

Session P – Bioengineering (Alphabetical)

An Optical System for Diagnosing and Monitoring Dermal Microvascular Health

Eric M.Chin

Mentors: Morteza Gharib, Danny Petrusek, and Alan Barr

We present dynamic microvascular refill (DMR), a novel measure of microvascular function. We assess microvascular refill in dynamic conditions by applying a series of pressures and monitoring changes in fingernail reflectance spectra. We hope to investigate differences in DMR between diagnosed diabetic and healthy normal populations; additionally, in diabetic populations, we seek correlation between DMR and HbA1c, a relatively long-term measure of blood glucose. Finally, we verify that the source is microvascular by validating against impedance plethysmography.

Characterization of a Breast Cancer Progression Model with a Hereditary Component

Adam Khan

Mentors: Thorarinn Gudjonsson, Magnús Karl Magnússon, and Saevar Inghorsson

Breast cancer affects 10% of women worldwide with 5-10% estimated to result from a hereditary component. Basal-like breast cancer is an aggressive subgroup of breast cancer that originates in the stem or progenitor cells of the breast and is associated with a negative prognosis. An epithelial cell-line A163 was established from a breast carcinoma of a patient who had a strong family history of breast cancer but none of the known genetic mutations typically associated with hereditary breast cancer. A163 was cultured *in vitro* in a serum-free culture medium H14 with 7 growth factors including the Epidermal Growth Factor (EGF). Previous studies have shown that ablation of EGF from culture medium followed by EGFR-1 amplification facilitates tumor progression. A163 exhibits a basal-like phenotype as shown by its expression of characteristic luminal and myoepithelial cytokeratins. Ablation of EGF from culture medium of A163 resulted in an EGF independent sub-line A163-S1 that retained the basal-like phenotype of A163. Interestingly, removal of EGF from H14 resulted in the over-expression of the Epidermal Growth Factor Receptor (EGFR-1) in A163-S1 further strengthening the basal-like phenotype. Understanding EGFR-1 over-expression in the A163-S1 breast cancer progression model may have important implications in the study of basal-like tumors.

Dynamic Microenvironments for the Study of Tissue Morphogenesis

Amy T. Lam

Mentors: Chin-Lin Guo, Wonhee Lee, and Mingxing Ouyang

The loss of robust assembly of tissue can lead to severe problems in embryonic development, tissue growth, and regeneration, sometimes leading to cancer. In order to better understand what constitutes healthy tissue morphogenesis, we must first understand what occurs at the cellular level. While there have been efforts to understand the intracellular mechanics of cellular morphogenesis and the effects of various static microenvironments, there have been very few that address the more realistic dynamic environment. In order to study these effects, we must first be able to create controllable microenvironments. We have been successful in creating a series of microenvironments using polydimethylsiloxane (PDMS), which we are able to pressurize with air, and thus actively change the topology of the device. In this experiment, we use Madin Darby canine kidney (MDCK) cells to test the feasibility of growing cells on our device; however, we plan on using undifferentiated mouse embryonic stem cells in the future. By culturing a sheet of cells on top of the device, we should be able to see the cell and tissue response to changes in the microenvironment.

Another Look at the Two-State System: Testing a New Theory of Non-Equilibrium Statistical Mechanics

Sarah Marzen

Mentor: Rob Phillips

Two-state systems are useful for modeling everything from the opening and closing of ion channels to ligand binding. Although the two-state system has already been well-studied, an in-depth analysis of a two-state system using Maximum Caliber, a relatively new theory of non-equilibrium statistical mechanics based in information science, reveals several insights into the nature of the two-state system and the foundations of Maximum Caliber. According to Maximum Caliber, understanding the dynamics of a two-state system is equivalent to understanding the 1-D Ising model; the theoretical calculations obtained from Maximum Caliber agree well with simulated data. Furthermore, Maxwell-like relations reveal a new relationship between changes of average state and changes in the average number of transitions with respect to rate constants. We raise and address possible resolutions to objections that Maximum Caliber is information science and not physics.

Single Molecule Dynamics of Concerted HIV-I Integration (*in vitro*) Using Tethered Particle Motion

Pradeep Ramesh

Mentors: David Baltimore, Rob Phillips, and Geoffrey Lovely

Concerted integration of retroviral genomic DNA by viral integration complexes (PIC) is necessary to propagate the viral infection. Previous methods of examining integration mechanics relied on reconstituting integration using traditional strand transfer assays and were consequently unable to elaborate on intermediate steps. We therefore propose a novel single-molecule approach to studying *in vitro* integration using a modified tethered particle motion set-up. The new approach enables (1) the biophysical characterization of purified integrase in real time; (2) the determination of any length-dependence limits of viral integrase; (3) the measurement of integration kinetics using fluorescently labeled viral substrates in real time; (4) the observation of DNA bending under its persistence length thus challenging the long-standing linked-chain polymer model; (5) the examination of potential integrase inhibitors and their effect on concerted integration reactions which has clinical relevance in the treatment of AIDS.

A Census of Lac Repressor at the Single-Cell Level

Dan (Linda) Song

Mentors: Rob Phillips and Hernan G. Garcia.

The quantitative study of gene expression *in vivo* calls for theoretical models that go beyond simple pictorial descriptions since the data currently reported on levels of gene expression is often quantitative. In particular, we explore thermodynamic models which predict the fold-change in gene expression as a function of concentration of the relevant transcription factors and strength of their interaction with DNA and the transcriptional machinery. Knowing the absolute number of transcription factors present inside the cells thus becomes of special interest as a way of dialing a knob of the machinery to test the predictions of these models. In this work, we quantified the absolute level of Lac repressor at the single-cell level *in vivo* using a recently developed fluctuation method. We are able to demonstrate the dependency of fold-change on the concentration of the repressors for the simple repression system, which is consistent with the prediction of the corresponding thermodynamic model. Furthermore, this work serves as a foundation for applying the same technique to more complex regulatory motifs such as DNA looping in which a precise number of regulators is crucial for explicitly testing the applicability of the thermodynamic models.