Curriculum Vitae

Robert C. Brewster

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Employment

2015–Present ..... University of Massachusetts Medical School, **Assistant Professor**,

2009–2015 ..... California Institute of Technology, **Postdoctoral Fellow** (with Rob Phillips), Research focused on: Quantitative dissection of bacterial transcription regulation

2007–2009 ..... Weizmann Institute of Science, **Postdoctoral Fellow** (with Sam Safran), Research focused on: Finite sized domains in soft and biological matter

Education

2005–2007 ..... University of California, Los Angeles. Department of Chemistry, **Doctorate in Physical Chemistry** Advisor: Alex Levine

2002–2005..... University of Massachusetts, Amherst. Graduate Department of Physics. Completed graduate physics curriculum and qualification.

1998–2002..... University of Massachusetts, Amherst. **Bachelor of Science in Physics.**

Research Highlights

Spanning constitutive gene expression space with base pair resolution (with Daniel Jones and Rob Phillips). How can we build promoters from scratch? If we wanted to predictively tune gene expression levels over many orders of magnitude, what are the base pair rules used to generate such promoters? Using a sequence-dependent RNA polymerase-DNA binding model from available deep-sequencing data, we have designed and created a library of constitutive promoters with predicted expression spanning over three orders of magnitude. Both the mRNA copy number and bulk protein expression from these promoters are in good correspondence with the predictions from our models of transcription. We view characterization of constitutive promoters as imperative for rational promoter design as they are the "chassis" on top of which complex regulation schemes are built. We have demonstrated this modularity in alternate regulatory schemes built upon our constitutive libraries.

The quantitative consequences of "overworked" transcription factors (with Franz Weinert and Rob Phillips). Genes are turned "on and off" by the binding and unbinding of proteins known as transcription factors on the DNA. Interestingly, in cases when a gene is located on a plasmid or on a viral genome in a cell, the number of these transcription factors can be smaller than the number of copies of the genes that need to be regulated by that transcription factor. This phenomenon, which we have dubbed the transcription factor titration effect, leads to enormous changes in the level of expression. Using video microscopy, we have characterized this interplay between transcription factor copy number and the demand for that transcription factor and demonstrated a parameter-free model of transcription can precisely predict the expression levels based on these quantities.

Stable and finite sized domain formation in biological membranes (with Sam Safran). Systems capable of forming finite-sized, equilibrium domains are of biological interest in the context of membrane rafts where it has been observed that certain cellular functions are mediated by small (several to tens of nanometers) domains with specific lipid composition that differs from the average composition of the membrane. These small domains are composed mainly of lipids with completely saturated hydrocarbon tails that show good orientational order in the membrane. The surrounding phase consists mostly of lipids with at least one unsaturated bond in the hydrocarbon tails which forces a kink in the chain and

inhibits ordering. In vitro, this phase separation can be replicated; however, the finite domains coarsen into macroscopic domains with time. We have proposed a mechanism, backed by theoretical calculations, where another lipid may stabilize these small domains by acting as a line-active component, akin to a surfactant stabilizing oil and water interface.

Current research

The relationship between expression noise and promoter architecture (with Daniel Jones and Rob Phillips). Often the variability in gene expression is cited as a parameter the cell may tune to gain evolutionary fitness. Simple predictive models of transcription provide predictions for how this variability depends on the details of the gene's regulation. However, recent studies have made the surprising claim that the cell to cell variability in expression is independent of these molecular details and is "universal" depending only on the mean level of expression and thus not an independent tuning parameter for the gene. Using mRNA FISH, we have measured the noise in expression from constitutive promoters whose mean expression level spans three orders of magnitude. Furthermore, we have measured the noise from these promoters as we put them under regulatory control and systematically vary the number of transcription factors in the cell and the strength with which those transcription factors bind. Our results show that noise can be tuned in a quantitatively predicted way by systematically controlling the molecular details of the regulatory DNA.

