PI Name	Institution	Title	Grant	Years Awarded	Funding Institute
Bloom, David M	Alces Technology, Inc.	Microcontact Printer For Ophthalmic Tissue Engineering	R43 EY015029	2 years	NEI

This project will demonstrate a prototype microcontact printing aligner for use in ophthalmic tissue engineering. The performance of the microcontact printing aligner will be refined and validated by experiments conducted in collaboration with the Stanford Ophthalmic Tissue Engineering Laboratory. Functioning much like traditional rubber-stamping methods, microcontact printing is widely used to print molecules, such as growth factors or extracellular matrix proteins, in well-defined locations at the nanoscale. This project focuses on microcontact printing applied to a tissue engineering solution for age-related macular degeneration. Using microcontact printing on autologous lens capsule, the Stanford Ophthalmic Tissue Engineering Laboratory is developing a replacement retinal pigment epithelium for treating age-related macular degeneration. Current microcontact printing techniques depend strongly on the skill and ability of the researcher. A prototype tool will be built to improve the reproducibility of microcontact printing. This will require investigation into and enhancement of the stamping materials and techniques. Additionally, this tool will allow for printing of multiple layers, with alignment between the layers. A tool for aligned microcontact printing is a significant enhancement over the current techniques, and something that is neither commercially available, nor easily developed in most biological research facilities.

Daniels, Robert H	Nanosys, Inc.	Nanowire DNA Hybridization Sensor	R43 CA101567	2 years	NCI

The aim of this program is to develop a novel genetic analysis platform that will provide multiple cost, time and sensitivity advantages over conventional technologies. The ability to diagnose the susceptibility to diseases such as breast or colorectal cancer provides valuable information for affected individuals that can lead to early detection and treatment of the disease. Current technologies for identifying genetic mutations and polymorphisms are limited in their diagnostic utility because of the complex sample processing, amplification steps and expensive detection instrumentation. Most genetic analyses is thus relegated to sophisticated and costly testing laboratories. The nanotechnology enabled sensor described in this proposal will be capable of highly selective and sensitive detection of nucleic acid hybridization in an array based format without the need for sample labeling or amplification. Furthermore, instead of expensive optical detection this nanosensor will detect hybridization by changes in electrical conductivity that can be made with a simple, low cost, low power instrument compatible with a point of care or even portable sensor platform. This detection system is based on the discovery that nanoscale materials such as nanotubes and nanowires can act as field effect transistors (FET's) at room temperature. This effect is termed NanoChemFET and it works because the conductive properties of exquisitely sensitive nanowires are modulated by charges on the analyte molecule that act like a gate voltage in a conventional field effect transistor. Because captured DNA or RNA would deliver a defined number of negative charges, proportional to the number of nucleotides brought into the vicinity of the nanowire, nucleic acids are well-suited to sensitive, quantitative analysis by a NanoChemFET sensor. As described in this proposal we will develop processes to controllably manufacture nanowires and assemble them on functional devices. We will use these devices to test the sensor performance for sensing and discriminating specific DNA sequences. In phase I of the program we will detect hybridization of a specific DNA sequence and we will determine the basal sensitivity and discriminatory capabilities of the sensor. This will provide the baseline from which to develop multiple genomic analysis applications including expression arrays, SNP genotyping and the detection of infectious agents in an exquisitely sensitive, rapid and cost effective manner.

Doranz, Benjamin J | Integral Molecular | A Cell-Free System For Assaying Ion Channel Function | R43 GM068322 | 1 year | NIGMS

Ion channels function as signaling intermediaries that are involved in nerve signal propagation, muscle contraction, cardiopulmonary regulation, and cell metabolism. Their importance is underscored by the existence of dozens of ion channel-specific drugs that treat important neurological and cardiovascular diseases. Despite their importance in human health, ion channels remain complex and difficult proteins to study. Ion channels are topologically complex membrane proteins that span the lipid bilayer of the cell several times, often form oligomers, and contain complex regulatory elements within their tertiary structure. Living cells are typically necessary for ion channel assays because the cellular lipid membrane maintains both ion channel structure and the differences in ion concentration that are the basis of cellular electrical signals. However, cells have inherent limitations for nano- and micro-scale applications. A better way to study ion channels is needed within microfluidic drug screening devices, for detecting ion and voltage changes in subcellular compartments too small to be measured directly, and for emerging nanotechnology applications. The purpose of this proposal is to enable the measurement of ion channel function on the nano-scale for application within microfluidic devices and at subcellular locations. We have developed a novel technology, the lipoparticle that makes it possible to present ion channels and other membrane proteins in native conformation in a stable, nano-scale format. In research that has spanned the past five years, we have defined the basic characteristics of lipoparticles as drug discovery tools. We have incorporated ion channels into lipoparticles, but have had no way of measuring their ability to function. We propose here to develop a nano-scale sensor for detecting ion channel function.

