

sophisticated process and expensive instruments. Hence, We propose the method to use optical anisotropy and scattered light intensity as indices for accurate and robust discrimination between single AuNPs and dimers using single-particle polarization microscopy in aqueous solution. This method is called a direct light scattering(DLS) method. This is homogeneous, rapid low-cost, sensitive method. We tested several way to deduce distance between two AuNP in the step of fabricating AuNP dimers and evaluated these AuNPs to utilize DLS method in colloidal solution. In this presentation, we report the result and conclusion about the influence of inter-particle distance in our experiment.

1640-Pos Board B591

Optically Tracing Electrical Synapses with Proton Channel-Based Voltage Sensing Protein

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The field of optogenetics has extensively improved over the last couple years allowing optical detection of single action potentials [CITE PAPERS]. In order to improve voltage sensors, a search strategy using the highly conserved S2 trans-membrane motif sequence identified 8 novel voltage sensing proteins. These proteins were fused to the fluorescent super ecliptic pHluorin A227D and transfected to HEK293 cells. The voltage sensing domain of a proton channel from liver fluke resulted in a sensor, Pado, which gave large optical signal and also responded to the induced pH change in the cell. This proton channel exhibits inhibition by extracellular Zn^{2+} and shifted response curves in differing pH environments. Higher buffering of the internal solution resulted in diminished pH dependent fluorescence change. With the ability to controllably manipulate pH of a cell, gap junctions can be traced through a network of HEK cells. Pado also provides a tool to screen fps for pH and voltage related activities. Ultimately Pado would make it possible to map electrical synapses and voltage activity simultaneously.

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Using Exploratory Data Analytics to Identify Deficiencies in mCherry Red Fluorescent Protein and Suggest Improvements

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Red fluorescent proteins are a valuable class of genetically encoded indicators for in vivo imaging. Even so, development of brighter variants has stagnated despite extensive efforts. To address this need, we have generated a library of over two hundred thousand simulated structures of mCherry fluorescent protein variants. The atomic positions of non-hydrogen atoms was then correlated with the planarity of the mCherry chromophore, a key determinant of molecular brightness. These studies revealed geometric instability of the mCherry chromophore. Further, we identified ten positional changes in mCherry side chains that strongly correlated with changes in the mCherry chromophore conformation. Site directed mutagenesis of key amino acids was then used to improve the mCherry quantum yield by approximately ten percent. These results suggest that a data analytics approach can be applied to structural engineering of fluorescent proteins for the first time.

1642-Pos Board B593

Design of a Theoretical Model to Identify Specific ssDNA Aptamers for Biosensing Applications

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Aptamers are single stranded DNA or RNA oligonucleotides which bind to proteins, small molecules, cells or organs with high affinity and specificity. Aptamers are designed to work in such a way that they only bind to their target proteins/cells, leaving other neighboring and non-targeting proteins/cells intact. This targeting efficiency makes them a promising solution to the problem associated with affinity and specificity of other bioactive agents, reducing harmful side-effects and costs. Target specific Aptamers can be produced by Systematic Enrichment of Ligands by Exponential Amplification, a method usually known as "SELEX Protocol". However, this SELEX protocol needs a large pool of ssDNA/RNA library containing $\sim 10^{14}$ unique ssDNA/RNA sequences. Selection of only one aptamer for a target takes 15-18 rounds of selection and each round can take 2-3 days, which makes the process very time consuming and costly. In our model, we are trying to identify specific aptamer sequence for a specific target prior to experimental SELEX by a purely theoretical approach. This is done by rigorous theoretical studies on aptamer behavior in different biological environments. A molecular mean field theory is used which takes into account the size, shape, electrical properties and physical configurations of the aptamers along with the size, shape and physical properties of other chemical species associated with a particular biological environment. Coupling between different interactions, acid-base equilibrium and inhomogeneous dis-

tribution of different species along with the minimization of strong electrostatic repulsions gives us the most stable structure of aptamers specific to the target. So, this model can be a very efficient tool for designing aptamers to use in wide range of biosensing applications.

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Experimental Determination of Transition Dipole Moment Directions in Representative Fluorescent Proteins

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Fluorescent proteins are the workhorses of biological molecular imaging. Important imaging modalities (such as polarization microscopy or FRET imaging) exploit anisotropic optical properties of fluorescent proteins. The anisotropy (directionality) of optical properties of fluorescent proteins is described by a vector, the transition dipole moment (TDM). Despite the importance of molecular TDM orientation for quantitative structural interpretation of many imaging experiments, experimental data on TDM direction in fluorescent proteins is very limited. Here we present the results of our optical measurements on crystals of representative fluorescent proteins, as well as mathematical interpretation of these results, yielding information on the orientation of TDMs within the investigated fluorescent protein molecules.

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Glycine Protects Hepatocytes through a Chloride Independent Mechanism

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¹Medical University of South Carolina, Charleston, SC, USA, ²Institute of Theoretical and Experimental Biophysics, Pushchino, Russian Federation. Cytoprotection by the amino acid glycine was first described more than two decades ago. Accumulated evidence shows that glycine protects against hypoxia, ischemia-reperfusion and several toxins in hepatocytes, renal tubular cells and other cell types. The exact mechanism of the protection is still under investigation and multiple models have been proposed. One working model is that the protection exerted by glycine is mediated by inhibition of ligand-gated chloride channels. In our study, we used a genetically encoded chloride biosensor to test this working model. Isolated rat hepatocytes infected with adenovirus for expression of the chloride biosensor were treated with the mitochondrial uncoupler, carbonyl cyanide *m*-chlorophenyl hydrazone (CCCP), in the presence and absence of 3 mM glycine. With and without glycine, intracellular chloride concentration increased upon addition of CCCP. Without glycine, CCCP led to swelling, membrane bursting and death of hepatocytes in Krebs-Ringer-Hepes buffer. By contrast in the presence of glycine, cell death was markedly delayed. Moreover instead of swelling, hepatocytes underwent cell shrinkage after CCCP. These findings suggest that glycine alters cell volume regulation after CCCP and prevents cellular swelling leading to plasma membrane rupture and consequent loss of cell viability. DK073336, DK037034 and 14.Z50.31.0028.

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Small Angle Neutron Scattering Studies of Glucose Oxidase Immobilized on Single Layer Graphene: Relevant to Protein Microfluidic Chip

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Chronic diseases like diabetes has become more pervasive. Blood glucose monitoring is a valuable tool in the health management and consequently point-of-care devices like glucometers has now become indispensable. Since, obtaining sufficiently accurate and reliable measurements are very essential, a promising solution is to incorporate enzymes into the biosensor [1]. Enzymes like glucose oxidase (GOx) are ideal for consistent measurements because of their specificity and the ability to electrochemically transduce from the enzymatic reaction. Consequently, the presence of glucose in blood plasma can be detected by the change in the potential of the electrode adjacent to GOx during catalysis of glucose [2]. Further, the oxidation overpotential of the electrode