The population genetics of competing plasmids (with Rob Phillips). Bacterial cells commonly carry and replicate small circular DNA segments known as plasmids. These plasmids typically encode several genes, often conferring some benefit to the cell such as generating proteins for antibiotic resistances. However, if cells are grown with two distinct but similar plasmids, over just several short generations, the cells will adopt either one of the plasmid but not both. In this case, the plasmid is not selected by fitness or evolutionary pressures but likely by random chance involved in winning the replication machinery for your own kind, increasing your population by one and making it more likely that your now larger population will win the replication competition going forward. Using video microscopy, I am exploring this competition in the theoretical context of population genetics, hopefully also shedding light on the mechanisms behind plasmid replication and segregation.

Awards

2013–2015 . . . . . Biology Division postdoctoral fellowship at the California Institute of Technology.

2008–2009 . . . . . Fulbright Foundation post-doctoral fellowship at the Weizmann Institute.

Publication List

1. Geoffrey A. Lovely, Robert C. Brewster, David G. Schatz, David Baltimore and Rob Phillips (2015) Single-molecule analysis of RAG-mediated V(D)J DNA cleavage, *PNAS*, **112(14)**, E1715-23

2. Daniel L. Jones*, Robert C. Brewster* and Rob Phillips (2014) Cell-to-Cell Variability in Gene Expression is Governed by Promoter Architecture, *Science*, **346 (6216)**: 1533-1536

3. Franz M. Weinert*, Robert C. Brewster*, Mattias Rydenfelt, Rob Phillips and Willem K. Kegel (2014) Scaling of Gene Expression with Transcription Factor Fugacity, *Physical Review Letters* **113**, 258101

4. Robert C. Brewster*, Franz M. Weinert*, Hernan G. Garcia, Linda Song, Mattias Rydenfelt and Rob Phillips (2014) The Transcription Factor Titration Effect Dictates Level of Gene Expression, *Cell* **156(6)** Research highlighted in:

   See manuscript highlighted in:

   - "Tuning expression by numbers", *Nature Methods* **11(475)**, 2014
   - "Target competition: transcription factors enter the limelight", Karreth, FA, Tay, Y, Pandolfi, P, *Genome Biology* **15(114)**, 2014

5. Benoit Palmieri, Tetsuya Yamamoto, Robert C. Brewster and Samuel A. Safran (2014) Line active molecules promote inhomogeneous structures in membranes: Theory, Simulations and Experiments, *Advances in Colloid and Interface Science (issue in honor of Wolfgang Helfrich)* **208**

6. Robert C. Brewster*, Daniel L. Jones* and Rob Phillips (2012) Tuning Promoter Strength through RNA Polymerase Binding Site Design in *Escherichia coli*, *PLoS Computational Biology* **8(12)**, e1002811

7. YongSeok Jho, Robert C. Brewster, Samuel A. Safran and Philip A. Pincus (2011) Long-Range Interaction between Heterogeneously Charged Membranes, *Langmuir* **27(8)**, 44394446

8. Robert C. Brewster and Samuel A. Safran (2010) Line active hybrid lipids determine domain size in phase separation of saturated and unsaturated lipids, *Biophysical Journal Letters* **98**, L21-Ld3

9. Tetsuya Yamamoto, Robert C. Brewster and Samuel A. Safran (2010) Chain ordering of hybrid lipids can stabilize domains in saturated/hybrid/cholesterol lipid membranes, *Europhysics Letters* **91**, 28002

10. Robert C. Brewster, Philip A. Pincus and Samuel A. Safran (2009) Hybrid Lipid as Biological Surfactants, *Biophysical Journal* **97**, 1087-194

11. Robert C. Brewster, Gary S. Grest, and Alex J. Levine (2009) Effects of Cohesion on the Surface Angle and Velocity Profiles of Granular Material in a Rotating Drum, *Physical Review E* **79**, 011305

12. Robert C. Brewster, Philip A. Pincus, and Samuel A. Safran (2008) Self Assembly Modulated by Interactions of Two Heterogeneously Charged Surfaces, *Physical Review Letters* **101**, 128101

13. Robert C. Brewster, Leonardo Silbert, Gary S. Grest, and Alex J. Levine (2008) Relationship between interparticle contact lifetimes and rheology in gravity-driven granular flows, *Physical Review E* **77**, 061302