Elghanian, Robert | Nanoink, Inc. | DNA Nanoarrays Printed Via Dip Pen Nanolithography | R43 HG002978 | 1 year | NHGRI

The goal of this research is to develop novel, biologically functional DNA nanostructures that dramatically enhance the reproducibility, sensitivity, and spatial density of chip-based DNA assays. These nanostructures will improve applications ranging from point-of-care diagnosis to genomic arrays used in basic research by enabling the development of next generation screening technologies that are faster, more sensitive, more reliable, and possibly more cost effective than those presently available in the life sciences market. To accomplish the stated goals, Nanolnk will develop a DNA patterning methodology based on Dip Pen Nanolithography (DPN) to generate sub-micron sized features of DNA on solid surfaces. This multidisciplinary effort will involve life and physical scientists at Nanolnk, MEMs and instrumentation engineers at our fabrication facility, in addition to support from outside experts in the fields of DNA microarrays and microfabrication. DPN, built upon the technique of Atomic Force Microscopy (AFM), allows one to deposit materials uniformly in a direct-write fashion on surfaces with nanoscale spatial precision. This strategy offers significant advantages over current microarray printing technologies that suffer from poor spot to spot reproducibility in terms of size, shape, and oligonucleotide density, as well as reproducibility across microarray slides. Preliminary work has demonstrated that the DPN technique can be used to deposit 12mer synthetic oligonucleotides on surfaces with extremely uniform sub-100 nm to several micron scale features. The DNA nanostructures formed robust films and exhibited selectivity in binding to complementary oligonucleotides. Thus, DPN can be used to generate uniform features of synthetic DNA far smaller than can be obtained with other spotting or photolithography techniques. In Phase I, Nanolnk will demonstrate feasibility of the DPN-based approach for generating sub-micron scale DNA nanostructures on glass surfaces. The resulting nanostructures will be analyzed using existing fluorescence probe technology to provide benchmarking standards for comparison to conventional microarray assays. In addition, for applications in life sciences and biomedicine, it is desirable and advantageous in terms of speed and throughput to extend the serial patterning capability of DPN to a parallel methodology. Thus, concurrent with ink development and patterning optimization, microfabricated parallel multipen arrays will be explored as a means for faster, simultaneous writing of multiple DNA inks.

Garlich, Joseph R Comchem Technologies, Inc. Bone Targeting Of Degradable Drug-Filled Nanoparticles R43 CA101545 1 year NCI

A major limitation to using small injectable particles containing therapeutic drugs for controlled drug delivery is their quick removal from the bloodstream by mononuclear phagocytes mainly in the liver and spleen. Recent advances in how to make biodegradable nanoparticles invisible to such processes (stealth nanoparticles) using polyethylene glycol coatings have allowed for much longer circulation times which we hypothesize make possible new targeting strategies. We propose to target such stealthy drug-laden biodegradable nanoparticles to the hydroxyapatite surface of bone using covalent attachment of certain chelating groups to the surface of the nanoparticte. The drug we propose incorporating into the nanoparticle is known to protect cells from the harmful effects of chemotherapy and radiation therapy. If successful, our targeting technology could allow for selective delivery of protectant drug-containing nanoparticles to the bone whereupon biodegradation would liberate free active drug causing selectively high concentrations in the nearby contiguous bone marrow. Such selective protectant delivery to marrow could allow for more aggressive treatments of some cancers without the need for bone marrow transplants. The targeting technology proposed is a platform technology as it is amenable to substituting in other therapeutic drugs in the novel nanoparticles such as chemosensitizers or chemotherapy agents.

Glass, James Russell Seashell Technology, LLC Nanofabricated Particles For Biolistic Delivery R43 EB001624 1 year NIBIB

We propose the development of multi-functional composite particles for use as improved DNA delivery vehicles in biolistic applications. These particles will be fabricated using the controlled assembly of nanoparticles, polymers, and biomolecules via a layer-by-layer technique where the core and each subsequent layer add specific functionality to the final particle. The density, stability, surface area, and surface chemical composition will be controlled by the choice of material and dimensions of the core and surrounding shell layers. When these novel nano-fabricated devices are used in biolistic applications, the efficacy and reproducibility of the procedure will be greatly enhanced. These novel nanoparticles will be of broad interest to users of biolistic transfection systems in both academic and commercial research communities.

