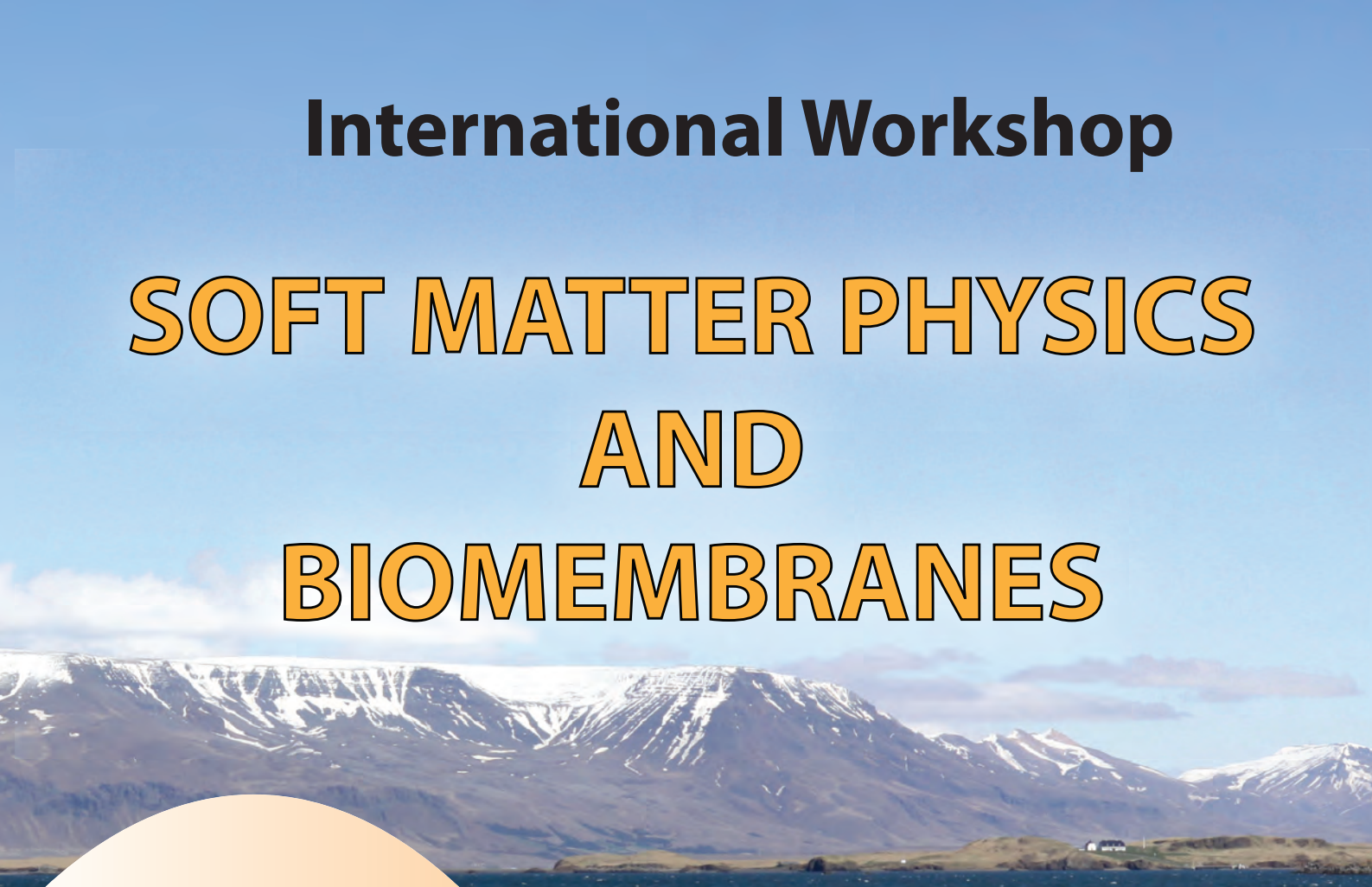
International Workshop

SOFT MATTER PHYSICS AND BIOMEMBRANES



Organizers: Jon Otto Fossum, NTNU, Norway
Aldo Jesorka, Chalmers, Sweden

Invited Speakers

Ilya Reviakine, Biomagune, Spain
Zbigniew Rozynek, NTNU, Norway
Bruno Goud, Institute Curie, France
Elisabeth Lindbo Hansen, NTNU, Norway
Annela Seddon, University of Bristol, UK
Dorthe Posselt, Roskilde University, Denmark
Ritva Serimaa, University of Helsinki, Finland
Ilpo Vattulainen, University of Tampere, Finland
Jan Skov Pedersen, Århus University, Denmark
Irep Gözen, Chalmers University of Technlogy, Sweden
Johanna Ivaska, Turku Center for Biotechnology, Finland
Tuan Phan Xuan, Chalmers University of Technlogy, Sweden
Fredrik Höök, Department of Physics, Chalmers, Sweden
Alar Ainla, Chalmers University of Technology, Sweden
Arne Skjeltorp, Institute for Energy Technology, Norway
Heloisa Bordallo, University of Copenhagen, Denmark
Pekka Lappalainen, University of Helsinki, Finland
Tomas Plivelic, MaxIV Laboratory Lund, Sweden
Iryna Mikheenko, University of Birmingham, UK
Pavlo Mikheenko, University of Oslo , Norway
Adrian Rennie, Uppsala Universty, Sweden
Olle Inganäs, Linköping University, Sweden

University of Iceland
Askja Building
Hall 132
Reykjavik - Iceland

May 21-24
2013

norden
NordForsk

Summer School & Workshop
“Soft Matter Physics And Biomembranes”

Jointly organized by the Nordic Network for Soft Matter Physics (SMP) and the Nordic Network for Dynamic Biomembrane Research (DBR).
The photo shows the conference participants at the historic site where R. Reagan and M. Gorbachev met at the Reykjavik summit in 1986. Group image & title image: Zbigniew Rozynek.

Location: University of Iceland: Askja Building, Hall 132, Reykjavik – Iceland, Dates: May 21-24, 2013

Organizers:

Jon Otto Fossum, NTNU, Norway - jon.fossum@ntnu.no (0047 91139194);
Aldo Jesorka, Chalmers, Sweden - aldo@chalmers.se (0046-734099801).

Transport between hotels and conference location is organized by: Reykjavik Excursions EHF. Contact: (Phone: +354 580 5400).

Schedule

21.05. 2013
11.30/17.00 Arrival, Transfer to the hotels by Reykjavik Excursions EHF
Pre-arranged shuttle busses leave at 11.00 and 16.30. Look for the “Nordforsk” sign.
17.45 Bus transfer to Restaurant *Einar Ben*
18.00 Dinner at Restaurant *Einar Ben* (Corner Veltusund/Hafnastraeti, www.einarben.is)
19.30 Bus transfer to Lecture Hall
20.00 Welcome note (Jon Otto Fossum & Aldo Jesorka)
20.10 Poster Relay
Every poster is presented in a 2 min talk! Talk first, ask questions later!

Please bring your poster presentation on a memory stick in MS Powerpoint format!
21.00 Poster session, Discussions
22.30 Bus transfer to the hotels

22.05.2013
08.40 Departure to Lecture Hall
Session I -- Chair: Jon Otto Fossum & Aldo Jesorka
09.00 Introduction by **Jón Atli Benediktsson**, Prorector of academic affairs, University of Iceland
09.10 **Johanna Ivaska**, Turku Center for Biotechnology, Finland
Integrin traffic and cross-talk with activity regulation and signaling
09.30 **Olle Inganäs**, Linköping University, Sweden
Energy and charge storage in conjugated polymer/biopolymer composites
09.50 **Dorthe Posselt**, Roskilde University, DK-4000 Roskilde, Denmark
Kinetics of structural reorganizations in multilamellar photosynthetic membranes monitored by small angle neutron scattering
10.10 **Adrian R. Rennie**, Uppsala University, Sweden
Colloid Physics of Water Purification - Learning about using Seeds from Trees as a New Technology
10.30 Coffee Break
Session II -- Chair: Kent Jardemark
11.00 **Heloisa Bordallo**, University of Copenhagen, Denmark
Neutrons: the key to understanding hydrogen bonds and improving our quality of life
11.20 **Pavlo Mikheenko**, University of Oslo, Norway
Magnetic flux avalanches in superconducting films
11.40 **Kristijan Leosson**, University of Iceland, Iceland
Polymer waveguide platform for highly integrated biophotonics

12.00 Lunch, University Cafeteria (walking distance ~ 5min)
Session III -- Chair: Ilpo Vattulainen
14.00 **Alar Ainla**, Chalmers University of Technology, Sweden
Lab on a membrane: A toolbox for reconfigurable 2D fluidic networks
14.20 **Bruno Goud**, Institute Curie, France
Mechanics of the Golgi apparatus and membrane trafficking probed by intracellular optical micromanipulation
14.40 **Jan Skov Pedersen**, Århus University, Denmark
Phospholipid bicelles for protein solubilization investigated by SAXS
15.00 **Ilya Reviakine**, Biomagune, Spain
Hydrodynamic effects in laterally heterogeneous films studied by QCM(-D).

