Massachusetts Institute of Technology
MIT Grier Room 34-401, 50 Vassar Street, Cambridge, MA
November 16, 2004
4:00 – 6:00 P.M.

Lester Wolfe Workshop in Laser Biomedicine
Optical Methods in Managing Diabetes: Will Technology or Biology Succeed First?

3:30 P.M. Refreshments

4:00 P.M. Challenges and Opportunities in Managing Diabetes
David Nathan, MD, Director, General Clinical Research Center and the Diabetes Center, Massachusetts General Hospital; *Professor of Medicine, Harvard Medical School, dnathan@partners.org*

Diabetes mellitus represents the most common, severe chronic disease in the US and much of the world. There are 1,200,000 new cases of diabetes annually in the US. The long-term complications of diabetes cause more blindness, kidney failure and amputations than any other disease and contribute to as much as 40% of all heart disease. Fortunately, we have effective interventions to prevent or limit the long-term complications of diabetes and to prevent the epidemic form of diabetes, Type 2 diabetes. Managing diabetes effectively requires frequent glucose monitoring. Advances in such monitoring should improve the chronic levels of glycemia experienced by diabetic patients, lessen the burden of self-care, and ultimately decrease the morbidity and mortality of the disease.

4:15 P.M. Imaging Islet Cell Function: From Single Cells to Intact Islets
David Piston, PhD, Professor, Department of Molecular Physiology & Biophysics and Department of Physics, Vanderbilt University, dave.piston@vanderbilt.edu

The convergence of newly developed instrumentation and optical probes allows us to examine quantitatively dynamic processes within ever more complicated biological systems. By using quantitative fluorescence imaging methods such as fluorescence recovery after photo bleaching (FRAP) and Förster resonance energy transfer (FRET) of multi-colored GFPs fused to the glucose sensing enzyme glucokinase (GK), we have discovered that the location and activity of beta cell GK is acutely regulated by insulin. These findings provide a mechanism whereby the glucose sensing ability of the beta cell is tightly coupled to insulin signaling. We have also measured pancreatic b-cell metabolism during glucose stimulation of pancreatic islets by quantitative two-photon NAD(P)H imaging. We have developed methods to delineate quantitatively the NAD(P)H signals from the cytoplasm and mitochondria, and show that the metabolic response of these two compartments are differentially stimulated by glucose and other metabolites. Absolute levels of NAD(P)H were determined using two-photon excited fluorescence lifetime imaging (FLIM). These findings elucidate the relative contributions of glycolytic and citric acid cycle metabolism in the normal and diabetic insulin secretion pathways.

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4:50 P.M. Diabetic Retinopathy: From Basic Science to Clinical Studies

Sven Bursell, PhD, Investigator in Section on Eye Research and Director, Joslin Vision Network; Associate Professor of Ophthalmology, Harvard Medical School, sven.bursell@joslin.harvard.edu

Diabetic retinopathy is the leading cause of new blindness in working age persons and leads to annual federal health care expenditures of $639 million. The ETDRS study showed that laser panretinal photocoagulation (PRP) was clinically effective in reducing the risk of vision loss to less than 5%. Despite this clinical benefit PRP destroys retina in an attempt to maintain vision with a number of associated adverse effects such as reduction in peripheral vision. Because PRP is a destructive therapy it becomes attractive to consider noninvasive, nondestructive therapies for the treatment of diabetic retinopathy. The Joslin Diabetes Center's basic research has focused on identifying potential therapeutic agents such as protein kinase C-b inhibitors. Results show that protein kinase C (PKC) activation plays an important role in the development of early retinal abnormalities in diabetes and that PKC inhibitors are effective in ameliorating these physiological abnormalities. PKC activation mediates increased endothelin-1 expression resulting in early reductions in retinal blood flow. Retinal blood flow reduction has been measured in early diabetes in diabetic rats and mice and in diabetic patients with less than 10 years duration of diabetes. Additionally, in a transgenic mouse model over-expressing PKC-b, retinal blood flow is reduced, endothelin-1 expression is increased, and diabetic retinopathy lesion development in these animals is accelerated compared to wild type diabetic mice.

