594 *Chem. Mater.* **2006,** *18,* 594−597

New Role for Urea as a Surfactant Headgroup Promoting Self-Assembly in Water

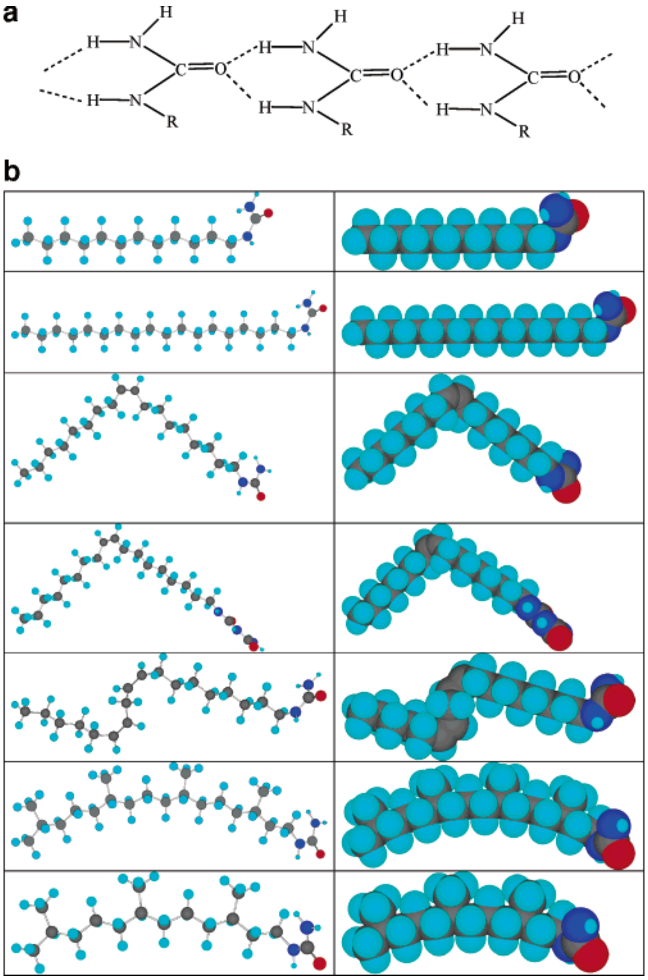
Celesta Fong,† Darrell Wells,† Irena Krodkiewska,†
Patrick G. Hartley,† and Calum J. Drummond*,‡

*CSIRO Molecular and Health Technologies, Private Bag 10,
Clayton, Victoria 3169, Australia, and CSIRO Molecular and
Health Technologies, P.O. Box 184, North Ryde,
New South Wales 1670, Australia*

Received October 13, 2005
Revised Manuscript Received December 15, 2005

Urea was the first organic molecule synthesized.[1] Since this historic event in 1828, the urea molecular unit has proven to be extremely versatile. Urea is used industrially as a fertilizer.[2] Urea inclusion compounds are used as host−guest materials to selectively separate molecular mixtures.[3] Polymers featuring urea functionality are employed as resins, microencapsulation media, biomaterials, and nonlinear optical materials (inter alia).[2,4] The urea moiety is frequently employed in solid-state molecular organic crystal engineering and in the construction of supramolecular assemblies.[5−6] Here, we present urea in a new role. We report the discovery that the strong intermolecular hydrogen bonding of urea can be ameliorated, through select structural modifications to hydrocarbon chain substituents, resulting in surfactant self-assembly and the formation of a lyotropic liquid crystalline (inverse hexagonal) phase that is thermodynamically stable in excess water. The nanostructured bulk phase can be dispersed in water to form nanoparticles ("colloidosomes"), and both the bulk and dispersed phase are potential new controlled/sustained release and structure-templating materials.

Chart 1. Urea Surfactant Structures[a]



[a] (a) Schematic depiction of the substituted urea intermolecular hydrogen-bonding network and (b) structural representations of the urea-based surfactants showing both ball and stick and space filling models of DDU, ODU, OU, OBU, LU, PU, and HFU, respectively. Parts of the Space filling models are "invisible" as a result of the zig-zag configuration of the chains in the space filling representations.

