Board B643

MBF, a binding factor regulated by transcription to target genes with functions which are important for bud growth and DNA synthesis, acts early in the cell cycle. Comprehensive understanding of MBF binding site is important for elucidating the complex events at the beginning of cell cycle. In this project, nuclear proteins were extracted from Saccharomyces cerevisiae cells which were grown for different time intervals after synchronization. A flow cytometer called FACScan was employed to ensure the cell populations are at distinct timepoints in the cell cycle. Surface plasmon resonance (SPR) based sensor, the Texas Instruments Spreeta, was then used to measure the binding of MBF binding site from RNR1 gene when exposed to nuclear proteins extracted at different timepoints. Highest DNA-nuclear proteins binding occurred at 15–30 minutes after synchronization. All other timepoints show similar DNA-nuclear proteins binding. Thus, we conclude:

1. Spreeta can be utilized to measure DNA-protein interactions.
2. Measurements verify that expression peak for binding of nuclear proteins from different time intervals after synchronization to MBF binding site occurs at the end of G1/S transition.
3. Measurements also verify that MBF acts as a repressor in other cell cycle phases.

1668-Pos Temperature dependent ionic conductance of OmpF: Effect of the confinement

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Board B644

The temperature dependence of the ion conductance through the outer membrane channel OmpF of *E. coli* was measured. For this a single trimer has been reconstituted into a planar lipid bilayer. We varied the temperature from −5 to 82 C and the salt concentration from 0.1 to 4M. The main contribution to the conductance stems from the ohmic part determined by the bulk conductance of the electrolyte for the given salt concentration and temperature. However, a temperature and salt dependent contribution is attributed to confinement effects. To understand such effects molecular dynamics simulations were performed. We observed a temperature dependent ion pairing that is enhanced inside of the nanopore. This could explain differences in the ionic conductance in confined environments.

1669-Pos Intrinsic Peroxidase-like Activity Of Ferromagnetic Nanoparticles

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Board B645

Nanoparticles that contain magnetic materials, such as magnetite (Fe_3O_4), are particularly useful for imaging and separation techniques. Since these nanoparticles are generally considered to be biologically and chemically inert, they are typically coated with metal catalysts, antibodies or enzymes to increase their functionality as separation agents. Here, we report that magnetite nanoparticles in fact possess an intrinsic enzyme mimetic activity similar to that found in natural peroxidases, which are widely used to oxidize organic substrates in the treatment of wastewater or as detection tools. Based on this finding, we have developed a novel immunoassay in which antibody-modified magnetite nanoparticles provide three functions: capture, separation and detection. The stability, ease of production and versatility of these nanoparticles makes them a powerful tool for a wide range of potential applications in medicine, biotechnology and environmental chemistry.

Biotechnology & Bioengineering

1670-Pos Extracellular Matrix Stiffness Influences Behavioral Decisions in Adult Neural Stem Cells

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Board B646

Neurodegenerative disorders such as Alzheimer's Disease, Parkinson's Disease, Huntington's Disease and others are devastating illnesses that result from the death of specific populations of cells in the central nervous system (CNS). Adult neural stem cells (ANSCs), which are present in the CNS throughout human life, have significant promise for protecting and regenerating tissue affected by such diseases. However, to date they have exhibited very limited differentiation in vitro into phenotypes that are attractive for neuroregeneration. Soluble factors known to control neural stem cell differentiation into neurons during development exert different effects on ANSCs; therefore, it is important to comprehensively explore alternate signals of the stem cell microenvironment that may exert effects on ANSC differentiation into target phenotypes. Here we explore the effect of mechanical cues from the extracellular matrix (ECM) on behavioral decisions of ANSCs. We created two-dimesnional polyacrylamide substrates with stiffnesses that span six orders of magnitude ($<$10 Pa to $>$100 kPa) as verified by micro- and macroscopic rheometry. We then covalently conjugated each substrate with the ECM protein laminin and cultured rat hippocampal ANSCs on these substrates in growth or differentiation media. We examined the relationship between ECM stiffness and the differentiation trajectories of ANSCs, measured by the expression of specific differentiation markers, cell morphology and multicellular architecture. Our results illustrate the importance of biophysical cues from the ECM in sculpting the development and assembly of ANSCs.

1671-Pos Optimization of Silver Colloid-Deposited High-Throughput Screening Plates for Rapid and Sensitive Cardiac Risk Assessment

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Board B647

In a typical hospital setting, cardiac marker immunoassays are usually run on serum (after blood separation) and can take $>$ 15 minutes to process per step per marker; the entire cardiac marker screening process therefore can take up to 1 hour. Commercially available tests for cardiac markers offer results from whole blood in approximately 15 minutes. However, these systems measure one sample at a time and have high initial and maintenance costs. In this regard, the development of new ultra-fast ($<$ 30 seconds) and sensitive immunoassays for cardiac markers, that can predict an AMI accurately, earlier and more economically, will significantly benefit human health.

We recently reported the application of a platform technology, namely "Microwave-Accelerated Metal Enhanced Fluorescence (MAMEF)" to a model protein assay in HTS well plates, where low concentrations of a target protein were detected in less than 30 seconds[1]

Here we present our findings on the reproducible deposition of silver colloids to high throughput screening (HTS) wells and their subsequent use in rapid cardiac marker assays, based on the microwave acceleration technology[1]. In this regard, we have successfully deposited silver colloids onto HTS wells already modified with cardiac marker specific capture antibody, without affecting the functionality of the antibody. Subsequently, we demonstrate that silver-colloid deposited HTS wells can be used in cardiac marker immunoassays for rapid and ultra-sensitive cardiac risk assessment.

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1672-Pos Microfluidic Emulsions As Picoliter Vessels For High-throughput Screening

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Board B648

We have developed a suite of microfluidic tools to study biochemical and biological phenomena in picoliter volumes at rates of thousands of reactions per second. We make and manipulate aqueous drops of a monodisperse water-in-oil emulsion within the channels of microfluidic devices. We can precisely control the size of drops from ten to tens of microns in diameter, and each drop acts as an independent picoliter-volume reaction vessel. We are able to split, recombine, and sort drops, and in this way perform many of the same experiments commonly done within multi-well plates, but at orders of magnitude higher rates and many orders of magnitude smaller volumes per reaction. Here we will present our work applying these tools to problems requiring extremely high-throughput screening: functional screening of enzyme catalysis, and also DNA sequence screening of microorganisms directly from the environment.

1673-Pos Single Cell Experiments Using Droplet Based Microfluidics

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Board B649

Microfluidic devices are used to produce monodisperse aqueous emulsion droplets in a continuous oil phase. The drops serve as individual, picoliter sized compartments for cells and enable us to study organisms on the single cell level while providing valuable statistical information. In addition, because the droplets encapsulate the cells within a confined volume, we are able to detect and screen cells which secrete specific molecules, as they can easily reach detectable concentrations within the droplets.

