3rd SFB1032 (MGK) Blockseminar

Prof. Oliver Lieleg (TU München)

Mucin glycoproteins for biomedical applications

All wet epithelia in the human body are covered with mucins, high molecular glycoproteins which constitute the main component of mucus. Those mucins combine a broad spectrum of interesting properties: they prevent bacterial adhesion to surfaces, lubricate surfaces and prevent wear and form hydrogels with selective permeability properties that e.g. block viral penetration.

In my lab, we purify those mucins from the mucosa of pig stomachs and generate mucin-based materials for different biomedical applications. I will discuss our approach to use mucins as components for wound gels [1], as biolubricants [2] or as components in artificial filter systems. I will also discuss strategies to tune the properties of mucins and mucin hydrogels by chemical and physical modifications, e.g. to achieve local penetration of mucus by artificial microswimmers [3].

[1] Nowald et al., *A Selective Mucin/Methylcellulose Hybrid Gel with Tailored Mechanical Properties,* revision submitted

[2] Crouzier et al., *Modulating Mucin Hydration and Lubrication by Deglycosilation and Polyethylene Glycol Binding*, Advanced Materials Interfaces, (2015)

[3] Walker et al., *Enzymatically Active Biomimetic Micropropellers for the Penetration of Mucin Gels*, Science Advances, in print

Prof. Irene A. Chen (University of California, Santa Barbara)

Information in the RNA World of early life.

The storage and propagation of biological information is one of the hallmarks of life. However, early replicating systems faced several difficulties related to information. How much information could be stored? Was that information meaningful for function? Does the ability to replicate information well interfere with functional activity? Using experimental systems of nucleic acids, we find that chemical mechanisms can resolve some of these issues, clearing the path for the emergence of replicators during the origin of life.

Dr. Maier S. Avendaño Amado (Harvard Medical School)

Painting life with DNA.

Understanding biological complexity at the nanoscale requires the development of new and more advance optical tools. Current super-resolution techniques offer unprecedented spatial resolution, however, a precise identification and quantification of multiple molecules that cannot be spatially resolved remains challenging. Together with my group of work I have developed and validated a quantitative multiplexing super-resolution approach, based on programmable autonomous blinking of a nucleic acid probe (technique named DNA-PAINT). I will first discuss the general principles for obtaining multiplexed 2D and 3D super-resolution imaging, for integer molecular counting, and for detecting specific endogenous protein interactions in cells. In the second part, I will present some biological applications of this new imaging approach for *in situ* visualization of single-copy regions of the genome in mouse fibroblasts, and finally for analyzing the complex interactions of 5 receptor tyrosine kinases (RTKs) simultaneously within their native cellular context in a breast cancer cell line.

Prof. Chase Broedersz (LMU München)

Cells in gels: Mechanosensing and active stress generation

Large-scale force generation is essential for biological functions such as cell motility, embryonic development, and muscle contraction. In these processes, forces generated at the molecular level by motor proteins are transmitted by disordered fiber networks, resulting in large-scale active stresses. While these fiber networks are well characterized macroscopically, this stress generation by microscopic active units is not well understood. We theoretically study force transmission in these networks, and find that local active forces are rectified towards isotropic contraction and strongly amplified as fibers collectively buckle in the vicinity of the active units. This stress amplification is reinforced by the networks' disordered nature, but saturates for high densities of active units. In the last part of this talk I will talk about the role of network disorder for the mechanosensing function of cells. In particular, I will show theoretical results to set physical bounds on the accuracy of mechanosensing of a cell migrating in a 3D tissue.

Dr. Nicolas Garreau de Loubresse (Wyss Institute at Harvard University)

Rethinking molecular engineering in structural biology

The classical structural biology techniques—X-ray crystallography and nuclear magnetic resonance—have given us a chemistry-level understanding of biological processes and enabled rational drug design. Over the last three years, advances in both software and hardware have revolutionized structure determination by cryo-electron microscopy, with the resolutions obtained beginning to rival those from X-ray crystallography. Together, these techniques have transformed the way we understand biological mechanisms but a large fraction of macromolecules still remains out of reach because of technical limitations. I will discuss the current and future development of molecular engineering applied to structural biology and present our progress in developing new tools to expand the capabilities of structural biology techniques using DNA nanotechnology.

Dr. Alex Borodavka (Astbury Centre for Structural Molecular Biology, University of Leeds)

Viruses as self-packing nano-containers: shedding light on the conformational dynamics of viral RNAs during genome encapsidation

RNA viruses are ubiquitous pathogens that infect organisms from every kingdom of life. Despite significant progress in structural biology, mechanisms of RNA virus genome packaging and assembly remain poorly understood. We have developed fluorescence correlation spectroscopy based assembly assays to monitor the conformations of viral genomes during assembly of simple ssRNA viruses. We have shown than interactions of RNA with cognate coat protein (CP) in two model viruses, cause a rapid collapse of their genomic RNAs during early stages of assembly. The collapse is caused by CP binding at multiple sites on the RNA, and is facilitated by additional protein-protein contacts. The specificity in RNA-CP interactions observed at low concentrations reflects the packaging selectivity in these viruses usually seen in vivo. RNA compaction by CP and cation-induced RNA condensation are distinct processes, implying that cognate RNA-CP contacts are required for assembly nucleation at low concentrations while charge neutralisation may work at higher concentrations in rather non-specific manner.

A different class of RNA viruses with segmented dsRNA genomes, including reoviruses and rotaviruses, possess segmented genomes, comprising 10-12 distinct double-stranded RNA molecules that are packaged into each virion. There is no plausible model explaining the highly efficient mechanism of this molecular process. Non-structural proteins encoded by these viruses are required for segment assortment in these viruses. Biophysical characterisation of conformational dynamics of large viral RNAs reveals fundamental mechanisms underpinning genome packaging and assembly in simple RNA viruses, as well as highlighting the role(s) of a non-structural RNA-binding protein in more complex dsRNA viruses.