Gu, Gang Molecular Nanosystems, Inc. Nanotube-Based Electronic Biosensors R43 EB001576 2 years NIBIB

The goal of the project is to develop a novel sensor technology platform based on carbon nanotube electronic sensor device, which could be integrated into a biochip and used for detection and analysis of biomolecules in samples from blood, saliva and other body fluids, as well as studies of protein-protein and protein-small molecule interactions in the research laboratory. The fundamental principle of the technology has been demonstrated by Professor Hongjie Dai's group at Stanford University, showing that the electrical resistance change of a nanotube can be detected when a biological event such as antigen-antibody binding occurs on the surface of the nanotubes. In the proposed electronic sensor array each nanotube sensor will be chemically functionalized and immobilized with biomolecules to provide selectivity and specificity for detection of various analytes. The detection scheme is based on changes in the nanotube's electrical conductance due to changes of the electrostatic environment upon analyte binding. Such detection system requires no expensive detection equipment, such as lasers, or fluorescence labeling of analytes. Instead the detection is based on a simple direct electrical readout. The objective of this program is to demonstrate the feasibility of a nanosensor providing the sensitivity and selectivity for detection of a biological marker, which is integrated into an electrical circuitry for easy readout. The specific aims include: (1) the fabrication of electrical devices containing semiconducting single-walled carbon nanotubes; this involves controlled growth of nanotubes on wafers by chemical vapor deposition and construction of electrodes and electrical circuits on wafers by semiconductor technology, (2) the functionalization of the nanotubes in the electrical devices for immobilization of biomolecules onto the nanotubes. A variety of functionalization strategies will be investigated to minimize non-specific adsorption and to maximize selectivity. Successful demonstration of the proposed nanotube-based biosensor will have a significant impact on a number of commercial sectors and help establish the United States as leader in the emerging field of nanotechnology.

Henderson, Eric R | Bioforce Nanosciences, Inc. | Microfabricated Deposition Tools For Creating Nanoarrays | R43 EB000613 | 2 years | NIBIB

The goal of this proposal is to design and test novel microcantilever devices for the purpose of improving deposition of biomaterials in ultraminiaturized arrays. BioForce NanoSciences, Inc. (http://www.bioforcelab.com) is an industry leader in the development of next-generation biomolecular screening arrays, or NanoArrays, with patented technology permitting the construction of biochips with sub-micron spatial addresses. These nano-scale arrays allow a thousand or more molecular tests to be carried out in the same surface space occupied by a single state-of-the-art microarray spot. This format offers dramatic savings in reagent cost that accompanies ultra-miniaturization, and more importantly, it provides a realistic and sought-after platform for performance of array-based analyses in applications involving extremely small quantities of sample material where uses of current microarray formats are not feasible. These applications include single-cell protein profiling, prenatal diagnostics, forensic testing, high-throughput drug screening, and minimally invasive clinical diagnostic testing. Commercialization of this technology requires highly reproducible molecular deposition that is not feasible with current deposition tools. This research project will advance this technology through the following specific aims: 1. Design and construct front and back loading quill- and aperture-type single, and multiplexed deposition tools; and 2. compare iterative deposition-wash cycle with pre-load, continuous flow deposition methods using the devices designed in aim 1.

Israel, Barbara A | Platypus Technologies, LLC | Nanostructured Substrates For Cell Assays | R43 GM069026 | 2 years | NIGMS

The long-term goal of this Phase I SBIR is to develop optimized components (nanostructured substrates) for use in the creation of a novel class of rapid, liquid crystal based cellular assays of enumeration, proliferation and function. There is a need for new technologies that are less labor intensive, consume fewer cells and reagents, can be performed in less time, allow for separation of chemotaxis from chemokinesis, provide better control over spatial and temporal aspects of the delivery of concentration gradients and are amenable to high throughput formats. Platypus Technology (tm) can be the foundation for such assays to accelerate basic cell research, drug discovery, and evaluation of anti-cancer drugs. The combination of nanostructured substrates with liquid crystals requires no labels or additional reagents to report the presence of cells. It will provide a unified platform for many diverse cellular assays. In this proposal, we will fabricate substrates with nanoscale topography by three methods: nanoscale molding, abrasion of polymeric materials and oblique deposition of thin films of gold. We will comparatively evaluate the ability of these substrates to: align liquid crystals in the presence of non-specific adsorption of proteins in cell media, support cell functions of epithelial, fibroblast and vascular endothelial cells, report cell numbers, and retain functional stability during a six-month storage period. At the completion of these studies we will have optimized the core component of a new technology for enumeration and functional evaluation of cells.