Here we present a high-throughput assay for monoclonal antibody screening. We encapsulate hybridoma cells in drops and incubate them for several hours to allow for antibody production. In order to detect the desired antibodies we co-encapsulate beads, which are coated with the particular target antigen, with the cells. This technique leads to attachment of solely the desired primary antibodies to the surface of the beads. A fluorescent secondary antibody is used to visualize the binding of the primary antibody. In this case, the fluorescence is localized on the bead surface, whereas it is diffuse in the droplet if the desired primary antibody is not present. We are able to detect this fluorescent signal in real time and sort the drops containing cells that produce the desired antibody. The cells can be recovered alive and cell lines for monoclonal antibody production can be established.

Currently, the production of monoclonal antibodies against a specific antigen is still very time and cost consuming. This novel technique is likely to reduce the time with for the screening process from several weeks to a few days and will therefore be very valuable for biomedical applications.

1674-Pos Local Heating of Phospholipid Bilayers with Gold Nanoparticles

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Board B650

Specific and direct delivery of drugs and bio-molecules to cells through the membrane is of great interest in biophysics and medicine but has been highly inefficient so far. Gold nanoparticles in particular show great promise for directing molecular transport through cell membranes. Ranging in size between 10–100 nm, they exhibit a unique plasmon resonance, resulting in increased light scattering and energy absorption. In this paper, we discuss the possibility of exploiting light induced heating of gold nanoparticles [1] for increasing the membrane permeability for large molecules.

Giant unilamellar vesicles (GUVs), 10–60 μm in diameter, provide a model system for the cellular membrane and were prepared from 1,2-Dioleoyl-sn-Glycero-3-Phosphocholine (DOPC) via the electroformation method. The plasmon resonance of gold nanoparticles incorporated into vesicle membranes can be imaged with dark field microscopy. The nanoparticles were prepared in various shapes (rods, spheres, cubes), sizes (20–100 nm) and with different surface chemistries (cetyl trimethylammonium bromide - CTAB and citrate). We find that these surfactants play an important role in the vesicle adhesion efficiency. CTAB forms bilayers around the gold and is readily incorporated into the GUV membranes. A too high concentration of gold nanoparticles results in vesicle rupture due to osmotic stress. Furthermore, we investigate the heating of the GUV-gold complexes by illumination with laser light. Increasing the laser intensity leads to rupturing of the bilayers. The intensity required for rupture is highly dependent on nanoparticle size and the number of gold nanoparticles in close proximity. This model system is also being used to quantitatively study the transport of biologically active molecules across the lipid membrane through specific and local cell heating.

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1675-Pos Gold Nano-Stoves for Microsecond DNA Melting Analysis

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Board B651

In diagnostics, medicine, genetics and biophysics, the melting analysis of DNA is a very important tool. Currently, typical time scales for a melting analysis range from several minutes up to one hour. This is due to the fact that the system, including the heating element, must be in thermal equilibrium during the measurement in most techniques reported so far [1], [2]. This severely limits the speed of the temperature ramp. A faster detection of the DNA melting point is highly desirable especially for high throughput DNA analysis. An additional requirement for a practically relevant DNA analysis method is the successful identification of mutants of the target DNA.

DNA bound gold nanoparticle aggregates are used as light absorbers to convert optical energy into thermal energy only locally. We exploit the characteristic plasmonic properties of such aggregates to optically induce and detect the melting of double stranded DNA. The results of pulsed optical experiments show that heating on a microsecond timescale is sufficient to melt DNA molecules. Only one single laser pulse is needed to distinguish between a perfectly matching target and a target with a single base pair mismatch. The clear identification of both target DNAs is possible even in a 1:1 mixture of the targets. Therefore we provide a very fast method for mutant identification with potential for multiplex and high throughput applications.

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1676-Pos High-Throughput Screening of Ion Channels in Lymphocytes for Diagnosis of Multiple Sclerosis

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Board B652

We present a high throughput assay to quantify functional activity of K+ ion channels in T lymphocytes from human blood, and we use this technique to screen ion channel activity for the diagnosis of multiple sclerosis (MS). Both early diagnosis and treatment of MS remain formidable challenges, but an emerging drug target for therapy is the voltage-gated Kv1.3 ion channel. Kv1.3 is expressed in high numbers on MS-specific T cells, and blocking these channels improved the course of an MS-model disease in rats. Here, we developed an automated, high-throughput assay capable of measuring Kv1.3 in >200 T cells from blood in parallel (a throughput ~20-fold higher compared to traditional techniques). We used this assay to screen Kv1.3 activity in T cells from 20 patients with MS and 20 age- and gender-matched healthy controls. Patients with progressive forms of MS (i.e. with chronic neurological symptoms, $N = 6$) had 25% higher average Kv1.3 activity in CD4+ T cells compared to healthy controls or relapsing-remitting MS patients ($P < 0.001$). By designing metrics that focused on CD4+ T cells with high Kv1.3, we were able to develop thresholds to diagnose correctly progressive MS patients with 100% sensitivity and 95% specificity. We were not able to correctly diagnose relapsing-remitting (RR) MS patients,

possibly due to the fact that all RR patients were in remission at the time of blood draw. Other factors measured, including Kv1.3 currents in CD8+ T cells and flow cytometry activation markers, did not show significant differences between MS and control populations. Measuring functionally high levels of Kv1.3 ion channel activity in CD4+ T cells in blood, therefore, may be a useful clinical diagnostic for quantifying T cell-mediated inflammation in diseases such as MS.

1677-Pos Smart Tissue Diagnosing Scalpel

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Board B653

We report on the development of a Smart Tissue Diagnosing Scalpel (STDS) designed to offer additional diagnostic/control functionalities to the surgeon. The STDS is capable of performing smart in situ multi-spectral diagnostics of the tissue under the scalpel and of controlling tissue removal in real time. We have demonstrated the feasibility of STDS by fabricating a miniature, pen-style scalpel delivering multiple focused laser pulses for tissue removal and illumination, simultaneously collecting reflected/scattered light for multi-spectral analysis. We also integrated smart algorithms and a graphical user interface into the scalpel for real-time tissue diagnostics and control of tissue removal in real time. The STDS prototype was demonstrated on multiple skin phantoms, and chicken breasts, where it made 1 mm incisions with a <0.5 s response time in diagnosing blood concentration $\sim$4 mm below the incision point. Recent developments include testing with animal models with a fully automated, ruggedized, miniature system capable of submicron diagnosis of tissue regions 5 mm in diameter around the incision point and our attempts at additional functionalities such as subdermal coagulation for bloodless surgery and efficient tissue debridement. The successful completion of our research will result in the first multifunctional scalpel that is capable of performing real-time tissue diagnostics and error-free surgery. STDS will perform in situ tissue diagnostics during surgery and allow the surgeon to control laser parameters including wavelength, amplitude, and pulse modulation for precise tissue removal with minimal secondary damage. The unparalleled precision of STDS in diagnosis and tissue removal will enable zero-error, bloodless surgery. The flexible fiber optic platform can be easily integrated with robotic systems for current and future remote telesurgery applications.

