM imaging. Above a threshold concentration, these fibers form a network yielding the stable hydrogel.

Small-angle X-ray scattering measurements performed on the DPUPC organogel in cyclohexane confirmed the presence of repeat periods of 4.6 ± 0.2 nm at 50 °C. This repeat period is consistent with the hydrophobic chains oriented toward the organic solvent with hydrophilic headgroups in close contact. This supramolecular assembly arises from the chain−chain hydrophobic interactions, π-stacking forces, and hydrogen-bonding interactions between the uridines. In nonaqeous solvents the U−U enthalpy for self-association is estimated to be −3.6 kcal/mol, and such energies are sufficient to create supramolecular structures.[36] The infrared spectrum of the organogel shows a carbonyl stretch at 1712 cm^{-1} (C_2=O associated stretching vibration), confirming U−U hydrogen bonding in the DMUPC supramolecular assembly.

UV−Vis and Circular Dichroism (CD) Spectroscopies. UV−vis and CD spectroscopies were used to study the optical

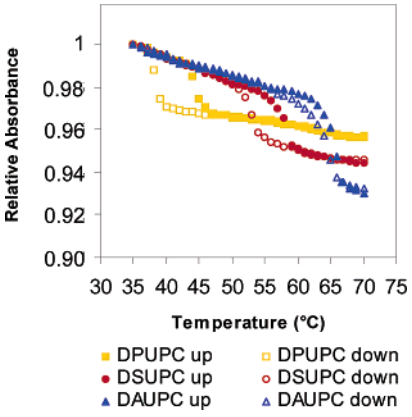

Figure 7. Relative absorbance for saturated nucleoside phosphocholine amphiphiles at 260 nm versus temperature. "Up" and "down" refer to the heating and cooling traces, respectively.

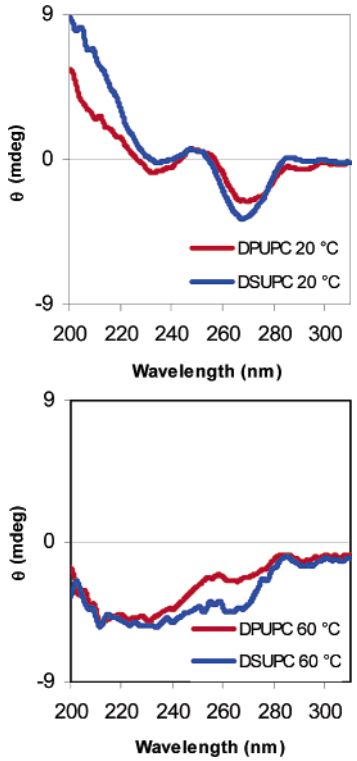

Figure 8. CD spectra of DPUPC (red) and DSUPC (blue) at (a, top) 20 °C below T_m and (b, bottom) at 60 °C above T_m.

properties of the supramolecular assemblies in aqueous media. To determine if the nucleoside headgroup of this amphiphile was affected during the transition from the solid to the liquid crystalline state, the absorbance at 260 nm of the uridine chromophore was monitored as a function of temperature. As shown in Figure 7, a hypochromic effect was observed above the phase transition temperature for the amphiphiles. This result indicates a different organization of the nucleoside in the fiber and bilayer states.

The high-resolution transmission electron micrograph (Figure 4g) shows a helical pitch present in the fibers, and thus CD was next used to investigate these supramolecular assemblies. The CD spectra of DPUPC and DSUPC fibers (1×10^{-4} M) below the T_m and the gelation concentration, but above the

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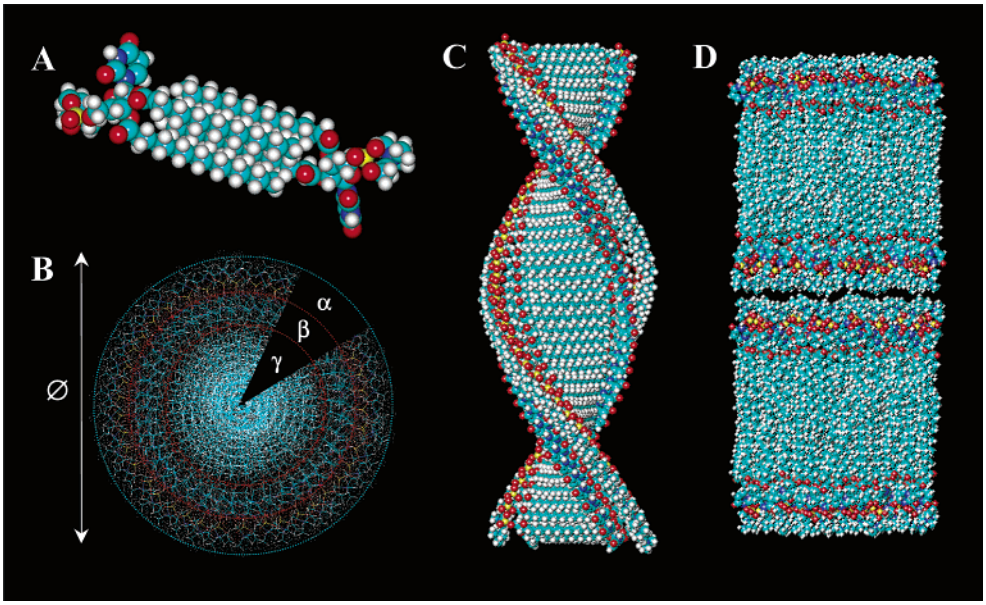

