# SUMMARY OF NEW AWARDS

## BIOENGINEERING RESEARCH PARTNERSHIPS (BRP)

## FY 2000

The following text provides a summary of new Bioengineering Research Partnerships (BRP) grants awarded during Fiscal Year 2000 by the BECON member institutes and centers in response to program announcements PAS-99-010 and PAS-00-006. The objective of the BRP program is to support basic bioengineering research addressing important biological or medical problems with the work being done by a multidisciplinary research team which applies an integrative, systems approach to develop knowledge or methods to focus on the project objectives.

Funded grants are listed below in alphabetical order by the principal investigator's (PI's) last name. Other information provided for each grant includes PI affiliation, project title, grant number, funding organization, and a brief summary of the project.

1. **Principal Investigator:** **Brown, Thomas** **Affiliation:** University of Iowa
   **Project Title:** Nonlinear Computational Biomechanics of The Hip
   **Grant Number:** 5-R01-AR-46601-01 **Funding Organization:** NIAMS
   **Abstract:**

Disorders of the hip comprise a substantial fraction of current musculoskeletal disease burden. Complex nonlinear mechanical phenomena pervade many aspects of treatment of hip disease and injury including total hip arthroplasty, intra-articular fractures, osteonecrosis, and developmental dysplasia. The focus of this research partnership is in applying nonlinear finite element formulations to address unquantified mechanical phenomena that are clinically recognized as being crucial to patient outcome. Building on previous and ongoing finite element work, new computational formulations will be developed to tackle nonlinearities currently limiting the accuracy of numerical simulations in five clinically important areas. The first two areas involve leading complications of total hip arthroplasty with regard to abrasive wear of polyethylene and dislocation. The third area involves intra-articular fractures of the acetabulum and involve estimating residual cartilage contact stress elevations accompanying attempts at surgical restoration of articular surface congruity. The fourth area involves osteonecrosis and includes computationally characterizing a new animal model (the emu) and using the models for in-vivo testing of computationally optimized placement of a novel head-preserving implant device. The fifth application area is surgical management of developmental hip dysplasia. The partnership will bring together a critical mass of surgeons and engineers to achieve clinically-grounded advances.

2. **Principal Investigator:** **Deluca, Carlo** **Affiliation:** Boston University
   **Project Title:** Harnessing Motoneuron Activity: From Lab To Clinic
   **Grant Number:** 5-R24-HD-38585-01 **Funding Organization:** NICHD
   **Abstract:**

This project is aimed at developing an automatic system for decomposing the electromyographic (EMG) signal into the constituent action potentials corresponding to the firing of individual motor units activated by motoneurons. This system will be an outgrowth of an existing rudimentary system which has enabled the performance of various novel investigations that have provided new insight into motor control. The proposal is composed of five projects. The first will be deign-driven and describes the design and development of the new system which has knowledge-based algorithms at its heart for decomposing the signals. The other four projects will be hypothesis-based and will address basic science questions and clinical applications that will reveal the utility of the system. Project 2 will address the modifications which occur in the firing of motor units as a function of aging and the benefits that can be restored by exercise. Project 3 will address the phenomenon of motor unit substitution which will be useful in ergonomic work environments and the rehabilitation of patients with peripheral nerve injury ahd spinal cord injury. Projects 4 and 5 are clinical studies which will explore the use of quantified neutromotor activity and will study patients with acute ataxia.

3. **Principal Investigator:** **Frazier, Albert** **Affiliation:** Univeristy of Utah
**Project Title:** Integrated Sample Preparation for Genomic Analysis in Micro Device Format
**Grant Number:** 5-R01-ES-10846-2- **Funding Organization:** NIGMS
**Abstract:**

A consortium of four research centers will conduct a joint investigation to study and create a functional integrated sample preparation device. The strategy of this front-end sample preparation micro device is to produce a research tool that has the flexibility to be integrated with a number of downstream analysis objectives; i.e., either sequencing or genotyping. The proposed micro-scale sample preparation system is composed of three main micro compartments which include: (1) sample introduction combined with cell sorting and collection, (2) cell lysis, recovery of the nucleic acid material of choice (DNA or mRNA), and sample cleanup via solid-phase extraction or affinity capture, and (3) elution of the material to an amplification micro chamber and subsequent amplification. The expected applications address national health care by providing effective miniaturized instruments for biochemical analysis. This research should provide critical insight into portable biochemical analysis systems.