Kalkhoran, Nader M Spire Corporation Nanophase Ceramics As Bone Implant Coatings R43 AR049657 2 years NIAMS

The overall objective of the proposed study is to create a new family of "smart" orthopedic and dental implant coating materials that enhance new bone formation over existing implants. "Smart" coating materials are necessary to selectively increase bone cell function while, at the same time, inhibit functions of competitive cells that lead to soft, instead of bony, tissue formation. Such osseointegration provides mechanical stability to an implant in situ, minimizes motion-induced damage to surrounding tissues, and is imperative for the clinical success of bone implants. In this manner, the health relatedness of the proposed project is to increase bonding between an implant and juxtaposed bone so that a patient who has received joint or dental replacement surgery may quickly return to a normal active lifestyle. Furthermore, the present proposal aims to increase the service-life of an orthopedic material by creating materials that form a strong, long lasting, bond with juxtaposed bone. The material proposed in the present study as a more effective orthopedic implant coating is: nanophase hydroxyapatite doped with yttrium. Nanophase ceramics are intriguing new material formulations since they possess grain sizes less than 100 nm in diameter. For this reason, nanophase ceramics simulate the grain size and topography of bone. Hydroxyapatite doped with yttrium coatings may increase initial absorption of calcium from serum leading to select protein adsorption to enhance bone cell function. This is speculated since yttrium has a high affinity for calcium. When not used as an implant coating, previous studies have determined that nanophase hydroxyapatite doped with yttrium increases bone cell function over existing hydroxyapatite formulations. The present study will build upon these results by using a novel technique to coat a currently utilized bone prosthetic material (titanium) with nanophase hydroxyapatite doped with yttrium. For Phase I studies, the ability of the titanium coated nanophase hydroxyapatite doped with yttrium to promote new bone synthesis and limit competitive cell function will be determined using in vitro cellular models. Specifically, osteoblast (bone-forming cells) and fibroblast (cells that have been associated with competitive soft tissue formation) function will be determined on the proposed coated materials. The specific aims of this proposal are therefore to combine previously designed materials that enhance new bone formation with a novel technique that will transform these bioactive materials into a practical bone prosthetic coating. Undoubtedly, design criteria used in the proposed study to investigate new coating techniques coupled with a new coating material could have great impact in the development of the "next-generation" of orthopedic implants with an improved ability to bind to juxtaposed bone.

Kim, Jinseong Lynntech, Inc. Array-Based Biological Sensor Using Nano-Pores R43 AI056623 2 years NIAID

Immunoassay is probably the most commonly used technology for the detection and quantification of biomolecules in the diagnosis and management of disease. Among many immunoassay methodologies, biosensors based on direct immunoassay have been attractive because of their ideal advantages such as rapid detection, real-time analysis, regeneration of sensing device or reproducible inexpensive disposable, no pretreatment, and so on. On the other hand, emerging nanotechnology creates functional materials, devices, and systems through control of matter at nanomater scale and exploit novel properties and phenomena at the same level. Coupling the nanotechnology with the immunoassay, Lynntech is proposing development of an array-based biological sensor detecting biologically relevant molecular and physical targets in samples from blood, saliva and other body fluids, or for use in the research laboratory (purified samples), clinical specimens and in the living body. The sensor consists of multiple antibodies on nano-sized pores with mechanical stability to monitor multiple proteins and molecules with minimized sample volume simultaneously so that the sensor can be used as a biomarker. Phase I effort will demonstrate the feasibility of the proposed technology by fabricating a single nanopore, attaching antibodies, evaluating the nanopore sensor, and fabricating nanopore arrays. Further development of the technology toward miniaturized biomarker will be included in the Phase II effort.

Lancelot, Robert Arryx, Inc. Optical Trapping R44 RR017152 2 years NCRR

Manipulation of objects on a microscopic scale can be done conveniently with a device known as a laser or optical tweezers. While laser trapping was originally devised for trapping Rayleigh particles (i.e. particles much less than the wavelength of the incident light) it's ability to manipulate biological particles such as macromolecules, viruses, microtubules and chromosomes offers great practical potential. In addition a laser tweezers can be used to fabricate small-scale devices such as microscale motors, pumps and switches. Many systems of interest require multiple optical traps and several methods have been developed to achieve multiple trap configurations. However currently available trapping systems can produce at most only a few independent traps. Recently, Grier and Dufresne conceived of a new solution for achieving a multi-trap system. In their method a hologram is used to alter a single laser beam's wave front. The wave front is altered so that the downstream laser beam forms a large number of individual laser beams with relative positions and directions of travel fixed by the exact nature of the hologram. The hologram can be calculated from a user specified pattern of desired trap positions. During the Phase I work we have successfully developed a commercial prototype of a holographic optical tweezers, which is capable of deploying up to 200 independent laser traps. As part of this work we have designed an easy to use laser tweezers with an imaging system, computer interface, sample chamber, and optical system. Field-testing at the Whitehead Institute and the University of Maryland has resulted in those institutions initiating purchase negotiations (the University of Maryland has purchased an instrument). In addition field-testing programs at the University of Chicago, Albert Einstein College of Medicine, and Drexel University are being negotiated. We now propose to make the holographic optical tweezers a commercial product by developing manufacturing standards for components and incorporating a greater variety of imaging modalities. We will also develop methodology to facilitate use of existing applications of laser trapping in a user friendly way.

