

Center for Biomedical Engineering

The mission of the Center for Biomedical Engineering (CBE) is to combine engineering with molecular and cellular biology to develop new approaches to biomedical technology with applications to medicine and biology. CBE continues to play a leading role in the discovery of hydrogel scaffolds for MIT's activities in tissue engineering and cell culture substrates. Recent initiatives have focused on the structure and function of biomembrane proteins for applications in sensors, specifically for olfaction. New collaborations with the Harvard School of Dental Medicine, Boston University Medical School, and Harvard Medical School have focused on the creation of a Boston-area-based center of excellence in osteoarthritis. In addition, fundamental discoveries in cellular and molecular mechanics and mechanobiology by CBE faculty and students have enabled critical advances for applications in musculoskeletal and cardiovascular tissue repair and regeneration. To maintain intellectual leadership in the still rapidly growing biotechnology industry-academic paradigm, both in Boston and nationwide, innovative approaches are needed to stimulate fundamental research and to facilitate timely translation of new discoveries into the health care sector. With these goals in mind, CBE continues to identify new opportunities and connections with industry that are aligned with its set of core research thrusts. The center continues to maintain a well-used base of core research facilities that are provided at minimal user cost to the MIT community. Taken together, our aim is to pursue multidisciplinary biomedical research and create an outstanding training environment for a new generation of students/leaders in biomedical and biological engineering.

Major Research Areas

CBE's core faculty members represent a variety of academic units, primarily within the School of Engineering, but with substantial participation from School of Science faculty; collaborating faculty from Harvard Medical School, Harvard School of Dental Medicine, Boston University Medical School, and Colorado State University (CSU) Medical School; and investigators at Ludwig-Maximilians University in Germany. These faculty participate in multi-investigator programs focusing on CBE's primary research areas: (1) cell and tissue engineering; (2) biomaterials and hydrogel scaffolds for regenerative medicine and drug delivery; (3) membrane protein structural biology, design, and fabrication; and (4) mechanobiology (the effects of physical forces on cell and tissue regulation of organ degeneration and repair). Together, these research thrusts have direct applications to cardiovascular and musculoskeletal physiology, pathology, tissue regeneration and repair, drug discovery, environmental and biohazard sensors based on membrane proteins, and nanobiotechnology. CBE maintains a broad funding base with support from the Department of Health and Human Services (55%), industry (10%), the Department of Defense (25%), and a variety of other public (e.g., National Science Foundation) and private sponsors. CBE faculty members participate in many interdepartmental programs as well as collaborative interactions with other universities and industry research laboratories.

Major Research Initiatives

CBE researchers have expanded research activities initiated in the context of a large ongoing Bioengineering Research Partnerships grant from the National Institutes of Health (NIH)/National Institute of Biomedical Imaging and Bioengineering centered on bone, cartilage, myocardium, and liver tissue. In collaboration with the CSU Clinical Sciences Department, a major study using self-assembling peptide hydrogel scaffolds was just completed involving the repair of cartilage defects in the knees of horses. The Food and Drug Administration has stated that this equine animal model for joint disease is the best animal model for ultimate application to human clinical needs, since potential biomaterials can be evaluated in a defect of clinically relevant size in an animal undergoing strenuous exercise. This blinded study will be decoded after all of the retrieved tissue analyses, which involve follow-on quantification of tissue biochemical (MIT), biomechanical (MIT), immunohistochemical (CSU), and histological (CSU) properties, are completed. The *in vitro* studies leading to this clinical equine trial involved four CBE faculty laboratories at MIT and a basic science laboratory at CSU. All of the peptide hydrogels used in this study are MIT-patented and licensed to industry (3DM), based on discoveries in CBE laboratories. These same hydrogel scaffolds are now also under study at Pfizer (North Cambridge campus, for applications in protein delivery and tissue regeneration) and at Olympus (bone tissue engineering).