14. Leonardo Silbert, Gary S. Grest, Robert C. Brewster and Alex J. Levine (2007) Rheology and Contact Lifetimes in Dense Granular Flows, *Physical Review Letters* **99**, 068002

15. Robert C. Brewster, James Landry, Gary S. Grest, and Alex J. Levine (2005) Plug flow and the breakdown of Bagnold scaling in cohesive granular flows, *Physical Review E* **72**, 061301

* denotes shared first authorship

Teaching and Short Course Assistant Positions

- Kavli Institute for Theoretical Physics: Evolutionary Cell Biology course 2015.
  - TA for Rob Phillips. Designed and ran two week project studying expression noise in *E. coli* under a wide array of regulation scenarios.
- Marine Biological Laboratory: Physiology summer course 2015.
  - TA for Matlab tutorial bootcamp. Assisted students in an introductory course in the use of Matlab to analyze microscopy data in the biological sciences. Subjects included: Image segmentation, particle tracking, signal quantification, simple simulations and modeling.

  - TA for Rob Phillips. Designed and ran two week project titled "Quantifying transcription through the cell-cycle in bacteria under diverse growth conditions", where two students used single-cell mRNA FISH to study how transcription changes over the course of the cell-cycle under a variety of growth conditions and environmental challenges. The measurements were compared against predictions from a simple theory of transcription to examine the robustness of the model under drastically different growth conditions.
- Marine Biological Laboratory: Physiology summer course 2014.
  - TA for Matlab tutorial bootcamp. Assisted students in an introductory course in the use of Matlab to analyze microscopy data in the biological sciences. Subjects included: Image segmentation, particle tracking, signal quantification, simple simulations and modeling.

- TA for Rob Phillips. Designed and ran two week project titled "Direct Imaging of Plasmid Competition in Living *E. coli*", where two students used advanced live-cell imaging techniques to directly image plasmids diffusing in living *E. coli* with 20ms resolution. The students were able to measure the diffusion constant of plasmids in a living cell and resolve several regimes of diffusion associated with the geometry of confinement within the cell.

- Marine Biological Laboratory: Physiology summer course 2012.
  - TA for Rob Phillips. Designed and ran two week research project titled "The Nature of Noise in Gene Transcription". Using single mRNA FISH students studied how transcriptional noise depends on gene copy number and growth rate.
- Marine Biological Laboratory: Physiology summer course 2011.
  - TA for Rob Phillips. Designed and ran two week research project titled "Transcriptional noise in Constitutive Gene Expression", where students used single mRNA FISH to explore the transcriptional output from a library of constitutive promoters and characterized the mean and noise as a function of the relevant molecular parameters, such as RNAP binding energy to the promoter.
- Marine Biological Laboratory: Physiology summer course 2010.
  - TA for Rob Phillips. Designed and ran two week research project titled "Noise in the Central Dogma", where students used single mRNA FISH and widefield fluorescence microscopy to explore the contributions of each step along the central dogma to the overall noise of the process of producing proteins.
- UCLA: Upper level Chemical Thermodynamics, Fall 2006 and Winter 2007.
  - Assisted with lectures and taunt a discussion section in an advanced physical chemistry course on thermodynamics.
- UCLA: General and Organic Chemistry Laboratory I, Spring 2006.
  - Taught two lab sections in an introductory chemistry laboratory.
- UCLA: Chemical Energetics and Change, Winter 2006.
  - Taught two discussion sections in an introductory course covering phase behavior, chemical equilibrium and chemical kinetics.
- UCLA: Chemical Structure, Fall 2005.
  - Taught four discussion sections in an introductory course on the structure of molecules with an introduction to quantum mechanics.
- UMASS: Research Assistantship, Summer 2003 - Summer 2005.
  - Research on granular flow through large-scale granular dynamics simulations.
- UMASS: General Physics Laboratory I, Spring 2003.
  - Taught four general physics laboratory sections designed for Engineers and other science majors
- UMASS: Tutor for high school physics and calculus, Fall 2001 - Spring 2002
  - Tutored two high school students in honors physics and calculus
- UMASS: Society of Physics Students outreach program, Fall 1999 - Spring 2002
  - Visited local elementary schools to teach physics through heavy use of hands on demonstrations

References

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