Figure 9. Proposed model illustrating DNA-like helical fiber and multilamellar organizations in water formed by DPUPC. (A) Two molecules of DPUPC forming the basic repeat unit of the helical structure. (B) Top view of the strand presented in (C) with a diameter ∅ of 4.2 ± 0.5 nm: α is hydrophilic domain (phosphocholines), β corresponds to uridine packing, and γ is the hydrophobic core. (C) Drawing of the fiber (for helical pitch = 7.0 ± 0.5 nm). (D) Drawing of a multilamellar, L_α, organization.

critical aggregate concentration, are shown in Figure 8. The CD spectra of both DPUPC and DSUPC at 20 °C show a positive and a negative inflection at 250 and 270 nm, respectively. The spectra of both DPUPC and DSUPC in water are different than that of uridine, as expected. The CD spectrum of the fiber formed by the nucleoside amphiphile originates from the chiral fiber aggregates. The aggregates interact differently with left and right circularly polarized light, indicating that a chiral supramolecular structure has been formed. The CD profiles obtained are temperature dependent, indicating again different organizations below and above the phase transition temperature. Above the T_m, the CD signal disappears as the fibers rearrange to form lamellar structures. It is hypothesized that internucleobase stacking plays an important role in stabilizing the helical structure. To confirm the base-stacking interactions in the fiber assemblies, we measured the UV spectra of DPUPC fibers and uridine at 20 °C. The molar absorptivity, ϵ, at 260 nm for the DPUPC fibers is 7642 compared with that for uridine (ϵ = 9401), again supporting a different environment of the nucleobase in the fibers compared to the lamellar structure and a stacking arrangement of the pyrimidine bases.

Proposed Self-Assemblies in Water. Molecular models for the fibers and multilamellar assemblies formed with the uridine phosphocholines are proposed in Figure 9. The structures were created using Hyperchem and are based on the above experimental data. For the fibers, a left-handed helix organization arising from amphiphile self-assembly is proposed below the T_m. This helical arrangement is governed by uridine base stacking and hydrophobic interactions. In Figure 9B,C, the α domain represents the hydrophilic moieties (phosphocholines), the β section corresponds to the π stacking of the uridines, and γ is the hydrophobic core with a maximum chain−chain association to minimize water contact (i.e., helical structure). The nucleoside phosphocholine amphiphiles assemble into lamellar structures above the T_m, and Figure 9D shows two compact bilayers. This model has a small intermembrane space and interpenetration of lipophilic chains leading to a low *d* value for the total membrane thickness, consistent with data collected from the SAXS experiments.

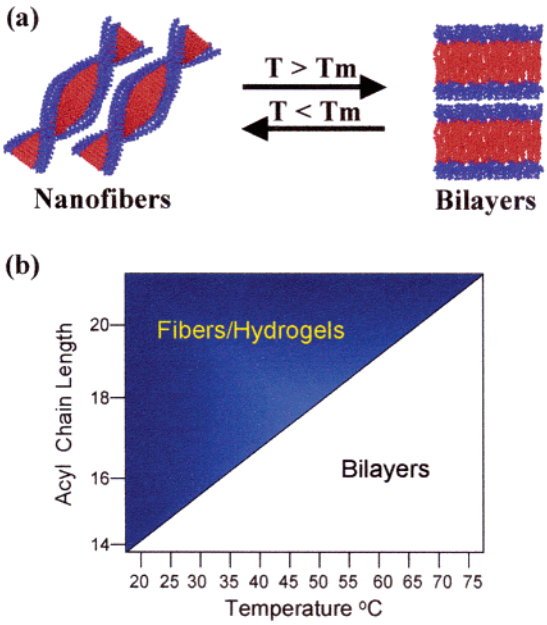

Figure 10. (a) Illustration of the thermoreversible helical−bilayer transition and (b) phase diagram at 1 atm for the transformation between fibers and bilayers.