1678-Pos Coarse-Grained Molecular Dynamics Simulations of Carbon Nanotube Interactions with Detergent and Lipid Bilayers

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Board B654

The interactions between carbon nanotubes and detergent molecules/lipid bilayers is of potential importance in a range of biophysical and bionanotechnological applications. For example, it has been proposed that carbon nanotubes may be utilised in drug and gene transport, cancer therapy, and as biosensors.

However, an obstacle for the usage of carbon nanotubes is their affinity for one another, making it difficult to disperse them as individual tubes in aqueous solution. Recent experimental studies have shown that lysophospholipids (single-tailed phospholipids) provide superior solubility for carbon nanotubes compared to the solubility provided by nucleic acids, proteins, and other surfactants. Here we investigate the interaction of lysophospholipids with single-walled carbon nanotubes via coarse-grained molecular dynamics. We present compelling evidence that the mechanism of adsorption of these detergents onto a carbon nanotube is dependent upon detergent concentration. Furthermore, the chirality of the carbon nanotube influences the detergent wrapping angle for low detergent concentration. These findings advance our understanding of the mechanism of carbon nanotube solubilisation via detergent molecules.

We have also performed preliminary coarse-grained molecular dynamics simulations of carbon nanotubes interacting with lipid bilayers. Our motivation for this work is in the potential use of carbon nanotubes in delivering various cargoes to cells.

1679-Pos A High Bandwidth, Low Noise System for Single Ion Channel Measurements

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Board B655

Electronic Bio Sciences has developed a high bandwidth, low noise system for performing single ion channel recordings across planar lipid bilayers. Currently available ion channel measurement systems are limited by the head stage amplifier voltage noise acting on the capacitance at the amplifier input. This capacitance results in an increase in current noise that scales linearly with frequency (f) and as $f^{3/2}$ with bandwidth. In current ion channel measurement systems the input capacitance is dominated by the area of the lipid membrane or the shunt capacitance through the wall of the device supporting the lipid bilayer. The area of the bilayer and the effects of shunt capacitance can be dramatically reduced ($>10^5$ times) by employing the glass nanopore membrane (GNM) developed by the University of Utah [1]. To take advantage of the low capacitance and dielectric loss of the GNM, we have integrated an optimized discrete component differential amplifier with feedback to a GNM coupled to a compact fluid flow system. The resulting system has achieved RMS noise levels of $<$ 0.5 pA at 10kHz bandwidth and $<$ 8 pA at 100kHz bandwidth. The AC coupling of the amplifier provides considerable immunity to vibration and electrical interference, and the small area of the lipid bilayer enables membrane lifetimes of days to weeks without the need for vibration isolation. This new platform will open up the potential for making very high bandwidth electrophysiological measurements that were not previously possible.

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1680-Pos Tunable Affinity Ligands for the Separation of Proteins and Biomacromolecular Complexes

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Board B656

The success of solid-phase biopolymer synthesis has opened the door for the facile synthesis of chain molecules, including mixed oligomers, that cannot be synthesized in living systems. Tunable Affinity Ligands (TALs) are a class of such chain molecules that are rationally designed to partition among conformational states with binding affinities that can be modulated in response to environmental stimuli. In the construction of nucleotide-based TALs, for example, non-natural bases and linkers with a variety of designed chemical functionalities can be incorporated into either natural or unnatural backbones. Synthetic polymer chains can be inserted between polynucleotide regions to provide flexible hinge regions with tunable target binding and release properties. Reactive chemistries can be used to incorporate a diverse array of functional groups, including amino acids, oligopeptides and a variety of synthetic polymers. TALs can be designed with regions that are neutral, zwitterionic, negatively and/or positively charged with target-specific implications. In this presentation we will demonstrate the utility of TALs for the separation of high-value target proteins. A unique feature of these separations is the ability to bind and elute under mild, non-denaturing conditions. The combination of our application-directed molecular technology with convective interaction media (e.g., monolith technology) provides a powerful tool for analytical and preparative applications involving the separation of biologically important peptides, proteins, macromolecular complexes and cells.

1681-Pos The Effect of MRET Activated Water on Microbiological Culture *Eschrichia Coli K-12 a*nd on Complex Microbiological Associations

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Board B657

The article relates to detailed observation of the effect of MRET activated water with the modified molecular structure, physical and electrodynamic characteristics on metabolic activity and growth of conditionally pathogenic microbiological culture *Escherichia coli K-12 (E.coli)* and on metabolic activity of complex microbial associations. The microbiological investigation described in this article was conducted at Kiev Institute of Microbiology and Virology of Ukrainian Academy of Science. After 25 hours of experiment in **aerobic** environment it revealed significant inhibition of growth of *E.coli*: about 30 times in nutrient medium MRET activated for 30 minutes and about 300 times in nutrient medium MRET activated for 60 minutes. The metabolic (reductant) activity of *E.coli* reduced up to 3 times in 30 minutes activated water and up to 1.6 times in 60 minutes activated water during the first 6 hours of experiment in **aerobic** environment. In **anaerobic** environment the metabolic activity of *E.coli* practically did not change. In order to simulate the environmental conditions similar to the conditions in the digestive system the test in **anaerobic** environment was conducted on metabolic activity of complex microbial associations. It revealed that MRET water substantially increased reductant activity of microbial associations during the first several hours of experiment.

1682-Pos Coarse grained molecular dynamic simulations of C60 and its derivatives

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Board B658

The biophysical mechanism of the fullerene C60, together with its water-soluble derivatives has an important impact in biomedical applications, especially in the field of drug delivery. It is therefore of vital importance to fully characterise the influence of pristine C60, and its functional derivatives on biological systems. We will focus specifically on the interactions with biological membranes which contain a number of membrane proteins. Both experimental evidence and atomistic molecular dynamic simulations have shown that increasing the functional groups on C60 decreases its cytotoxic effects. Studies using atomistic simulations have provided us with detailed interactions of a handful of molecules of C60 (and its derivatives) with model membranes, but do not focus on the aggregated versions of C60, which are found in solution. Here, we report on coarse grained molecular dynamics simulations performed on pristine C60 and derivatives. Coarse grained simulations allow us to speed up the simulation time by reducing the complexity of the system and thus we are able to simulate multiple molecules of C60. We investigate what effects unimolecular and aggregated molecules of pristine C60 and its functionalised versions have on a model cell membrane (dipalmitoylphosphatidylcholine or DPPC). We have systematically functionalised C60 to address its cytotoxic effects with the lipid bilayer. We also look at the interactions of C60 with membrane proteins embedded in DPPC. The proteins chosen were ones which had pores large enough to accommodate C60. These simulations were performed to ascertain the degree to which C60 would "prefer" to enter the pore of the protein rather than the bilayer.

