Final Report

Grant Number: ER46323

Title of Award: **Electrostatically Self-assembled Amphiplexes** (PI: Helmut H. Strey)

Date: 3-May-2010

Period Covered: 31-August-2006 to 15-February-2010

Accomplishments

1. Design and construction of a combinatorial material science X-ray scattering beamline at NSLS

Our amphiplex system and also many other soft matter materials (e.g. personal care products, detergents, emulsions) consist of many different components. In order to optimize the structure and performance of these materials for specific applications the phase diagram has to be scanned as a function of composition. The difficulty with this approach is that, because of the enormity of the available phase space, often many thousand samples with varying compositions have to be investigated.

One possible solution to this problem is to scan through phase space using a combinatorial approach. This approach has been successfully employed in molecular biology, drug discovery and recently in material science. In collaboration with Elaine DiMasi from Brookhaven National Labs, we have developed a combinatorial material science X-ray scattering setup at the National Synchrotron Light source (NSLS) located at Brookhaven National Labs (BNL) for structure determination in soft matter systems. The development consists of a robotic liquid handling system commonly used in molecular biology using 96 or 384 well plates for the preparation of samples with varying composition.

We designed a motorized stage that can hold up to 3 well plates and that will allow us in an automated fashion collect X-ray scattering data of up to 1152 individual samples in one run. For samples that contain organic molecules that are incompatible with standard plastic well plates we developed 96-well plates made from Teflon whose top and bottom can be pressure sealed using mylar sheets.

Fig. 1: NSLS X-ray scattering setup to test our sample preparation procedures. Using a x-y translation stage X-ray scattering pictures were obtained for samples in different wells. Using this setup, we successfully determined phase diagrams of ESAs as a function of composition.

During the reporting period, we developed software that aids in the design of experiments to determine phase diagrams of a mixture of multiple liquid components. For that we chose to use “Python” as a programming language because it is open-source and will allow us to create an

open analysis platform for other X-ray scattering users. We wrote “Python” scripts that allow the user to enter the number of components, their physical parameters such as specific density and concentration, and the range over which each component should be varied in the phase diagram. The scripts are then designing the plate layouts taking into consideration the resulting sample thickness (to optimize scattering intensity), patterns that minimize the number of individual pipetting steps for multi-pipetters. In addition to the plate layout, the scripts also create XML files that contain the sample position, plate number and composition in each well. That allows to later automate X-ray scattering data analysis and construction of phase diagrams.

2. Membrane assembly

We have been studying a model system of charged lipid bilayers and oppositely charged polyelectrolytes that aggregate into membrane stacks. This system serves as a planar model for the more complicated polyelectrolyte-surfactant amphiplexes with mostly curved interfaces. The intermembrane distance in within the stacks is measured by X-ray scattering as a function of charge density of lipid bilayer and polyelectrolyte, polyelectrolyte molecular weight, and salt concentration. In addition, we are studying to what extent counterion release [1] is responsible for the polymer induced attraction using thermodynamic and electrochemical methods.

Fig. 2: Schematic picture and concept of oppositely charged polyelectrolyte induced bridging of two charged surfaces.

The simplest model system for the study of polyelectrolyte induced self-assembly is the assembly of charged lipid bilayers using oppositely charged polyelectrolytes. In this study we used different molar ratios of Dioleoyltrimethylammoniumpropane (DOTAP) and Dioleoylphosphatidylcholine (DOPC) complexed with different molecular weights of poly (acrylic acid) (PAA).

We investigated the effect of molecular weight of the polyelectrolyte and salt concentration (from 10mM to 500mM NaCl) on the polyelectrolyte layer thickness (this thickness is given by the lamellar repeat spacing minus the DOTAP:DOPC membrane thickness of 39Å [2]). As predicted by theoretical models [3-5], the lamellar spacing decreases as the membrane charge density increases. Interestingly, at charge densities lower than 50% and 500mM NaCl (see Fig.3) we observed a coexistence of two lamellar spacings, one larger and one smaller than at lower salt concentrations. Further studies have to be performed to fully understand this system.

Fig.3: Poly (acrylic acid) layer thickness in membrane stacks assembled from PAA and a 1:1 mixture of DOTAP and DOPC as function of Debye screening length. The thickness is calculated from the X-ray lamellar spacing minus the bilayer thickness of 39Å

Fig.4: Lamellar Repeat spacing of membrane stacks assembled from PAA and a mixture of DOTAP and DOPC as function of lipid surface charge density. At Low charge density, a coexistence of two lamellar phases is observed. The polyelectrolyte thickness can be calculated from the X-ray lamellar spacing minus the bilayer thickness of 39Å.