Substituted ureas have been reported to form linear hydrogen-bonding networks via the carbonyl group of one urea and the anti-hydrogen atoms of the nitrogen atoms of a neighboring urea in the solid state (bifurcated hydrogen bonding; shown in Chart 1a).[7] The thermotropic behavior of *n*-alkyl substituted ureas and, in particular, the crystal to isotropic liquid-phase transition (melting point) is governed by this very strong intermolecular hydrogen-bonding interaction. The melting point of *n*-alkyl ureas is essentially independent of the alkyl chain length from methyl (C1) to docosanyl (C22) because the van der Waals chain−chain interaction is dominated by urea hydrogen-bonding interaction.[7−10] Urea has a very high water solubility (e.g., >20 mol dm^{-3} at 25 °C), but the strong preference, in the medium and long hydrocarbon chain substituted ureas that have been studied up until now, for homo urea hydrogen bonding rather than hetero urea−water interactions has meant that there have been no previous reports of urea surfactants that self-assemble in water. The effective hydrophilicity of the

---

* To whom correspondence should be addressed: e-mail, calum.drummond@csiro.au.

† CSIRO Molecular and Health Technologies, Clayton, Victoria.

‡ CSIRO Molecular and Health Technologies, North Ryde, New South Wales.

(1) Chattaway, F. D. Isolation and synthesis of urea. *Chem. News J. Ind. Sci.* **1909**, *99*, 121.

(2) Mavrovic, I.; Shirley, A. R., Jr.; Coleman, G. R. Urea. *Kirk-Othmer encyclopedia of chemical technology*; John-Wiley & Sons: New York, 1998.

(3) Harris, K. D. M. Meldola lecture: understanding the properties of urea and thiourea inclusion compounds. *Chem. Soc. Rev.* **1997**, *26*, 279−289.

(4) Henningsen, J. Polyurea coating − technology and uses. *Mater. Perform.* **2001**, *40*, 34−36.

(5) Coe, S.; Et al. Molecular Symmetry and the design of molecular solids: the oxalamide functionality as a persistent hydrogen bonding unit. *J. Am. Chem. Soc.* **1997**, *119*, 86−93.

(6) MacDonald, J. C.; Whitesides, G. M. Solid-state structures of hydrogen-bonded tapes based on cyclic secondary diamides. *Chem. Rev.* **1994**, *94*, 2383−2420.

(7) Hashimoto, M.; Tajima, F.; Eda, K.; Yamamura, K.; Okazaki, T. Exploration of plastic-like phases in alkylurea systems. A thermal and X-ray study. *J. Mol. Struct.* **2005**, *734*, 23−33.

(8) DeBeer, E. J.; Buck, J. S.; Hjort, A. M. The relative anesthetic effects of some aliphatic ureas. *J. Pharmacol.* **1934**, *52*, 216−222.

(9) Buck, J. S.; Hjort, A. M.; Ide, W. S.; DeBeer, E. J. 1-***n***-Alkyl-5,5-ethyl-isobutyl barbituric acids. *J. Am. Chem. Soc.* **1938**, *60*, 461−462.

(10) Wells, D.; Drummond, C. J. Nonionic *n*-hexyl, *n*-heptyl and *n*-octyl urea surfactants: some physicochemical properties. *Langmuir* **1999**, *15*, 4713−4721.

10.1021/cm0522681 CCC: $33.50

Table 1. Thermotropic Transitions for the Neat Urea Surfactants Determined by DSC and the Temperature Regions where Lyotropic Phases Were Observed by Optical Microscopy.

surfactant[a]	melting point (T_{onset}, T_{max}; °C) [DSC transition (kJ/mol)]	window of phase existence (°C)	
		H_{II}	L_{II}
DDU	95, 103 [−42.0]		
ODU	101, 105 [−38.5]		
OU	79, 81 [−26.6]	61 to 110	64 to >95
OBU	99, 105 [−26.9]	85 to 107	87 to >107
LU	72, 79 [−29.0]	57 to 92	59 to >99
PU	62, 64 [−4.9][b]	<rt to 93	26 to >99
HFU	61, 62 [−5.8][b]	<rt to 63	<rt to >99