1683-Pos Photophysics of Cis-Trans Isomerization in Synthetic GFP Chromophores

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Board B659

In the last decade the *Green Fluorescent Protein* (GFP) and its mutants revolutionized molecular biology and became the most widely used genetically-encodable fluorescent probes. Recently new photoswitchable GFP mutants[1] and homologs[2] were also developed that opened the way to novel studies in the cellular environment and may even lead to GFP applications within information and communication technology. To date the detailed photophysical processes at the basis of photoswitching have not been fully clarified.

The use of synthetic GFP chromophores is a promising way to shed light on the photophysical processes underlying the photochromism of the respective proteins. Here, we investigate the photochromic behavior of synthetic chromophores in solution under different protonation states. We measured the variation of spectral properties before and after photoconversion, and compared them with accurate quantum chemistry calculations on various chromophore conformations. The results show that the observed chromophore spectral changes are associated with cis-trans isomerization.

The impact of these results on the description of the photophysics of photochromic GFP mutants will be discussed.

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1684-Pos A Microfluidic Platform for the Culture & Analysis of Single Cells

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Board B660

In modern biology, it is often assumed that populations of cells are composed primarily of average cells; cells that do not deviate significantly from an observational mean. This assumption is empirically convenient and until recently was necessary due to technological limitations. However, it is possible that ignorance of cellular individuality may lead one to draw incorrect conclusions, especially when the population under study is heterogeneous. Cells that exhibit significant deviation from the mean behavior can reveal important information which would be normally obscured by ensemble averaging techniques.

We have developed an array of microfluidic analytical techniques capable of studying the biochemistry of single cells [1,2,3]. Our current effort focuses on the development of a microfluidic device capable of sustaining a cell culture of a unicellular microorganism, Synechococcus, which can be resolved at the single-cell level. In our microchip, cells are captured hydrodynamically via a pressure-driven cross-flow of nutrient media. With efficient manipulation of the cellular microenvironment, the individuality of the cells' adaptive responses to stress conditions such as nutrient deprivation can be studied quantitatively using fluorescence microscopy. The design of imaging system with controlled illumination source as well as the use of different pumping mechanisms is described.

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1685-Pos A Platform For Controlling And Varying The Chemical Environment Around And Within Cells

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Board B661

A platform for controlling and varying the chemical environment inside and around biological cells has been developed. The platform comprises a computer-controlled motorized scanning stage and a microfluidic device that generates a laminar flow with a varying solute content. With the scanning stage, a cell can be rapidly translated with high precision between the different solution environments in the laminar flow. By designing the laminar flow and programming the scanning stage suitably, nearly arbitrary chemical waves can be generated around the cell. In order to enable controlled concentration variations not only around cells, but also inside them, we combine the platform with permeabilization of the cell membrane through electroporation with microelectrodes. We are currently investigating the possibility to use this system for titration of intracellular receptors and studies of enzyme activity. For this we have chosen the intracellular inositol 1,4,5-trisphosphate (IP_3) receptor and the enzyme alkaline phosphatase as model systems.

1686-Pos Microstructured Picoliter Cavities As An Approach Towards Highly Parallel, Low Noise Recording Of Single Ion Channels

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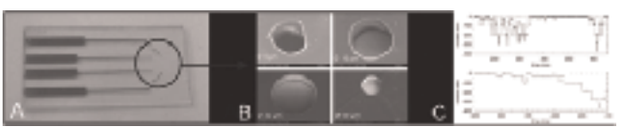
Highly parallel, low noise electrophysiological recordings of single ion channels in biological cell membranes are of interest both for basic research and drug development. Here, a microsystems approach is presented (see Fig.1A) which greatly simplifies the recording configuration and optimizes the electrical parameters governing noise. The lipid bilayer is formed on a picoliter cavity generated within a microstructured photochemical resist acting as a dielectric. On the bottom of each cavity a nanoporous Ag/AgCl microelectrode is placed (see Fig.1B).

Using standard photolithographical techniques this design allows for the generation of many such setups on one chip in a microarray, and is therefore well suited for highly parallel single channel recordings. Low-noise recordings (<1 pA rms @ 10 kHz) of currents mediated by alamethicin are shown as a first proof of principle (see Fig.1C) illustrating the potential of this novel approach towards high-throughput measurements of single ion channels.

Fig.1: ***A:*** *Complete microchip, with leads individually addressing each cavity.* ***B:*** *SEM images of cavities with different diameters.* ***C:*** *Current trace of a single alamethicin channel in a painted bilayer.*



1687-Pos Control of Cell Proliferation and Adhesion Dynamics by Microscale Topographical Cues

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Board B663

Regeneration of myocardial tissue through the use of synthetic scaffolds requires strategies to promote cardiomyocyte attachment while minimizing fibroblast proliferation and scar formation. Previous studies have demonstrated that a synthetic platform consisting of an array of microscale polydimethylsiloxane (PDMS)-based pillars ("micropegs") can accomplish both of these goals, yet the mechanism through which micropeg attachment inhibits fibroblast proliferation has remained a mystery. We hypothesize that this suppression of proliferation is related to the ability of fibroblasts to generate high contractile forces against the micropegs, which provide a three-dimensional attachment point for the cells. We show that when NIH 3T3 fibroblasts are cultured on a micropeg-based scaffold, a cell attached to a micropeg is statistically less likely to proliferate than its counterparts on flat regions of the scaffold; this difference is abrogated when cells are prevented from stressing the micropegs by pharmacologically inhibiting rho-associated kinase (ROCK) and myosin light chain kinase (MLCK). To gain mechanistic insight into these phenomena, we used time-lapse imaging to track lengths of tethers between the micropegs and flat control surfaces and in the presence and absence of MLCK and ROCK inhibitors. In the absence of the inhibitors, cells cultured on flat substrates displayed shorter maximum tether lengths than those attached to micropegs. MLCK inhibition significantly reduced maximal tether lengths of all cells, with the shortest average tethers observed on the flat substrates. Conversely, ROCK inhibition resulted in longer tethers overall, with the longest tethers observed on the micropeged substrates. These findings support a model in which attachment to the micropegs regulates the ability of the fibroblasts to generate strong contractile forces, correlating with a reduced propensity to proliferate.