4. **Principal Investigator:** **Fredberg, Jeffrey** **Affiliation:** Harvard School of Public Health
**Project Title:** Micromechanics of Airway Smooth Muscle Cells in Culture
**Grant Number:** 5-R01-HL-65960-2- **Funding Organization:** NHLBI
**Abstract:**

Acute narrowing of the airway lumen in asthma is driven by myosin motors that exert their mechanical effects within a cytoskeletal scaffolding that is both deformable and in a continuous state of remodeling. The mechanical properties of that scaffolding are not well defined. This is a multi-disciplinary design-directed bioengineering project to fill that gap of knowledge. We will develop a micromechanical technology to measure the rheological properties of adherent living airway smooth muscle cells in culture, and the time-course of mechanical changes that occur in response to contractile stimuli or after genetic manipulation of cytoskeletal proteins. Ligand-coated ferromagnetic microbeads are bound to the cytoskeleton, and oscillatory mechanical torques are then applied to the bead by a sinusoidally-varying external magnetic field. Resulting oscillatory bead motions deform the cell, and can be determined by measuring changes of the remanent magnetic field due to bead rotations or, alternatively, by direct observation of oscillatory bead displacements using light microscopy; these are complementary detection methods each with special advantages. This technology becomes, in effect, a micro-rheometry system that can probe - in cell culture conditions - contractile responses and underlying cellular rate processes over time scales as short as tens of milliseconds to as long as hundreds of seconds. Thus, it measures mechanical properties of cells using deformation times (and stress magnitudes) that span the physiological range. We will develop this technology and then use it to test the hypothesis that the contractile response of human airway smooth muscle cells in culture is attenuated by overexpression of heat shock protein 27 (HSP27) dominant negative mutants. This hypothesis bears upon a question whose importance has been identified only recently, namely, the stability of the cytoskeleton of the airway smooth muscle cell and the role of CSK stability in airway narrowing in asthma.

5. **Principal Investigator:** **Gilbert, Charles** **Affiliation:** The Rockefeller University
**Project Title:** Imaging Activity in Visual Cortex At The Cellular Level
**Grant Number:** 5-R24-EY-12896-2- **Funding Organization:** NEI
**Abstract:**

The project proposes to combine advances in optics, molecular probes, and gene therapy techniques to monitor neural activity in the visual cortex of awake, behaving animals with single cell resolution. Two-photon imaging makes it possible to visualize fluorescent cells lying several hundred microns under the cortical surface and to minimize the photodynamic damage to the cells. Fluorescent proteins will allow visualization of details of cell morphology, and the fluorescence can be linked to neural activity. Adenovial vectors will be developed to insert genetic constructs that code for these proteins into the genome of cortical cells allowing one to label large numbers of cells with a sparse distribution and with minimal damage to the cells. The imaging instrumentation will be merged with the animal recording devices for monitoring activity tied to visual stimuli and animal behavior. Probes that reflect neural activity will be determined and the imaging technique will be adapted to studies in behaving animals. The applications of this work include the study of morphological changes in cells, the biophysics of neuronal integration, the neural basis of learning and higher order cognitive function, and patterns of gene expression in the intact brain.

6. **Principal Investigator:** **Greenberg, Robert** **Affiliation:** Second Site, LLC
**Project Title:** Development/Testing of Artificial Retinas for the Blind

**Grant Number:** 5-R24-EY-12893-2- **Funding Organization:** NEI

**Abstract:**

This proposal is to develop a long-term, implantable retinal stimulator for patients blinded by outer retinal degenerations. Using technologies developed by the Alfred E. Mann group of companies over the past 30 years for implantable stimulators, the investigators will develop a chronic retinal stimulator and associated external hardware for use both in research and as a clinical device. To achieve this goal, several areas of research are needed. In this work, academia will collaborate with industry to accomplish the basic research necessary to make a chronic retinal prosthesis a reality. Areas of basic research that the project will focus on include electrode geometry and electrode material selection, surgical attachment of the retinal implant, low-power electronic circuit designs, and hermetic packaging. Each of these areas needs additional research for the creation of an optimal chronic retinal prosthesis which will enable persons blinded by outer retinal degenerations to regain the most important loss they have suffered – the loss of mobility. The aim of this proposal is to complete the design and manufacture of a retinal prosthesis and associated external hardware and test it chronically in animals so that an investigational device application can be made to the FDA in preparation for a clinical trial.