[a] IUPAC nomenclature for the surfactants is 1-dodecyl urea, 1-octadecyl urea, *cis*-octadec-9-enyl urea, *cis*-octadec-9-enyl biuret, *cis,cis*-octadec-9,12-dienyl urea, 3,7,11,15-tetramethyl-hexadecyl urea, and 3,7,11-trimethyl-dodecyl urea, respectively. [b] Liquid crystal to isotropic liquid-phase transition.

headgroup is a key determinant of nonionic surfactant phase behavior in water.[11−12]

In this work, three strategies were employed in an attempt to ameliorate the strong preference for solid-state crystallinity imposed by the intermolecular urea hydrogen bonding and thereby generate lyotropic liquid crystalline formation in water. The strategies were targeted at expanding the lateral packing and disrupting the order of the hydrocarbon chain region. The first strategy was to introduce positional isomerism, that is, to move the urea headgroup away from the end of the alkyl chain. For other surfactant molecules this leads to a lowering of the solid-state melting point and the Krafft temperature in water, for example, positional isomers of dodecyl β-D-glucosides.[13] In the case of urea surfactants, positional isomerization was found to increase the melting point, and no lyotropic liquid crystalline phases were observed. This is interesting in its own right and will be discussed elsewhere.[14] The second and third strategies were to introduce unsaturation and isoprenoid branching into the alkyl chain, respectively. Both of these strategies resulted in the formation of an inverse hexagonal (H_{II}) lyotropic liquid crystalline phase that is stable in excess water.

Five H_{II} phase forming urea-based surfactants, namely, oleylurea (OU), linoleylurea (LU), oleylbiuret (OBU), phytanylurea (PU), and hexahydrofarnesylurea (HFU) are reported. Structural representations of these surfactants are given in Chart 1b. Differential scanning calorimetry (DSC) and cross-polarized optical microscopy were used to determine the phase transitions of the neat surfactants in the temperature range from 0 to 120 °C. The phase transitions are provided in Table 1. Dodecylurea (DDU) and octadecylurea (ODU) are also included for comparison.

The urea surfactants have very low water solubility. The maximum solubility of the urea surfactants is less than 0.003 wt % (the minimum amount of surfactant observable by our experimental techniques) in the temperature range from 0 to 100 °C.

Water penetration experiments as a function of temperature, with cross-polarized optical microscopy, were performed to determine whether lyotropic liquid crystalline phases were formed. There was no surfactant−water phase formation, as expected, for DDU and ODU. Water penetration experiments for each of the other neat surfactants exhibited similar phase formation. HFU can be employed to exemplify what happens. A hexagonal phase developed immediately at the interface on contact with water at ambient temperature (24 °C). It was assigned as an inverse phase structure on the basis of the very low solubility of HFU in water. Further, its formation at the surfactant−water interface demonstrates stability of the H_{II} phase in excess water. Formation of this anisotropic phase was initially slow, and the characteristic fanlike texture for a hexagonal phase became evident on standing overnight as the H_{II} phase broadened into the neat surfactant phase. A mobile isotropic L_{II} phase also developed between the H_{II} phase and the neat phase. The isotropic phase was assigned as an inverse micellar L_{II} phase because of the compositional proximity to the H_{II} phase and the low viscosity (bubbles of air trapped within the phase were mobile and spherical in geometry). The development of the phases at this stage is shown in Figure 1a. It appears that the viscosity of the H_{II} phase and the transport route through a hydrocarbon matrix inhibit the diffusion of water as the development of this phase into the isotropic L_{II} band was slow even at higher temperatures. The H_{II} phase melted to an isotropic phase at 62 °C but readily reformed on cooling.

The phase progression for the other urea surfactants was similar, but with the unsaturated chain surfactants, a temperature above room temperature needed to be attained before H_{II} or L_{II} phase formation was observed. Table 1 summarizes the phase transition temperatures determined from the water penetration scans.

