Experimental Report

11/01/2014

Proposal:	**9-10-1282**	**Council:**	10/2012
Title:	Extending the surface/bulk phase link from flexible synthetic polymer/surfactant systems to stiff biomacromolecule/surfactant systems		

This proposal is continuation of: 9-10-1191

Research Area: Soft condensed matter

Main proposer: **VARGA Imre**

Experimental Team: VARGA Imre
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Samples: hexadecyltrimethylammonium bromide
d33-hexadecyltrimethylammonium bromide
DNA
sodium bromide

Instrument	Req. Days	All. Days	From	To
FIGARO Adsorption troug	4	4	06/05/2013	10/05/2013

Abstract:

Polyelectrolyte/surfactant mixtures adopt a variety of structures at static air/liquid interfaces. Systems have been previously classified into type 1 involving strong complex adsorption and thick layers and type 2 forming thin layers and a surface tension peak. Recently we have shown on FIGARO that a surface tension peak characteristic for type 2 systems occurs after unlimited precipitation of surface-active materials both in systems characterized as type 1 and type 2. These results highlight the needs to collect data under well-defined bulk conditions for a range of strongly interacting systems and to examine the link between the interfacial properties and non-equilibrium effects in the bulk both for synthetic polymers and biomacromolecules such as rigid DNA or protein molecules. To re-evaluate the interactions governing the interfacial properties of the DNA/CTAB system, we propose to measure the surface excess and interfacial composition in the phase separation region with respect to two well-defined bulk states. This work will also contribute to the basis of a new generalized model to describe P/S systems at interfaces with respect to their bulk phase behavior.

Final Report for Figaro Experiment #9-10-1282

Scientific background. Polyelectrolyte/surfactant (P/S) mixtures adopt a variety of structures at static air/liquid interfaces, ranging from monolayers,[1] through trilayers[2] and pentalayers,[3] to multilayers.[4] In a review by Taylor et al., systems were classified into two sorts: *type 1* involving strong complex adsorption resulting in thick layers of several nanometers or more, and *type 2* forming thinner layers while tending to exhibit the surface tension peaks.[5] An example in the former category is sodium poly(styrene sulfonate)/dodecyltrimethyl-ammonium bromide (PSS/DTAB) which forms 60-Å trilayers, and an example in the latter is poly(diallyldimethylammonium chloride)/sodium dodecyl sulfate (Pdadmac/SDS) which forms compact layers and exhibits a sharp cliff edge peak in its surface tension isotherm. Recently, we have shown that in the case of flexible, synthetic polyelectrolytes the interfacial properties of both the *type 1* and *type 2* systems can be described unambiguously in terms of the colloid stability of the bulk P/S complexes, highlighting that a key parameter in the understanding of these system is their slowly evolving non-equilibrium nature. Our motivation for this experiment was to extend our recent results to the field of rigid biomacromolecule-complexes to shed light on how the interfacial properties of these systems are also governed by the bulk colloid stability of the these complexes.

Recent advances. Recently, we demonstrated in *J. Phys. Chem. Lett.* that the surface tension peak for the Pdadmac/SDS system (*type 2*) is produced only after the slow precipitation of most of the surface-active material out of the solution.[6] We then showed in a *J. Phys. Chem. B* paper resulting from work carried out on FIGARO (see report #9-12-1081), that under the same conditions as the peak forms, a sharp drop in the surfactant surface excess in the precipitation region (see fig. 1A) can quantitatively account for the feature (see fig. 1B).[7] This calculation with reference to pure surfactant data was based on the simple assumption that there is no polymer left in the liquid after the precipitation process has finished, and its excellent matching to the experimental data compounds the non-equilibrium nature of the system.

