CPP 25: Biomaterials and Biopolymers II (joint session BP/CPP)

Time: Wednesday 9:30–12:45 Location: H43

Topical Talk CPP 25.1 Wed 9:30 H43

Processing of recombinant proteins for biomaterials applications: about spider silk and more — •Thomas Scheibel — Universität Bayreuth, Lehrstuhl Biomaterialien, 95440 Bayreuth, Germany

Proteins reflect one fascinating class of natural polymers with huge potential for technical as well as biomedical applications. One well-known example is spider silk, a protein fiber with excellent mechanical properties such as strength and toughness. During 400 million years of evolution spiders became outstanding silk producers. Most spider silks are used for building the web, which reflects an optimized trap for flying prey. We have developed biotechnological methods using bacteria as production hosts which produce structural proteins mimicking the natural ones. Besides the recombinant protein fabrication, we analyzed the natural assembly processes and we have developed spinning techniques to produce protein threads closely resembling natural silk fibers. In addition to fibers, we employ silk proteins in other application forms such as hydrogels, particles or films with tailored properties, which can be employed especially for biomaterials applications.

CPP 25.2 Wed 10:00 H43

Nano-confined protein anchors, structured by STED lithography, probed by dSTORM. — •Richard Wollhofen[1], Moritz Wiesbauer[1,2], Kurt Schilcher[2], Jaroslaw Jacak[1,2], and Thomas A. Klar[1] — [1]Johannes Kepler University, Linz, Austria — [2]Upper Austria University of Applied Sciences, Linz, Austria

The ability to place individual proteins onto nano-confined structures plays a constantly growing role in bioscience, from basic studies in biology to development of nanosensors. One of the possibilities to generate sub-micrometer sized structures is direct laser writing (DLW) lithography. The resolution of DLW can be enhanced by stimulated emission depletion (STED) for assembly of polymeric structures down to several tens of nanometers [1]. Using a pulsed 780nm laser for two-photon DLW and a 532nm laser for STED, we are able to obtain structure sizes of down to 55nm and manufacture two clearly separated lines with 120nm distance [2]. The structures show good biocompatibility and allow an easy biofunctionalization with proteins down to the single protein level. We use direct stochastic optical reconstruction microscopy (dSTORM), which enables determination of protein density at a nanoscale level [3]. Combining STED lithography with dSTORM allows us to produce and characterize biocompatible structures, applicable to many biological assays. [1]J. Fischer et al.,Adv.Mat.,Vol.22,Nr.32,pp.3578-3582(2010); [2]R.Wollhofen et al.,submitted; [3]S. van de Linde et al.,Photochem.&Photobiol.Sc.,Vol.8,Nr.4,pp.465-469(2009);

CPP 25.3 Wed 10:15 H43

Influence of direct laser written three-dimensional topographies on osteoblast-like cells — •Judith K. Hohmann[1], Erik H. Waller[1], Rainer Wittig[2], Rudolf Steiner[2], and Georg von Freymann[1] — [1]Physics Department and Research Center OPTIMAS, University of Kaiserslautern — [2]Institute for Laser Technologies in Medicine and Metrology (ILM) at the University of Ulm

Biological cells react to various signals of their environment. While biochemical pathways have been investigated for decades, the influence of physical characteristics of the cellular environment has only been studied in the very recent past [1]. Especially information on the interaction with three-dimensional structures is barely available, since common chemical and/or physical surface treatments (e.g. acid-etching, sand blasting) lead to randomly shaped surface topographies. In general, results generated in such two-dimensional systems can hardly be transferred to natural, three-dimensional conditions.

Our well-defined three-dimensional templates are fabricated by direct laser writing and coated with titanium dioxide via atomic layer deposition. This allows us to provide biocompatible substrates.

We aim at understanding the relation between various three-dimensional structures and viability parameters of osteoblastic cells. To observe cellular behavior, SaOs-2 osteosarcoma cells are seeded onto the structures in order to test proliferation, morphology, adhesion and differentiation via fluorescence and staining techniques. These results might lead to novel dental implant surfaces which promote osseointegration. [1]Nikkhah et al. Biomaterials 33 (2012) 5230-5246

CPP 25.4 Wed 10:30 H43

Biocompatibility of Fe-Pd ferromagnetic shape memory alloys - influence of surface roughness and protein coatings — •Uta Allenstein[1,2,3], Yanhong Ma[2], Ariyan Arabi-Hashemi[2], Stefan G. Mayr[2,3,4], and Mareike Zink[1] — [1]Division of Soft Matter Physics, Institute for Experimental Physics I, University of Leipzig — [2]Leibniz-Institute for Surface Modifications (IOM) — [3]Translational Center for Regenerative Medicine (TRM), University of Leipzig — [4]Faculty of Physics and Earth Sciences, University of Leipzig

Recent decades have seen a huge turn in implantology and biomaterial development towards regenerative medicine. The approach in orthopedic surgery is no longer to just replace damaged tissue by a passive implant that evokes the least possible interference with biological tissue, but rather to provide active stimulation and actuation. Fe-Pd ferromagnetic shape memory alloys are a promising new class of smart materials with a unique set of properties ideal for biomedical applications, including superelasticity, magnetically switchable strains and biocompatibility. In this study the latter was shown by in vitro experiments with NIH 3T3 fibroblasts, MCF 10A epithelial cells and HOB osteoblasts on vapor-deposited single crystalline $Fe_{70}Pd_{30}$ thin films and roughness graded polycrystalline splat-quenched samples. Proliferation, adhesion and morphology were assessed on substrates of different surface roughness and different adhesive coatings, such as fibronectin, laminin and poly-L-lysine, as well as RGD peptides.