7. **Principal Investigator:** **Huse, William** **Affiliation:** Novasite Pharmaceuticals, Inc.
**Project Title:** Drug Discovery of Large-Scale Variant Targets By Hts
**Grant Number:** 5-R01-AI-48517-2- **Funding Organization:** NIAID

**Abstract:**

This partnership will develop a novel HTS system for Drug Discovery capable of screening 10 3 variant targets simultaneously in real time, at little additional costs relative to one-target screening systems. We will use this instrumentation to develop a novel approach to Drug Discovery via large scale generation and screening of variant targets, centered on identifying ligand-receptor interactions at large scale. The advantage is a combinatorial explosion in the number of ligand- receptor interactions explored relative to one-receptor screening approaches (from 10 3 to 10 8, for 10 3 libraries). Our aim is the discovery of an anti-inflammatory and immunosuppressive agent, by discovering an agonist for the cannabinoid-2 G protein coupled receptor (GPCR). We have set up a general expression system that efficiently transfects one single variant target cDNA per cell in a massive transfection step. We use a cell-based GPCR functional assay based on Ca2+ sensitive fluorescence dyes. In the first Specific Aim, we will develop a novel fluorescence microscope imaging system capable of visualizing (screening) thousands of individual cells (variant targets) simultaneously in one well at High Throughput. This system brings HTS to a new level, where each cell is a functional assay by itself, the physical limit for a cell-based assay. For lead optimization, another component aims at developing a special HTS flow cytometry system to sort cells with significantly lower EC5Os than the wild type receptor. The variant GPCR present in these isolated cells, identified by PCR, represents a mutation that enhances the potency of a given lead. This data will be analyzed by computational molecular models, attempting to match the variation of chemical moieties within the lead compound with the variation of amino acid residues within the receptor to guide docking procedures. Bioinformatic analysis of these biochemically-derived computational models will be used to translate the identified amino acid changes that enhance the lead's potency into mirror-image modifications proposed on the chemical compound that will guide the lead optimization process.

8. **Principal Investigator:** **Intaglietta, Marcos** **Affiliation:** University of California - San Diego
**Project Title:** Bioengineering Design of Artificial Blood
**Grant Number:** 5-R24-HL-64395-2- **Funding Organization:** NHLBI

**Abstract:**

This proposal is aimed at designing, developing, and producing an economic oxygen-carrying plasma expander based on modified molecular human hemoglobin engineered with properties that insure the maintenance of microvascular function leading to improved survival and tissue oxygenation relative to the blood for treatment of trauma victims within 48 hours of injury. The program includes production of purified hemoglobin from red blood cells by means of a modified, self-contained plasma fractionation centrifuge that directly produces the necessary molecular modifications and a unit of artificial blood ready for use. The molecular modification to be pursued is surface modification with polyethylene glycol and other modifications that will result in molecules with large radius. The program encompasses all aspects of artificial blood production from obtaining the raw materials to the final commercial product and is aimed at establishing a blood transfusion technology that delivers a blood replacement material that is cost effective and as efficacious as blood.

9. **Principal Investigator:** **Jacques, Steven** **Affiliation:** Oregon Medical Laser Center
**Project Title:** Biomedical Optics for Medical Research And Clinical Care

**Grant Number:** 5-R24-CA-84587-2- **Funding Organization:** NIBIB
**Abstract:**
Biomedical optics is a field that uses light to interrogate tissues for diagnostic purposes and to treat disease and assist surgery and has applications in both biomedical research and clinical care. This proposal establishes a Biomedical Optics Laboratory on the campus of the Oregon Health Sciences University (OHSU) as a core research facility to support the interface of new optical technologies from the bioengineering network with projects in the medical research and clinical activities at OHSU. This proposal would provide funding for the bioengineering network to initiate new projects for translation to the medical center. The initial projects fall into the two general areas of tissue engineering (biomaterials development with emphasis on bone regeneration and biomaterial implants) and cancer detection and treatment (optical imaging of dysplasia and superficial cancer, optical fiber devices for dosimetry during photodynamic therapy), and photodynamic therapy as a tool in basic medical research. This research program complements a Biomedical Optics curriculum at the OHSU.