Lichtenhan, Joseph D Hybrid Plastics, LLC Nanocomposite-Based Dental Materials R44 DE014026 2 years NIDCR

It is the specific aim of this proposal to develop an advanced adhesive and dental restorative system that is more quickly placed and reliable than current products. Thus improving patient satisfaction and general health care. The method for achieving this objective is to design a dental bonding agent and restorative system that are structurally controlled at the nanoscopic (1-10nm) level through the macroscopic level. Natural tooth contains both nanoscopic and macroscopic length scales. The development of such a restorative system will maintain the continuum of structure from the natural tooth through the adhesive and restorative system thereby improving compatibility and physical properties. Nanoscopic building blocks based on the nontoxic polyhedral oligomeric silsesquioxane (POSS) Nanostructured TM Chemical technology will be utilized in combination with conventional macroscopic and nanoscopic metal oxide fillers to design a dental restorative which benefits from multi-length scale structural control (nano to macro). POSS-Monomers will be utilized as isotropic 1.5nm structural segments in the base restorative resin. In this capacity the resulting nanostructured resin will provide an enhanced filler-resin interaction and lower shrink characterists as demonstrated in Phase I. Improvement of the filler-resin interface will also result in improved mechanical properties and reduced degradation of the restorative system. The performance of restoratives are also governed by the nature and integrity of the inherent bondline formed with natural tooth. Here again the design of bonding agents which utilize 1.5nm monomeric POSS building blocks provide a more natural interface with the 2nm sized apatite crystals present in natural tooth and with the l nm-10nm structural segments incorporated into the proposed restorative. Overall better bonding to the tooth and restorative per Phase I findings. By utilizing both nano (lnm-10nm) and macroscopic length scales in a dental restorative and bonding system, a continuation of length scale and properties with those in natural tooth can be acheived. It is the intent of the proposed Phase II work to develop a prototype system for transition to Phase III.

Marr, David WM Metafluidics, Inc. Precision Microfluidic Contol For Nanobiotechnology R43 EB000952 2 years NIBIB

An automated microfluidic chip-top flow image cytometer and cell sorter has been developed that is capable of precisely distinguishing and directing both cells and colloidal particles. This approach relies on the natural separation maintained by laminar microfluidic streams and the ability of applied fields, specifically optical manipulation techniques, to translate cells and particles between these streams. These microdevices have the potential to perform chip-top microbiology more rapidly and with less associated hardware and preparation time than any other techniques currently available. Furthermore, the optical actuation approach allows for seamless integration with widely applied fluorescence labeling and detection schemes. Single chip-top devices that employ this technology will be capable precisely handling individual cells and microspheres and will be poised to make a considerable contribution to molecular biology on the nanoscale, and therefore molecular therapeutics in the coming decades. The specific aims of this proposal include 1) The expansion of the existing cytometry configuration to accommodate multiple sorting fates by using an optical trap to precisely position particles and cells in channels and allowing laminar flows to direct them to the proper outlet channel 2) The integration of fluorescence based detection for rapid discrimination among cell phenotypes, binding events and particle-bound compounds 3) The increase of sorting rates through optimization of detection schemes, an increase in the number of simultaneous traps, and through parallelization of multiple devices upon the same chip 4) The realization of a working prototype capable of handling cells, performing multiple bead-based analyses and rapidly recovering and interpreting the results of these tests.

Maryanski, Marek J MGS Research, Inc. 2-Phase Nanogels For Quantitative 3D Radiation Dosimetry R43 CA094540 2 years NCI

MGS Research has pioneered the development of polymer gel dosimeters, using both MRI and optical CT scanning. Our goal in Phase I will be to prove the feasibility of an innovative two-phase nanostructured polymer gel for quantitative 3D radiation dosimetry. The new gel structure can be described as a "single-hit 3D photon counter" that can be expected to be linear with dose and to be dose-rate independent. Moreover, it will be characterized by greater reproducibility, accuracy and stability of the dose response. As a result, the nanostructured polymer gels will be more suitable than the existing single-phase gels for the quantitative 3D radiation dosimetry and high-precision quality assurance, which are very much needed in intensity modulated and 3D conformal radiation therapy.