1688-Pos Smart MRI Contrast Agents based on Superparamagnetic Iron Oxide Nanoparticles and Functional Nucleic Acids

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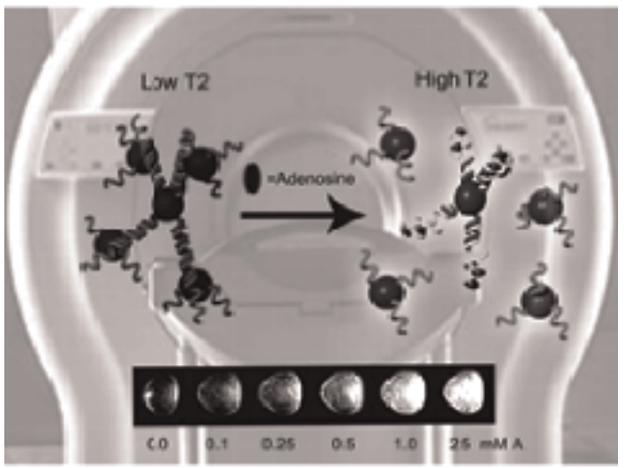
Board B664

Magnetic Resonance Imaging is a powerful method for non-invasive three-dimensional imaging of cells and human bodies. One active area of research in this field is development of novel MRI contrast agents, particularly smart agents for small molecules or biomolecular markers. Superparamagnetic iron oxide nanoparticles (SPIOs) are new class of contrast agents for MRI. Due to their biocompatible and biodegradable properties they have, recently, been widely used for research and clinical purposes. Functionalization of SPIOs with several biomolecules is possible due to the polymer coating at the outer shell. These properties of SPIOs make them great candidate for targeted agents for in vivo applications. Herein we report the first example of smart MRI contrast agents based on the aptamer technology and superparamagnetic iron oxides which could be applicable to any molecule of choice. We have built a smart "turn-on" MRI contrast agent for a small molecule, adenosine, which generates a bright signal as the analyte is recognized in the environment. We have also built a contrast agent for a protein, human alpha thrombin, which generates dark signal with MRI as the thrombin is detected in the system.



1689-Pos Nanoengineered Polymer Capsules: Tools for Controlled delivery and Site Specific Manipulation

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Board B665

Hollow nanometer-sized containers are of increasing interest in nanotechnology, since they can protect proteins, enzymes or drugs from hostile surroundings and provide an optimal microenvironment. Here we report on functionalized nanocapsules as intracellular reporters providing a new tool in cell biology. Cell active molecules, hormones, enzymes or reporter molecules may be hidden from the outside, protected against chemical and biological degradation, targeted to specific compartments inside a cell and released in a controlled manner. For example we loaded capsules with antigenic peptides and inject the capsule with electroporation. We describe here the laser-triggered release of peptides into the interior of a cell which is followed by their binding to MHC class I molecules, and the subsequent movement of the peptide-class I complex to the plasma membrane. In a separate project magnetic fluorescent liposomes were prepared by hydration of dried film of Egg-PC and Rhodamine-B labeled fluorescent lipids with a magnetic fluid (8nm average diameter iron citrated nanoparticles in buffer solution). The iron oxide nanoparticles were prepared by co-precipitation of a mixture of iron chloride salts. Citrate ions were adsorbed at the surface to provide a colloidal stability between pH 4 to pH 10. In a further series we prepared hydrophobic superparamagnetic nanoparticles and entrapped then in the liposomal bilayer. This technique bypasses the step of gel filtration. Further, these magnetoliposomes are coated with alternating polymer polyelectrolyte layers, resulting in magnetoliposome capsules. These capsules are introduced into CHO or Vero cells by either electropermeabilization or microinjection.

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1690-Pos Smooth Muscle-like Cells Induced by TGF-β1 from Adipose Tissue Derived Stromal Cells

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Board B666

Adipose tissue derived stromal cells (ADSCs) have been investigated as a good source of seed cells for tissue engineering. While it takes a long time for their being induced into smooth muscle-like cells. In the present study, we present a new method to rapidly induce ADSCs into smooth muscle-like cells.

ADSCs were isolated from adults after confirmed consent. They were identified to have the phenotype similar to that of bone marrow stromal cells detected by flowcytometry. And they could be induced to differentiate into osteogenic and adipogenic lineage cells in vitro. Furthermore, these cells were also induced to differentiate into smooth muscle-like cells by TGF-β1. Treatment with 5 ng/ml of TGF-β1 for 3 days, cell morphology changed from a fibroblast-like shape to a flattened and enlarged shape, and cells expressed multiple markers of smooth muscle cells, such as smooth muscle α-actin (α-SMA) and myosin heavy chain (MHC) identified by immuno-fluorescence. There isn't significant difference in the expression of α-SMA on the 3rd day and the 14th day demonstrated by western blotting. For gene transcription studies, six well-known genes which are markers of leiomyogenic differentiation (α-SMA, MHC, SM22, smoothelin, calponin, caldesmon) were studied by RT-PCR. Upon induction, transcription of α-SMA and MHC is increased on the 2nd and 3rd day, which is consistent with the results of protein expression.

Smooth muscle-like cells cultured on chitosan-gelatin composite films with varied ratios (w/w: 4/1, 3/2, 2/3) exhibit normal morphology and grow well. Proliferation of the cells increased with the increase of gelatin content. ADSCs could be induced to differentiate into smooth muscle-like cells on the films of chitosan-gelatin composite, and the cells grew well, still highly expressed α-SMA on the 7th day.

1691-Pos Blood Flow Mapping Through Spatial Temporal Image Correlation

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Board B667

We are continuing to develop an optical technique that can be used as the basis of a device to measure blood flow in the brain during

neurosurgery. This technique is based on detecting and analyzing optical fluctuations and will directly measure the velocity of micron-scale particles_such as red blood cells. It will complement existing technology in that it can be used on arterioles less than 1mm in diameter: a category of blood vessels in which it has been difficult to obtain quantitative flow measurements. The ability to detect the direction as well as the magnitude of flow and map these parameters over an area also sets this technique apart.

The ability to measure blood flow is important during neurosurgery and can be crucial for patient survival. Applications of blood flow measurement include detecting residual flow after aneurysm clipping and determining that flow has been reestablished after temporary blood vessel clamping. Surgical blood flow measurements are also useful to detect and correct hyper-fusion after artery transplant. Our optical technique could be helpful in any of these situations in which small blood vessels are present.

The analysis technique for fluid velocity measurement employed here is Spatial Temporal Image Correlation, a technique borrowed from fluorescence imaging in cells. This method is based on the fact that blood, when viewed on a small enough scale, is an inhomogeneous substance. Individual blood cells passing between a near-infrared light source and a detector will cause fluctuations in the transmitted optical signal. The speed at which the blood cells are traveling can be determined from these optical fluctuations. We have performed a series of computer simulations and experiments on phantom systems to test this techniques ability to map complicated flow patterns.

