264-Pos Board B143
Engineering the protein-nanoparticle interface
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Nanoparticles have been conjugated to proteins to create unique imaging agents, multifunctional particles, and drug delivery vehicles. However, the biggest barrier for the success of these applications is understanding the interface of biomolecules with nanoparticles. Often conjugation of proteins and DNA with nanoparticles results in protein denaturation and non-specific adsorption, which are due to the many non-covalent interactions at the inorganic-biological interface. While development of new biological applications of nanoparticles has garnered a great deal of attention, the protein-nanoparticle interface has remained poorly characterized. As a result, insufficient understanding of the interface has limited the capabilities of nano-bio hybrids.
We present work in which we study the interface between inorganic nanoparticles of Au and CoFe2O4 and the protein cytochrome c, which is covalently linked to the nanoparticle. We devise a method to site-specifically label the protein, minimizing non-specific adsorption. We study the effect of nanoparticle ligand, nanoparticle material, and protein labeling site on the structure of the protein. Biophysical techniques such as quantitative gel electrophoresis, circular dichroism, and optical spectroscopy are used to characterize the structure of the protein in the conjugate. These experiments allow us to understand the chemical interactions involved in non-specific adsorption, and come up with general design rules for optimal conjugation. We determine that nanoparticle labeling generally destabilizes the motif containing the labeling site, and that when the nanoparticle is labeled on certain motifs, protein denaturation is not recoverable.

265-Pos Board B144
Combining Microfluidics, Electrophysiology, and Fluorescence Detection to Study Drug Transport Across Biomembranes
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The goal of this project is to develop a chip-based functional assay for studying multi-drug resistance (MDR) transporter proteins. The proposed assay combines electrophysiology with fluorescence spectroscopy on a microfluidic platform to yield new information about MDR transporter functions and their role in cancers. This apparatus and the information gained will facilitate drug screening for molecules that mediate or bypass MDR transporter mechanisms. Here, we highlight the progress made thus far in developing this chip-based assay.

266-Pos Board B145
A Realistic Model For The Water-amorphous Silica Interface: Insights Into The Electrical Double Layer And Bioengineering Applications
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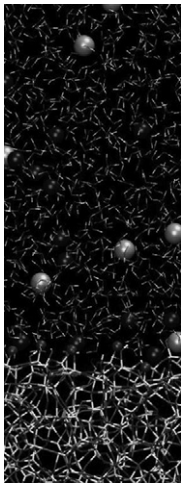
The physical and chemical properties of the amorphous silica-water interface are crucial for fundamental understanding of electrical double layer and electrokinetic phenomena, and for various applications including chromatography, sensors, metal ion extraction, and the construction of micro- and nanoscale devices for biomedical applications. The model reported here, which includes both dissociated and undissociated silanol groups on the surface, is a step toward a practical microscopic model of this important system. Our calculated value for the heat of immersion, 0.3Jm-2, falls within the range of reported experimental values (0.2-0.8Jm-2). The silica surface is characterized by hydrophilic and hydrophobic regions, depending on the statistical variations in silanol group density. This, and other properties, have been successfully benchmarked against ab initio MD simulations. We report structural and dynamical properties of the electrical double layer for various ionic strengths, testing venerable theories like the Gouy-Chapman-Stern model. We are extending our model to allow simulation of proteins, nucleic acids and other polymers near the surface.



267-Pos Board B146
1-d Lipid Bilayers On Nanotube And Nanowire Templates: Properties And Device Applications
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One-dimensional nanomaterials present an exciting opportunity for creating functional biologically-inspired structures because they have unique materials properties, dimensions comparable with the typical size of biological assemblies or individual molecules, and geometry suitable for integration into functional devices and assemblies. We have integrated carbon nanotubes and silicon nanowires with phospholipid bilayers in a ''one-dimensional lipid bilayer'' assembly in which a nanowire or a nanotube is shielded by a continuous fluid lipid membrane. We will discuss the structure and properties of this bio/nanomaterial assembly, as well as its electrochemical characterization and application in bio/nanoelectronic devices utilizing functional membrane proteins.

268-Pos Board B147
Examining the Role of Neuregulin-1 in Synaptogenesis Using Microfluidics
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Determining the molecular mechanisms behind synaptogenesis (synapse formation and maintenance) is of great importance for understanding higher brain function as well as disease states such as Alzheimer's and muscular dystrophy. Neuregulin-1 (NRG-1), a nerve derived protein, was isolated based on its ability to stimulate new acetylcholine receptor (AChR) formation on muscle. This molecule has been hypothesized to enforce the high density of AChRs on the post-synaptic membrane in neuromuscular synapses, however, its role in vivo has been difficult to study due to the early death of NRG and ErbB mutants. Therefore, we have developed a microfluidic system, mimicking a synapse by focally delivering nerve derived proteins to a cell chamber containing myotubes, to study synaptogenesis. Also, the device's focal delivery capacity coupled with patterning of the culture chamber surface allows us to ask questions with spatial variables. As a platform for our AChR kinetics studies, we have examined complex, aneural AChR clusters found on muscle and first reported by Kummer et al. in 2004. These features are good models of in vivo post-synaptic areas, because they have similar topologies, protein expression and developmental patterns. After staining with fluorescent bungarotoxin, we have found that neuregulin decreases the half-life of receptors in pretzels by 21.4%. We have also confirmed that NRG-1 increases receptor insertion into pretzels. Afterwards, we examined the extent NRG-1 activation travels in the long, multi-nucleated muscle cell using mRNA in situ hybridization.