15.20 Coffee Break
Session IV -- Chair: Jan Skov Pedersen
15.50 **Fredrik Höök**, Chalmers University of Technology, Sweden
Label-free biomolecular interaction analysis and equilibrium-fluctuation-based single-molecule studies of cell-membrane mimics
16.10 **Zbigniew Rozynek**, NTNU, Norway
Active structuring of clay colloidal armour on liquid drops
16.30 **Hongxia Zhao**, University of Helsinki, Finland
Membrane-sculpting Bin-Amphiphysin-Rvs (BAR) domains *generate stable lipid microdomains*

17.00 Transfer Viking Village (http://www.fjorukrain.is/en)
17.30 Dinner at Viking Village
19.00 Transfer to Reykjavik city center / hotel

23.05.2013

Session V -- Chair: Adrian R. Rennie

08.30 Departure to Lecture Hall
09.00 **Irep Gözen**, Chalmers University of Technlogy, Sweden
Thermal Migration of Molecular Lipid Films as Contactless Fabrication Strategy for Lipid Nanotube Networks
09.20 **Arne Skjeltorp**, Institute for Energy Technology, Norway
GIAMAG magnets for materials separation
09.40 **Elisabeth Lindbo Hansen**, NTNU, Norway
An orientationally ordered glass of soft colloidal platelets
10.00 **Ilpo Vattulainen**, University of Tampere, Finland
Concerted dynamics of lipids with membrane proteins
10.20 Coffee Break

Session VI -- Chair: Ritva Serimaa

10.45 **Tuan Phan Xuan**, Chalmers University of Technlogy, Sweden
Formation of spherical-like, strands-like and rod-like particles and their structural building up. The case of β-lactoglobulin and nanocrystalline cellulose.
11.05 **Annela Seddon**, University of Bristol, UK
Control of Highly Ordered Three Dimensional Biological Nanostructures
11.25 **Ritva Serimaa**, University of Helsinki, Finland,
Structures of natural polymer based materials using x-ray and neutron scattering and imaging methods
11.45 Final Note -- **Jon Otto Fossum, Aldo Jesorka**
12.00 Excursion to Blue Lagoon, Lunch (*Lunch self-organized at the BL*)

Bring bathing clothes, a towel will be provided!
~17:00 Return to Reykjavik city center/hotel

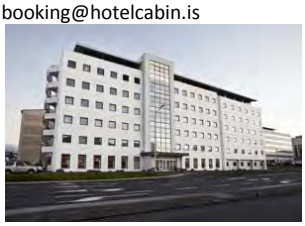
24.05.2013

08.00 Departure to airport (busses are arranged, exact time will be announced)
Everyone not leaving on this date will receive a transport voucher for the airport shuttle.

Accommodation:

Hotel Cabin		Hotel Klettur	
Zbigniew Rozynek	Aldo Jesorka	Anna Kim	Hans-Hermann Gerdes
Elisabeth Lindbo Hansen	Ilya Reviakine	Ilona Wegrzyn	Ivan Rios-Mondragon
Arne Skjeltorp	Kent Jardemark	Mehrnaz Shaali	Xiang Wang
Pawel Sobas	Oscar Jungholm	Alar Ainla	Dominik Frei
Pavlo Mikheenko	Jon Sinclair	Pekka Lappalainen	Magnus Wigner Austefjord
Adrian Rennie	Jan Skov Pedersen	Yosuke Senju	Johanna Ivaska
Hauke Carstensen	jörn d kaspersen	Hongxia Zhao	Jeroen Pouwels
Maja Helsing	Dorthe Posselt	Riina Kaukonen	Elisa Närvä
Tomas Plivelic	murillo longo martins	Bruno Goud	Elina Mattila
Ana Labrador	Heloisa Bordallo	Fredrik Höök	
Sophie Canton	Ritva Serima		
Tuan Phan Xuan	inkeri kontro		
Irep Gözen	Ilpo Vattulainen		
Olle Inganäs	Karol Kaszuba		
Fredrik Bäcklund	Pekka Postila		
Niclas Solin	Jon Otto Fossum		
Annela Seddon	Iryna Mikheenko		

Cabin Hotel, www.hotelcabin.is
Borgartúni 32, 105 Reykjavík
00354 5116030
Reception open 24h
booking@hotelcabin.is



Klettur Hotel, www.hotelklettur.is
Mjölnisholt 12-14, 105 Reykjavík
00354 4401600
Reception open 24h
booking@hotelklettur.is

Talks

Integrin traffic and cross-talk with activity regulation and signaling

Johanna Ivaska

University of Turku, Turku Centre for Biotechnology and VTT Medical Biotechnology

Endocytic trafficking of integrins has an important role in cellular motility and cytokinesis. Integrins are constantly endocytosed from the cell surface and recycled back to the plasma-membrane to facilitate the dynamic regulation of cell adhesion. Recruitment of integrin cargo to the endocytic machinery is regulated by the small GTPase Rab21, but the detailed molecular mechanisms are yet unknown. Furthermore, it is unclear at present whether endocytosed integrin cargo have signalling functions in the endosomes. I will describe our new findings related to this. In addition, the distinct trafficking pathways of active-ligand bound and inactive integrins will be described.

Energy and charge storage in conjugated polymer/biopolymer composites

Olle Inganäs
Biomolecular and organic electronics, IFM
Linköping, Sweden
(ois@ifm.liu.se)

As renewable electrical energy becomes cheaper and more abundant, plentiful and cheap solar electricity will be available at midday, but need also to be used at midnight. This requires new means of scalable electrical energy storage. In biological systems quinone compounds carry the flow of electrons and protons building the pH gradient, useful for ATP-synthase and all subsequent bioenergetics. Quinones can also be generated in lignin derivatives, and lignin is biopolymer number two on Earth.

We have found ways of incorporating black liquor from paper processing into polypyrrole/lignin electrodes where the dominant charge storage is due to the quinone electrochemistry, and where the electronic polymer polypyrrole form the leads to this redox site [1]. This can double the charge density, compared to polypyrrole electrodes. Introducing more quinone species can further improve the charge density in these composite materials to levels found in inorganic cathode materials suitable for Li-ion batteries. Addition of inorganic redox species in the electrode further improves the redox window, capacitance and charge density. However, the redox process of the biopolymer composites requires water environments, limiting the voltage, energy and power density. The other advantage is however that of scalability and cost.

[1] G. Milczarek and O. Inganäs, Science **335** (6075), 1468 (2012).

Kinetics of structural reorganizations in multilamellar photosynthetic membranes monitored by small angle neutron scattering

Dorthe Posselt[1], Gergely Nagy[2,3,4], László Kovács[5], Renáta Űnnep[3], Ottó Zsiros[5,] László Almásy[3], László Rosta[3], Peter Timmins[4], Judith Peters [4,6,7], and Gyõzö Garab[5]

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[4] *Institut Laue-Langevin, BP 156, F-38042, Grenoble Cedex 9, France*
[5] *Institute of Plant Biology, Biological Research Center, Hungarian Academy of Sciences, POB 521, H-6701, Szeged, Hungary*
[6] *Universit´e Joseph Fourier Grenoble I, UFR PhITEM, BP 53, F-38041, Grenoble Cedex 9, France*
[7] *Institut de Biologie Structurale Jean Pierre Ebel CEA-CNRS-UJF, F-38027, Grenoble Cedex 1, France*

In higher plants, the photosynthetic pigment–protein complexes are embedded in the thylakoid membranes, which are located in the chloroplast, and are surrounded by an aqueous matrix, the stroma. We have performed transmission small-angle x-ray and neutron scattering on thylakoids freshly isolated from spinach or pea and suspended in an aqueous medium under near physiological conditions. A broad peak at $q^* \sim 0.02$ Å^{-1} correponds to a repeat distance, RD, of 294 Å±7 Å in spinach and 345 Å±11 Å in pea ($RD = 2\pi/q^*$) . The repeat distance is strongly dependent on the osmolarity and the ionic strength of the suspension medium, as demonstrated by varying the sorbitol and the Mg^{++}-concentration (Posselt *et al*, 2012). The repeat distance decreases when illuminating the sample with white light. The change is reversible and using time-resolved SANS we have investigated this effect on a seconds-to-minutes time scale (Nagy *et al*, 2011, Nagy *et al*, accepted). The structural changes observed are associated with functional changes, as e.g. evidenced by the observation that addition of an uncoupler prohibits the light-induced structural changes, strongly indicating that the light-induced changes are driven by the transmembrane proton gradient.