Small-angle X-ray scattering (SAXS) was performed on HFU−water and PU−water samples to provide additional confirmation that the anisotropic lyotropic liquid crystalline phase was a hexagonal phase. The SAXS patterns are shown in Figure 1b and display peaks consistent with the 1:$\sqrt{3}$:$\sqrt{4}$ reflections of the hexagonal phase. The reflection at (100) dominates the scattering patterns and corresponds to an observed *d* spacing of 29.3 Å for the HFU system and 39.4 Å for the PU system. These *d* spacings equate to a hexagonal lattice parameter and distances between water channel radii of 34 Å for the HFU system and 45.5 Å for the PU system. Why the reflection at (100) is considerably stronger than the other reflections is not known, but we note that this has also been observed in other inverse hexagonal systems.[15]

---

(11) Seddon, J. M. Structure of the inverted hexagonal (H_{II}) phase, and nonlamellar phase transitions of lipids. *Biochim. Biophys. Acta* **1990**, *1031*, 1−69.

(12) Laughlin, R. G. *The aqueous phase behaviour of surfactants*; Academic Press: London, 1996; Chapter 10.

(13) Boyd, B. J.; Drummond, C. J.; Krodkiewska, I.; Weerawardena, A.; Furlong, D. N.; Grieser, F. Alkyl chain positional isomers of dodecyl β-D glucoside: thermotropic and lyotropic phase behaviour and detergency. *Langmuir* **2001**, *17*, 6100−6107.

(14) Wells, D.; Fong, C.; Drummond, C. J. Nonionic urea surfactants: Influence of hydrocarbon chain length and positional isomerism on the thermotropic and lyotropic phase behaviour. *J. Phys. Chem. B*, in press.

(15) Fong, C.; Krodkiewska, I.; Wells, D.; Boyd, B. J.; Booth, J.; Bhargava, S.; McDowall, A.; Hartley, P. G. Submicron dispersions of hexosomes based on novel glycerate surfactants. *Aust. J. Chem.* **2005**, *58*, 683−687.