10. **Principal Investigator:** **Jain, Rakesh** **Affiliation:** Massachusettes General Hospital
**Project Title:** Integrative Biology of Tumor Angiogenesis, Invasion And Metastasis
**Grant Number:** 5-R24-CA-85140-2- **Funding Organization:** NCI
**Abstract:**
Now that numerous important genes associated with tumor angiogenesis, invasion, and metastasis have been discovered, the grand challenge is to understand their function in intact animals. A second major challenge is to integrate and apply this knowledge to cancer prevention, detection, and treatment. The proposed work aims at meeting these challenges with a new, more precise quantitative, integrative, and multi-disciplinary bioengineering approach. The approach builds on (1) genetically engineered mice to visualize gene expression, (2) in vivo models to visualize molecular and cellular events, (3) computer-assisted in vivo spectroscopy to quantify gene expression and function, and (4) mathematical modeling to integrate the resulting information. Using this technology, four critical aspects of tumor metastasis will be investigated: angiogenesis, invasion, hematogenous metastasis, and lymphangiogenesis and lymphatic metastasis. This work offers a new paradigm for integrative studies of the dynamics of gene expression and function in cancer that will facilitate translation of knowledge about the molecular biology of cancer into effective cancer prevfention, detection, and treatment strategies.

11. **Principal Investigator:** **Koller, Manfred** **Affiliation:** Oncosis, Inc.
**Project Title:** Laser Cell Processing for Basic And Clinical Research
**Grant Number:** 4-R44-RR-15374-2- **Funding Organization:** NCRR
**Abstract:**
Photosis is a technology platform that incorporates high-speed optical scanning of biological samples, image analysis, and computer-controlled laser-irradiation of specific targets within the sample for the purpose of inducing a biological response. Specific cells to be treated within a mixed population are identified by parameters such as size, shape, fluorescence, or other distinguishing features. Once identified, individual cells are targeted with a laser to induce a desired response, such as cell death, optoporation (for gene transfer), or even inactivation of a specific mRNA transcript within the cell. The current beta1-prototyle system can process hundreds of millions of cells in an hour under sterile conditions, making it useful for several research and clinical applications. Photosis has many potential uses, and this project brings together a number of institutions and researchers to investigate and define the possible applications of this novel technology. In its current configuration, the instrument uses a single color for cell detection and a laser to induce necrosis in every targeted cell. Additional applications will be developed, some of which will require modifications to the system design and building of new prototypes. The prototypes will be placed at four partnership sites where the basic and clinical applications research will be carried out, including: 1) in vivo study of purified stem cell subpopulations in the xenogeneic fetal sheep transplant model; 2) human clinical trials to assess NHL purging in autologous stem cell transplantation; 3) purification of genetically- modified stem cells and T-cells expressing a selectable transgene, as well as selective transduction of specific cells in a mixture via optoporation; and 4) accurate mRNA expression profiling from purified primary human prostate cancer cell populations. The proposed work will result in several types of novel bioengineering instrumentation for advancing the state-of-the-art in cell processing. These instruments will be used in this program to advance basic and clinical research in stem cell biology, cancer, immunology, and genomics.

12. **Principal Investigator:** **Long, Richard** **Affiliation:** Western Michigan University
**Project Title:** Blind Pedestrians' Access To Complex Intersections
**Grant Number:** 5-R24-EY-12894-2- **Funding Organization:** NEI
**Abstract:**

The objectives of this proposal are (1) to use the strengths of a multi-disciplinary team to understand the perceptual and cognitive requirements of negotiating complex intersections without vision and with low vision, (2) to design and test engineering and training solutions to problems of information access that are currently known and that are identified in the course of this research, and (3) to produce materials about the problems and solutions that are useful to transportation engineers, individuals with visual impairments, and rehabilitation personnel. This study will focus on intersections that are complex by virtue of their size, shape, and/or signalization. Particular areas to be investigated by the research team members include transportation engineering, characteristics of pedestrian signals, acoustics research concerning the perception of moving sound sources as related to street crossing, auditory motion display for research and training, eye gaze strategies, and mental effort during street crossings.