In a project area initially funded by the Defense Advanced Research Projects Agency (DARPA), CBE researchers are studying membrane protein molecular structure and a class of G-protein coupled receptors (GPCRs) that are specifically involved in human olfaction. Unlike mammalian vision and audition, which function by employing only a handful of different receptors, olfaction needs several hundred different receptors to provide the brain with the stimuli corresponding to various scents. With about 400 olfactory receptors, humans can recognize many thousand distinct odors, even without training. Critical applications are wide ranging, from issues involving national security and contraband identification to related receptors that are involved in many cellular and environmental signaling events. However, the mechanisms of underlying primary olfaction are not well understood. There is currently no atomic-level resolution for the structure of an olfactory receptor. One of the crucial bottlenecks is the difficulty of finding a surfactant that maintains olfactory receptor function and stability. Our interdisciplinary CBE team has become expert in olfactory receptor production and the technology for stabilizing and integrating membrane proteins. By June 2010, 12 olfactory receptors had been produced *en masse* using short designer lipid-like peptides as surfactants to solubilize and stabilize each receptor. Circular dichroism showed that purified olfactory receptors had alpha-helical secondary structures. Microscale thermophoresis ligand-binding measurements suggested that the receptors were functional and bound their odorants. These receptors were thoroughly assayed and used to confer selectivity to three independent, label-free binding detection/signal transduction technologies. Speckle interferometry, dielectric spectroscopy, and parallel resistometry devices (portable and scalable) were deployed in the DARPA presentation and shown to be capable of single odorant detection at concentrations comparable to the lowest limits of detection by trained dogs. On the basic science side, the newly discovered peptide surfactants demonstrated their usefulness as a new class of surfactants for olfactory receptors and possibly other GPCRs and diverse membrane

proteins. This project involved the collaboration of two teams of CBE researchers along with industry (Nano Temper Technologies GmbH, Germany) and investigators at Ludwig-Maximilians University.

CBE researchers have further advanced the design of special self-assembling peptide scaffold applications in cell culture and regenerative medicine. The functionalization of hydrogel scaffolds with biologically active motifs presents many possibilities for the design of synthetic biological materials for 3D cell culture; reparative, regenerative medicine; and tissue engineering. New discoveries have involved functionalized scaffolds of short peptides containing matrix metalloproteinase-2 (MMP-2) cleavable motifs designed to promote fibroblast proliferation and accelerated 3D cell migration independent of scaffold mechanical stiffness. These scaffolds are based on the peptides VEVK9 (Ac-VEVKVEVKV-CONH₂) and VEVK12 (Ac-VEVKVEVKVEVK-CONH₂), which were directly coupled to motifs containing two RGD binding units, cell adhesion sequences of laminin, and an MMP-2 cleavable sequence. These scaffolds were demonstrated to promote proliferation of periodontal ligament fibroblasts. Moreover, when combined with the MMP-2 cleavable motifs, the peptide scaffolds significantly accelerated the fibroblasts' migration. The recently published results suggest that the interaction between integrin receptors of fibroblasts and cell adhesion motifs of the scaffolds stimulated MMP-2 production by the fibroblasts to accelerate migration via cell-mediated proteolysis of the scaffold, similar to the manner in which migration would occur through the native extracellular matrix. These peptide scaffolds may be particularly useful in tissue engineering and regenerative medicine for periodontal tissue reconstruction. Related peptide scaffolds demonstrated enhanced biosynthesis of type I and type III collagens, which are critically important matrix protein components of the periodontal ligament, thus promoting wound healing and periodontal tissue regeneration.

In an exciting new project area, CBE researchers are designing nanofiber-based hydrogels for slow and sustained release of protein drugs. Controlling cellular microenvironments is thought to be critical for the successful application of biomaterials for regenerative medicine strategies. A class of RADA16 self-assembling peptides that have been successfully used for tissue engineering applications is now being used for these drug delivery applications. Understanding the fundamental mechanisms of protein mobility within, and ultimately release from, this nanostructured system is a critical aspect of controlling cellular activity. Also, in a recently published study, the CBE team has shown that designer peptide scaffolds facilitate slow and sustained release of bioactive growth factors that are relevant to tissue synthesis and repair. Multiple diffusive mechanisms within the hydrogels were observed for several human growth factors—fibroblast growth factor-2 (FGF-2), vascular endothelial growth factor (VEGF), and brain-derived neurotrophic factor (BDNF)—depending in part on the net positive or negative charges located at the C-terminus of the peptide nanofibers. In some cases, two populations of diffusing molecules were observed at the molecular level: one diffusing fully within the solvent and another exhibiting hindered mobility. Thus, the results suggest that growth factor mobility can be regulated by both steric and electrostatic interactions between the growth factors and the scaffold nanofibers. Moreover, assays using adult neural stem cells were employed to assess the functional release of active

FGF-2 for up to three weeks. The results provide not only evidence of long-term molecular release from self-assembling peptide scaffolds but also inspiration for other slow molecular release strategies for clinical applications.