Nagy, G., Posselt, D., Kovács, L.,Holm, J.K., Szabo, M., Ughy, B., , Timmins, P., Rétfalvi E., Rosta, L., and Garab, G, Biochemical Journal, 436, 2011, 225–230

Posselt, D., Nagy, G., Kirkensgaard, J.K.K., Holm, J.K., Aagaard, T.H., Timmins, P., Rétfalvi E., Rosta, L., Kovács, L. and Garab, G., Biochimica et Biophysica Acta - Bioenergetics 1817, 2012, 1220-1228

Gergely Nagy, László Kovács, Renáta Űnnep, Ottó Zsiros, László Almásy, László Rosta, Peter Timmins, Judith Peters, Dorthe Posselt and Gyõzö Garab, accepted for publication in European Physical Journal E

Colloid Physics of Water Purification - Learning about using Seeds from Trees as a New Technology

Adrian R. Rennie, Maja S. Hellsing, Materials Physics, Uppsala University, Sweden.
H. M. Kwaambwa, School of Health Sciences, Polytechnic of Namibia, Windhoek, Namibia.
Bonang Nkoane, Fiona Selato, Dept. of Chemistry, University of Botswana, Gaborone, Botswana.

Provision of clean water is an essential requirement for health and a major priority throughout the world. An important first step in purification is usually flocculation of particulate impurities so that the majority of mineral particles, plant residues and bacteria are removed by filtration or sedimentation. On a village scale, the crushed seeds from Moringa oleifera have been used as a coagulant. The seed protein is the active ingredient in this respect and is readily available from sustainable sources. The seeds are edible and accepted as safe to use. Scattering experiments (reflection and small-angle scattering) provide valuable information about how the protein binds to impurities and flocculation occurs. The results suggest how the purification process may be optimised and extended to larger scale purification plants. The results of a co-operation between the Universities of Uppsala and Botswana and the Polytechnic of Namibia will be described.

Kwaambwa, Hellsing & Rennie (2010) *Langmuir* **26**, 3902-3910.
Kwaambwa & Rennie (2012) *Biopolymers* **97**, 209-218.

Neutrons: the key to understanding hydrogen bonds and improving our quality of life

Heloisa N. Bordallo

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bordallo@nbi.ku.dk

Hydrogen bonds are ubiquitous to our bodies and the world around us. Although most hydrogen bonds exhibit weak attractive forces, with a binding strength about one-tenth of a normal covalent bond, they are very important, for without them our daily lives would be impossible. If we could see inside ourselves at the molecular level we would observe a marvellous display of chemical reactions taking place, keeping the body healthy. When a foreign drug enters our inner world, it can interfere with these reactions via mechanisms common to solution chemistry --- including hydrogen bonding, dipole-dipole interactions, charge-transfer and covalent bonding --- with (unpredictably) beneficial, benign or catastrophic consequences. Clearly, understanding the structure of a drug in terms of its hydrogen bonds and their interaction with our body chemistry is vital to the challenge of designing new and improved therapeutic drugs.

In our physical (outer) world, hydrogen bonds are just as important. Without them, for instance, cement would crumble and it would not be possible to use this magic material in such diverse applications as moulding into different shapes and sizes to build skyscrapers, bridges, superhighways and houses, or to repair our teeth and keeping them healthy.

In this lecture I will show that Inelastic Neutron Scattering and DFT calculations are powerful instruments for probing matter. Together, they make it possible to follow and understand many problems related to hydrogen bond interactions between molecules in physics, chemistry and biology.

N. Tsapatsaris, S. Landsgesell, M.M. Koza, B. Frick, E.V. Boldyreva, H.N.Bordallo. Polymorphic drugs examined with neutron spectroscopy: Is making more stable forms really that simple? *Chemical Physics* Accepted (2013)

H. N. Bordallo, B. Zakharov, E.V. Boldyreva, M. Johnson, M. M. Koza, T.Seydell, and J. Fischer. Application of Incoherent Inelastic Neutron Scattering in Pharmaceutical Analysis: Relaxation Dynamics in Phenacetin. *Molecular Pharmaceutics*, **9**, 2434-41 (2012)

Magnetic flux avalanches in superconducting films

P. Mikheenko[1], A. J. Qviller[1], J. I. Vestgården[1], S. Chaudhuri[2], I. J. Maasilta[2], Y. M. Galperin[1,3] and T. H. Johansen[1,4]

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The flux distribution and flux propagation in superconductors shows features of soft matter behaviour. The flux distribution obeys models similar to those describing sand piles. It also shows appearance of abrupt avalanches, which are common for piles of sand or other granular materials. The electromagnetic interactions between individual 'particles' carrying magnetic flux in superconductors (vortices), moderated by their pinning, are, however, very different from the interactions between the particles in granular materials. This makes the soft vortex matter a very specific substance. For example, the avalanches that interrupt smooth flow of vortices in thin superconducting films deliver flux into the interior of the sample with staggering speed up to 100 kilometers per second.

We report investigation of this phenomenon using magneto-optical imaging (MOI) that allows visualizing magnetic flux. Moreover, we combine this technique with electrical measurements. Measuring electrical pulses created during the propagation of avalanches gives information on nanosecond time scale, which is not possible in routine MOI.

It will be explained what new insight these measurements could give. The exotic types of avalanches will be demonstrated and methods of the protection from the avalanches will be suggested.

Hydrodynamic effects in laterally heterogeneous films studied by QCM(-D).

Ilya Reviakine

CIC biomaGUNE, San Sebastian, Spain

Quartz crystal microbalance, or QCM(-D), is widely used to study soft, heterogeneous interfaces in aqueous environment. It is an acoustic technique based on measuring resonance frequency and dissipation of a quartz crystal oscillating in a shear-thickness mode. In this talk, we will examine how these measured parameters are related to the properties of laterally heterogeneous interfaces—such as those that form when proteins or liposomes adsorb to a surface (of an inorganic material). In such systems, hydrodynamic effects related to the flow of water around the particles become important, as does the motion of the particles around the particle-surface contact regions. These effects can be modeled with finite element method (FEM) calculations, reproducing a number of experimental observations—such as the non-linear relationship between frequency shifts and surface coverage of particles and transient maxima in dissipation observed in some systems. The conclusion that emerges from experimental results and FEM calculations is that dissipation in laterally heterogeneous films is related not to the internal viscoelastic properties of the adsorbed particles but rather to the geometry of their attachment to the surface. Furthermore, the relationship between the dissipation and the frequency shifts, revealed through the analysis of the *Df* ratio, allows the effects of surface coverage to be separated from those of particle size: *Df* ratio decreases as a function of surface coverage but increases as a function of particle size. We use this effect to study liposome deformation in a model-free fashion.

LAB ON A MEMBRANE: A TOOLBOX FOR RECONFIGURABLE 2D FLUIDIC NETWORKS

Alar Ainla, Irep Gözen, Bodil Hakonen and Aldo Jesorka

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Supported molecular phospholipid films are versatile model membrane architectures, which are valuable to mimic fundamental properties and features of the plasma membrane at reduced complexity. Double bilayer, single bilayer as well as monolayer films can be formed on solid supports, providing enhanced stability and improved accessibility by probing techniques. Supported membranes can cover an extensive area homogenously, which greatly facilitates modification, observation and imaging. Two-dimensionality and fluidity allow their utilization in micro- and nanofluidic devices, which supports functional studies of membrane proteins, and promotes the development of membrane-based chemistry, sensing and separation. Here we introduce a microfluidic toolbox to write 2D nanofluidic networks composed of supported phospholipid membranes, and dynamically modify their connectivity, composition, and local function. We demonstrate how such networks are conveniently generated and locally restructured, and show how various design possibilities such as diffusional barriers and hydrodynamic trapping points can be used in a "lab on a membrane" to directly address biomembrane functions and properties, or to perform membrane-assisted studies of molecular interactions.

