The SAXS measurements were performed with the GTPase domain of human Septin 2 (SEPT2G) at 0.5 and 1 mg/mL and temperatures from 4 to 45°C. At 4°C, our results demonstrate that SEPT2G is self-aggregated as a dimer at 0.5mg/mL, whereas dimers coexist with cylinder-like aggregates (36 nm-long and 12 nm-cross section) at 1mg/mL. At this temperature, the protein does not evolve over one hour of observation.
As the temperature was increased to 15°C we verified that, initially coexisting with the protein dimer and cylinders, a small amount of larger aggregates is also present in solution. However, the number of very large aggregates increases with time concomitantly with the decrease of cylinder amount in the solution. Analyzing the samples at 37°C it's not possible to observe cylinders anymore and the amount of dimers decreases from 50% to 20% in less than 1 hour. For 45°C this effect is even more accentuated: the percentage of dimers is only 6% in solution.
In conclusion, our results showed the coexistence of dimers of SEPT2G with small fibers and larger aggregates in solution that evolve not only with concentration and temperature but also with time.
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1942-Pos Board B712
Packing of Synthetic Polyelectrolytes in Viral Capsids
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Viruses are natural examples of ''nanomachines'' combining remarkable structural and functional properties which should inspire the design of artificial systems. Virus-based nanotechnology takes advantage of the natural circulatory and targeting properties of viruses in order to design therapeutics and vaccines that specially target tissues of interest in vivo. Viruses can act as nanocontainers and encapsulate synthetic materials for delivery applications. The conformation of encapsulated genome, the self-assembly mechanisms of protein capsid and the interactions between capsid and packed polyelectrolyte are still unclear. The aim of our project is to study the packing of synthetic polyelectrolytes into viral capsids devoid of genom. As already shown in previous investigations using cryoelectron microscopy [1], cowpea chlorotic mottle virus (CCMV) is capable to encapsulate polystyrene sulfonic acid (PSSA). In this work, we use the same system to probe the effect of the polymer molecular weight on its own conformation inside the capsid as well as on the capsid size and shape. The SANS combined with the contrast variation method allow to probe the structure of each component separately, and their mutual interactions as well. We present our first results and show how the capsid adjusts its size in order to accommodate the polyelectrolyte to encapsulate. Quite surprisingly, the capsid becomes smaller with polyelectrolyte in its interior than without any cargo.
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1943-Pos Board B713
Temperature Dependent Dynamics of Cytochrome P450cam from Elastic Incoherent Neutron Scattering
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The derivation of mean-square displacements from elastic incoherent neutron scattering (EINS) of proteins is examined, with the aid of experiments on camphor-bound cytochrome P450cam and complementary molecular dynamics simulations. It is shown that a q^4 correction to the elastic incoherent structure factor (where q is the scattering vector) can be simply used to reliably estimate from the experiment both the average mean-square atomic displacement (MSD) of the non-exchanged hydrogen atoms in the protein and its variance. The molecular dynamics simulation results are in broad agreement with the experimentally-derived MSD and its variance derived from EINS on instruments at two different energy resolutions, corresponding to dynamics on the ~100 ps and ~1 ns timescales. Significant dynamical heterogeneity is found to arise from methyl-group rotations. The easy-to-apply q^4 correction extends the information extracted from elastic incoherent neutron scattering experiments and should be of wide applicability.

1944-Pos Board B714
Probing Protein-Water Dynamics using Neutron Scattering on a Fully Deuterated Green Fluorescent Protein
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The role of hydration shell in dynamics and function of proteins remains a topic of active research. We employ neutron scattering spectroscopy to compare directly the dynamics of Green Fluorescent Protein (GFP) to the dynamics of its hydration water by measuring regular (hydrogenated) GFP in D2O and deuterated GFP in H2O. Analysis of the neutron scattering spectra demonstrates diffusive-like motion of water, while protein atomic motions at the same time scale appear to be localized. Another difference appears at lower temperatures: GFP dynamics exhibits first onset at T~100-120K due to methyl group rotation, while hydration water shows dynamic onset at T~170-190K associated with the glass transition temperature. Detailed analysis also reveals that hydration water suppresses GFP dynamics at lower temperatures. The obtained results present a novel view on coupling of protein and solvent dynamics.

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X-Ray Reflectivity and Diffuse Scattering Study of Effect of Ca2+ on Cushioned Lipid Bilayer
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The effects of Ca2+ on bio-membranes are of interest because of their role in several biological processes, including facilitating the membrane fusion process. In a recent study, we have investigated a cushioned [1] DPPC bilayer in water and in different concentration CaCl2 solutions via fluid exchange. We used a polymer (PAA) cushion deposited on a single crystal silicon substrate. Using a 22 keV X-ray beam at the beamline of Sector 8 at the Advanced Photon Source, we obtained high resolution reflectivity from the bilayer and cushion through water, and were also able to study the grazing incidence x-ray diffuse scattering. The effect of Ca2+ on the bilayer structure has been obtained from the reflectivity analysis, which shows the Ca2+ ions binding to the headgroups of outer leaflet of the DPPC bilayer. The diffuse scattering is used to obtain the physical properties such as the surface tension, bending modulus, etc. Combining both, we present a complete picture of how Ca2+ affects a DPPC bilayer in water at room temperature as a function of Ca2+ concentration.
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Emerging Single Molecule Techniques II

1946-Pos Board B716
Nerve Growth Cones as Sensing, Amplifying and Filtering Modules: A Single-Molecule and Microfluidic Approach
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Nerve growth cones (GCs) are chemical sensors that convert graded extracellular cues into oriented axonal motion. Ensuring a sensitive and robust GC response to directional signals requires the ability to amplify and filter external gradients. However, our knowledge of how these signal processing tasks are performed at the single cell level remains sparse. This is largely due to the limitations of conventional guidance assays that have precluded systematic measurements of the GC output response to variable input gradients. We developed a novel shear-free gradient-generating microfluidic device with a simple architecture that greatly facilitates the interface of cultured neurons with microcircuits. With this device, we probed the information-processing capabilities of single GCs during GABA directional sensing. By measuring at the single molecule level the polarization of GABAA chemoreceptors at the GC membrane as a function of the external GABA gradient, we found that GCs act as: (i) signal amplifiers over a narrow range of concentrations, (ii) low-pass temporal filters with a cut-off frequency independent of stimuli conditions. Furthermore, thanks to quantitative computational modeling, we related these systems-level properties to the saturable occupancy response and the lateral dynamics of GABAA receptors, and, thereby, provided an integrative view of individual GCs as sensing devices.