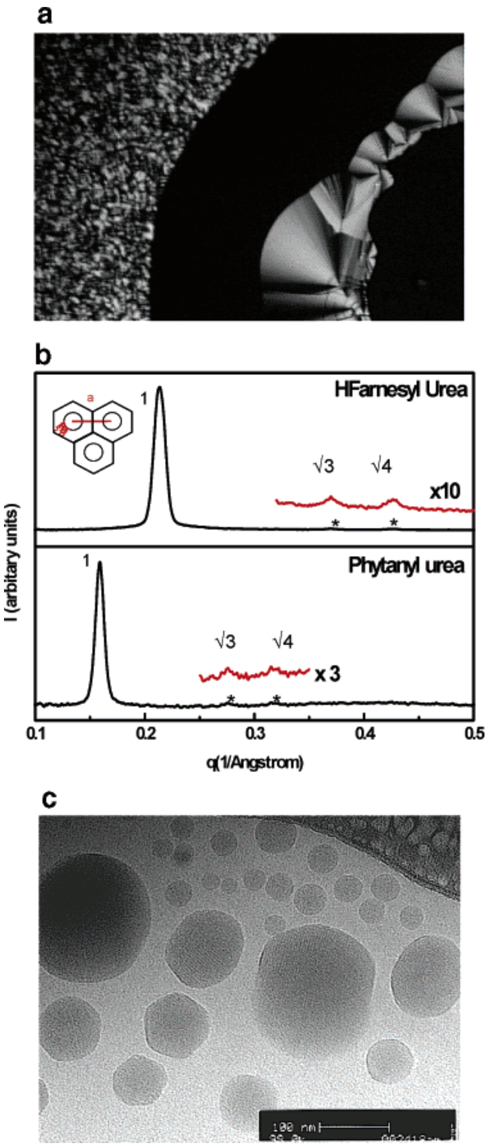


Figure 1. Inverse hexagonal phase formation. (a) Optical microscope image of a water penetration scan for HFU held at 24 °C. Observation under cross-polarized light at 150× magnification allowed identification of the phases formed by the visible birefringent texture, or lack thereof. The water penetrates from right to left with the phase progression anistropic H_{II}, isotropic L_{II}, and then anistropic smectic liquid crystalline neat surfactant. (b) SAXS pattern for a HFU−water system and PU−water system, where the analyzed lyotropic liquid crystalline phase had been equilibrated in excess water. For the HFU−water system scattering peaks are located at 29.3 (s), 17.0 (w), and 14.7 (w) Å and are indexed as 100, 110, and 200, respectively; a = 34 Å. For the PU−water system scattering peaks are located at 39.4 (s), 22.8 (w), and 19.9 (w) Å and are indexed as 100, 110, and 200, respectively; a = 45.5 Å. (c) Cryo-transmission electron microscopy image of the colloidosomes formed upon dispersion of the bulk H_{II} phase of HFU. The particles are sterically stabilized with Poloxomer 407. The scale bar is 100 nm.

The evidence, including SAXS, supports the conclusion that the thermodynamically stable lyotropic liquid crystalline phase formed by the urea surfactants in excess water consists of a hexagonally close-packed arrangement of parallel cylindrical self-assembled aggregates. Hydrated headgroups are situated in the core of the aggregates and are arranged within a continuous nonpolar matrix composed of hydrocarbon chains. The honeycomb structure of the H_{II} phase has a long-range order which is two-dimensional.

Formation of the H_{II} phase in water has been reported previously for a number of lipids possessing two hydrocarbon chains, especially those with phosphatidylethanolamine hydrogen-bonding headgroups.[11] The formation of a H_{II} phase from a single chain surfactant is relatively rare. Glyceryl monolein, unsaturated chain homologues, and phytantriol are examples that exhibit the H_{II} phase at temperatures higher than room temperature.[16−18] We know of only two other studies[15,19] of single chain surfactants, namely, 1-glycerol phytanyl ether,[19] oleyl glycerate,[15] and phytanyl glycerate,[15] where an inverse hexagonal phase has been observed at room temperature and below. Interestingly, possession of a headgroup with the ability to hydrogen bond is a common feature of all these H_{II} forming amphiphiles.

For the reported urea surfactants, the H_{II} phase is formed in preference to other lyotropic liquid crystalline phases because of the interplay between local and global constraints. The local constraint is primarily a function of the interfacial curvature, which is governed by the effective shape of the urea surfactants, with the combination of the small interfacial area occupied by the urea headgroup (<0.28 nm^2)[10,20−21] and either the "kink(s)" in an unsaturated chain or the "banana shape" induced by an isoprenoid repeat structure resulting in an inverse wedge-shaped molecular geometry. This results in the formation of cylindrical aggregates of urea surfactants. In terms of the critical packing parameter (CPP) model popularized by Israelachvili et al., CPP > 1 (CPP = v/a_0l_c, where v is the volume of the hydrocarbon chain, a_0 is the area of the headgroup, and l_c is the effective chain length).[22] The global constraint is dictated by the volume fraction of urea surfactant in the surfactant−water mixtures, and depending on surfactant concentration we either observe an isotropic solution of inverse cylindrical micelles (L_{II}) or hexagonal packing of these micelles (H_{II}).

For many years, thermodynamically stable lamellar phases in excess water have been dispersed to form liposomes.[23] More recently, thermodynamically stable bicontinuous cubic and inverse hexagonal phases in excess water have been dispersed to form nanostructured colloidal particles, "colloidosomes", named cubosomes and hexosomes.[24−25] As shown in Figure 1c, the H_{II} phase of the urea surfactants can also be dispersed to form colloidosomes. Discrete hexagonally shaped colloidosomes are observed alongside more spherical objects. We believe that the spherical objects

---

(16) Hyde, S. T.; Andersson, S.; Ericsson, B.; Larsson, K. A cubic structure consisting of a lipid bilayer forming an infinite periodic minimal surface of the gyroid type in the glycerolmonooleate water system. *Z. Kristallogr.* **1984**, *168*, 213-219.

(17) Misquitta, Y.; Cherezov, V.; Havas, F.; Patterson, S.; Mohan, J. M.; Wells, A. J.; Hart, D. J.; Caffrey, M. Rational design of lipids for membrane protein crystallization. *J. Struct. Biol.* **2004**, *148*, 169−175.

(18) Barauskas, J.; Landh, T. Phase behaviour of the phytantriol/water system. *Langmuir* **2003**, *19*, 9562−9565.