References.

Alar Ainla, Irep Gözen, Bodil Hakonen & Aldo Jesorka, *"Lab on a Membrane: a Toolbox for Reconfigurable 2D Fluidic Networks"*, **submitted manuscript.**

Alar Ainla, Gavin D. M. Jeffries, Ralf Brune, Owe Orwar & Aldo Jesorka *"A multifunctional pipette"*, **Lab on a Chip**, 2012, 12(22), 4605-4609.

Mechanics of the Golgi apparatus and membrane trafficking probed by intracellular optical micromanipulation

In vitro studies have shown that physical parameters, such as membrane curvature, tension and composition, influence the budding and fission of transport intermediates. Endocytosis in living cells also appears to be regulated by the mechanical load experienced by the plasma membrane. In contrast, how these parameters affect intracellular membrane trafficking in living cells is not known. To address this question, we have investigated the impact of a mechanical stress on the organization of the Golgi apparatus and on the formation of transport intermediates from the Golgi apparatus. Using confocal microscopy, we have visualized the deformation of Rab6-positive Golgi membranes applied by an internalized microsphere trapped in optical tweezers, and simultaneously measure the corresponding forces. Our results show that the force necessary to deform Golgi membranes drops when the actin cytoskeleton is disassembled or when myosin II activity is inhibited. We also show that the applied stress has a long-range effect on Golgi membranes and induces a sharp decrease in the formation of vesicles from the Golgi apparatus as well as tubulation of Golgi membranes.

Our results suggest that acto-myosin contractility strongly contributes to the local rigidity of the Golgi apparatus and regulates the mechanics of the Golgi apparatus to control intracellular membrane trafficking.

Phospholipid bicelles for protein solubilization investigated by SAXS

Jan Skov Pedersen, Grethe V. Jensen, Heriette G. Hansen, Sara K. Hansen, Thomas Vosegaard, Niels Christian Nielsen
Department of Chemistry and Interdisciplinary Nanoscience Center, Aarhus University, Aarhus, Denmark

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Mixed phospholipid micelles are widely applied in NMR studies of membrane proteins in solution, as they can solubilize them and be aligned in the magnetic field. Mixing of dihexanoyl phosphatidyl choline (DHPC) and dimyristoyl phosphatidyl choline (DMPC) in certain ratios leads to the formation of anisotropic micelles, called bicelles. It has been proposed that the DMPC molecules with relatively long C_{14} hydrocarbon tails constitute a flat bilayer, whereas the DHPC molecules with shorter C_6 tails form the rim of the bicelles [1,2]. Thus, according to this idealized picture, the DMPC/DHPC ratio determines the size of the bicelles. Although SAXS [3,4] and SANS [5,6] data have previously been published for this system, only limited analysis in terms of a geometric model for the shape of the bicelles has been done [5,6], and not at all for SAXS data. In this work, SAXS data were collected for a wide range of DMPC/DHPC ratios. Solutions applied for NMR measurements with 30 wt% were diluted to avoid structure factor effects in the SAXS patterns. Dilution with pure solvent, however, leads to an increased DMPC/DHPC ratio in the micelles, as DHPC has a relatively high solubility. Dilutions with solutions of different DHPC concentrations were performed to find the concentration which does not lead to a change in the micelle composition. For the correct concentration, the structure factor effects decrease upon dilution, whereas the form factor does not change. The SAXS data indicate a relatively complex phase diagram as a function of DMPC/DHPC ratio with different morphologies of the aggregates, which do not follow the suggested trends for the ideal bicelle model.

[1] Bian & Roberts, *Biochemistry*, **29**, 7928 (1990)
[2] Vold & Prosser, *J. Magn. Reson. B*, **113**, 267 (1996)
[3] Bolze *et al.*, *Chem. Phys. Lett.*, **329**, 215 (2000)
[4] Kozak, M., Domka, L. and Jurga, S., J. Mol. Struct., 846, 2007, 108-111.
[5] Nieh *et al.*, *Biophys. J.*, **82**, 2487 (2004)
[6] Harroun *et al.*, *Langmuir*, **21**, 5356 (2005)

Characterizing the Photoinduced Structural Dynamics in Fe(II) Spin Crossover Complexes

Sophie E. Canton

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Lund University, Sweden

(sophie.canton@maxlab.lu.se)

The development of photoactive devices based on nanomaterials requires mapping the dynamical structural changes induced in their building blocks upon photoabsorption. The growing family of spin crossover Fe(II) complexes continues to be intensively investigated in connection to their numerous practical applications that include data storage, displays or sensors .

In these molecules, an intricate balance between the electronic and steric interactions governs the spin multiplicity of the ground state, which is largely found to be the Low Spin (LS) state for the case of Fe(II) complexes. The added influences of the surrounding solvent and counterions, although well-documented, remain unexplained to date. Applying an external perturbation (e.g. UV-visible light) generally results in the transient population of the High Spin (HS) state. Characterizing the nature of this short-lived species is of paramount importance to eventually control the decay dynamics back to the ground state, hence the resulting functionality.

This talk will show that a combination of several synchrotron-based techniques can further the current understanding of how to control the ground state structure. In addition, with the advent of ultrafast laser synchronization with electron storage ring over the last decade, it has also become possible to implement optical pump- X ray probe set ups. They allow following in real time the coupled evolution of electronic and geometric structures of photoexcited Fe(II) complexes. Several examples will be discussed.

Label-free biomolecular interaction analysis and equilibrium-fluctuation-based single-molecule studies of cell-membrane mimics

Fredrik Höök, fredrik.hook@chalmers.se, Chalmers University of Technology, Gothenburg, Sweden

Measurements of ligand binding events on membrane protein receptors in a near-natural environment would display an advantage in mechanistic studies of membrane receptors. Furthermore, the residence time of drug-target interactions is being increasingly recognized as a key parameter in evaluating drug efficacy, but is hampered by the technical challenge to perform such studies on membrane proteins. With single-molecule sensitivity, such information can be gained for both high and low affinity interactions, and be used in both drug-screening and medical-diagnostic applications. Recent advancement in e.g. nanotechnology has led to a diverse set of tools offering single molecule sensitivity. However, to yield sufficient statistics within reasonable time scales, multiple single biomolecular binding events should preferably be probed simultaneously. For strong interactions, this may put constrains on the lowest concentration that can be detected, while for weak interactions, a high acquisition rate will also be required. I will present a single-molecule detection concept that in principle meets these requirements. The principle is based on the use of fluorescently labeled lipid vesicles as enhancer elements in total internal reflection fluorescence (TIRF) microscopy, making the concept compatible with analysis of both water-soluble and cell-membrane bound receptors. Focus will be put on how the concept is currently evaluated as a diagnostic assay for virus and biomarker detection[1] and explored in drug-screening applications[2]. I will also discuss our recent progress in label-free nanoplasmonics (localized surface plasmon resonance), pointing towards the realization of single-molecule detection without the use of fluorescent labels[3] and a new means of utilizing the two-dimensional fluidity of supported lipid bilayers for microfluidic-based membrane-protein chromatography applications[4] and label free imaging biomolecular with diffraction limited lateral resolution[5].

[1] Bally M, Gunnarsson A, Svensson L, Larson G, Zhdanov VP, Hook F: Interaction of Single Viruslike Particles with Vesicles Containing Glycosphingolipids. Physical Review Letters 2011, 107: # 188103: http://dx.doi.org/10.1103/Physrevlett.107.188103
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ACTIVE STRUCTURING OF CLAY COLLOIDAL ARMOUR ON LIQUID DROPS

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Keywords: clay mineral, structuring, electric field, colloids, pupil-like behaviour

Abstract: Adsorption and assembly of colloidal particles at the surface of liquid droplets are at the base of particle–stabilized emulsions [1] and templating [2]. Here we show that electrohydrodynamic and eletrorheological effects in leaky-dielectric liquid drops can be used to structure and dynamically control colloidal particle assemblies at drop surfaces, including electric-field-assisted convective assembly of jammed colloidal "ribbons", electro-rheological colloidal chains confined to a two-dimensional surface and spinning colloidal domains on that surface. In addition we demonstrate the size control of "pupil" like openings in colloidal shells. We anticipate that electric field manipulation of colloids in leaky-dielectrics can lead to new routes of colloidosome assembly and design for "smart armoured" droplets [3].