Inhibition of PKC activation has been evaluated clinically a phase I b single center, randomized, parallel, double masked placebo controlled trial. Twenty-nine patients with disease duration less than 10 years participated in the study with oral treatment of the PKC inhibitor for 30 days. Results from this study showed that treatment significantly ameliorated associated reductions in retinal blood flow and did not adversely affect retinal vascular leakage in diabetic patients with normal retinal vascular permeability. Potential beneficial results using PKC inhibition have now been extended into phase III clinical trials.

5:25 P.M. Optical Methods for Noninvasive Blood Glucose Analysis

Mark Arnold, Director, Optical Science and Technology Center and Professor of Chemistry, University of Iowa, mark-arnold@uiowa.edu

Optical methods are under consideration as a means to measure blood glucose non-invasively in human subjects. The concept is to pass a selected beam of light into the human body and then determine the level of glucose from an analysis of the resulting spectrum. This approach is noninvasive in that no sample is required—there is no need to collect a sample of blood or interstitial fluid. Clinically, noninvasive methods offer a number of critical features that render this approach attractive for monitoring glycemia in the treatment of diabetes. Specifically, pain-free and reagent-less noninvasive measurements may improve tight glycemic control by encouraging frequent measurements through the elimination of pain associated with collecting blood samples and reduction in the test costs. In addition, an optical noninvasive glucose monitor offers the potential for continuous glycemic measurements while avoiding the complications associated with implantable glucose biosensor technologies. These features drive the development of this technology.

The ability to determine the concentration of glucose from an analysis of spectra collected non-invasively from human subjects is not trivial. A high degree of selectivity is required to distinguish glucose from the large number of endogenous molecules within the human body. The chemical structure of the glucose molecule does not provide a uniquely distinguishing spectral feature over the wavelengths of light that can be used to probe inside the human body. Optical signals from glucose overlap those from other endogenous substances, thereby demanding sophisticated methods to extract the glucose information selectively.

Successful noninvasive blood glucose monitoring represents a great challenge and requires a detailed understanding of the chemistry, physics, physiology, and optics associated with the measurement. Different approaches will be reviewed and the current state-of-the art will be discussed.

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CIMIT MEETING SUMMARY 11/16/2004

The meeting was held in conjunction with the Lester Wolfe Workshop in Laser Biomedicine and focused on "Optical Methods in Managing Diabetes". David Nathan, MD, MGH reviewed the current status of diabetes care in a talk entitled "Challenges and Opportunities in Managing Diabetes". The long-term complications of diabetes cause more blindness, kidney failure and amputations than any other disease and contribute to as much as 40% of all heart disease. Current treatment of Type I diabetes is guided by the glucose hypothesis, namely that treatment that normalizes glucose levels will delay or eliminate most of these complications. Control of glucose levels requires frequent glucose monitoring, which currently is very demanding of patients. Advances in such monitoring should improve the chronic levels of glycemia experienced by diabetic patients, lessen the burden of self-care, and ultimately decrease the morbidity and mortality of the disease. In addition, external and implantable insulin pumps currently operate open loop, without feedback; the development of non-invasive continuous glucose monitors will not only allow closed-loop control of pumps but will also decrease the effort of patients using conventional insulin injection. Currently, most commercial monitoring devices are inaccurate at low glucose levels and are not widely used. A biological alternative to technological solutions is the pancreatic transplant, which is, however, limited by the shortage of donor organs. An alternative biological approach is transplantation of islet cells via a transhepatic stick; this procedure can clamp glucose levels near normal physiological ones.

