ROBERT R. PUCCINELLI

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EDUCATION:

2010 – 2014 **B.S. Physics,** Biophysics
B.S. Biological Sciences, Microbiology and Immunology
University of California, Merced
Cumulative GPA: 3.675, STEM GPA: 3.754

RESEARCH EXPERIENCE:

Research Associate II, Chan Zuckerberg Biohub Jun. 2017 – *present*
Rafael Gómez-Sjöberg, PhD; *Bioengineering Systems Development*

Chan Zuckerberg Biohub was launched by Mark Zuckerberg and Priscilla Chan with the intention to cure every disease within 100 years. The bioengineering team sits in a unique position as we support collaborative efforts between Stanford, UCSF, UC Berkeley, internal scientists and international collaborators by building instrumentation on-demand to address new scientific questions or enhance current scientific capabilities.

Independent Projects

- Collaborated with internal scientists to develop a high-density tube capping-decapping system to enable the use of tubes with unique IDs for storing and tracking sensitive samples. The first prototype was a semi-automated unit that reduced cap transfer time from 10 minutes to 2. The second prototype being built is a fully automated system that is designed to integrate with a custom automated cell culture system.
- Collaborated with protein scientists to develop a protein purification system to improve throughput while maintaining flow rate control at cost-sensitive prices. The success and demand for the system has led to continued development to make a fully automatable and parallelized system.
- Collaborated with members of the CZ Biohub microbiome initiative to organize instrumentation in a large anaerobic chamber and developed motorized axes to transfer materials across the workspace.

Contributions to Team Efforts

- Developed an autosampler that aliquots the output of a microfluidic device into PCR tubes
- Developed an electronic demultiplexer that selectively activates one of several LED light sources on a custom UV microscope using a single LED driver and PC-controlled IO
- Developed a cantilevered arm and plate chiller that collects and preserves rare cell populations acquired from a custom single cell picker system
- Developed a capillary holder mechanism for imaging in a tightly confined dual light sheet microscope
- Developed a digital, parallelized reagent degassing module for a custom in-situ transcriptomics microscope
- Developed Arduino code and a LCD module for an ultra low cost turbidy meter that was built and deployed to Uganda within 3 days to replace faulty equipment in a UC Berkeley CEND protein purification workshop at Makerere University

Additional Responsibilities

- Regularly conducted 3D printer training with FDM and SLA printers for internal scientists and external collaborators to enable them to prototype their own designs
- Co-designed and instructed the “Fab in Lab” minicourse at UCSF with the CZB bioengineering team

Life Sciences Research Professional, Stanford University Sep. 2014 – May 2017
Polly Fordyce, PhD; *Biophysical Interactions in Microfluidics*
High-Throughput Analysis of DNA-Protein Biophysical Interactions

My research in the Fordyce Lab was centered around employing the MITOMI microfluidic device, a multiplexer with >1500 sample chambers that mechanically traps molecular interactions, for high throughput investigations of transcription factor – DNA oligo binding equilibrium and kinetics to access fine resolution data that is not readily observed in other systems such as ChIP-seq. To accomplish these objectives, I assembled hardware for these systems, prepared the supplies consumed in each experiment, wrote software to manipulate and analyze image datasets and trained other members.

MATLAB Custom Script Implementation (Independently Written)

- Updated microfluidic control scripts to permit MATLAB to control microscope and microfluidic devices
- Automated microfluidic device control and time series imaging protocol
- Decoupled stacked TIFFs to isolate fluorescent images of data and flat-field corrected images with MATLAB scripts before importing ImageJ to stitched as a complete image
- Processed complete image datasets with automated, high-throughput GUI for high content array analysis of TF-DNA binding events for >1500 isolated chambers. Reduced analysis from 2hrs to 15mins per dataset.

Sample Preparation

- PCR extended basic helix-loop-helix TF DNA such as Max and c-Myc to include transcription/translation sites before cloning into a plasmid and transforming competent E. coli for expansion and sequencing
- Prepared DNA oligo libraries with a fluorescent tag before printing onto glass slides with DeRisi-style microarrayers with printing solutions that were optimized for our setups
- Fabricated two-layer MITOMI devices with PDMS and aligned to >1500 DNA spots on glass slide

Instrumentation Assembly

- Assembled, maintained and established communication for several epifluorescent microscopes with PCs employing open-source MicroManager microscope controlling software
- Constructed a DeRisi-style microarryer and established connections for computer driven operation
- Constructed microfluidic pneumatic setups and established connections for computer driven operation

Undergraduate Research Assistant, University of California, Merced Oct. 2011 – May 2013
Kara McCloskey, PhD; *Cardiac Tissue Engineering*
Influence of Nanoscale Topography and Electrical Stimulation on Stem Cell Fate

Stem cells that have not attained an adult-like state are unsuitable for clinical applications due to their propensity to revert to a multipotent state and induce tumorigenic growth. My research focused on differentiating mouse and human embryonic stem cells into immature cardiomyocytes and investigating the effects of surface topography and electrical pacing on advancing their maturation to an adult-like state. Generating mature cardiomyocytes would be a field-wide milestone in the path to restoring function to regions of the heart that has been irreversibly damaged by hypoxia.