Paulose, Maggie Sentech Corporation Remote Query 'Smart-Tube' Nanosensors R43 HD044334 2 years NICHD

We propose extending magnetoelastic sensor technology from the microscale to the nanoscale. This scaling of dimensions will enable the resultant sensor arrays to be placed within tube sidewalls, providing physical and chemical information of the tube interior (air or liquid) without interference of flow characteristics. Device sensitivity on the nanoscale should enable the instantaneous detection of single molecule binding events. The remote query nature of the sensor technology enables sensor information to be transmitted from the interior of the tube without the use of physical connections, avoiding possible contamination. Since the sensors transmit information via magnetic flux they are not susceptible to interference affects that might affect optical sensors, e.g. moisture condensation, smudges, misalignment, etc. Combining the best features of nanowire device fabrication and magnetoelastic sensor technology, the sensors should be able to distinguish mass changes of femto-grams but yet be inexpensive enough to be readily used on a disposable basis. It is anticipated that such sensors would find immediate application in clinical care, in effect creating a 'smart tube' technology. In Phase I, we will fabricate ordered magnetoelastic nanowire cantilever arrays, as described herein, of 1, 5, 10, and 50 cantilevers. Initially we will modify and extend the needed measurement electronics to monitor these uncoated nanocantilevers in air and liquid establishing baseline signal-to-noise values as a function of nanowire composition, properties and dimensions. The nanocantilevers will then be coated with a polymer of precisely controlled thickness, in incremental steps of less than 2 nm, to establish baseline sensitivity values and operating characteristics. In Phase II, the nanocantilevers will be coated with analyte specific layers and integrated with disposable tubes enabling their use as clinically relevant analyte and environmental sensors for, initially, neonatal monitoring.

Phaneuf, Matthew D | Biosurfaces | A Nanofibrous Biocomposite Small-Diameter Graft | R43 HL074771 | 1 year | NHLBI

There is no small-diameter (< 5mm internal diameter) vascular prosthesis clinically available that is capable of emulating the biological and physical properties of the normal arterial wall. The goal of this two-year phase I project, which unites a diverse group of industrial, biomedical and academic researchers, is to develop in vitro a novel nanofibrous bioactive small-diameter prosthetic vascular graft using electrospinning technology. The resulting vascular graft would possess both biological (antithrombogenic and mitogenic) and physical properties comparable to that of native artery, thereby improving graft patency. Our hypothesis is that the next generation of prosthetic arterial grafts will have to possess multiple structural and biological properties that mimic some of those processes inherent to native arteries in order to prevent complications such as thrombosis from occurring. A small-diameter nanofibrous biocomposite vascular graft will be electrospin from polyester (Dacron) and collagen, thereby possessing properties similar those of native artery. The potent antithrombin agent recombinant hirudin (rHir) and endothelial mitogen vascular endothelial growth factor (VEGF) will be covalently bound to collagen within the construct. The elastic properties of the electrospin polymer will provide circumferential compliance, with kink-resistance prevented by a thin braided Dacron mesh within the graft wall. The specific objectives are to: 1) develop electrospinning methodology for a Dacron/collagen composite graft (ESDC), 2) incorporate novel inner-wall reinforcement for ESDC, 3) synthesize novel small-diameter ESDC graft containing inner-wall reinforcement, 4) characterize physical properties of ESDC graft, 5) immobilize rHir and VEGF to ESDC graft, 6) examine surface antithrombin properties, 7) evaluate surface mitogenic properties and 8) assess surface rHirNEGF stability under simulated arterial flow conditions. Phase II of this project will evaluate this novel ESDC-rHir- VEGF graft in a canine carotid grafting model. Development of a bioactive small-diameter vascular graft would have a significant impact on small vessel repair and replacement. These grafts could be utilized in peripheral bypass as well as for coronary artery bypass, which have some 500,000 grafts implanted annually in the United States. Potentially, the annual market value for an "off-the-shelf' synthetic coronary artery bypass graft could exceed $1.5 billion.

Routkevitch, Dmitri | Nanomaterials Research, LLC | Novel Platform For Living Neural Networks | R43 NS045507 | 1 year | NINDS

Unlocking the mechanism of neural growth understanding and communication is needed for and treatment of many degenerative diseases, as well as for neural prosthesis and restoration of damaged neural connections. Living neural networks (LNN) can enable a broad array of new tools for neural research. Furthermore, LNNs, being capable of detect minute environmental perturbations, are an attractive target for chemical and biological sensing. However, producing reliable LNNs is challenging, and requires control over the neuronal growth and formation of synaptic junctions, high charge density high-resolution neuronal contacts, overall biocompatibility and reproducibility. Substrates for LNNs that would satisfy these requirements are not available. To address this opportunity we propose to use self-organized nanoporous alumina ceramic as a platform for guided growth and interfacing of LNNs. The core of the innovation is in the combination of several ideas: nanoengineering of the anodic alumina to provide tailored neuron/substrate interface; hybrid micro machining of patterns for neural growth guidance; using encapsulated nanoelectrodes arrays and routing the excitation/response signals to the bottom of the chip to provide soft high resolution electrical contacts to neurons. Proposed living neural networks have significant commercial potential. If realized, they could be used as tools for neural network research, disease studies and diagnostics, drug and toxin screening, and new generation of biochemical sensors.