13. **Principal Investigator:** **Majumdar, Sharmila** **Affiliation:** University of California - San Francisco
**Project Title:** Morphological And Functional Musculo-Skeletal Imaging
**Grant Number:** 5-R01-AG-17762-2- **Funding Organization:** NIA
**Abstract:**

This partnership will focus on the systematic study of the morphology and function of the musculoskeletal system in disease and health. The aim of this consortium is to improve medical care through bioengineering developments, and to facilitate close interactions between bioengineers, computer scientists, clinical investigators, basic scientists and corporate partners. This effort will expedite the development of clinically-relevant quantitative imaging tools and propel the technical advances from the laboratories into the operating rooms and clinics. We hypothesize that high resolution, fast magnetic resonance imaging techniques and positron emission tomography, combined with quantitative image analysis, processing and visualization, can provide new insights and clinically viable and relevant methods for objective evaluation of disorders of the musculo-skeletal system. The long-term objective of this partnership is to understand the link between morphology, function, biochemical changes and clinical symptoms in the musculo-skeletal system. An immediate objective is to develop, implement and optimize novel non-invasive imaging methods (magnetic resonance imaging: MRI and positron emission tomography: PET) that will allow us to depict the musculo-skeletal system, quantitate morphology, function, provide unique 3D visualization and graphical representations of function and morphology, as well as correlate these with biochemistry and clinical status. This research partnership is aimed at quantitating early degenerative changes in two clinical areas of emphasis: the knee and the spine. The specific goals are: (i) to develop quantitative morphological and functional markers for degenerative diseases of the spine, (ii) to develop quantitative morphological and functional markers for the degenerative changes in the knee and osteoarthritis.

14. **Principal Investigator:** **Peli, Eliezer** **Affiliation:** Stephens Eye Research Institute
**Project Title:** Engineering Approaches To Low Vision Rehabilitation
**Grant Number:** 5-R24-EY-12890-2- **Funding Organization:** NEI
**Abstract:**

This proposal applies novel engineering approaches to the problems of low vision rehabilitation. This will be accomplished by building prototype devices based on solid theoretical foundations that, eventually, will become marketable rehabilitation products. The devices will be tested critically using diverse patient populations with the help of clinical partners to determine the effects on function and on the quality of life. The work will develop and test both optical and electronic devices that implement three specific engineering approaches aimed at restoring at least part of the important interplay of central (high-resolution) and peripheral (wide-field) vision. The three engineering approaches are multiplexing, dynamic control of display, and image enhancement. The effort will show that various combinations of these approaches are possible and likely to be beneficial. The proposed assessment and testing will emphasize two approaches: a virtual environment for controlled and quantitative testing in the laboratory, and on-the-street evaluation for real-life determination of the effect and utility of the devices and techniques.

15. **Principal Investigator:** **Ratner, Buddy** **Affiliation:** University of Washington
**Project Title:** Engineered Cardiac Morphogenesis - Stem Cells And Scaffolds
**Grant Number:** 5-R24-HL-64387-3- **Funding Organization:** NHLBI
**Abstract:**

The long-term aims of this project are to produce tissue-engineering ventricular wall patches for myocardial repair, ventricular assist devices, and replacement ventricles. The research team collaboration will encompass three foci: (1) instructive tissue scaffolds using advanced biomaterial fabrication to engineer biodegradable matrices and meshes with controllable pore dimensions modified with receptor=specific molecules; (2) cell and developmental biology in which primary and stem cell-derived muscle and vascular cells will be studied on modified scaffolds to determine the optimal conditions for producing

functional muscle tissues and vascular networks; and (3) clinical science and animal models to test contractile ventricular patches in an injured heart model. Integration with host tissue and restoration of contractile function will also be developed. The "tube hearts" will be conditioned to pulsatile flow circuits, assessed for mechanical performance in vitro, and eventually grafted into aortas of synergistic rats for in vivo evaluation. Progress toward these goals should establish design principles necessary for constructing more complex ventricular devices.