CPP 25.5 Wed 10:45 H43

Sorption of proteins to charged microgels: characterizing binding isotherms and driving forces — •Cemil Yigit, Nicole Welsch, Matthias Ballauff, and Joachim Dzubiella — Soft Matter and Functional Materials, Helmholtz-Zentrum Berlin, Hahn-Meitner Platz 1, 14109 Berlin, Germany

We present a set of Langmuir binding models in which electrostatic cooperativity effects to protein sorption is incorporated in the spirit of Guoy-Chapman-Stern models, where the global substrate (microgel) charge state is modified by bound reactants (charged proteins). Application of this approach to lysozyme sorption to oppositely charged core-shell microgels allows us to extract the intrinsic, binding affinity of the protein to the gel, which is salt-concentration independent and mostly hydrophobic in nature. The total binding affinity is found to be mainly electrostatic in nature, changes many orders of magnitude during the sorption process, and is significantly influenced by osmotic deswelling effects. The intrinsic binding affinity is determined to be about 7 kT for our system. We additionally show that Langmuir binding models and those based on excluded-volume interactions are formally equivalent for low to moderate protein packing, if the nature of the bound state is consistently defined. Having appreciated this, a more quantitative interpretation of binding isotherms in terms of separate physical interactions is possible in future for a wide variety of experimental approaches.

CPP 25.6 Wed 11:00 H43

Diffusion and Adsorption of Proteins in Mesoporous Environments — •Sebastian Mörz[1] and Patrick Huber[2] — [1]Experimental Physics, Saarland University — [2]Materials Physics and Technology, Hamburg University of Technology

In the recent years, several studies discussed the encapsulation of biomolecules in mesopourous materials and its potential applications in e.g. protein chromatography or as novel means of controlled drug release. Both the diffusion of biomolecules under such confinement and the interaction with the surface of the host material are crucial to these applications.

In this study, we examine the adsorption of bovine heart cytochrome c onto the pore surface of the porous silica material SBA-15. Comparison between the folded and unfolded state of this protein allows us to separate the contributions from the different interaction mechanisms involved i.e. coulombic and hydrophobic interaction. Furthermore, we attempt a qualitative validation of the Stokes-Einstein equation for the diffusion of proteins in a porous anodized aluminum oxide membrane and its applicability for protein separation.

15 min break

CPP 25.7 Wed 11:30 H43

FACS-sorting of particles to reduce the data variance in Optical Tweezers assisted Dynamic Force Spectroscopy measurements — •Tim Stangner[1], David Singer[2], Carolin Wagner[1], Christof Gutsche[1], Olaf Ueberschär[1], Ralf Hoffmann[2], and Friedrich Kremer[1] — [1]Universität Leipzig, Institut für Experimentelle Physik I, Linnéstraße 5, 04103 Leipzig, Deutschland — [2]Biotechnologisch-Biomedizinisches Zentrum Leipzig, Fakultät für Chemie und Mineralogie, Deutscher Platz 5, 04103 Leipzig, Deutschland

By combining Optical Tweezers assisted dynamic force spectroscopy experiments with fluorescence activated cell sorting (FACS), we demonstrate a new approach to reduce the data variance in measuring receptor-ligand-interactions on a single molecule level by ensuring similar coating densities. Therefore, the carboxyfluorescein-labeled monophosphorylated peptide tau226-240[pThr231] is anchored on melamine resin beads and these beads are sorted by FACS to achieve a homogeneous surface coverage. To quantify the impact of the fluorescence dye on the bond parameters between the phosphorylated peptide and the corresponding phosphorylation specific anti-human tau monoclonal antibody HPT-104, we perform dynamic force spectroscopy and compare the results to data using unsorted beads covered with the non-fluorescence peptide analogue. Finally, we demonstrate that the data variance of the relative binding frequency is significantly decreased by a factor of 3.4 using presorted colloids with a homogeneous ligand coating compared to unsorted ones.

CPP 25.8 Wed 11:45 H43

Thermal vibrations reduce the efficacy of sacrificial bonds — •Soran Nabavi[1], Matthew J. Harrington[2], Peter Fratzl[2], Oskar Paris[1], and Markus A. Hartmann[1] — [1]Institute of Physics, Montanuniversitaet Leoben, Leoben, Austria — [2]Max Planck Institute of Colloids and Interfaces, Department of Biomaterials, Potsdam, Germany

Mussel byssal threads are a fascinating biological material combining high stiffness, toughness and extensibility. Experimental studies suggest that these outstanding properties are achieved by using so called sacrificial bonds (SBs) which are weaker than the covalent bonds holding the structure together and that can form and open reversibly [1]. The SBs break before the covalent bond rupture, providing hidden length and allowing for efficient energy dissipation.

