Experimental Report

Proposal: **9-13-534** **Council:** 4/2014

Title: Interactions of eggshell polypeptide-surfactant mixtures with keratinmonolayers studied by neutron reflection

This proposal is a new proposal

Researh Area: Soft condensed matter

Main proposer: **LU Jian Rena**

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Samples: Keratin and eggshell membrane polypeptides
d- and h-SDS

Instrument	Req. Days	All. Days	From	To
D17	0	4	19/09/2014	23/09/2014

Abstract:

This experiment represents our first effort of trying to study how preformed polypeptide-surfactant complexes adsorbed onto the model keratin substrate surface using neutron reflection. The success of the experiment will transform the current biocompatibility assessment and lead to new insight into molecular interactions underlying surface biocompatibility from the novel NR approach.

Relevant publications in the field:

Chen, C.X.; Hu, J.; Zhang, S.Z.; Zhou, P.; Zhao, X.C.; Xu H.; Zhao, X.B.; Yaseen, M.; Lu, J.R., Biomaterials 2012, 33, 592-603.
Han, S.; Xu, W.; Cao M.; Wang J. Xia, D.; Xu, H.; Zhao, X.; Lu, J.R., Soft Matter 2011, 8, 645-652.
Han, S.; Cao, S.; Wang, Y.; Wang, J. et al., Chem. Euro. J. 2011, 17, 13095-13102.
Wang, S.J.; Ge, X.; Xue, J.Y.; Fan, H.M.; Mu, L.J.; Li, Y.P.; Xu, H.; Lu, J.R., Chem. Mater. 2011, 23, 2466-2474.
Zhao, X.B.; Pan, F.; Xu, X.; Yaseen, M.; Shan, H.; Hauser, C.A.E.; Zhang, S.; Lu, J.R., Chem. Soc. Rev. 2010, 39, 3480-

Adsorption of Protein Hydrolysates-SDS mixtures onto Keratin substrates

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The harshness of surfactant solutions towards skin is a major concern when formulating personal care products. Although a number of methods have been developed over the years to assess the irritation potential of surfactant solutions to skin proteins, the zein solubilisation test is becoming widely adopted. In this case, surfactant harshness to proteins is assessed on its ability to denature and ultimately dissolve zein as it is otherwise insoluble. The BCA assay is then utilised to accurately determine the exact amount of the protein dissolved in solution. We are currently studying protein hydrolysates-surfactant mixtures seeking for application in personal care areas. However, the quantification of the irritation potential of this polypeptide has proved difficult as it is already soluble in aqueous solution. Seeking for an alternative approach to model the interaction between the polypeptide (and its mixtures with surfactants) with a skin model, we have developed a novel methodology by depositing a keratin film onto solid (silicon) substrate. Through controlling keratin concentration and spin coating conditions, we could control keratin film thickness and uniformity. It was further found that the keratin films could be dried and swollen repeatedly without compromising film structure.

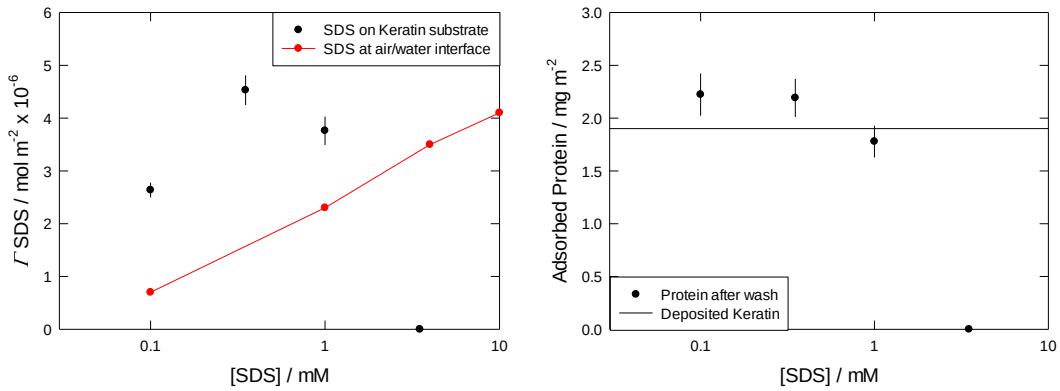

Figure 1 left: SDS adsorbed onto the keratin substrate from SDS-polypeptide mixtures. The SDS adsorbed amount at the air/water interface under same conditions is shown for comparison. Figure 1 right: Keratin remaining onto the silicon surface after treatment with SDS-polypeptide mixtures. Removal is only observed at [SDS] > 1 mM, whereas keratin desorption in presence of SDS solution already starts at the SDS concentration of 0.035 mM.

In this experiment we have studied the adsorption of 1 mg/ml polypeptide mixtures with a varying range of SDS concentrations onto the almost identical keratin layers prepared. The behaviour in presence of anionic surfactants is in fact relevant to personal care applications. We found in previous experiments that SDS solutions alone with concentration as low as 0.035 mM would start to remove the keratin from the silicon surface. Neutron reflection experiments have helped us to reveal the co-adsorption of polypeptide-SDS mixtures onto the keratin layer. The total protein adsorbed could be determined using d-SDS in D_2O (with the net polypeptide adsorbed being the difference from the amount of keratin pre-adsorbed). The amount of keratin removed after the treatment could be determined by replacing the bulk phase with pure water (D_2O). We found that with SDS concentration up to 1 mM the keratin layer was retained on the surface on its entirety. Only with SDS concentration approaching 3.5 mM and beyond the keratin layer was then removed sharply. This observation is important as it shows the ability of the polypeptide to reduce the harshness of surfactants to skin models. The amount of SDS deposited onto keratin was higher than what could be found at the air/water interface under similar conditions, suggesting that the surfactant reached the surface in the form of complexes with the polypeptide.

We are now going to perform parallel measurements with QCM-D to further complement the neutron reflection data. After these data are collected and analysed, we believe the study will be at publishable standard.