David Piston, PhD, Vanderbilt, spoke on "Imaging Islet Cell Function: From Single Cells to Intact Islets". His goals include using spectroscopic methods with high spatial resolution to understand the critical biological factors in development of normal pancreatic cells and, ultimately, to apply the knowledge gained to create functional beta cells and effective transplants. A number of quantitative fluorescence imaging methods such as fluorescence recovery after photobleaching (FRAP) and Förster resonance energy transfer (FRET) of multi-colored green fluorescent proteins (GFPs) fused to the glucose sensing enzyme glucokinase (GK) have been used and have led to the discovery that the location and activity of beta cell GK is acutely regulated by insulin. These findings demonstrate a mechanism whereby the glucose sensing ability of the beta cell is tightly coupled to insulin signaling. Methods to delineate quantitatively the NAD(P)H signals from the cytoplasm and mitochondria have been developed and show that the metabolic response of these two compartments is differentially stimulated by glucose and other metabolites. Absolute levels of NAD(P)H were determined using two-photon excited fluorescence lifetime imaging (FLIM). These findings elucidate the relative contributions of glycolytic and citric acid cycle metabolism in the normal and diabetic insulin secretion pathways.

Sven Bursell, PhD, Joslin Vision Network, spoke on "Diabetic Retinopathy: From Basic Science to Clinical Studies". Diabetic retinopathy is the leading cause of new blindness in working age persons. Laser panretinal photocoagulation (PRP) is clinically effective in reducing the risk of vision loss to less than 5%. Despite this clinical benefit, PRP destroys retina in an attempt to maintain vision with a number of associated adverse effects such as reduction in peripheral vision. Because PRP is a destructive therapy it becomes attractive to consider non-invasive, non-destructive therapies for the treatment of diabetic retinopathy. The Joslin Diabetes Center's basic research has focused on identifying potential novel therapeutic agents such as protein kinase C-b inhibitors. Results have shown that protein kinase C (PKC) activation plays an important role in the development of early retinal abnormalities in diabetes and that PKC inhibitors are effective in ameliorating these physiological abnormalities. Inhibition of PKC activation has been evaluated clinically a phase I b single center, randomized, parallel, double masked placebo controlled trial. Twenty-nine patients with disease duration less than 10 years participated in the study with oral treatment of the PKC inhibitor for 30 days. The results from this study showed that treatment significantly ameliorated associated reductions in retinal blood flow and did not adversely affect retinal vascular leakage in diabetic patients with normal retinal vascular permeability. The potential beneficial results using PKC inhibition have now been extended into phase III clinical trials.

Mark Arnold, University of Iowa, discussed. “Optical Methods for Noninvasive Blood Glucose Analysis”. While numerous such optical methods are being studied, selectivity is an ongoing problem with all of them. Direct measurements are based upon some intrinsic optical property of glucose, while indirect ones measure the effect of glucose upon some secondary process. Indirect approaches include sensing light scattering, skin autofluorescence and skin structure. Because glucose is not the only means by which these properties can be altered, selectivity is an issue and detailed in-vivo experiments must be performed. Direct glucose sensing, based on measuring intrinsic spectral properties of glucose, suffers from the presence of many biomolecules which can interfere. Vibrational properties of glucose can be exploited by either Raman spectroscopy or by infrared (IR) absorption spectroscopy. Near IR spectroscopy suffers from weak glucose absorption but does provide access to a unique glucose spectrum. Initial solution experiments using multivariate calibration were described. The method uses spectra of known concentrations of analyte to establish a correlation between concentration and spectral variance. However non-invasive glucose measurements which demonstrate an unambiguous correlation of signal changes with concentration have not yet been demonstrated. In-vivo experiments using a rat model were described; fiberoptics were used to measure the transmission through a pinch of tissue. Glucose was injected after a constant level had been established and the characteristic variance of the spectra above baseline determined. This variance was compared with that obtained using glucose solutions. The data are still somewhat preliminary, but do demonstrate spectral changes resulting from glucose injection in an animal model.

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