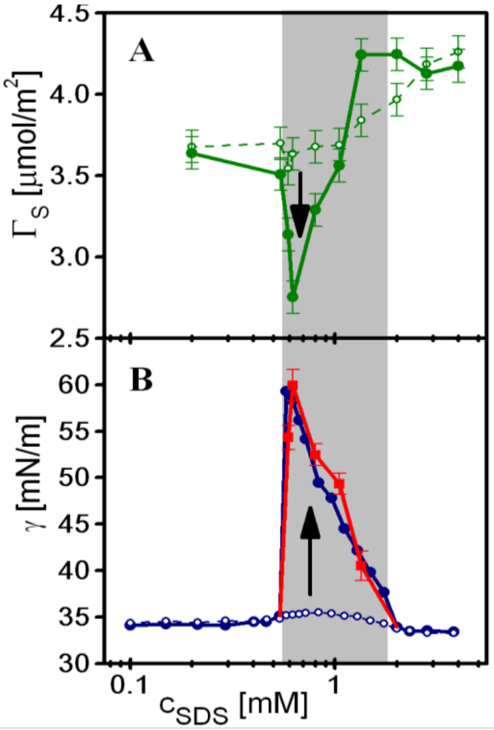


Figure 1. (A) NR measurements of the surface excess of SDS in Pdadmac/SDS mixtures, and (B) surface tension values measured (blue circles) and calculated from the NR data in panel A on the assumption that total removal of polymer from the liquid reduces the system to a depleted SDS solution in the precipitation region (red squares); fresh mixed (open symbols) & aged samples (closed symbols).

In a recent experiment on FIGARO (see report #9-10-1191), we went on to demonstrate that the same rules govern the behavior of the NaPSS/DTAB system (*type 1*): it can either exhibit a surface tension peak (*type 2* behavior) or only a surface tension plateau (*type 1* behavior) depending on the bulk conditions. These results clearly demonstrate the inadequate nature of the current equilibrium model for the interpretation of the interfacial properties of P/S systems. Our work shows that the surface tension peak is created only after samples are left to equilibrate for long enough time in each investigated system. Even though interfacial properties measured in these systems might reach steady state within minutes or hours, they likely represent a local air/liquid equilibrium corresponding to a bulk state far from equilibrium. Hence there is a need for data collected under well-defined bulk conditions in these systems. The widespread presence of P/S systems in biophysics, e.g., complexes involving rigid proteins or DNA molecules, shows that this topic merits urgent further work.

FIGARO experiment. We proposed to examine whether the interfacial properties of a rigid biomacromolecule (DNA) and oppositely charged surfactants (DTAB and CTAB) are governed by the same non-equilibrium principles like those of the flexible synthetic polyelectrolyte/oppositely charged surfactant systems. Like with our recent works on the Pdadmac/SDS and NaPSS/DTAB systems, we chose to gain such a perspective through the comparison of fresh and aged samples where the bulk aggregation states are extreme opposites; the latter being as close to equilibrium as can be achieved on practical timescales. Prior to the beam time application we had pre-characterized the bulk behavior of the DNA/DTAB and DNA/CTAB systems extensively using electrophoretic mobility and turbidity measurements with respect to the bulk composition, sample age and ionic strength. The bulk behavior of the systems closely resembles that of the flexible polyelectrolytes. Our question was: how does the interfacial composition change across the precipitation region in relation to the bulk phase behavior? An allocation of 4 days of beam time on FIGARO was awarded for us to find out.

Results & publication. The FIGARO experiment went according to plan with the measurement of fresh and aged DNA/DTAB and DNA/CTAB samples recorded in different isotopic contrasts. We chose to combine the DNA/DTAB results with those recorded previously on the synthetic P/S mixtures to write a Letter for urgent publication to Langmuir: "New method to predict the surface tension of complex synthetic and biological polyelectrolyte/surfactant mixtures".[8]

