Experimental report

15/05/2019

Proposal:	**9-10-1515**	**Council:**	4/2017
Title:	Synergistic effects in self assembly induced structuring of the water-air interface		
Research area:	Chemistry		

This proposal is a continuation of 9-10-1487

Main proposer:	**Mark RUTLAND**
Experimental team:	Georgia PILKINGTON Erik BERGENDAL Lucas KREUZER
Local contacts:	Philipp GUTFREUND

Samples: eicosanoic acid
18methyl eicosanoic acid
19methyl eicosanoic acid

Instrument	Requested days	Allocated days	From	To
FIGARO Langmuir trough	4	4	28/05/2018	01/06/2018

Abstract:

The hypothesis is that inducing packing constraints in the self assembly structure of fatty acids at the water-air interface can cause the liquid-air interface to dimple in a 3D fashion. This was spectacularly proven for the first time using FIGARO 2 days before this proposal deadline. The branched fatty acids in question are biologically interesting due to their barrier properties on hair and wool. These experiments may also cast light on their biological function, and possibly their bacteriostatic properties. The recent measurements showed an unexpected but unmistakable synergy effect in the extent of interfacial dimpling. To allow this effect to be understood and permit publication of this extremely important observation, we are applying for beam time to study the interfacial dimpling of mixtures of three different combinations of fatty acids. With the combination of fatty acids proposed and their different individual dimpling properties, a sufficiently large matrix will be generated to explain the mechanism of this striking synergistic effect.

Synergistic effects in self-assembly induced structuring of the water-air interface.

Background: The project addresses the vexed question of the surface orientation adopted by the branched fatty acids which form the main component of the lipid barrier on the outer surface of hair (and wool). The exact reason for the unusual structure of the fatty acid, and how the two terminal methyl groups arrange themselves at the monolayer interface is still debated. An obvious possibility for why the branched fatty acids is found there is their lower melting temperature than corresponding saturated fatty acids. The alternative natural mechanism to achieve this is through unsaturation (as in omega fatty acids or the lipids in cell membranes) but such molecules are easily oxidized when exposed to air, so that route is not available on the hair surface. The chain packing constraints imposed by the methyl group lead to different molecular areas and chain conformations. Other potential explanations for the presence of the branch are bilayer formation inhibition or improved registration of the available area per molecule to the density of cystein residues to which the fatty acids anchor on the underlying macromolecular structure. Such fatty acids moreover have been proven to possess bacteriostatic properties, and this might have implications for the known bacterial resistance of for example wool. It has also been speculated that the *anteiso* groups affect water structure or adhesion of foreign matter.

As part of a *systematic physicochemical study*[1] of the properties of such fatty acids (Figure 1), using a range of techniques including Vibration Sum Frequency spectroscopy, Langmuir Blodgett film studies and Atomic Force Microscopy it became apparent that deposited films of branched fatty acids formed well defined 2D domains (Figure 2). These domains were NOT observed for unbranched molecules (smooth homogeneous films were formed). The domain size is determined by the position of the branch. We speculate that the reason for domain formation is an intrinsic curvature of the liquid-air interface driven by the balance between the chain packing and the surface energy of the carboxylate-water contact area. A previous experiment at FIGARO (9-10-1484) has proven to strengthen this hypothesis. During this experiment, it was observed that the roughness of the water–air interface increased tremendously at increased fatty acid monolayer compression. Using low contrast hydrogenated fatty acids on a D_2O subphase emphasised that this increase in surface roughness must originate from the D_2O-interface. The observed roughness far exceeds what can be rationalised using capillary wave theory, suggesting a texturing of the water–air interface in accordance with the domains observed on deposited monolayers.

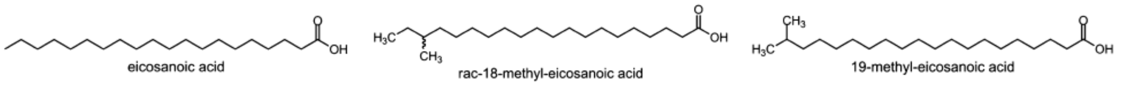

Figure 1 Studied molecules. Larger domains is observed with the 19-branch. Higher curvature is predicted for the 18-branch. Intermediate behaviour was observed for mixtures for 18-MEA and EA.

Results: To further give insight in the self-assembly and interfacial texturing of branched fatty acid monolayers, FIGARO was used for neutron reflectometry and grazing incidence small angle neutron scattering (GISANS). The neutron reflectometry included experiments for the completion of the mixture series of 18-MEA:EA, namely a 38:62 mixture, where preliminary fits are shown in Figure 3. This indicates that there is a sharp transition in the observed surface

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[1] Self-assembly of long chain fatty acids: effect of a methyl branch. Liljeblad, JFD, Tyrode, E; Thormann, E; Dublanchet, AC; Luengo, G; Johnson, CM; **Rutland, MW.** Physical Chemistry Chemical Physics 2014, 16 (33) 17869-17882.

roughness due to domain formation between 38 and 50 wt% 18-MEA in the mixture. Surface domains for deposited monolayers of the 18-MEA:EA 38:62 mixture are elongated and in the order of at least 200 nm along the long axis.