We investigated the effect of molecular weight of the polyelectrolyte and salt concentration (from 10mM to 500mM NaCl) on the polyelectrolyte layer thickness (this thickness is given by the lamellar repeat spacing minus the DOTAP:DOPC membrane thickness of 39Å). As predicted by theoretical models, the lamellar spacing decreases as the membrane charge density increases. Interestingly, at charge densities lower than 50% and 500mM NaCl we observed a coexistence of two lamellar spacings, one larger and one smaller than at lower salt concentrations. Recent theoretical calculations by Podgornik [6-8] can explain this phase coexistence by predicting two phases with different polyelectrolyte distributions as illustrated below.

Fig. 5: At low charge density we observe phase coexistence of two lamellar phases. Theoretical calculations by Podgornik predict two distinct phases with different polyelectrolyte density distributions.

3. Polyelectrolyte – oppositely charged surfactant – cosurfactant – oil complexes

As model system we are studying microemulsions formed from copolymers of poly (acrylic acid – acrylamide), cetyltrimethylammonium chloride, alcohols of different chain length (1-pentanol, 1-octanol), and dodecane (Fig.4). We have studied this system as function of composition (ratio between polyelectrolyte and surfactant is fixed by charge neutrality: for each surfactant charge there is one charged group on the polyelectrolyte chain), polyelectrolyte charge density, and salt concentration.

Fig. 4: Schematic representation of the molecules used for the quaternary complexes

Fig. 5 shows the phase diagrams of PAA, CTACl, Dodecane and Pentanol at 100mM, 300mM and 500mM NaCl. We find phases with lamellar, hexagonal and cubic structures and phase coexistence of those. Interestingly, we found long-range orderd cubic and hexagonal order at high oil content. To our knowledge, this is the first time that such high oil content was observed for hexagonal or cubic phases (usually long-range order is only found for oil contents below 20%). This finding is crucial for applications in which the oil phase is solidified by cross-linking to fabricate solid nanoporous membranes for use in filtration and possibly fuel cells.

Fig. 5: Phase diagram of pentanol and dodecane relative to the CTACl weight [wt %] content at 100 mM (upper left), 300mM (upper right) and 500mM (lower right) NaCl salt concentration.

In general, at a given pentanol concentration the phase sequence is dominated by lamellar – hexagonal – cubic with increasing dodecane concentration. Such a phase sequence is similar to what is observed in a diblock copolymer system when starting from a 50:50 diblock and increasing one monomer relative to the other.

We also observed a swelling of the unit cell of each phase as the dodecane concentration is increased. The unit cell parameter a , for the hexagonal phase increase from 68.3 °Å to 108.2 °Å, for the lamellar phase increase from 42.3 °Å to 54.5 °Å, while for the cubic phase increase from 142.6 °Å to 164.1 °Å.

Fig.6: Swelling of the hexagonal unit cell parameter a as function of dodecane (300mM NaCl, left) and pentanol (100mM NaCl, right)

Fig. 6 gives a more detailed account of that swelling. At a fixed pentanol concentration (Fig. 6, left) the phases swell until they reach a plateau. This plateau can be explained by the fact that pentanol is needed for swelling of the surfactant micelles to counteract the intrinsic spontaneous curvature. As the dodecane concentration is increased more pentanol will dissolve in dodecane and becomes unavailable to affect the surface properties. Further studies will be needed to confirm this hypothesis.

Fig. 7: Small angle X-ray diffraction spectrum of the ESA complex at 80 [wt %] Dodecane and 10 [wt %] Pentanol respect to the surfactant, (100 mM NaCl salt). A total of 11 Bragg peaks of the cubic $Pm3n$ phase were observed. The ratio of the reciprocal spacing with corresponding plot of s , ($s = 1/d_{hkl}$) versus $(h^2 + k^2 + l^2)^{1/2}$ is given.

Fig. 7 and Fig. 8 show an example of a $Pm3n$ and $Ia3d$ cubic phase. In some instances we observed 11 Bragg peaks of the $Pm3n$ phase whose peak positions all fit on a straight line to show that we indeed observe a cubic phase. Even with that many peaks it is still difficult to determine whether the $Pm3n$ phase is bicontinuous or not. In the literature there are two types of $Pm3n$ phases found in soft matter systems: an elongated micellar one and a fully connected bicontinuous phase. The task of reconstructing in detail the composition of the unit cell is made even more difficult by the fact that we don't even know exactly what components the unit cell contains. This is because our phases precipitate out of water and their water content is uncertain. In addition, there is the possibility that the polyelectrolyte and/or surfactant and/or the cosurfactant partitions into the water phase.