First we modeled the surface tension of aged Pdadmac/SDS and NaPSS/DTAB mixtures in the phase separation region where they form a depleted surfactant solution. The surface tension is determined by the surfactant chemical potential, which in this case is simply determined by the free surfactant concentration. NR data from FIGARO were important in the latter case to show that no polymer remained at the interface. The surface tension isotherm for the pure surfactant is converted to that of the aged mixture by transforming the concentration scale of the pure surfactant to that of the free surfactant in the mixture, after a correction of the bulk binding isotherm. Previous work on a related P/S mixture showed that complexes lost their colloidal stability (at the left edge of the phase separation region) with 60% surfactant binding, and only a few percent excess binding occurs after stoichiometric binding. Therefore we apply an empirical correction in the derivation of the free surfactant concentration where surfactant binding increases linearly from 60% on the left edge of the phase separation to 100% at and beyond the point of charge neutrality.

The input parameters are the surface tension of the pure surfactant (γ_{surf}), bulk polymer concentration in terms of monomer units (c_{poly}), bulk surfactant concentration at the left edge of the phase separation region (c_{left}), and bulk surfactant concentration at point of charge neutrality (c_{neut}). No surface measurements of the P/S mixtures are required. The surface tension of the mixture, γ_{mix}, may then be expressed as

$$\gamma_{mix}(c_{surf}) = \gamma_{surf}(c_{surf} - kc_{poly})$$

where $k = 0.6 + 0.4\frac{(c_{surf} - c_{left})}{(c_{neut} - c_{left})}$ for $c_{surf} < c_{neut}$ and $k = 1$ for $c_{surf} \geq c_{neut}$.

The complexity of the problem has thus been reduced to a single empirical equation. Fig. 2 shows the predicted surface tension of aged Pdadmac/SDS (blue) and NaPSS/DTAB mixtures using the model (red), and the agreement with the experimental data is excellent.

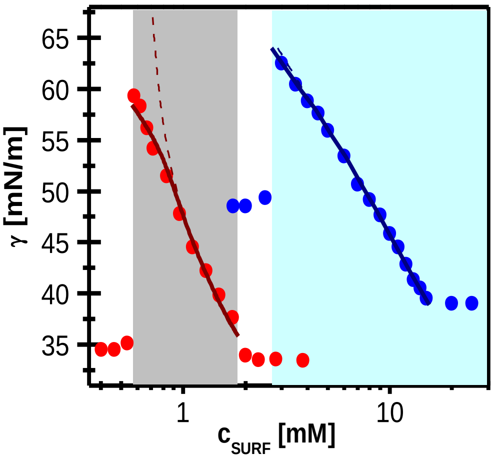


Figure 2. Surface tension isotherms of 100 ppm, 100–200 kDa Pdadmac/SDS solutions in 0.1 M NaCl after 3 days (red) and 100 ppm, 1 MDa NaPSS/DTAB solutions after one month (blue). The blue and grey backgrounds are the phase separation regions for the two mixtures, respectively. The lines are models in the phase separation region as described in the text: dashed lines with stoichiometric binding and bold lines with a correction before charge neutralization.

We then applied the model to the data recorded in this FIGARO experiment on DNA/DTAB solutions in 10 mM NaBr. Fig. 3A and 3B shows the optical density and surface tension, respectively, of freshly mixed samples as well as those aged for 3 days. The phase and surface tension behavior of the DNA/DTAB system closely resemble those of the synthetic P/S mixtures: compositions around charge neutrality of the bulk complexes are initially turbid yet become transparent with time while a sharp surface tension peak is produced.

Calculation of the surface tension of aged DNA/DTAB samples based on our model matches the experimental data. The only significant deviation comes at the left edge of the phase separation region, yet after this sample had been centrifuged the data point converged on the model, which shows that at the phase boundary the precipitation timescale is very slow. The interfacial composition of the supernatant of aged and centrifuged samples was measured using NR, again showing no polymer at the interface in the phase separation region, and the surface excess conversion to the surface tension of the depleted surfactant solution again gives good agreement with the data. We concluded that our model of the surface tension of P/S mixtures also works for biomacromolecule/surfactant systems.