Colloidal particles can bind strongly to fluid interfaces and assemble into thin layers. Monodisperse colloidal beads can form 2D ordered colloidal crystal monolayers [4] and poly-disperse and anisotropic particles form amorphous shells [5]. This effect is currently much studied in relation to particle-stabilized "Pickering" emulsions [1] where particle coatings on droplets effectively prevent droplet coalescence and produce very stable surfactant-free emulsions. Solid colloidal capsules; colloidosomes, can also be produced by fusing or linking the colloidal particles at the surface of Pickering emulsions droplets [6].

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Membrane-sculpting Bin-Amphiphysin-Rvs (BAR) domains generate stable lipid microdomains

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Bin-Amphiphysin-Rvs (BAR) domain proteins are central regulators of many cellular processes involving membrane dynamics. The BAR domain is a dimeric α-helical protein motif, which can sense/generate membrane curvature to stimulate the formation of membrane protrusions or invaginations in cells. Depending on the geometry of the lipid-binding interface and oligomerization properties of the domain, BAR superfamily domains can sense/generate either positive (BAR and most F-BAR domains) or negative membrane curvature (most I-BAR domains) as well as stabilize planar membrane sheets. Here we report that, in addition to regulating membrane geometry, BAR domains can generate extremely stable lipid microdomains by 'freezing' lipid dynamics. This is a general feature of BAR domains because the yeast endocytic BAR/F-BAR domains, the I-BAR domain of Pinkbar, and the eisosomal BAR protein Lsp1 induced phosphoinositide-clustering and halted lipid diffusion, despite differences in mechanisms of membrane interactions. Lsp1 displays comparable low diffusion rates *in vitro* and *in vivo*, suggesting that BAR domain proteins also generate lipid microdomains in cells. These results uncover a conserved role for BAR superfamily proteins in regulating lipid dynamics within membranes. Stable microdomains induced by BAR domain scaffolds and specific lipids can generate phase boundaries and diffusion barriers, which may have profound importance in diverse cellular processes.

Thermal Migration of Molecular Lipid Films as Contactless Fabrication Strategy for Lipid Nanotube Networks

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Motion of individual molecules along thermal gradients, known as thermophoresis, thermomigration, or the *Soret effect*, involves the movement of independent molecules or particles within a mixture along a temperature gradient. On a much larger size scale, in droplet microfluidics or biological cells, temperature-directed migration is an area of significant scientific and technological interest. In between single molecules and biological systems resides the domain of organized molecular assemblies, where the collective, rather than the individual, behavior dominates the physical and chemical properties. In this so-called mesoscale regime, which is one of the key areas of research in nanoscience and technology, thermophoresis or other modes of temperature-directed transport had never been experimentally observed. We show for the first time that an organized ensemble of molecules, in our case a phospholipid double bilayer membrane adhered on an appropriately engineered solid support, can exhibit thermomigration along a temperature gradient, generated on a microscale substrate. We believe that our findings will stimulate the development of new manipulation techniques for soft matter on the mesoscale. In particular the optical fabrication of nanotube interconnected vesicles is a valuable alternative to the previously reported techniques. The controlled placement of pinning sites, e.g. by surface nanofabrication techniques, can potentially enable design and automated fabrication of vesicle-nanotube networks, which can greatly facilitate the construction of nanoscale models for communication and transport studies in biology and information technology.

Nordforsk network workshop on "Soft matter physics & biomembranes" in Reykjavik Iceland 21-24 May, 2013

Abstract

GIAMAG magnets for materials separation

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Abstract

A new design of a magnet system denoted **GIAMAG*** (**GI**ant **MA**gnet field **G**radient) has been realized with an unprecedented value of the product of the magnetic field strength B and the field gradient ∇B. This is crucial for rapid extraction of e.g. magnetic particles in dispersions as the magnetic force acting on magnetic particles is

$F \sim B \times \nabla B$.

Existing magnet systems can just pull magnetic microparticles from solutions, whereas GIAMAG can extract magnetic particles down to nanosizes.

A review will be given of the principle design of the magnet and various possible applications.

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* www.giamag.com

An orientationally ordered glass of soft colloidal platelets

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Abstract: Colloidal dispersions of anisometric particles can display dynamical arrest and ordering phenomena involving not only translational but also rotational degrees of freedom. We show that orientational order can develop in glassy colloidal dispersions of soft platelets submitted to a slow concentration increase from evaporation. Our model system of Laponite (LRD) platelets in deionized water has been extensively studied for its dynamical arrest transitions, and the existence of an underlying isotropic-nematic transition, possibly masked by the slow dynamics, has been debated. We use small-angle x-ray scattering, dynamical light scattering and birefringence observations to characterize our samples, and discuss whether evaporation effectively causes a 'quench' into an orientationally ordered state that traps the system on very long timescales or if, conversely, evaporation acts to move the system closer to an underlying equilibrium state that does indeed possess orientational order.

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Concerted dynamics of lipids with membrane proteins

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Abstract

We discuss how the diffusion of membrane proteins and lipids depends on molecular crowding. The topic is highly important since lateral diffusion is one of the most significant dynamic processes in cell membranes, as it governs a variety of phenomena such as formation of membrane protein complexes and self-assembly of functional nanoscale membrane units. Further, given that native membranes are rich in proteins, it is quite evident that crowding may play a decisive role in lateral diffusion. Here we consider these topics from a molecular perspective using atomistic and coarse-grained simulations where we vary the protein concentration over a wide range, starting from dilute systems and extending to membranes that are rich in proteins. We demonstrate the importance of understanding the concerted nature of molecular diffusion as well as the profound influence of crowding on diffusion. Besides this, we discuss the limitations of molecular simulations in sampling dynamic processes whose characteristic times are not short compared to times that one can currently simulate.

Formation of spherical-like, strands-like and rod-like particles and their structural building up. Case of β-lactoglobulin and nanocrystalline cellulose.

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The modulation of structural properties of biopolymer aggregates offers a potential to tune its functional properties. We will present results that show how the structural properties of a golublar protein (β-lactoglobulin) and a rod like polysaccharide (nanocrystalline cellulose) can be modulated by changing pH or by adding salts.

Stable suspensions of spherical protein particles (microgels) are formed upon heating β-lactoglobulin (β-lg) in pure water within a narrow range of the pH between 5.75-6.1 close to the isoelectric point of the protein (pI). The particles have a radius of about 60 to 200 nm and a density of about 0.15 $g.mL^{-1}$. At higher pH β-lg forms short curved strands with a diameter of about 5 nm and a length of about 50 nm. At protein concentrations above about 50 $g.L^{-1}$ the microgels, or strands, randomly associate into self-similar aggregates with a size that increases with increasing concentration until the system gels. Interestingly, larger and denser microgels are formed in the presence of Ca^{2+} at neutral pH, compared to the case in pure water close to pI. The amount of calcium bound to the proteins was determined and the results suggest that the crucial parameter for microgel formation is the net charge density of the native proteins. The conditions for the formation of strands, spheres microgels or fractal protein aggregates will be discussed in details. A proposed mechanism for the formation of these different structures as function of medium conditions will be presented.

Cellulose rod-like nanocrystals (NCC) are obtained from sulfuric acid hydrolysis of cellulose fibers. In this process negatively (surface) charged NCC particles are formed and it results in a perfectly uniform dispersion of the particles in water via electrostatic repulsion. In this study, the dynamic properties of NCC suspensions were investigated using polarized and depolarized dynamic light scattering (DLS, DDLS). Translational (D) and rotational (Θ) diffusion coefficients in dilute suspensions were measured and gave values of D = 5860 μs = and Θ □□□□□□□□□□□□□□. With the use of Broersma relations, the rotational and translational diffusion coefficients lead to values of the average length L = 170 nm and the cross-section diameter d = 17 nm. The static properties of NCC suspensions were also studied. The evolution of the structure factor Sq at high q didn't follow a q^{-1} dependence as expected in the case of rod like particles, which was due to the polydispersity. The stability and self-assembly of these NCC particles were examined in different solvent and salt conditions and will also be

presented.