Tissue Culture

- Maintained multiple lines of mouse and human stem cells including thawing, feeding, passing and freezing of cultures so that excess cells were always ready for experiments in the lab
- Directed differentiation of multiple lines of stem cells towards the myocardial lineage with physical techniques, such as embryoid body formation, chemical techniques, such as scheduled growth factor exposure and feeding, and environmental techniques, such as electrical stimulation
- Cultures were labeled with fluorescently labeled with antibodies and imaged or FACS sorted

Additional Responsibilities

- Trained incoming students on cell culture, stem cell culture, and fluorescent microscopy to provide the with a basic skill set for their own projects
- Drafted experimental design for the project, directed and oversaw progress of the project, and discussed troubleshooting measures with my post-doctoral mentor
- Prepared and maintained reagent and media inventory

Undergraduate Research Assistant, University of California, Merced Jan. 2013 – Oct. 2014
Ajay Gopinathan, PhD; *Computational Cellular Biophysics*
Influence of Nanoscale Topography on Actin Dynamics

f-Actin is a cytoskeletal element that plays significant roles in cell motility, endocytosis and defining cell shape by polymerizing against regions of the cell membrane with positive curvature. For my physics senior thesis, I modeled actin polymerization events at the cellular scale with a 2D, coarse-grained, nearest-neighbor Monte Carlo simulation whereby parameters for membrane-surface binding events, membrane curvature and elasticity, in addition to actin polymerization were used to evaluate cell shape modulation over time. This has predictive power for applications in tissue engineering where nano- and microscale topography is utilized to influence stem cell differentiation with cell shape.

Computational Modeling

- Constructed model for actin dynamics at the cellular level de novo by considering resistive and propulsive forces in the system including membrane bending, membrane adhesion and actin polymerization
- Generated different modes of actin dynamics such as cell spreading and cyclical actin wave generation by tuning parameters in the model
- Model was applied to different surface topographies to evaluate their influence on the localization of actin polymerization and cell shape
- Extracted relevant energetic and rate constants from the literature or set to a neighboring value

PUBLICATIONS:

Diversification of DNA binding specificities enabled SREBP transcription regulators to expand the repertoire of cellular functions that they govern in fungi. Del Olmo Toledo V, Puccinelli R, Fordyce PM, Pérez JC. *PLos Genet.* 2018 Dec 31; 14(12):e1007884.

Single-cell transcriptomics of 20 mouse organs creates a Tabula Muris. Tabula Muris Consortium; Overall coordination; Logistical coordination; Organ collection and processing; Library preparation and sequencing; Computational data analysis; Cell type annotation; Writing group; Supplemental text writing group; Principal investigators. *Nature*. 2018 Oct; 562(7727):367-372.

An Open-Source, Programmable Pneumatic Setup for Operation and Automated Control of Single- and Multi-Layer Microfluidic Devices. Brower K, Puccinelli R, Markin CJ, Shimko TC, Longwell SA, Cruz B, Gómez-Sjöberg R, Fordyce PM. *HardwareX*. 2018 Apr; 3:117-134.

Optimized Sequence Library Design for Efficient In Vitro Interaction Mapping. Orenstein Y, Puccinelli R, Kim R, Fordyce P, Berger B. *Cell Systems.* 2017 Sep; 5(3):230-236.

GRADUATE TEACHING EXPERIENCE:

Fab in Lab, UCSF Summer 2019
Instructors: Joseph DeRisi and Wallace Marshall
Teaching Staff: Joanna Cabrera, Rafael Gómez-Sjöberg, Eric Lam, Paul Lebel, Robert Puccinelli, Kevin Yamuchi
BP 219

Microfluidic Device Design Laboratory, Stanford Winter 2017
Instructors: Polly Fordyce and Stephen Quake
Teaching Staff: Kara Brower, Diego Oyarzun, Robert Puccinelli, Adam White
BIOE 301D/GENE 207

POSTER PRESENTATIONS:

September 2016 **2016 Stanford Department of Genetics Annual Retreat**
Poster Presentation: Influence of Flanking Sequences in Transcription Factor - DNA Binding
Robert R. Puccinelli and Polly M. Fordyce

September 2015 **2015 Stanford Department of Genetics Annual Retreat**
Poster Presentation: Resolving functional networks in a highly proficient enzyme
C. Markin, R. Puccinelli, C. Guegler, F. Sunden, I. AlSadhan, P. Fordyce, D. Herschlag

March 2014 **2014 Undergraduate Research Poster Competition, UC Merced**
Poster Presentation: Modeling Actin Dynamics on Nanoscale Topography
Robert R. Puccinelli and Ajay Gopinathan
**Certificate of Achievement awarded for best poster presentation*

March 2013 **Student Research Poster Competition, UC Merced**
Poster Presentation: Roles for Electrical and Topographical Cues in the Maturation of Derived Cardiomyocytes.
Robert R. Puccinelli, William S. Turner, Kara E. McCloskey

August 2012 **Sixth Annual Summer Research Symposium, UC Merced**
Poster Presentation: Combinatorial Effects of Nanoscale Topography and Electrical Stimulation in Stem Cell Fate
Robert R. Puccinelli, William S. Turner, Kara E. McCloskey

AWARDS/HONORS:

2014 Graduation with Honors, *University of California, Merced*
2014 Outstanding Student Award, *University of California, Merced*
2014 Best Poster Presentation, *UC Merced Research Week 2014*
2007 Eagle Scout, *Boy Scouts of America*