Fig. 8: SAXS pattern of cubic *Ia3d* phase at 100 mM NaCl salt for the binary PAA-CTACl complex.

3. Cross-linking of ESAs using a polymerizable oil

In recent years, tremendous progress has been made in the synthesis of solid nanoporous materials taking advantage of molecular self-assembly. The research on such materials is motivated by the promise as optical materials, catalyst support and filter membranes. Successful synthesis of mesoporous silica and block copolymer materials has resulted in materials and membranes of exquisite order and uniformity.

We are pursuing a slightly different strategy using ESAs. In this project, ESAs serve as templates for solid nanoporous structures with tunable pore size. Similar strategies have been reported in the literature resulting in solid lamellar structures. We believe that by our choice of monomer and templating system we will have access to a larger variety of structures that are reproduced with higher fidelity. In order to fabricate solid nanoporous materials we are forming ESAs in which the oil is replaced by a polymerizable monomer.

For our monomer we chose dicyclopentadiene (DCPD) with added 2.8% of 5-ethylidene 2-norbornene to make the monomer liquid at room temperature. The advantage of this monomer is that the bulk density of the polymerized material is identical to the bulk density before polymerization. This avoids the common problem of breakup of the material after polymerization that is caused by residual stresses resulting from shrinkage. Dicyclopentadiene is often used in the reaction injection molding manufacture of such large-scale parts as golf cart bodies, sports equipment, and for high-temperature chemically resistant pipes.

During this period, we have mapped out ESA phase diagram with dicyclopentadiene as the oil component in order to identify desirable structures (e.g. bicontinuous cubic phases) for separation materials. We have measured phase diagrams of CTACl, PAA, dicyclopentadiene with various cosurfactants (pentanol, octanol, Brij 52, Brij 56, Brij 76, Dimethyldioctadecylammonium chloride, SPAN 80) at 300mM NaCl to explore for hexagonal and cubic phases with high incorporation of monomer.

We then polymerized some of those samples using ring-opening metathesis polymerization (ROMP) using Second-generation Grubbs' catalysts at a 1:2500 ratio catalyst/DCPD. Second generation Grubbs' catalyst, compared to other metal-catalyst systems, is more tolerant to water and ions and has been successfully used in emulsion and microemulsion polymerization.

Fig. 5: Polymerized samples of various compositions placed in a 96-well plate for X-ray measurements.

Fig. 6: (upper) X-ray scattering data of CTACl, PAA, DCPD (% is wp with respect to CTACl) and 2% Brij 52; (lower) same samples as in the upper panel but polymerized.

Fig. 7: Lamellar samples of CTACl, PAA, DCPD and pentanol before and after polymerization. In general the unit cell shrinks slightly after polymerization.

We found that some of the phases (lamellar, hexagonal and cubic) phases can be polymerized. Some samples (mostly lamellar and hexagonal) polymerized into hard solid samples whereas other samples polymerized were soft and spongy. So far, we have not been able to obtain polymerized cubic samples. As seen in Fig. 6, all unpolymerized cubic phases change into the hexagonal phase during polymerization. As a general trend we observe a slight shrinkage of the unit cell after polymerization. This shrinkage is enhanced when the samples are washed in Water and/or Triton X-100 (removes surfactant/cosurfactant and PAA).

Papers

Michael L., Strey H.H., Measurement of Phase Transition Free Energies in Polyelectrolyte-Surfactant Complexes, *Macromolecules* 2010, 43,4379-4383.

Vensa Stanic, Mancuso M., Wong W., DiMasi E., Strey H.H., Phase Diagram of Electrostatically Self-assembled Amphiplexes, *Macromolecules* 2011,44(18),7423-7429.

People and support

Helmut H. Strey, Professor, 13% effort (3 years)
Vesna Stanic, Postdoc, 100% effort (3 years)
Theresa Cargioli, Graduate student, 100% effort (2 years)
Belle Lederer, Research assistant, 20% effort (1 year)
Matt Mancuso, Undergraduate student
Alex Bruno, Undergraduate student

Cost status

All funds are spent.