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1692-Pos Optical Tweezers with Microfluidic Temperature Control System

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Board B668

Temperature control is an important ingredient for manipulating single molecules and living cells in optical tweezers system. We set up an optical tweezers system and directed the temperature controlling laser spot into the microfluidic channel to control the temperature of a localized area. Due to the higher optical absorption coefficient of the coated metal, it will create a localized hot spot when illuminated with laser. Applying photolithography and sputter deposition methods we can create metallic micropatterns on coverslip. To observe the temperature effects, we added polystyrene beads in the laser trap area. Because of the thermophoretic depletion and convection (1), polystyrene beads accumulated rapidly in circular ring shape as laser spot began heating the coverslip substrate. By coating different metals, such as platinum and gold, they produced different temperature gradients and induced different accumulation rates. In this paper, we will present a temperature control system based on above study. Note that micropatterns could be easily integrated into microfluidic channel by photolithography method, so it doesn't require extra stages or holders to control the temperature stably (2). As the application, the integrated system could be applied to manipulate living cell in 37°C environment for long time observation in the microfluidic channel. This kind of system is also useful for study thermodynamic behavior of single molecule when isothermal condition is required.

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1693-Pos Surface Immobilisation And Characterisation Of An Engineered Protein Interface For Cell Growth

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Board B669

The interaction of thiols with gold is well understood, thus if proteins are modified to contain an active sulphur, such as a cysteine, it is possible to immobilise them on gold surfaces. In this study we have modified a vector protein to contain one cysteine residue, and shown using BIAcore and AFM analysis that it binds to gold. Further modification of the core protein has produced biologically active motifs that alter the behaviour of osteoblastic progenitor cells. The aim of this study was to show that immobilisation of this engineered protein on gold does not affect the functionality of the bioactive sequences. Two peptide motifs were incorporated in our scaffold protein, one from osteopontin, a known cell adhesion motif involved in osteogenesis and a second, BMP2-derived sequence which regulates osteoblast behaviour by activating the SMAD signalling pathway. The binding of these proteins to the gold surface was not significantly affected, as shown by BIAcore analysis. Surfaces created with the protein containing the osteopontin motif supported better osteoblast adhesion than vector control. The binding of osteoblasts to the surfaces changed with varying osteopontin concentrations. The surfaces modified with the BMP2-active motif initiated appropriate signalling cascades and the cells were able to form bone nodules without other osteogenic supplements in the media. Using uCP it is possible to direct specific areas of the surface to be cell adhesive whilst other areas are non-adhesive and using a GFP-tagged protein it is possible to show that the cells bind to the pillars of protein and not the surrounding gold or backfill molecules. This indicates that this technology could be used in the production of surfaces requiring display of specific motifs at controlled concentrations.

1694-Pos Optical Trapping Of Nanoshells Near Resonance

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Board B670

Nanoshells, nano-scale particles with a dielectric core and metallic coating that exhibit tunable plasmon resonances, have been used in photo-thermal tumor ablation therapy and for Stimulated Emission Raman Scattering spectroscopy. For these applications, it may be desirable to localize the nanoshells. One approach to localization is optical trapping. Theory predicts that the trapping force may be three to fifty times larger for trapping laser wavelengths near the plasmonic resonance than for wavelengths far off resonance. 1 We are investigating the optical trapping of nanoshells, and the possible enhancement of trapping due to the plasmon resonances. The resonance absorption of the nanoshells can be tuned by adjusting the ratio of the radius of the dielectric core, r1, to the overall radius, r2, which includes the thickness of the metallic coating. 2 Using back focal plane detection, we measure the trap stiffness of optical tweezers, from lasers at 784 nm, 853 nm, 980 nm, and 1064 nm, for trapped nanoshells with several different r1/r2 ratios. In addition, we measure single nanoshell absorbance spectra so that the resonance is not blurred by the dispersion in nanoshell size characteristic of a bulk sample.

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1695-Pos A Novel Method To Investigate Regulation And Expression Of Transport Proteins On A Functional Level In Mammalian Cells

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Board B671

Investigation of regulation and functional expression of transport proteins is time-consuming and in many cases of limited significance. Typical methods are based on m-RNA detection, antibody-mediated protein detection or radioactive uptake into cells or vesicles. Here we present a fast and robust method for electrical measurements of plasma membrane transport proteins using the SURFE^{2}R technology (IonGate Biosciences, Germany). This technology is suitable for electrical measurements of transport processes mediated by membrane proteins in many different membrane preparations such as purified plasma membranes, intracellular vesicles or proteoliposomes. We developed a protocol for fast and efficient adsorption of membranes onto a solid supported membrane (SSM) using whole cells. This protocol drastically reduces preparation effort and the amount of cells necessary for SSM measurements. Cells are directly centrifuged onto an SSM sensor and subsequently flushed at high flow rates in order to disrupt the cells. The remaining membrane fragments are strongly coupled to the sensor surface and can be examined for transport activity. Proteins within the membrane fragments are activated by solution exchange, and resulting transport currents can be detected down to single turnover precision.

Due to the integration of the membrane preparation step into the measuring device, time from cell culture to measurement is only minutes. We will present data demonstrating the application of this technology to characterize different PepT1 cell lines having inducible promotors in terms of background activity, expression level and time course of induced expression.

The technology is also applicable to clone picking, making direct functional testing of large numbers of cell line candidates possible.

1696-Pos Direct Detection of Membrane Channels from Gels using Water-in-oil Droplet Bilayers

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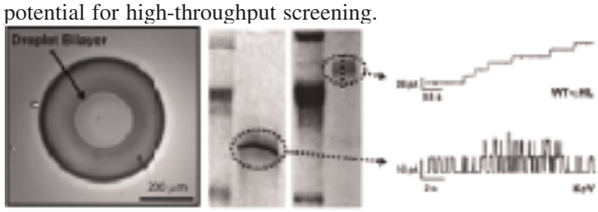
Board B672

We describe a new water-in-oil method of creating artificial lipid membranes, where bilayers are formed between aqueous droplets and hydrogel supports immersed in a lipid-oil solution. By scanning the droplet-on-hydrogel bilayers (DHBs) over the surface of SDS-PAGE gels we are able to insert and directly examine membrane proteins from their localised electrophoretically separated bands using single-channel recording. Using this technique we are able to examine low levels of endogenous protein from cell extracts without the need for over-expression. We also use DHBs to detect small molecules from hydrogels using the protein nanopore α-hemolysin.

Droplet bilayers show substantially enhanced stability compared to conventional planar lipid bilayers, and only require nanolitre volumes. Bilayer size and position can also be controlled during an experiment.