In this study computer simulations are used to investigate the effect of SBs on the mechanical properties of a single polymeric chain. Special emphasis was put on the interplay of covalent and sacrificial bonds and the effect of thermal vibrations that have been largely overlooked in the description of SBs so far. In a simple setting with only one SB it is found that molecular chain fluctuations reduce the efficacy of SBs. Even for SBs with rather high binding energies of ˜1 eV backbone fluctuations lead to a rupture of SBs before external loading sets in. Thus, the theoretical strength of SBs is reduced more than a factor of two. This effect increases with increasing polymeric chain length and with increasing temperature.

[1] M. J. Harrington et al., J. Struct. Biol. 167, 47 (2009)

CPP 25.9 Wed 12:00 H43

Benchmarking the water-peptide interaction — •Sucismita Chutia, Mariana Rossi, and Volker Blum — Fritz-Haber-Institut der MPG, Faradayweg 4-6, 14195 Berlin

The interaction between water molecules and the hydration sites of peptides is critical for any quantitative modeling of solvated peptides. We address this interaction for the successive hydration of two peptides for which accurate experimental reference data exist: Ac-Ala_5-$LysH^+$ (non-helical) and Ac-Ala_8-$LysH^+$ (helical). In particular, finite-temperature Gibbs reference water binding energies ΔG_0 and equilibrium constants are known [1,2]. However, earlier force-field predicted preferred water binding sites do not agree with one another. We present an exhaustive first-principles study (density-functional theory based on the van der Waals corrected PBE functional) that demonstrates [3]: (i) There is a close competition between possible hydration sites (protonated carboxyl group or ammonium group). The preferred first hydration site breaks an intramolecular bond of the ammonium group in the unsolvated molecule. (ii) Calculated ΔG_0(T) are in remarkable agreement with experimental data. Lowest-energy H_2O H-bond networks are predicted for up to five H_2O molecules, and the connection to the solvated state is explored by ab initio molecular dynamics with up to 152 H_2O molecules. [1] Liu, Wyttenbach, Bowers, IJMS. *236*, 81 (2004) [2] Kohtani, Jarrold, JACS. *126*, 8454 (2004) [3] Chutia, Rossi, Blum, JPCB DOI: 10.1021/jp3098268

CPP 25.10 Wed 12:15 H43

Biomolecular translocation through nanopores: from statistics to real DNA conformations — •Maria Fyta[1], Simone Melchionna[2], Sauro Succi[3], and Efthimios Kaxiras[4] — [1]Institut für Computerphysik, Universität Stuttgart, Germany — [2]IPCF-CNR, Università La Sapienza, P.le A. Moro 2, 00185 Rome, Italy — [3]IAC-CNR, Via dei Taurini 19, 00185 Rome Italy — [4]Department of Physics and School of Engineering and Applied Sciences, Harvard University, Cambridge MA 02138, U.S.A

We apply a multiscale computational scheme to model a biomolecule translocating through a narrow pore, an intensively studied subject due to its variety of applications such as ultra-fast DNA sequencing. The model uses a mesoscopic lattice Boltzmann method to treat the solvent and a Molecular Dynamics scheme to deal with the biomolecule. Our first results involve an anonymous polymer translocating in pure water. We have obtained important insight into the statistics and dynamics of the process. The translocation time exponent compares well with the experimental values, while we were able to monitor multiconformational translocation. As a next step, we include electrokinetic effects, i.e. ions, as well as a realistic quantum-mechanically derived potential for double stranded DNA. We are now able to reveal in more detail the structural conformations of the DNA molecule as well as the ion distribution within the pore. The results also provide a qualitative and quantitative understanding of the ionic conductance and DNA blockade as compared to the experiments. Our conclusions also involve the effect of the pore geometry in the DNA translocation process.

CPP 25.11 Wed 12:30 H43

Driving forces in corneocyte expansion: a geometric perspective — •Myfanwy Evans[1] and Roland Roth[2] — [1]Institut für Theoretische Physik, Universität Erlangen-Nürnberg, Deutschland — [2]Theoretische Physik, Universität Tübingen, Deutschland

The arrangement of keratin in corneocytes, the dead cells in the outer layer of mammalian skin, are likely a highly ordered packing of helical filaments. This specific geometric arrangement allows the exotic physical property of cell expansion on prolonged exposure to water to occur in a mechanically stable and reversible regime. We examine the solvation free energy of a water-like solvent filling the volume of the corneocytes around the hydrophilic keratin fibres by the morphometric approach to energy landscapes. We find that the energy minimisation drives the system to absorb water and expand where water is available. During this expansion, the elastic energy in the keratin intermediate filaments increases, and the balance of the two forces forms a natural limit for the expansion process and the system maintain full reversibility.