Control of Highly Ordered Three Dimensional Biological Nanostructures

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Biological lipids, such as those that make up the cell membrane have a wide range of self-assembly behaviour which can be exploited for both biophysical studies and the fabrication of nanoscale biomaterials. This work demonstrates how the self-assembly behaviour on the nanoscale can be tuned to produce soft, biocompatible materials with tailored structures. Furthermore, by using x-ray scattering techniques under dynamic conditions such as flow, these materials can be ordered to give structures with a high degree of alignment, in essence creating a crystal-like structure from a soft material. By considering the manner in which we prepare these materials, we can access bulk structures with a range of phases, as well as thin films with a high degree of order. Further to this work, new methods of coupling x-ray scattering with nanolitre volume microfluidics and in-line spectroscopy will be discussed. This has potential applications in the high throughput production of template materials, growth of protein crystals for crystallography, as well as deepening our understanding of the mechanisms underlying the behavior of biological liquid crystal phases.

Structures of natural polymer based materials using x-ray and neutron scattering and imaging methods

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Plant cell wall may be considered as a nanocomposite of cellulose, hemicelluloses and lignin. In the cell wall cellulose chains are aggregated to form partially crystalline microfibrils which are further agglomerated into bundles. The weakly ordered structures of lignin and hemicelluloses are still under debate. Such structures of plant cell wall may be present also in natural polymer based materials like pulp, microcrystalline cellulose, and cellulose whiskers.[1]

X-ray and neutron scattering and imaging methods are powerful tools for structural characterization of wood cell wall and natural polymer based materials. Results on recent small and wide angle scattering and microtomography studies on wood cell wall and the enzymatic hydrolysis of wood based nanocellulose to fermentable sugars will be reviewed.[2] Possibilities to enhance these studies at the new nanofocus x-ray scattering beamlines at synchrotron facilities will be discussed.

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Posters

Organization of small molecules in protein wires

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Oligothiophenes such as sexithiophene (6T) are an important class of materials for use in organic electronics and photonics applications but difficult to work with due to poor solubility and sensitivity to different aggregation states. It would therefore be valuable to have an effective dispersion agent that also results in a controlled packing and orientation of 6T. In previous work, we have developed a method of preparing self-assembled protein nano wires functionalized by small hydrophobic molecules in order to obtain functional materials. We have now found that when incorporating 6T into insulin protein nano wires, the thiophene molecules readily disperse within the protein aggregates in such a way that the 6T molecules become orienTed along the fiber axis. The protein dispersion of 6T not only results in an increased 6T emission intensity compared to the solid state, but the emission has also been shown to be polarized.

Structure and self-organisation in magnetic liquids

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Abstract:
Ferrofluids are magnetic liquids that contain nanometer sized magnetite particles. They are commercially used in many different applications, e.g. in high-end loudspeakers or as liquid seals. Furthermore ferrofluids are object of present research.
Here we present a new route of addressing self-organisation in a magnetic liquid. The basic idea behind it is that ferrofluid is used as solvent for micrometer sized ferromagnetic and diamagnetic particles. The effective magnetic behaviour of the particles is altered since they replace ferrofluid in a certain volume. This effect can be seen analogue to the Archimedes principle.
The approach described above makes it possible to tune the magnetic properties of the micrometer sized particles by changing the concentration of nanometer sized particles in the solvent and thus the effective magnetic behaviour of the large particles. Due to the magnetic interaction, the larger particles arrange themselves in lattices. By changing the ferrofluid concentration the magnetic susceptibility of the solvent is changed and the effective susceptibility of the larger particles is altered. Therefore the interaction between them is tuneable and different structural arrangements can be created. Here we have studied the phase transition from cubic to hexagonal ordering while continuously increasing the magnetic susceptibility of the solvent. The positions of the particles were visualised by particle specific dyes and the use of an optical microscope. The individual particle positions were evaluated automatically and a model to quantitatively explain the results is presented.

NEW APPROACH TO FABRICATING JANUS AND PATCHY PARTICLES

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Keywords: clay mineral, self-assembly, electric field, colloidosome, Janus particle

Abstract: Emulsions are generally stabilized by the use of surfactants, but solid particles dissolved in one phase can also stabilize emulsions. This is known as Pickering emulsion. Colloidal particles adsorb strongly at liquid interfaces, and this has been exploited to stabilize emulsions and to produce colloidosomes [1]. Clay minerals and other nanoparticles are known to produce very stable Pickering emulsions [2].
We present a simple and robust method to produce clay-based Pickering emulsion. We also show how to fabricate shells composed of two hemispheres of different colloidal particles, Janus shells, by electrocoalescence of two oil-in-oil emulsion drops covered by clay and polymer colloidal particles.
The method of Janus particle fabrication presented here is entirely dependent on drop electrocoalscence dynamics and the Taylor circulation flow, resulting in a ribbon formation prior to the coalescence [3]. Therefore the liquids should be chosen such that that Maxwell time in the drop is longer than that of the surrounding liquid. In principle, any non-polar liquid emulsion system could be used; provided that the drop and surrounding liquid has the right Maxwell times for producing electrohydrodynamic flow.

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Phase Behaviour of Colloidal Mixtures Under Shear

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Mixtures of colloidal particles of essentially two different sizes are used in technological applications such as film formation on surfaces, paper coatings, etc. Dispersed latex particles provide an excellent model system for study of mixtures with long range interactions. We will report changes in structure that are observed when concentrated monodisperse charged particles, that on their own would form crystals, are mixed. Small-angle neutron scattering experiments show that dispersions of charge stabilized latex particles of a single size form face centered cubic structures [1]. If salt is added, the interactions are screened and more liquid like structures are seen. In mixed dispersions of small and large latex particles, the small particles melt the structure of the large, whilst retaining their overall structure [2].

The rheology of bimodal dispersions of colloidal particles has been discussed by a number of authors [3,4] and some of these studies have used small-angle neutron scattering to investigate single components in shear cells. Different mechanical response of components of samples can give rise to shear banding and to macroscopic phase separation. The complexity of behaviour of colloidal dispersions and mixtures has been reviewed from a fundamental perspective [5]. There are specific ideas about the influence of shear on mixtures [6]. Understanding the flow behaviour of mixtures of colloidal particles is crucial to many practical applications where the end structure is determined by process conditions. Despite a number of studies on mixtures of chemically different particles, there has been little work on what would appear to be an excellent model system with particles that are chemically similar and differ only in size.

In order to probe this effect we have recently performed RheoUSANS experiments on mixtures of small and large latex particles to attempt to determine the structure of clusters formed and the effect of shear on cluster formation. The samples at these concentrations display only modest changes in viscosity but large changes in structure under flow. Preliminary results show some rather interesting behaviour in that a regime of low/high/low shear does not produce scattering that is the same in both low shear rate states. Going back to low shear does not return the system to the same structure.

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A multifunctional pipette for localized drug administration to brain slices

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We have developed a superfusion method utilizing an open-volume microfluidic device for administration of pharmacologically active substances to selected areas in brain slices with high spatio-temporal resolution. The method consists of a hydrodynamically confined flow of the active chemical compound, which locally stimulates neurons in brain slices, applied in conjunction with electrophysiological recording techniques to analyze the response. The microfluidic device, which is a novel free-standing multifunctional pipette, allows diverse superfusion experiments, such as testing the effects of different concentrations of drugs or drug candidates on neurons in different cell layers with high positional accuracy, affecting only a small number of cells. We here demonstrate the use of the method in electrophysiological recordings of pyramidal cells in hippocampal and prefrontal brain slices from rats, determine the dependence of electric responses on the distance of the superfusion device from the recording site, and document an approximately 30 fold gain in solution exchange time, as compared to whole slice perfusion. Localized solution delivery by means of open-volume microfluidic technology also reduces reagent consumption and tissue culture expenses significantly, while allowing more data to be collected from a single tissue slice, thus reducing the number of laboratory animals to be sacrificed for a study.