In a collaboration involving Brigham and Women's Hospital and Ipsen Pharmaceuticals, and with grants from NIH and the Massachusetts Life Sciences Center, CBE researchers discovered another important means of delivering growth factors for tissue repair. Here the goal was to design a modification of the growth factor protein itself to enable retention of the protein within tissues while maintaining bioactivity. CBE faculty and students focused on insulin-like growth factor-1 (IGF-1), which stimulates cartilage repair but is not a practical therapy because its small size leads to its rapid release from the body. The CBE team modified IGF-1 by adding a heparin-binding domain and showed that this fusion protein (HB-IGF-1) stimulates sustained extracellular matrix synthesis in cartilage. The newly published discoveries focused on the mechanism by which HB-IGF-1 is retained in such connective tissues, and the team further tested whether HB-IGF-1 provides sustained growth factor delivery to cartilage in vivo and to human cartilage explants. Results showed that intra-articular injection of HB-IGF-1 led to sustained delivery through binding to the high density of negatively charged chondroitin sulfate chains of the matrix proteoglycan aggrecan. Thus, electrostatic interactions were important to the mechanistic understanding of this problem. Most importantly, in vivo studies involving intra-articular injection in rat knees showed that HB-IGF-1 was retained in articular and meniscal cartilages, but not in tendon, consistent with enhanced delivery to chondroitin sulfate-rich cartilage. Finally, HB-IGF-1 but not IGF-1 was also retained in human cartilage explants. Thus, modification of growth factors with heparin-binding domains may be a new strategy for sustained and specific local delivery to tissues such as cartilage that contain a high density of ionized fixed charge groups within their matrix. Tissue fixed charge can therefore provide a depot-like local, sustained delivery of IGF-1 to desired cells. These results are also leading to new interactions with Merrimack Pharmaceuticals (Cambridge, MA) focusing on targeted delivery of growth factors for bone and cartilage pathologies. In addition, a new Boston-area-based center of excellence in osteoarthritis in collaboration with Harvard Medical School and Boston University Medical School will likely include expansion of these efforts.

Core Facilities

One of the critically important missions of CBE is to maintain and expand a set of central core research facilities. These core facilities are made available to faculty, staff, and students, MIT-wide, at no or minimal user cost, and are particularly relevant for CBE's major research areas. These facilities include the new Attana 200 dual-channel, label-free, temperature-controlled, continuous-flow system for analysis of molecular interactions (e.g., kinetics, affinity and off-rate screening) and a Rigaku X-ray diffraction facility for protein and molecular crystallography and analysis. In addition, CBE continues to maintain the widely used Applied Biosystems (AB) 7900HT fast real-time 384-well plate quantitative polymerase chain reaction (qPCR) instrument and associated peripherals and a NanoDrop ND-1000 Spectrophotometric Analyzer to measure RNA quality (which is useful in assessing samples prior to amplification using the AB 7900HT qPCR facility). An Alpha Innotech Gel Imaging Facility has been added for quantitative analysis of

electrophoresis gels, along with a multiphoton microscopy facility, a Cressington Quick-Freeze Deep Etch facility to prepare specimens for follow-on electron microscopy, and a BiaCore 2000 surface plasmon resonance instrument to quantify binding reaction constants between molecules and between molecules and surfaces. CBE also continues to run a large cell, tissue, and organ culture facility including four six-foot biosafety cabinets and eight incubators. These are available to faculty and students MIT-wide who would otherwise not be able to explore new ventures in biomedical engineering involving living cells because of a lack of specialized facilities in their own laboratories (including an array of associated instruments and peripherals needed for maintaining and experimenting with living cells and tissues). All of these instruments are located on the second- and third-floor laboratories of CBE in NE47 (500 Technology Square).

UROP Activities

CBE continues to connect outstanding undergraduate students in several departments at MIT to laboratories associated with the center as well as laboratories at Boston's Beth Israel Deaconess Medical Center working in cancer-related research. CBE provides logistical and administrative support to ensure the continued success of this long-standing partnership.

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More information about the Center for Biomedical Engineering can be found at <http://web.mit.edu/cbe/www/>.