(19) Hato, M.; Minamikawa, H.; Salkar, R. A.; Matsutani, S. Alkylglycosides with an isoprenoid-type hydrophobic chain can afford greater control of aqueous phase structures at low temperatures. *Langmuir* **2002**, *18*, 3425−3429.

(20) Glazer, J.; Alexander, A. E. The monolayer polymorphism of hexadecyl and octadecyl urea. *Trans. Faraday Soc.* **1951**, *47*, 401−409.

(21) Hunter, D. S.; Barnes, G. T.; Godfrey, J. S.; Grieser, F. Octadecylurea monolayers. *J. Colloid Interface Sci.* **1990**, *138*, 307−313.

(22) Israelachvili, J. N.; Miller, D. J.; Ninham, B. W. Theory of self-assembly of hydrocarbon amphiphiles into micelles and bilayers. *J. Chem. Soc., Faraday Trans. II* **1976**, *72*, 1525−1568.

(23) *Liposomes: a practical approach*, 2nd ed.; Torchilin, V., Weissig, V., Eds.; Oxford University Press: Oxford, 2003.

are a result of preparing the colloidosomes by melting the urea surfactant and then dispersing this material to make spherical emulsion droplets in water. These emulsion droplets are then cooled to form the liquid crystalline particles (see Supporting Information for details of the particle preparation). The nature of the cooling process appears to be such that some emulsion droplets "crystallize" as hexosomes and others "freeze" as nanostructured spherical particles. It should be noted that Johnsson et al.[26] have recently reported a heat treatment procedure to reproducibly obtain hexagon-shaped particles, hexosomes, for a dispersion of the H_{II} phase in a diglycerol monooleate/glycerol dioleate−water system. We expect this procedure to be transferable to the urea surfactants.

The new group of self-assembling urea surfactants possesses the simplest peptide containing headgroup that has been found to have sufficient hydrophilicity to result in lyotropic liquid crystalline phase formation. By analogy with other H_{II} phase forming surfactants, the self-assembling urea surfactants may ultimately find application as sustained release drug delivery vehicles, either as depots of the bulk phase or as colloidal particle formulations,[27] and nonviral gene delivery agents.[28] Structure templating in the generation of mesoporous materials and polymers is another potential use.[29]

Acknowledgment. The authors thank Ben J. Boyd for preparing the HFU dispersions, Alisdair McDowall for obtaining the cryo-transmission electron microscopy images, and James Booth and Suresh Bhargava for assistance with the SAXS measurements. This work was partially supported by an AusIndustry R&D Start grant and Soltec Pty, Ltd. (now MaynePharma). C.J.D. is the recipient of an Australian Research Council Federation Fellowship.

Supporting Information Available: Details of synthesis, characterization, and dispersion of the urea-based surfactants (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

CM0522681

---

(24) Gustafsson, J.; Ljusberg-Wahren, H.; Almgren, M.; Larsson, K. Cubic lipid-water phase dispersed into submicron particles. *Langmuir* **1996**, *12*, 4611−4613.

(25) Gustafsson, J.; Ljusberg-Wahren, H.; Almgren, M.; Larsson, K. Submicron particles of reversed lipid phases in water stabilized by a non-ionic amphiphilic polymer. *Langmuir* **1997**, *13*, 6964−6971.

(26) Johnsson, M.; Lam, Y.; Barauskas, J.; Tiberg, F. Aqueous phase behaviour and dispersed nanoparticles of diglycerol monooleate/glyceryl dioleate mixtures. *Langmuir* **2005**, *21*, 5159−5165.

(27) Drummond, C. J.; Fong, C. Surfactant self-assembly objects as novel drug delivery vehicles. *Curr. Opin. Colloid Interface Sci.* **2000**, *4*, 449−456.

(28) Koltover, I.; Salditt, T.; Radler, J. O.; Safinya, C. R. An inverted hexagonal phase of cationic liposome-DNA complexes related to DNA release and delivery. *Science* **1998**, *281*, 78−81.

(29) Schuth, F. Non-siliceous mesostructured and mesoporous materials. *Chem. Mater.* **2001**, *13*, 3184−3195.