16. **Principal Investigator:** **Shain, William** **Affiliation:** Wadsworth Center
**Project Title:** Brain Prostheses: Tissue Compatibility & Integration
**Grant Number:** 9-R01-NS040977-05 **Funding Organization:** NIBIB
**Abstract:**

Nanofabricated neural prosthetic devices provide tremendous potential for furthering understanding of central nervous system (CNS) function and treating CNS disease and injury. Such devices will permit precise localization of targets and control of electrode function. However, the success of these devices is currently limited by reactive biological responses. This proposal is aimed at comparing and contrasting events underlying early and prolonged responses observed following prosthetic insertion to develop strategies for successful design and use of neural prosthetics. Signaling events that produce these responses will be identified along with the sources of these signals, and technology advances in prosthesis insertion, design, and pharmacological delivery will be used to control these events. Cell and organic cultures, developmental staging, and mouse genetic models will be used to test experimental hypotheses developed from observations in adult rats. Prostheses will be made using nanofabrication techniques, and surfaces will be modified using chemical, physical, and topographic methods. Results from these experiments will provide important new information for the intelligent design of improved biomaterials and micro-devices to control dynamic biological events in the CNS and to insure successful long-term performance of neural prosthetic devices.

17. **Principal Investigator:** **Snyder, Alan** **Affiliation:** Penn State University - Hershey Medical Center
**Project Title:** Biomedical Applications of Electroactive Polymers
**Grant Number:** 5-R01-HL-65959-2- **Funding Organization:** NHLBI
**Abstract:**

The objective of this partnership is to refine materials and establish methods for application of electroactive polymers in prosthetics and interventional medical devices. Electroactive materials are materials that change shape when exposed to an electric field. They are attractive as actuators because of their high energy density - the amount of energy that can be imparted to a load for a given volume or mass of active material. Electroactive materials, chiefly the piezoceramics, have found important uses in a variety of industrial, consumer and military systems, as well as in ultrasonic transducers for medical imaging, flow measurement and therapy. The piezoceramics have not been successfully applied, however, as actuators in other medical devices. A new class of electroactive polymers has recently been discovered, which make possible the development of devices using forms that would not have been practical using available materials. These materials remain flexible, can readily be formed into a variety of shapes, and provide much larger shape changes than do previously available materials. This will make possible the development of devices using forms that would not previously have been practical. Two target application areas have been chosen: (1) next-generation prosthetic blood pumps for treatment of end-stage heart disease, and (2) advanced instrumentation for minimally invasive surgery, particularly for use in confined spaces such as the thorax. These disparate applications share the need for very compact, efficient and uncomplicated means of actuation. Both suffer today from the need for bulky actuation mechanisms that must remain physically distinct from the parts which pump blood or manipulate tissue. The technology to be developed under this program will blur the lines between structure and actuator, leading to modes of therapy that are not currently available. The Materials Research partner will work to optimize electroactive polymers, which have been developed thus far for military applications, for use in the target medical devices, and develop methods for fabrication of the required multilaminate actuator materials. As these materials are fundamentally different from active materials of actuating mechanisms used by engineers in the past, the Mechanical Engineering partner will work to develop new design methodologies for use with the new materials. The Bioengineering partner will develop prototype devices to demonstrate the potential of the technology and lay the ground work for full development of new devices.

18. **Principal Investigator:** **Soper, Steven** **Affiliation:** Louisiana State University
**Project Title:** The Design And Fabrication of Novel Micro-Instrument Platforms for Performing Genetic-Based Analyses
**Grant Number:** 5-R24-CA-84625-2- **Funding Organization:** NCI

**Abstract:**
The focus of this project is to fabricate novel micro-instrument platforms targeted at detecting mutations in genes associated with certain cancers (colon and breast). The mutational analysis tool to be developed will detect point mutations at a level of l mutant DNA in approximately 100 normal DNA's. The devices will be fabricated using high aspect ratio micromachining in plastics (PMMA) and LIGA processing. The ability to effectively use these PMMA materials is based on the use of near-IR fluorescence readout which alleviates much of the background signal arising from autofluorescence of the substrate or matrix interferences.