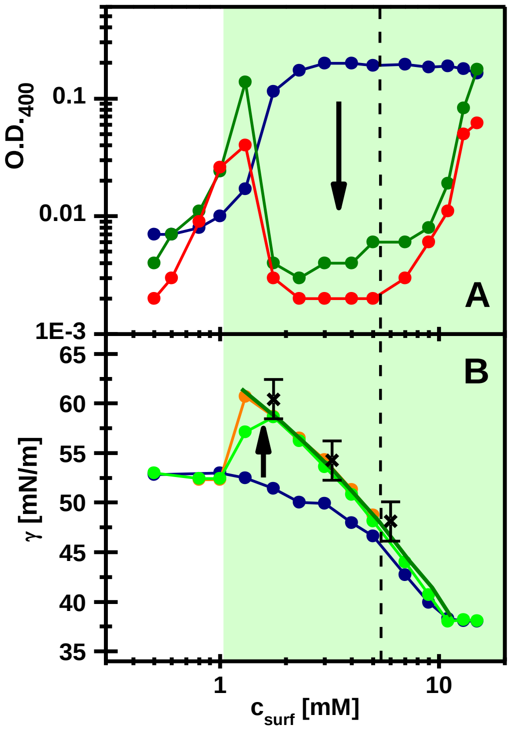
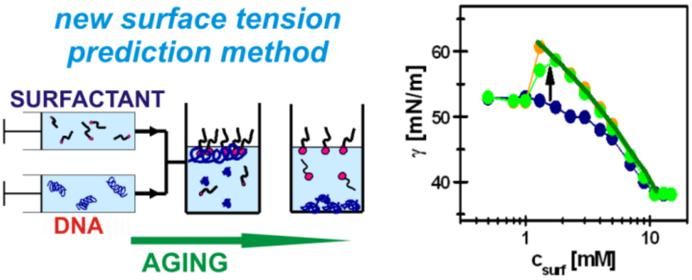


Figure 3. (A) Optical density at 400 nm of DNA/DTAB solutions in 10 mM NaBr as a function of the bulk surfactant concentration measured immediately after mixing (blue), after one day (green), and three days (red). (B) Surface tension measured immediately after mixing (blue), after 3 days (green), and after centrifugation (orange); calculation from NR data of depleted surfactant solutions (black). The green background is the phase separation region and the vertical dashed line marks charge neutrality. The line is the model in the phase separation region described in the text.

In this experiment we applied a novel method of determining the interfacial composition using two isotopic contrasts recorded in air contrast matched water. This new approach reduces uncertainty in the data from the choice of interfacial structure applied and was possible due to the high strength at low Q of FIGARO. There is insufficient space to go into more details here but the approach is outlined clearly in the Supporting Information of our Letter[8] and will be used broadly in the future. Note that data remaining from this FIGARO experiment on the DNA/CTAB system were not included in the publication but are likely to be the subject of an additional paper in the near future.



References. [1] Staples, Tucker, Penfold, Warren & Thomas, *Langmuir*, **2002**, *18*, 5147–5153; [2] Taylor, Thomas & Penfold, *Langmuir*, **2002**, *18*, 4748–4757; [3] Vongsetskul, Taylor, Zhang, Li, Thomas & Penfold, *Langmuir*, **2009**, *25*, 4027–4035; [4] Penfold, Tucker, Thomas & Zhang, *Langmuir*, **2005**, *21*, 10061–10073; [5] Taylor, Thomas & Penfold, *Adv. Colloid Interf. Sci.*, **2007**, *132*, 69–110; [6] Campbell, Angus-Smyth, Yanez, Tonigold, Nylander & Varga, *J. Phys. Chem. Lett.*, **2010**, *1*, 3021–3026; [7] Campbell, Yanez, Angus-Smyth, Nylander & Varga, *J. Phys. Chem. B*, **2011**, *115*, 15202-15213; [8] Abraham, Campbell & Varga, *Langmuir*, **2013**, *29*, 11554-11559.