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STRUCTURE DETERMINATION OF ALFA-SYNUCLEIN OLIGOMERS

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In biological systems self-assembly is a central process for formation of complexes of biological macromolecules and between biomacromolecules and small molecules. Unwanted aggregation of proteins after misfolding occurs in a number of neurodegenerative disorders such as Parkinson's, Creutzfeldt-Jakob's and Alzheimer's [1].

Parkinson's disease (PD) is connected with the presence of large protein aggregates within the brains of those affected with the disease. The main protein component of these is aSN which is natively unfolded. Recombinant human aSN has been shown to form filaments or fibrils under physiologically relevant conditions with similar structure to those of filaments extracted from PD affected brains and other aSN deposition diseases [2, 3]. Therefore, aSN is believed to play a central but not fully understood role in the development of PD.

The cytotoxic state of the aggregated aSN protein is likely to be an oligomeric intermediate structure [4], reported to have the shape of a torus [5]. The oligomer is thus believed to be able to incorporate into the membrane and thereby lyse the host cell by creating holes in the membrane. Using Small-Angle X-ray Scattering we investigate the structure of these oligomer species with and without the fibrillation inhibitor Epigallocatechin gallate (EGCG) which is able to prevent the aSN oligomers from disrupting membranes.

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Lipids changing the structure of the Human Epidermal Growth Factor Receptor

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A number of structural studies have suggested lipids to be an integral component of membrane proteins [1-4], their role being to stabilize or even modulate protein conformation and hence their function. The epidermal growth factor (EGF) receptor is a membrane protein composed of the ligand-binding extracellular domain, a single transmembrane segment, and a large kinase domain. It has been established that activation of the EGF receptor is modulated by the lipid composition of a membrane. Depletion of cholesterol from plasma membranes leads to the hyper-activation of EGFR, whereas the cellular GM3 gangliosides have been found to inhibit its activation [5-6]. These biochemical observations raise an intriguing question about the structural mechanism governing the activation process. Unraveling this issue is very difficult through experiments, which simply lack the proper resolution. We tackled the problem by extensive atomistic molecular dynamics simulations [7]. We investigated the influence of lipid composition, and in particular the role of GM3 gangliosides on the dynamics of the nearly full-length EGF receptor chain, thereby shedding light on the mechanism of how the receptor's activity can be inhibited at the molecular level. Our results show a substantial impact of lipids on the receptor structure particularly on its extracellular region. We observed clearly different EGFR structures in two different membranes in the presence and absence of GM3. These differences were particularly profound in the receptor's region that is responsible for its dimerization and thus activation. The performed simulations suggest that under the influence of GM3, the receptor adopts a conformation which either slows down or even inhibits the dimerization process of the EGFR. Our simulations highlight lipid specific conformational changes and offer a rational explanation for the previously conducted biochemical studies [5].

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Myosin-X promotes breast cancer invasion and spreading under regulation of mutant p53

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Mutations of the tumor suppressor *TP53* increase tumor cell invasion and metastasis with several mechanisms including increased targeting of integrins to the plasma membrane. Here we demonstrate a role for the filopodia-inducing motor protein Myosin-X (Myo10; encoded by *MYO10*) in mutant p53–driven cancer invasion. Myo10 is required for breast cancer cell invasion and dissemination *in vitro* and *in vivo.* The pro-invasive functions of Myo10 are dependent on its ability to transport β1-integrins to the filopodia tip. Introduction of mutant p53 promotes Myo10 expression in cancer cells and pancreatic ductal adenocarcinoma in mice, whereas suppressing endogenous mutant p53 using RNAi attenuates Myo10 levels and cell invasion. Myo10 is predominantly expressed at the invasive edges of clinical breast carcinomas, where high Myo10 expression correlates with the presence of *TP53* mutations and is associated with poor survival. These data identify Myo10 as a new downstream component for mutant p53–driven invasion and demonstrate that plasma-membrane protrusions, like filopodia, may serve as specialized metastatic engines.

A New Multifunctional Pipette for Localized Single-Cell Superfusion

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Microfluidics has become an important technology in studies of biological cells. Recent developments have initiated a transition from closed-channel devices to new concepts which de-couple cell cultures from the fluid-handling circuitries and thereby enable several beneficial features of microflows in open-volumes [1,2].

Previously, we reported a device for highly localized superfusion, thermed the "Multifunctional Pipette", which we fabricated in a soft polymer material [3]. This device has already been used for a variety of single cell applications. However, some challenges still remain associated with the material such as absorption of hydrophobic compounds and manufacturing scalability.

Here we present a novel miniaturized version of the multifunctional pipette fabricated in a hard photo-patternable polymeric material. This particular material, SU-8, was chosen due to its favorable chemical and mechanical properties.

The miniature multifunctional pipettes were fabricated in a multilayer photolithography approach, using a bonding method adapted from Agirregabiria et. al [4]. The bonding mechanism was shown to be reliable and the pipettes were tested to withstand pressures up to 0.95 bar.

The miniature multifunctional pipette is currently being used in studies of network communication between astrocytes in cultures. Further investigations include applications in the research fields of neuropharmacology, cardiac muscle, stem cells and release of biological substances from single cells.

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Characterization of S-layer coated liposomes using SAXS

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Many bacterial strains have a crystalline protein surface layer (S-layer) on their surface. As S-layers tend to self-assemble into two-dimensional, porous layers on surfaces with suitable properties, they are of interest in many medical and biotechnological applications. Suggested applications for S-layers include microsieves and coatings. Some S-layers facilitate adhesion of bacteria to surfaces. In particular the S-layer protein SlpA of *Lactobacillus brevis* ATCC 8287 facilitates adhesion to human intestinal cells. [1,2] The crystal lattice formed by SlpA has previously been characterized with small angle X-ray scattering (SAXS). [3]

Liposomes are hollow aggregate structures that phospholipids form when dispersed in aqueous solutions. The use of liposomes in biomedical and medical applications is highly diverse, and liposomes can be used to enhance drug delivery by encapsulating biologically active molecules in the internal aqueous lumen or in the lipid bilayer. Of particular importance for the present research is that S-layer coatings have been found to stabilize liposomes and improve their ability to retain the active molecules e.g. against thermal shock and pH change. [4]

S-layer reassemblies on liposomes have previously been studied using other methods, such as cryo-electron microscopy [4,5] but to our knowledge, SAXS has not been applied to these kinds of systems. Therefore, in this work we have investigated the possibility to immobilize reassemblies of SlpA on neutral and positively and negatively charged liposomes. Phospholipids were hydrated with the soluble fraction of SlpA and monolamellar liposomes were prepared by the extrusion technique. The samples were characterized by SAXS with measurements done at beamline I911-SAXS in MAX-lab, Sweden.

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MAX IV Laboratory promoting Research in Soft Matter Physics and Biomembranes

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The MAX IV Laboratory has been established in 2010 to include both the operation of the present MAX-lab facilities (MAX I, II, III) and the new MAX IV project in Lund, Sweden. The new MAX IV 3 GeV storage ring is foreseen to be operative in 2016. The overall goal of the MAX IV Laboratory is to be an outstanding facility for research in a remarkable scientific and social environment.

A recent upgrade on the capabilities of MAX IV laboratory to study nanostructured materials has been done with the construction of the new multipurpose SAXS beamline I911-4 [1]. Such facility has been running since April 2011 and it's serving a broad scattering community of around 50 groups in Scandinavian and Europe in general. Mostly soft matter science projects (polymers and biological materials) are studied at the station.

In the present work we will show recent examples of research produced at I911-4 as well as in other stations of MAX-lab. Studies on muscles [2], bio-based materials [3] and inorganic nanostructured system [4,5] are described. The latter ones make efficient use of a broad spectrum of techniques available at MAX IV laboratory.

New and future developments at the I911-4 station as well as the perspective toward the new facilities coming at MAX IV are outlined. Common developments in close collaboration with the users' community are encouraged.

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Biomembranes for recovery of precious metals

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Cytoplasmic membranes of some bacterial species, such as *Desulfovibrio desulfuricans*, *Desulfovibrio fructosovorans* and *Escherichia coli* were used for recovery of Platinum group metals. Intact resting cells were employed. The outer membrane of the bacteria is highly transparent for ions and complexes of Pd and Pt. The nucleation of metal particles takes place on [Fe] and [Ni-Fe] hydrogenases present in the periplasmic space and on the inner membrane. The metals are reduced in the form of nanoparticles.