19. **Principal Investigator:** Stephanopoulos, Gregory **Affiliation:** Massachusettes Institute of Technology
**Project Title:** Linking Genomics To Function Via Metabolic Phenotyping
**Grant Number:** 5-R01-DK-58533-2- **Funding Organization:** NIDDK
**Abstract:**
Recombinant strains with well defined genetic backgrounds are often found to exhibit small functional differences despite specific changes at the genetic level while in other cases, single gene alterations result in profound phenotypic variations. Although a first step in explaining such macroscopic differences is to probe the full detail of the expression phenotype by genome-wide expression measurements, transcription data alone are insufficient to elucidate the actual metabolic state of a cell and its functions. The latter require information about intracellular metabolic fluxes, which constitute fundamental determinants of cell physiology and excellent metrics of cell function. "Metabolic phenotyping" is the process and methods of determining intracellular fluxes as determinants of the cellular metabolic state. Combined with transcription data, the investigators provide a complete framework for analyzing the effect of drugs and studying disease. This project integrates the expertise of three participating laboratories for the purpose of combining metabolic and expression phenotyping to elucidate central carbon and lipid metabolism in model mouse hepatoma and hepatocyte cultures. Determination of intracellular fluxes will follow a systems approach termed metabolic reconstruction whereby the entire metabolic network is configured such as to best represent macroscopic rate and isotopic label distribution measurements made by GC-MS. Of particular attention are issues of observability, redundancy, and solution stability to ensure method feasibility and accuracy of the results. Differential transcription data will be obtained by DNA microarrays for mouse genes involved in central carbon metabolic, gluconeogenic and lipid biosynthetic pathways, as well as for other genes with particular expression variability that will be identified in the course of the research. Bioinformatics methods and programs, developed over the past 12 years will be deployed for this purpose. The general goal of the research is to identify relationships between the metabolic phenotype as defined above and the transcriptional state as defined by expression data of consequence in pathways important to diabetes. The broader contribution of this research is to extend the paradigm of holistic transcriptional investigation introduced by DNA microarray technologies to the study of metabolic level processes by metabolic phenotyping. As such, it holds the promise of identifying most, if not all points in metabolism affected by the action of drugs or genetic modifications thus guiding future programs of drug development and gene therapy.

20. **Principal Investigator:** Sweeney, HL **Affiliation:** University of Pennsylvania
**Project Title:** Bioengineering Research Partnership - Muscular Dystrophy
**Grant Number:** 5-R01-AR-47292-2- **Funding Organization:** NIAMS
**Abstract:**
The goal of this partnership is to utilize a number of aspects of bioengineering in order to develop tools and therapeutics for the treatment and monitoring of muscular dystrophies. The project is collaboration between three investigators and includes the following areas of bioengineering: 1) cell and tissue engineering, 2) imaging and 3) therapeutics. Collectively we will delineate factors that when expressed in muscle may slow that rate of degeneration that is concomitant with either the complete (Duchenne muscular dystrophy) or partial (Becker muscular dystrophy) loss of dystrophin. These studies will utilize the mdx mouse as the animal model for dystrophin deficiency. The long-term goal is to gain the understanding and tools necessary to develop adeno-associated (AAV)-based gene therapy for Duchenne and Becker muscular dystrophies. Three parallel lines of investigation (each directed by one of the three investigators) are proposed: (1) a dissection the mechanical role of dystrophin and muscle adhesion proteins; (2) an assessment of the functional benefits of restoring adhesion molecules to dystrophic muscle using recombinant adeno-associated virus gene delivery; and (3) development of non-invasive methods for monitoring therapeutic benefits of dystrophin gene transfer.

21. **Principal Investigator:** Weiss, Shimon **Affiliation:** Lawrence Berkeley National Laboratory
**Project Title:** Development of Q-Dots As Biological Probes
**Grant Number:** 3-R01-RR-14891-1-S1 **Funding Organization:** NIBIB

**Abstract:**
The long-term goal of this project is to develop semiconductor nanocrystalline fluorescent probe (q-dots) technology that will provide biomedical research with better tools for diagnosis of diseases and biomedical techniques and instruments necessary for basic research of cellular and molecular structure and function. This effort includes q-dot synthesis, bio-conjugation techniques, dedicated optical instruments, and unique imaging technologies. Optimized protocols for q-dot synthesis will be developed to obtain desired optical, physical, and chemical properties; and various spectroscopic measurements will be used to characterize q-dots. The utility and new possibilities afforded by q-dot technology will be demonstrated by studying protein trafficking and assembly in living cells and by physically mapping genes. A large number of distinct markers will also be physically mapped on chromosomes and combined DNA molecules, and the kinetics of chromosome pairing during the meiotic prophase will be monitored.