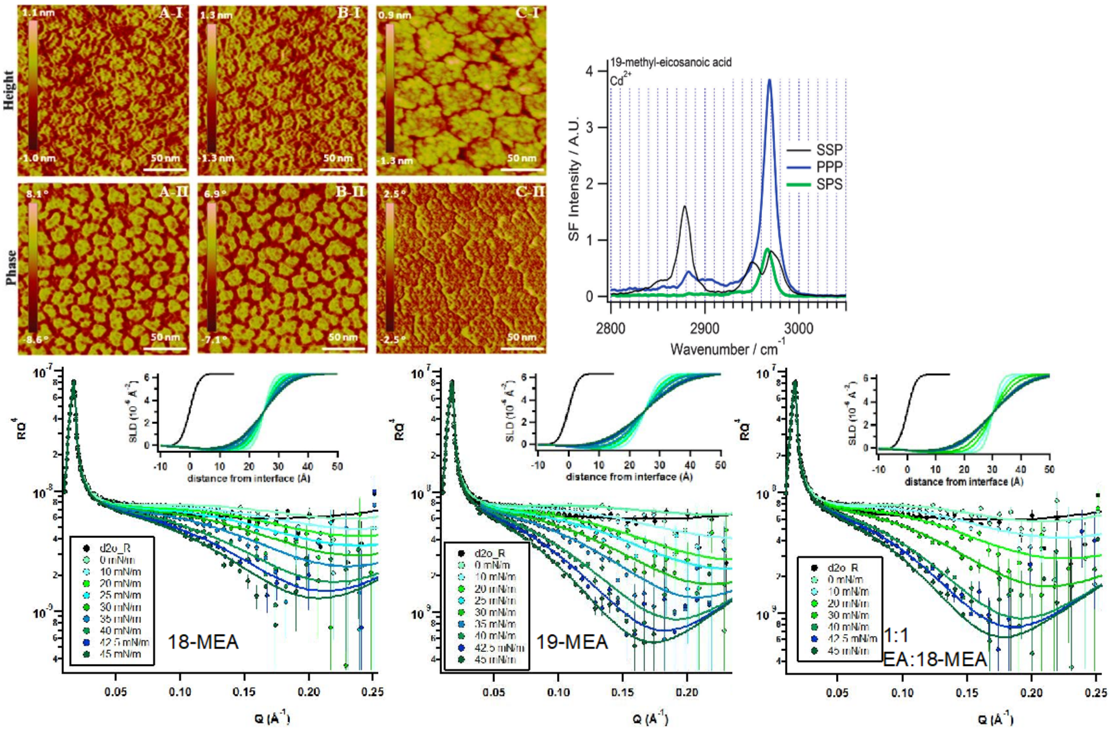

Figure 2 Left: AFM images of branched samples showing domains. Right: VSF spectrum of 19 –MEA. (It demonstrates more chain disorder than for unbranched molecules). Bottom: specular neutron reflectivity data (dots) from experiment 9-10-1484 shown together with fits (lines) from the SLD profiles in the inset. They indicate that at higher pressure (darker) the topography of the liquid-air interface increases. At low surface pressure (brighter) the topography is indistinguishable from pure D_2O. Furthermore, mixing the branched chain 18-MEA with its straight analogue induces a larger effect of the observed surface roughness. This agrees with larger surface domains formed for this mixed system compared to the neat 18-MEA.

Figure 3 also shows the first data and appurtenant preliminary fits for mixed system of the two branched fatty acids, 18-MEA and 19-MEA. This mixture does not show the same type of strong synergistic effect as the 18-MEA:EA 50:50 mixture, possibly explained by a decreased packing efficiency due to the staggered methyl branch placements. The observed surface roughness is more in the order of what could be expected by capillary waves, however, for the previous systems of compact monolayers, this has shown to not be a satisfactory explanation. This is presently under investigation in relation to AFM imaging of a series of deposited monolayers of the mixed branched system.

Having one day of allocated beamtime in the backlog of the 9-10-1484 proposal, this was used to examine the possibility of grazing incidence scattering of monolayers (or water structure due to the presence of monolayers) at the water–air interface. Side reflections in the beam guides have previously made this difficult, combined with the low expected signal of a monolayer of hydrogenated lipids. Side reflections were still present, necessitating a reduction of the flux of an already intensity constrained method. Monolayers were stable for the acquisition time of 18 hours, owing to the addition of $CdCl_2$ in the subphase and relevant pD-adjustments. It was not possible to observe any increased scattering for (hydrogenous) 18-MEA at two surface pressures investigated with GISANS. However, there was an unambiguous increase in intensity from off-specular scattering observed for the mixed system of 18-MEA and deuterated EA. Even though this system is expected to show larger domain formation, and has previously been shown the largest increase in surface perturbation with NR, it is difficult to distinguish the attribution of this signal to texturing of the water–air interface, or to texturing of the higher contrast contributing d-EA (shown in *Figure 4*). This highlights the two possible explanations for the absence of GISANS signal for the hydrogenated 18-MEA on D_2O:

- There is no texturing of the water–air interface due to packing constraints of branched fatty acids. This would mean that chain staggering would induce the low roughness observed with NR, an irregular roughness at the molecular scale. This is, however, contradicted by the synergistic effect of the mixed system, together with its compelling agreement with AFM imaging of deposited monolayers.
- With a system proven to be very flux demanding, and approaching (or crossing) the resolution limit of GISANS, a single wavelength reflectometer optimised for GISANS would be needed to definitively determine if the proposed hypothesis is correct.

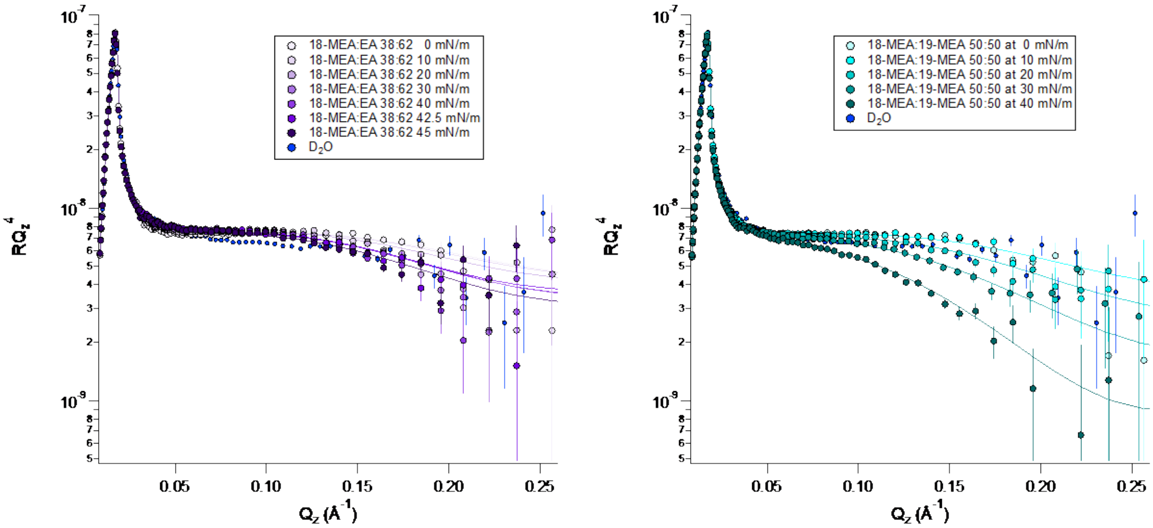

Figure 3 Neutron reflectometry of mixed branched and straight chain fatty acid 18-MEA:EA 38:62 (left) and the two branched fatty acids 18-MEA:19-MEA 50:50 (right). Data is shown as markers and preliminary fits as solid lines. D_2O subphase is shown in both images is blue markers.

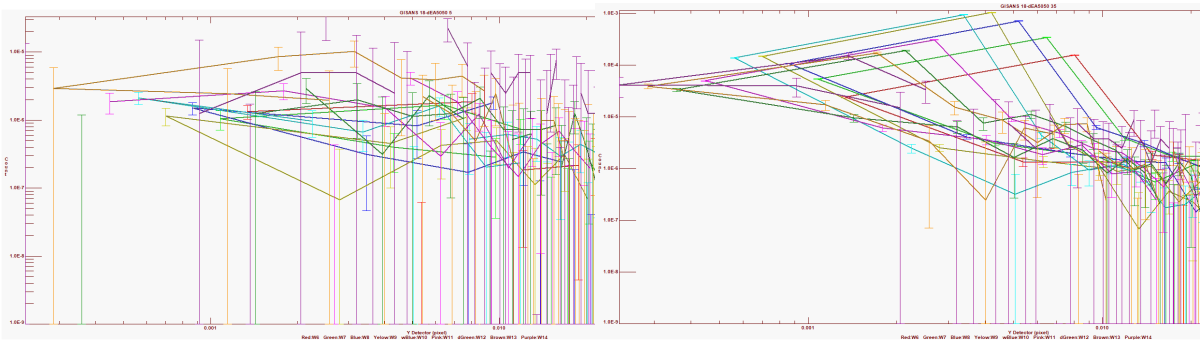

Figure 4 Qy-cuts of GISANS of 18-MEA:dEA 50:50 collected at a D2O interface at 5 mN/M (right) and 35 nM/m (right). The parasitic signal is clearly seen in 35 mN/m as it is only visible on one side of the detector. The absolute intensity is what distinguishes the data at low and high intensity, showing that there is an increase in the off-specular scattering at higher surface pressure.