Hydrogel scanning with droplet bilayers provides a new method for the discovery and char–acterisation of ion-channels with the potential for high-throughput screening.



1697-Pos Target Smart Delivery: Multi-Compartment Carriers Containing Hollow Au Spheres with Release Triggered by NIR Laser

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Board B673

vesosome, a new multi-compartment structure consisting of drug-loaded liposomes encapsulated within another bilayer, is a promising drug carrier with better retention and stability compared with unilamellar liposomes. The current vesosome preparation takes advantage of the interdigitated phase of saturated lipids, which causes lipid bilayers to form flat, open sheets at low temperature, that close to form large unilamellar vesicles at higher temperatures. During this closure, the interdigitated sheets encapsulate other lipid vesicles or colloidal particles to form and thereby become the outer membrane of the vesosome. It has been shown that by adding the triblock copolymer to the interdigitated sheets made from dipalmitol-phosphatidylcholine, the vesosomes are improved to be in a proper size for clinical use. Meanwhile, hollow gold nanospheres with absorbance wavelength in the visible and near infrared (NIR) range are good candidates to trigger the content release from drug carriers. Vesosomes containing hollow gold nanospheres in their aqueous interior compartments have been verified by cryogenic electron transmission microscopy. Furthermore, 6-carboxyfluorescien has been encapsulated together with gold spheres to investigate the release from vesosomes irradiated with femetosecond laser pulses. Substantial amounts of carboxyfluorescein were released in response to only a few laser pulses. Our results suggest vesosomes containing hollow gold nanospheres, intrinsically with large loading capacity, are useful for laser induced delivery of therapeutic agents and other applications of lasers in biological systems.

1698-Pos Fully Automated High Throughput Membrane Formation

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Board B674

Reconstitution of pore and channel proteins in artificial lipid bilayer membranes is of considerable interest scientifically and also has sensing and pharmaceutical discovery/screening applications. Traditionally freestanding lipid bilayers are made individually by a skilled operator, rendering them unsuitable for high throughput applications. We are developing an automated, highly parallel platform for lipid bilayer formation to address these shortcomings. This system creates lipid bilayers by using gravity as a driving force to bring two self assembled monolayers in contact with each other, spontaneously forming a quality lipid membrane. This is an adaptation of the inverted phase method of Funakoshi et al to a vertical orientation in which a low density lipid-containing solvent phase floats atop an aqueous phase. The dissolved lipids in the organic phase self assemble on the aqueous phase causing a lipid monolayer to form at the interface. Dispensation of an aqueous droplet within the upper solvent phase results in the sinking of this droplet until it rests on the initial interfacial boundary and forming a lipid bilayer. Since gravity drives the organization and association of these phases, the only process required is fluid dispensation, which is easily done robotically. These membranes are high quality (Gohm resistances) and support the incorporation and measurement of channel proteins at the single molecule level. The reproducibility and yield of the membranes are very high. We report our initial measurements with this system as well as our work to develop it as a platform for high throughput screening of ion channels.

1699-Pos Analysis Of The Biophysical Properties Of Sodium Channel Nav1.5 Using An Automated Electrophysiology System

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Board B675

In recent years, automated patch clamp systems have been rapidly adopted by pharmaceutical and biotechnology companies involved in ion channel drug discovery. The applications of this technology have been for primary screening of directed libraries, hit-to-lead programs, compound profiling/lead optimization and cardiac safety studies. The amplifiers used by the PatchXpress® 7000A are based on a proven conventional patch clamp amplifier, the MultiClamp 700A. Therefore the PatchXpress 7000A has the ability to perform detailed biophysical characterization of ion channels. In our laboratory, we used a sodium channel to test the accuracy and limitations of PatchXpress 7000A on collecting biophysical parameters.

One of the limiting factors in the characterization of ion channels with rapid kinetics is the series resistance Rs. The PatchXpress 7000A reduces voltage errors and speeds voltage control in two different ways: by pneumatically minimizing Rs, and by electronically compensating the residual resistance. In our studies, we pneumatically reduced Rs below a threshold value and applied 80% series resistance compensation. With Rs effectively minimized by these methods, we recorded currents from the fast cardiac sodium channel (Nav1.5) on the PatchXpress 7000A. The measured biophysical properties, such as current-voltage (I-V) relationship, conductance-voltage relationship (G-V), inactivation time constant and half-inactivation potential are in good agreement with published values obtained in conventional patch-clamp. In summary, PatchXpress 7000A provides a fast, convenient and reliable way of characterizing ion channels, producing data of similar quality but at much higher throughput than conventional patch-clamping.

1700-Pos Proteomic Analysis to Identify Candidate Genes Influencing High-Density Lipoprotein Particle Size in Obese Individuals

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Board B676

Obesity is associated with a significant risk for cardiovascular co-morbidities. This effect is primarily mediated by an atherogenic

dyslipidemic profile, specifically a preponderance of small, dense high-density lipoprotein (HDL) particles. However, the molecular basis for the altered HDL particle size distribution in obesity is poorly understood. A genetic analysis in a family-based cohort of 2207 individuals identified a strong quantitative trait locus (QTL) (LOD = 3.15) for HDL median particle diameter on human chromosome 12. The interval contains 213 annotated genes, none of which have a known role in cholesterol or lipid metabolism.

Here, we describe a proteomic analysis using tandem mass spectrometry in conjunction with an enzymatic labeling technique to identify differentially expressed proteins in HDL particles. Recent research suggests that the HDL proteome is altered in dense HDL particles.

We isolated HDL fractions from plasma samples using non-denaturing fast protein liquid chromatography, with the use of a single Superose 6 30/100 GL column. A chloroform extraction procedure allows for the efficient isolation of lipid-embedded or associated proteins from HDL particles compatible with subsequent mass spectrometric analysis. Preliminary data will be presented on quantitative profiling of the HDL proteome. Using proteomic data, in conjunction with pathway and protein interaction analyses, we will highlight connections between alterations in protein expression and genes in the QTL region on human chromosome 12.