269-Pos Board B148
A Simulation Study of Carbon Nanotube Interactions with Designed Amphiphilic Peptides
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There is great interest in exploiting the novel properties of carbon nanotubes (CNTs) for use in biology and medicine. For example, CNTs have potential application in drug delivery, cancer and gene therapy, and as biosensors. However, prior to their usage we need to develop methods to overcome the hydrophobicity-induced aggregation of CNTs. Recently, designed synthetic peptides have proven effective at dispersing CNTs. This approach has the significant advantage that the nature of the peptides coating the CNTs can be controlled by specifying the amino acid sequence. Hence, peptides can be designed such that the peptide/CNT complex may target specific tissue. One such designed synthetic peptide, nano-1,[1] folds into an amphiphilic α-helix in the presence of CNTs and leads to CNT dispersion. Here we implement molecular dynamics to investigate the self-assembly of nano-1 onto CNTs, using both a coarse-grained and atomistic approach. Using this multi-scaled method, we show that nano-1 interacts with CNTs in a preferential orientation. Furthermore, the charged surfaces of nano-1 facilitate inter-peptide interactions within the peptide/CNT complex, promoting helix stability.
[1]. Dieckmann, G.R. *et al.* Controlled assembly of carbon nanotubes by designed amphiphilic peptide helices. *J. Am. Chem. Soc.* **125,** 1770-1777 (2003).

270-Pos Board B149
Interaction of Fullerol C60(OH)20 with Nucleic Acids
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Functionalized fullerenes have received much attention during the recent decade in view of their potential use in in vivo imaging, drug transport, and even functioning as the drugs themselves as HIV-1 protease inhibitors, antioxidants, and neuroprotective agents [1]. Yet, the use of functionalized nanoparticles in

such applications is shadowed by the lack of reliable information on their fundamental interaction mechanisms with biological systems. This entails not only specific atomistic interaction mechanisms between individual molecules but also transformation and non-covalent functionalization of the nanoparticles used within a biological organism. For interactions between nanoparticles and RNA or DNA, interference with their regular functioning or induction of damage by, e.g. photocleavage, may offer new routes for biomedical engineering - but potentially also for a number of unwanted effects in cells and bacteria.
We have used atomistic molecular dynamics simulations to study the interaction of polyhydroxylated fullerenes C60(OH)20, i.e. fullerols, with nucleic acids ssRNA, ssDNA and dsDNA. The nucleic acids are modeled by oligomers of 20 arbitrary nucleotide (bp) sequences. The modeling provides atomic-scale information on the binding modes of the fullerols, as well as local structural deformation of nucleic acids by the fullerol binding. The data obtained from the simulations is compared to spectroscopic measurements on solutions containing such nucleic acids and fullerols.
[1] T. Da Ros and M. Prato, Chem. Commun., 1999, 663-669.

271-Pos Board B150
A Microdevice To Indicate Human Neural Stem Cell Differentiation Potential
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''*Stem cells*'' is a term used to describe primal cells capable of differentiating into a number of specialised cell types. Stem cells have potential importance for a range of clinical and cell-based therapies. However, major difficulties lie in identifying these stem cells from differentiated offspring due to lack of appropriate biomarkers.
Dielectrophoresis (DEP) is a non-invasive technique utilizing the induced motion of particles in non-uniform electric fields. Particles experiencing such forces can be made to exhibit a variety of motions including attraction to, and repulsion from, regions of high electric field by changing the frequency of the applied electric field. The main factors influencing the electrical properties of a cell are the surface charge, the membrane capacitance and the conductivity of the cytoplasm which combine to form an electrophysiological ''fingerprint'' of the cell.
Human neural stem cells (HuNSCs) are of interest because of their potential use for treatment of central nervous system injuries and disease. In this study we investigated two types of HuNSCs, which differ in their ability to generate neurons and glia, using DEP in a microdevice system. We have determined the relative contributions of the cells' membrane and cytoplasmic compartments to their overall behaviour in DEP. The results showed that there are significant identifiable differences in the specific capacitance of the membranes of both types of HuNSCs. Furthermore, we have found that the electrophysiological properties of the HuNSC populations changed over time, which correlated with the changing differentiation potential of the cells.
The work demonstrated a novel, label-free technique that predicted the neurogenic potential of HuNSCs by means of detecting differences in the membrane compartment between neurogenic and gliogenic human NSCs.