By changing the temperature, the nucleoside amphiphiles form fibers or bilayers. This transformation between the two supramolecular structures is fully reversible (Figure 10). For example, heating the DPUPC hydrogel dissolves the fiber network/hydrogel and vesicles begin to form in solution. The changes can be observed visually. Figure 10 illustrates this thermoreversible phenomenon between the two supramolecular assemblies and includes a graph of temperature vs nucleoside

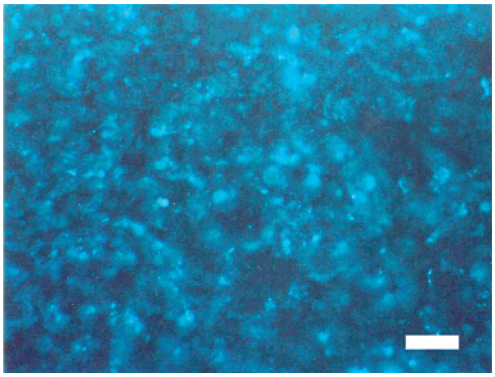

Figure 11. Fluorescence microscopy image of DSUPC hydrogel loaded with calf thymus DNA (bar = 8 μm).

chain length which depicts the regions where bilayers and fibers will be formed, respectively.

DNA Entrapment in the Hydrogels. The hydrogels created by the nucleoside amphiphiles are of potential use for the delivery of therapeutics. In fact, an amphiphilic hydrogel that acts as a temporary scaffold for delivery of DNA or RNA would be of interest, since current methods to delivery nucleic acids using synthetic vectors occur with poor efficiency. Given the composition of these amphiphiles, the propensity to have molecular recognition interactions, and the observation that some of the nucleoside phosphocholine hydrogels are stable at 37 °C, we determined if nucleic acid could be entrapped within the hydrogel. Given the constraint that the hydrogel must be stable at a physiological temperature, we selected DSUPC for these studies. DSUPC forms a hydrogel in water below its T_m (T_m 53.0 °C) at 5% w/w. Two different hydrogel samples were prepared, one without DNA and one with DNA, and subsequently characterized at room temperature using electron and fluorescence microscopies. The DSUPC hydrogel sample (5% w/w) without DNA was similar to DPUPC, which is presented in Figure 4. The TEM and SEM images showed a fiber network with microcavities of 1−10 μm in size. The second hydrogel (DSUPC, 5% w/w) was prepared in the presence of linear calf thymus DNA (0.0125% w/w) and a DNA-binding fluorophore (Hoechst 33258, 0.00312% w/w). As shown by fluorescence microscopy, condensed particles of DNA were entrapped in the hydrogel network (Figure 11). The DNA particles are a few microns in size.

Conclusions

A synthetic route to uridine phosphocholine analogues of conventional glycerol phosphochlines is described which provides easy access to a wide range of nucleoside based phosphocholine amphiphiles. Physicochemical experiments demonstrate that these nucleoside amphiphiles possess unique properties compared to their conventional glycerol analogues. Light and electronic microscopic (TEM, SEM) studies indicate that in aqueous solutions these nucleoside phosphocholine amphiphiles self-assemble into liposome-like bilayer structures for the "fluid" phase above T_m and DNA-like helical fibers in the crystalline solid state below T_m. Differential scanning calorimetry experiments show similar melting temperatures for the uridine phosphocholine amphiphiles as for the natural glycerol or ribose based phosphocholines, but with substantially smaller enthalpies. X-ray diffraction studies on DOUPC and DPUPC samples at temperatures above the melting temperatures reveal the presence of lamellar phases roughly 2 nm more compact than the natural analogues. High-resolution TEM images show fibers with a diameter of 4.2 nm with an apparent helical pitch of 7.0 nm, consistent with the SAXS results. When the concentration of fibers is increased, a hydrogel forms with micron-sized cavities. UV and CD spectroscopic experiments suggest $\pi-\pi$ stacking and a helical organization in the fibers. Moreover, these materials are thermoresponsive, and fibers or bilayers can be formed by simply changing the temperature. Of the nucleoside phosphocholine amphiphiles investigated, DPUPC exhibits ambidextrous gelation and forms gels in water and cyclohexane. This property is a consequence of different molecular organizations: in water below the T_m, the hydrogel is created by a network of fibers, whereas in cyclohexane a continuous multilamellar phase stabilizes the organogel above T_m. The hydrogels are stable, and DNA can be entrapped with these structures. The results with these new amphiphiles provide further incentive to synthesize, characterize, and evaluate new small molecules that assemble into functional materials. Such materials are likely to be of interest for medical and tissue engineering applications. In summary, the supramolecular assemblies prepared in this study illustrate the importance of molecular structural complexity on amphiphile self-assembly.

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Supporting Information Available: Complete experimental details: ^{1}H, ^{13}C, and ^{31}P NMR, FAB-MS, MDSC, TGA (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

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