Here we demonstrate unusual properties of the nanoparticles, in particular ferromagnetism and spin-polarized state of Pd nanoparticles revealed by a range of techniques such as SQUID magnetometry, x-ray magnetic circular dichroism, muon scattering and magneto-optical imaging.

We argue that use of naturally occurring biomembranes is not only important for industrial-scale recovery of precious metals but also gives supported onto organic matter stable nanoparticles with unique properties that could be used in various nanotechnology applications.

Encapsulation of Paclitaxel into a composite based on iron oxides, hydroxyapatite and chitosan for breast cancer treatment

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Paclitaxel is a diterpene with recognized antitumor activity and very unique action mechanism that has proven to be effective against ovarian and breast tumor. However, it's known that its effectiveness directly depends on structural conformation, which can be modified when the drug is encapsulated. In this project, composites based on iron oxide nanoparticles (maghemite (γ-Fe_2O_3) and/or magnetite (Fe_3O_4) and also manganese and zinc ferrites $Mn_{(1-x)}Zn_xFe_2O_4$), hydroxyapatite and chitosan, containing encapsulated paclitaxel for drug delivery systems for breast cancer treatment, are synthesized.

Since the drug's effectiveness directly depends on structural conformation, which can be modified when the drug is close to ceramic and polymeric materials present in the composites we propose the study of the dynamic of these materials with inelastic neutron scattering, since it is a rarely used approach and can also indicate modifications in these molecules even if they are not detectable by direct structure measurements, such as neutrons and/or X-ray diffraction.

Proton transfer at the Q_o-site of the cytochrome bc_1 complex suggested by atomistic simulations

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Cytochrome (cyt) bc_1 complex, which is an integral part of the respiratory chain and related energy-conserving systems, has two quinone-binding cavities (Q_o- and Q_i-sites), where the substrate participates in electron and proton transfer. Due to its complexity, many of the mechanistic details of the cyt bc_1 function have remained unclear especially regarding the substrate binding at the Q_o-site. In this work we address this issue by performing extensive atomistic molecular dynamics simulations with the cyt bc_1 complex of *Rhodobacter capsulatus* embedded in a lipid bilayer. Based on the simulations we are able to show the atom-level binding modes of two substrate forms: quinol (QH_2) and quinone (Q). The QH_2 binding at the Q_o-site involves a coordinated water arrangement that produces an exceptionally close and stable interaction between the cyt *b* and the iron sulfur protein subunits. In this arrangement water molecules are positioned suitably in relation to the hydroxyls of the QH_2 ring to act as the primary acceptors of protons detaching from the oxidized substrate. In contrast, water does not have a similar role in the Q binding at the Q_o-site. Moreover, the coordinated water molecule is also a prime candidate to act as a structural element, gating for short-circuit suppression at the Q_o-site.

DIRECT ELECTRON-BEAM NANOPATTERNING OF TEFLON AF SURFACES FOR SITE-SELECTIVE FORMATION OF MOLECULAR PHOSPHOLIPID FILMS

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Teflon AF is a family of amorphous copolymers containing fluoroethylene and dioxole groups. Its splendid properties such as low surface energy, high optical transmission, chemical resistance and low autofluorescence, have made it a desirable surface for the fast generation of molecular phospholipid films, which are being evaluated for biosensing and single molecule spectroscopy. The possibility of confinement of chemical species to a surface-adhered 2-dimesional film, while keeping them mobile within the structure, circumvents many problems of volume-based flow systems (Czolkos et al. 2011).

Patterning the Teflon AF by common photolithography is limited to a few specialized processes with micrometer resolution, and it is still difficult to get nano-structured Teflon AF surfaces. It has been shown that a thin film of Teflon AF can be directly patterned by electron beam lithography without the need of further chemical development (Karre et al., 2009), where degradation of the fluorinated dioxole groups by electron beam radiation changes the hydrophobicity of the exposed area.

We have established that electron beam-exposed Teflon AF features far lower hydrophobicity, effectively preventing the spreading of phospholipid monolayers. By taking advantage of this functional difference, we established a nanostructuring protocol by means of electron beam frame exposure around a desired nano-scale region. The frame exposure separates desired surface areas of high hydrophobicity by a region of low hydrophobicity, confining the lipids in the framed surface areas. By applying this effective nanopatterning strategy on Teflon, we could successfully achieve guided monolayer lipid film formation on 75 nm wide lanes, which can be used as a new platform for single molecule studies.

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CO_2 adsorption and intercalation in aerogels and clay materials studied by SANS

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Geological storage of CO_2 in deep sedimentary rocks is widely proposed to reduce CO_2 content in the air and reduce the greenhouse effect. [1,2] To implement an effective and safe CO_2 injection on a larger scale, evaluation of the aquifer and overlying caprock by determination of their trapping capacity is needed. For this evaluation small angle neutron scattering (SANS) and pressure-composition-Temperature (pcT) studies have been made. The relevant geological structures may show large variations in composition (sandstone in a sedimentary basin, caprock, clays). Furthermore, CO_2 trapped in porous materials relies on different mechanisms of confinement that act on different time scales. Some important factors to consider are: 1) an impermeable caprock that keeps the fluid underground (*supercritical* CO_2 fluid); 2) the solubility of the CO_2 in the water; 3) adsorption into clay nanopores and intercalation into clay structure; 4) chemical reactions that bind the carbon in mineral form to the rock.

The studies were divided in two parts. In the first part, tporous Vycor glass and aerogel served as standard samples, and synthetic clays (sodium fluorohectorite and Laponite RD) were measured subsequently. In contact with sub-critical and supercritical (sc) CO_2 , porous Vycor glass (porosity ~28%) and aerogel (porosity ~96%) demonstrate two-phase and three-phase behaviour, respectively.

In the case of the aerogel, and unlike the Vycor + $scCO_2$ system, the change of I(q) vs. CO_2 pressure reaches a maximum and decreases at higher pressures. This behavior indicates the presence of a third "phase" – CO_2 of high density – adsorbed to the surface of the nanopores, in line with what has been observed earlier by Melnichenko et al. [3]. The synthetic clays: sodium fluorohectorite NaFH and Laponite RD behave similar to the Vycor glass + $scCO_2$ system. NaFH represents a two-phase system, although showing small "positive" deviation from linear dependence. Laponite also represents two-phase system.

In the second part the studied LiFH clay was surface modified using the organic long chained cation, CTAB, where the CTAB replaces the inorganic cations between the clay platelets, forming 4CEC LiFH. After modification the d_{001} spacing between the clay sheets increased from 1.2 up to 3.1 nm. These studies allowed us to check the intercalation ability of the clay.

In addition to the SANS studies mentioned above, recently pressure-composition-Temperature measurements have been performed in order to obtain a better understanding of CO_2 adsorption and intercalation in different clay materials.

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Functionalization of Protein-wires with hydrophobic materials

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We have recently developed novel methodology to prepare protein nano-wires incorporating various hydrophobic materials.[1,2] Examples are protein wires functionalized with phosphorescent organometallic complexes, fluorescent organic small molecules, as well as magnetic nanoparticles. In this poster the preparative method[1,2] and some applications[3,4] of the materials will be explained.

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A Microfluidic Temperature Probe

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A microfluidic pipette, operating by a hydrodynamically confined flow (HCF), can be utilized as a positionable open-volume and fluorescence based temperature measurement device suitable to support microscopy experiments. Using the solution switching capability of the device, we used two fluorescent rhodamines, which exhibit different fluorescent responses with temperature, and made ratiometric intensity measurements. The device primarily re-circulates a solution of the temperature-responsive fluorophore Rhodamine B (RhB), which is well known to exhibit an inverse dependency of its fluorescence emission intensity on temperature. We alternate RhB solution with Rhodamine 6G (Rh6G), which does not exhibit a dramatic dependence on temperature. By making a comparative analysis of the ratio of fluorescence intensity obtained from either solution as the temperature is changed, we are able to exclude all environmental factors such as pipette position, microscope and detector settings, and heating source variances. Furthermore, we utilized fluorescent thermometer to evaluate the temperature during thermal activation of heat-sensitive TRPV1 ion channels in single CHO cells, measured as a YO-PRO-1 uptake assay.

Reference: *Wegrezyn et al., Sensors 2013, 13(4), 4289-4302; doi:10.3390/s130404289*