272-Pos Board B151
A Spectroscopic Monitoring Module Based on a Ceramic Microfluidic Platform
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A 3-dimensional mesofluidic biological monitoring module has been successfully designed and fabricated using a low-temperature co-fired-ceramic (LTCC) technology. This mesofluidic device consists of a network of microchannels and a spherical mixing cavity. The selection of appropriate commercially available ceramic tapes has been done with regard to their biocompatibility performance. Specific processing procedures required for the realization of such complex structure are demonstrated. Three dimensional numerical flow simulations have been conducted to characterize the concentration profiles of liquids at a specific measuring port and verified by experiment. The module was successfully applied to study complex chemical reaction kinetics complemented by mathematical modelling.

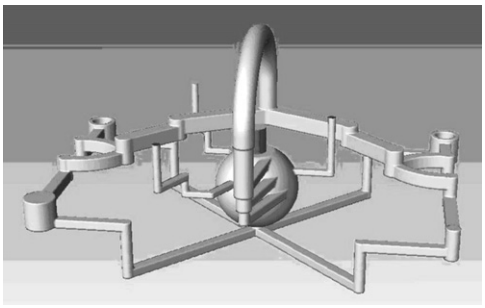

Fig. 1. Scheme of the monitoring module

273-Pos Board B152
Dynamics of Calcitriol Uptake and Signaling using Conjugated Quantum Dots
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Properties exhibited by Semiconductor Nanoparticles (Quantum Dots, (QDs)) make them powerful tools for the use as fluorescent probes for real time imaging. They are characterized by enhanced photostability, and high quantum efficiency compared to conventional organic dyes. Calcitriol, the active form of VitaminD_3, belongs to the group of steroid hormones. Past research lacked to develop active Calcitriol conjugates that are stable over an extended period of time to follow its cellular uptake and intracellular dynamics. In order to design an active conjugate only one hydroxy group within the structure of Calcitriol can be used for coupling to the QD. We successfully developed a bioactive Calcitriol-QD conjugate and imaged the uptake and the dynamics of Calcitriol into cells in real time. Our data show that it is stable for at least 48h at RT. We determined its interaction with the cell membrane and accumulation in the cell nucleus. VitaminD_3 can have both preventive and therapeutic effects by controlling cell growth, the cell cycle, apoptosis, and differentiation - a role greater than earlier views that focused on bone health and maintenance of calcium homeostasis. Epidemiological studies have found a significant association between low serum levels and low dietary intake of VitaminD_3 and the incidence, degree of malignancy, metastases, and mortality of cancers of the breast, prostate, colon, and ovaries. VitaminD_3 when bound to its receptor appears to have significant protective effects against the development of cancer. Based on this research, it has been proposed that taking VitaminD_3 could lower the cancer risk by 50% in colon cancer, and by 30% in breast and ovarian cancer. The mechanism for VitaminD_3's chemoprevention is not well-defined, but understanding how it works would provide vital information for targeting populations at high risk for developing hormone-dependent cancers.

Genome Packaging & Manipulation I

274-Pos Board B153
The Role of Electrostatics in Sequence Dependent Nucleosome Stability Analysis
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Nucleosome stability is strongly sequence dependent; yet the conformation of the DNA in the nucleosome is sequence independent. The nucleosome conformation is determined only by the histone octamer, and there is an energy penalty for deforming free DNA into this conformation. By comparing all atom molecular models and coarse grained elastic rod based models to experimentally determined free energies, we demonstrate that the contributions to the free energy of nucleosome formation arise from two sources: electrostatics and the local material properties of DNA.
The primary contribution to electrostatic energy in the nucleosome comes from the highly charged yet conserved phosphate backbone of DNA. Since all DNA sequences assume the same conformation in the nucleosome the electrostatic energy associated with the nucleosome conformation of DNA is largely sequence invariant. Thus the electrostatic energy contribution to nucleosome stability is defined not by the conformation of DNA in the nucleosome but by the conformation of free DNA. The free conformation rather than the bound conformation determines the binding free energy.
The material properties of DNA (elastic and van der Waals energies) behave quite differently. These energies are strongly dependent upon the sequence and conformation of the DNA. The conformation of free DNA tends to minimize these energies. When DNA binds the histone octamer and assumes the nucleosomal DNA conformation, the elastic and van der Waals energies associated with such deformation are also strongly sequence dependent. If the DNA sequence possesses intrinsic conformational properties that match those of the nucleosome or if the sequence is suitably flexible then the energy penalty is lower than for sequences which do not possess such characteristics. Both the free and bound states contribute to binding free energy.