1701-Pos New Robust Method for the Detection of Acetylcholine Esterase Activity in Biological Materials

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Board B677

Acetylcholine esterase (AChE) degrades the neurotransmitter acetylcholine (ACh) thereby regulating the activity of cholinergic neurons and muscle. Toxicologically, AChE is a primary target of organophosphate and carbamate insecticides and nerve agents. Present methods for the detection of AChE activity are either costly, time-consuming, or they depend on colorimetric measurements of acetylcholine analogues.. This work describes the development of a robust system for measurements of AChE activity in biological materials via ACh-sensitive electrodes and the SURFE^{2}R Technology (IonGate Biosciences, Germany). This technology is based on solid supported membranes and allows electrical measurements of transport processes mediated by membrane proteins. To produce ACh-sensitive electrodes, cell membranes containing the organic cationic transporter 2 from rat (rOCT2) were adsorbed to the gold surface of sensors. The measurements with the rOCT2 biosensors were performed with the SURFE^{2}R One setup. To allow the detection of ACh degradation through AChE, the measuring protocol was optimized to differentiate between ACh and choline in the solutions. Subsequently, the concept for detecting AChE activity was tested with recombinant AChE. As expected, kinetics of the ACh degradation depended on the applied AChE amount. Further experiments demonstrated the inhibition of AChE by the organophosphate paraoxon and the reactivation of the paraoxon-treated AChE by the antidote obidoxime. Finally, the protocol was adapted for measuring esterase activity in fresh pig blood. Also here, the kinetics of ACh degradation depended on the amount of applied blood. Moreover, blood esterase was inhibited by paraoxon and reactivated by obidoxime.

The described system uses ACh-sensitive electrodes to directly determine ACh in solutions. Hence, the system is independent of the optical properties of test solutions and allows the analysis of AChE activities in biological materials without laborious preparations.

1702-Pos A Novel Hybrid Myo-Electronic Interface For Single Cell Electric Capacitive Stimulation On Myotubes To Modulate Muscle Plasticity In Vitro

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Board B678

We employed semiconductor bioelectronics to design an artifical neuromuscular junction prototype which achieves focal electric capacitive stimulation (ECS) of muscle cells and together with agrin stimulation represents a model to study muscle plasticity and synaptogenesis. For this purpose we set up a model of electrical interfacing of individual muscle cells and silicon microstructures, as well as the assembly of elementary hybrid systems made by myotubes networks and semiconductor microelectronics which are able to reduce classical electrolytic effects of MEA technology and to achieve a higher spatial resolution. The myo-electronic junction is employed to stimulate muscle cells via ECS at different development stages and induces cytosolic calcium transients. Slow calcium waves are evoked on myoblasts while fast [Ca2+]i transients are induced on myotubes and muscle fibers. The hybrid junction elicits via chronic ECS a programming of muscle cells inducing muscle contraction maturation and muscle plasticity effects, such as NFAT-C3 nuclear translocation. In addition, in the presence of agrin, chronic ECS induces a modulation of AchR clustering which simulates an in vitro synaptogenesis.

1703-Pos Glucose Monitoring Using Dielectric Spectroscopy

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Board B679

Dielectric spectroscopic techniques have been increasingly used for studying the interaction of electromagnetic waves with biological materials. The dielectric properties of biomaterials provide information on the water content, charge content and physiological state of the samples. Changes in the level of glucose in blood result in

significant variations in their dielectric characterization in the radiofrequency range. Measurements can be made using a probe and an automatic network analyzer. This work proposes the use of dielectric data for glucose level monitoring. The results will enhance the development of instrumentation and methodology for effective glucose monitoring.

1704-Pos Light-controlled Manipulation Of Membrane Potential In Cells Using Qdot® Nanocrystals

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Board B680

Long-term, real-time monitoring of cellular activity is crucial for cell biology. Because of their noninvasive nature, optical approaches are best suited for this task. However, the ability to activate cells by changing their membrane potential without pharmacological or chemical intervention is a huge challenge for optical cell-based assays.

To address this problem, we are developing a light-controlled stimulation platform with semiconductor nanoparticles (quantum dots or Qdot® nanocrystals) utilizing their unique physical and photochemical properties. This platform is based on a photovoltaic mechanism generating free charge carriers (e.g. electrons) in nanoparticles upon light illumination. When photo-excited nanoparticles are placed in close proximity to a cell, free charge carriers create the cumulative electromagnetic field, thus modulating the cell membrane potential.

In our experiments, cells were cultured on top of multilayers of nanoparticles deposited on a glass substrate. Nanoparticles did not produce any adverse effects on cellular morphology and physiological responses. We were able to repeatedly depolarize the membrane in non-excitable cells and generate a light-triggered action potential in excitable cells upon light illumination.

Since the absorption spectrum of semiconductor nanoparticles is broad, they can be excited by any wavelength shorter than their emission maxima, thus making them compatible with various optical recording methods. We have designed an integrated optical assay that combines optical stimulation (via a nanoparticle-coated substrate) and optical recording (via Fluo-4). We performed these experiments on both hippocampal neurons and NG108 cells and demonstrated the increase of intracellular calcium concentration as a result of light-controlled depolarization.

In summary, in contrast to other methods of cellular stimulation, our nanoparticle-based optical activation platform allows to stimulate cells physiologically and repeatedly in a manner compatible with optical methods of registration thus providing a complete solution for cell-based studies.

1704.1-Pos Long Term Culturing Of Primary Adult Human Liver Cells Reveals An Existence Of A New Type Of Stem Cell In The Adult Human Liver

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Board B681

Growth patterns and interactions of adult primary human liver cells (PHLC) in long term culture condition have never been investigated, since there was no suitable culture medium to do such experiments. After preparing an original special medium for this purpose, growth patterns, interactions and protein and gene expression in PHLC had been observed in 4 months. The results of this experiment show that Epithelial to Mesenchymal Transition (EMT) in Hepatocytes occurred in a small amount. But there were mesenchymal type of cells, which are negative to CD34, CD45, CD14 and Albumin and had enormous ability to multiply under growth factors and cytokines. These cells go to quiescent condition and organize Embryonic Body (EB) like Liver Body (LB) during serum deficiency and again multiply in increased serum content of culture medium. They also answer to Embryonic signal peptides BMP-2,-4 stimulation by expressing Hepatocyte specific genes (Albumin, Transthyretin, alfa-1-Antitrypsin). A part of these cells positively immunostain to human Albumin in cell culture and in "Liver condition" in SCID mice liver. All these results bring to suggestion an existence of embryonic Mesoendodermal origin of stem cells in the adult human liver, which under special circumstances could differentiate to Hepatocytes and other tissue specific cells of Mesoendodermal origin. Excellent growth of these new Stem Cells in the special cell culture medium makes it possible to use these cells for bioengineering of tissues in vitro condition.

Molecular Mechanics & Force Spectroscopy

1705-Pos A Holographic Optical Tweezers Setup For Force Measurements On Biomaterials

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Board B682

In recent years, optical tweezers have proven their suitability in the field of single-molecule experiments. Micrometer-sized particles can be trapped and used as handles to manipulate the even smaller molecules, and the picoNewton forces that can be exerted and measured with this non-invasive technique lie in the force range of many biomolecular properties and events, including for example the mechanical forces exerted by molecular motors. When probing more complex systems, however, such as red blood cells or protein networks, the 3D character of such materials calls for more flexibility in manipulating trapped particles. With holographic optical