Wang, Y A | Nanomaterials And Nanofabrication Labs | Kits For Converting Nanocrystals To Bio-Reagents | R43 GM069065 | 2 years | NIGMS

This NIH SBIR program intends to ultimately convert high quality colloidal nanocrystals, mainly semiconductor nanocrystals and with some concerns on noble metal nanocrystals, to standard bio-medical reagents. This program is based on our patent-pending technologies for ligand chemistry and bio-conjugation chemistry for colloidal nanocrystals. Colloidal nanocrystals are nanometer-sized fragments of corresponding bulk crystals, which are typically synthesized and manipulated in solution. Their strong size dependent properties and flexible solution-phase processability make them as ideal candidates for many bio-medical applications, such as bio-medical labeling tags, drug delivery carriers, MRI enhancing reagents, etc. The two key challenges for such applications are the commercial scale production of high quality nanocrystals and their reliable bio-conjugation chemistry. The first issue is being addressed by two NSF SBIR programs in this company. This NIH SBIR program will address the second issue, bio-conjugation chemistry. The success of this program will provide academic and industrial scientists/engineers as well as medical workers stable, reliable, inexpensive and simple means to apply high quality nanocrystals in their research, development and other daily duties. This goal will be accomplished by design and development of three complementary series of ligand chemistry and bio-conjugation chemistry kits for the bio-medical applications of colloidal nanocrystals.

Wharton, J Tim | Lynntech, Inc. | An Integrated Optical DNA Detector For Portable PCR | R43 AI056738 | 1 year | NIAID

The goal of this Phase I project is to develop a real-time nanoparticle-based optical detector for polymerase chain reaction (PCR) amplified DNA that is integrated into a portable PCR unit for use in low technology environments. The detection of target DNA oligonucleotides (ON) from PCR amplification conventionally involves either electrophoresis/staining or fluorescence indicators. Electrophoresis analysis generally takes several hours to complete and fluorescence requires the use of large, expensive equipment that limits the detection method to laboratory settings. We propose the use of nanoprobes whose visible-light absorption properties change dramatically in the presence of the target ON in the PCR solution. The optical detector has been shown to distinguish between target ON and ON with only a single mismatched base pair. A simple optical detector system, based on an LED and a phototransistor, will be integrated into a completely automated miniature PCR device already in advanced stages of engineering at Lynntech with the goal of producing a prototype detector for use in low technology environments by minimally trained individuals that will produce test results in minutes. The device can be used for in vitro medical diagnostics, detection of biological warfare agents, detection of food pathogens, etc.

Wharton, J Tim | Lynntech, Inc. | Novel Nanostructures For Topical Photodynamic Therapy | R43 CA103268 | 2 years | NCI

The objective of this proposal is to synthesize and evaluate novel, carbon-based nanostructure photosensitizers (PS) for the topical treatment of a variety of non-melanoma, pre-malignant and malignant cutaneous lesions, inflammatory dermatoses, and localized bacterial and fungal infections by topical photodynamic therapy (PDT). Our concept is based on a recently discovered family of molecules that have shown a remarkable ability to sensitize singlet oxygen under a variety of conditions. The visible light-absorbing photosensitizer has been shown in previous work to exhibit no dark toxicity. It has also been shown by our preliminary in vitro work to be internalized into a murine cancer cell line and a gram-positive bacterium where significant phototoxicity was observed upon irradiation. It is easily "tailored" by the methods of organic synthesis, which has the potential of leading to a combinatorial library of PS for different applications. After topical application of the PS to the affected area followed by a period of incubation, the area is selectively irradiated with light of appropriate wavelength and intensity. This causes the formation of reactive oxygen species (ROS) by the catalytic action of the nanostructure PS on molecular oxygen. The ROS cause a loss of viability in the affected cells.

Xiao, Danny | Inframat Corporation | Nanostructured Membrane For Implantable Glucose Sensors | R43 GM063287 | 2 years | NIGMS

Long-term implantable biosensors have been commercially unavailable because a common problem associated with biosensors is protein deposition and fibrous encapsulation known as biofouling limiting device lifetime. Implantable biosensors can benefit greatly from improving membrane and coating layer structures using functional materials on biomembrane surfaces that reduce protein adsorption and the subsequent inflammatory response. Inframat Corporation proposes to demonstrate the feasibility of a novel nanostructured membrane for implantable glucose sensors. The proposed nanostructured membrane is expected to control tissue/glucose sensor interface interactions, thereby reducing protein adsorption while increasing biocompatibility for the sensor. This will extend glucose sensor lifetime. The proposed program consists of preparing a protein-resistant nanostructured membrane, evaluating its physical properties, and confirming improved performance in-vitro for glucose sensor applications. This program is based on our extensive experience in nanotechnology, particularly in nanostructured materials and coatings. Inframat is collaborating with Dr. George Wilson of the University of Kansas to evaluate the nanostructured membranes. The nanostructured membrane system can be used for a variety of implantable devices including biosensors, stents, hip and knee implants, and drug delivery systems. Anticipated socio-economic benefits of extending sensor lifetime include lower overall health costs to the nation, and improved quality of life.

Xiao, T Danny | Inframat Corporation | Antibody Encapsulated Electrochemical Biosensor For GABA | R43 NS046088 | 2 years | NINDS

Gamma-aminobutyric acid ("GABA") is a main mammalian nervous system inhibitory neurotransmitter, playing important roles in neural function and dysfunction. Accurate real time measurement of GABA will greatly accelerate discoveries on the key role of GABA in motor disorders including Huntington's disease and Parkinson's, seizures, myoclonic discharges, and alcohol addiction. Current technology for extracellular measurement of GABA focuses on microdialysis of the cerebro-spinal fluid, followed by liquid chromatography combined with pre-/post column derivatization. Liquid chromatography-based measurements are not continuous, while microdialysis is an invasive procedure causing neuronal death and reactive gliosis with poor spatial-temporal resolution. INFRAMAT proposes to demonstrate feasibility of exploiting functionalized biomaterials to dramatically improve biosensor performance in high sensitivity GABA detection, via a "competitive enzyme immunoassay" strategy, electrochemically measuring non-electroactive GABA neurotransmitters. The proposed technology will adopt IMC's wet chemical synthesis to produce antibody-linked nanoparticles, immobilize functionalized nanoparticles onto transducers to obtain high sensitivity biosensors for real time monitoring GABA concentrations, and enable in-vitro testing of the biosensor based functionalized nanoparticles in artificial cerebro-spinal fluid. This nano-engineered approach will enable biosensors for continuous and direct measurement of GABA concentrations, and can be extended to multiple channels to obtain a spatial-temporal distribution in brain slice and cell culture preparations.

Yun, Wenbing | Xradia, Inc. | X-Ray Phase Microscopy Of Biological Samples In 3D | R43 GM071090 | 1 year | NIGMS

Xradia Inc. has recently developed the world's first X-ray tomographic microscope with a demonstrated 3D resolution of 70 nm for the semiconductor industry. It uses 5.4KeV X-rays from a rotating anode source. A complete data-set is collected and analyzed automatically in 8 hours, while a single 2D image is acquired in 10 minutes. The specific aim of this proposal is to upgrade this microscope so it can operate in phase contrast and dark-field mode for biomedical applications at 70 nm resolution in Phase I, and 30 nm resolution in Phase II. (Phase contrast is needed since biological specimens show very little amplitude contrast in the microscope). In Phase I a phase ring will I be designed, fabricated, incorporated into the microscope, and used to demonstrate 3D Zernike phase contrast imaging on yeast. In Phase II new optical elements will be developed to reach the 30nm resolution goal. The proposed microscope is unique in that it outperforms all other 3D X-ray microscopes in resolution and throughput, yet it uses a rotating anode X-ray source, rather than a synchrotron. It is designed to image single cells, cell cultures, or predetermined regions within tissue sections by "virtual sectioning". The specimens may be frozen hydrated, or fixed and labeled using conventional methods. These capabilities will make the instrument a n important complement to confocal microscopes (offering higher resolution) and electron microscopes (offering thick-specimen capability and simplified specimen preparation).

Zhang, Zongtao | Inframat Corporation | Electrophoretic Nano-HA Coating For Improved Adhesion | R44 AR047278 | 3 years | NIAMS

This proposed phase II program is expected to produce prototype hydroxyapatite ("HA") coated bone implants with significantly extended lifetime, fabricated using a room temperature electrophoretic deposition process. Novel HA nanocoatings with significantly increased adhesion strength and corrosion resistance under simulated body fluid in-vitro have been demonstrated in the Phase I work. The proposed Phase II program emphasizes developing prototype implant devices to rapidly commercialize this Phase I nanotechnology. The benefits of exceptional coating to substrate bond strength enable expansion of the HA nanocoatings market to hips, knees, and dental applications. Achieving 100% crystallinity and density at the HA substrate interface with electrophoretic deposited HA nanocoatings assures no degradation during implant service. Functionally graded HA nanocoatings can be generated at the HA tissue interface, thus promoting optimal bioactivity. The Phase II specific aims include scale up of the experimental HA bath nanocoating composition to pilot-scale production of prototype medical implants; demonstration of the HA nanoparticles coating on prototype medical implants, such as hip, knee, and dental implants; and demonstration of superior performance of the coated prototype devices with extended lifetime in-vivo, using an animal model. Phase II participants include Spire Corporation and the University of Texas at San Antonio.