Gresham Lab Strategic Review

January 2015
What our lab studies

Our lab studies adaptive evolution, control of cell growth and regulation of mRNA degradation.

Our approach integrates high-throughput genome-scale methods and computational biology with microbiology and molecular biology.
Published 2 peer reviewed research papers and 2 peer reviewed commentaries
Steff passed quals and joined the lab
Sam joined the lab
Benjy and Jungeui successfully defended PhD theses (PhD’s completed in 5.33 years)
Jungeui interviewed for post-docs with Vidal, Altschuler/Wu, Lieb and Iacobuzio-Donahue labs. Accepted post-doc in Iacobuzio-Donahue lab (MSKCC)
High School students Wei and Anise spent their second summer in the lab mentored by Niki
Victoria graduated and joined Sevinc’s lab
David applied for tenure
Lab represented at:
- NYU Abu Dhabi Genome Meeting (David)
- SMBE (Naomi and David)
- RNA meeting (Niki and David)
- ASM Experimental Evolution meeting (Jungeui and David)
- GSA Yeast Genetics meeting (David)
- EMBL Experimental Approaches to Evolution in Yeast and other species (Naomi and David)
- NYAPG meeting (Naomi, Sam David)

Awards
- Benjy awarded the Steve Kazianis award
- Jungeui awarded the Fleur Strand award
- Christina awarded DURF and Jack Sitt Scholar award
The functional basis of adaptive evolution in chemostats

David Gresham & Jungeui Hong

Department of Biology, Center for Genomics and Systems Biology, New York University, New York, NY, USA
Determination of in vivo RNA kinetics using RATE-seq

Benjamin Neymotin, Rodoniki Athanasiadou and David Gresham

RNA published online August 26, 2014
De-Novo Learning of Genome-Scale Regulatory Networks in *S. cerevisiae*

Sisi Ma¹, Patrick Kemmeren³, David Gresham⁴, Alexander Statnikov¹,²*¹

¹ Center for Health Informatics and Bioinformatics, New York University Langone Medical Center, New York, NY, United States of America, ² Department of Medicine, New York University School of Medicine, New York, NY, United States of America, ³ Molecular Cancer Research, Center for Molecular Medicine, University Medical Center, Utrecht, The Netherlands, ⁴ Department of Biology, New York University, New York, NY, United States of America
The enduring utility of continuous culturing in experimental evolution

David Gresham a, Maitreya J. Dunham b

a Center for Genomics and Systems Biology, Department of Biology, New York University, New York NY, USA
b Department of Genome Sciences, University of Washington, Seattle WA, USA
NSF Specific Aims

The functional basis of genetic interactions underlying quantitative trait variation

Objective 1: Select mutants with increased growth rate in nitrogen-limited environments and identify all acquired genetic variation.

Objective 2: Quantify the individual effect of acquired genetic variants in mutants and test whether the combined effect of genetic variants is explained by an additive or non-additive model.

Objective 3: Generate a testable model of the functional relationships underlying different genetic interaction classes.
NSF Specific Aims

The functional basis of genetic interactions underlying quantitative trait variation

**Objective 1:** Select mutants with increased growth rate in nitrogen-limited environments and identify all acquired genetic variation. We will perform long-term selection experiments in nitrogen-limited environments and identify spontaneous mutants with increased growth rates. We will quantify the growth rate of mutants in the same condition and identify all acquired genetic variation using high throughput sequencing. **Completion of objective 1 will result in a collection of 54 mutants for which we have 1) quantified growth-rate (our quantitative trait of interest) and 2) identified all acquired mutations.**

**Objective 2:** Quantify the individual effect of acquired genetic variants in mutants and test whether the combined effect of genetic variants is explained by an additive or non-additive model. We will measure the phenotypic effect of individual acquired mutations and pairwise combinations. We will perform statistical tests to define the class of genetic interaction for each pair of QTL tested. **Completion of objective 2 will result in the rigorous definition of interactions between QTL as either additive or one of the three possible classes of epistatic interactions.**

**Objective 3:** Generate a testable model of the functional relationships underlying different genetic interaction classes. We will identify the functional relationships that underlie different genetic interaction classes and experimentally test our model. **Completion of objective 3 will result in a model that predicts the type of interaction between QTL on the basis of functional relationships between genes and tests that model.**
NIH RO1 specific aims

Regulation of Quiescence in Eukaryotic Cells

• **Aim 1.** Define the conserved quiescence program.

• **Aim 2.** Determine how signaling pathways coordinately regulate initiation of quiescence.

• **Aim 3.** Test the role of regulated variation in mRNA stability as cells enter quiescence.
NIH RO1 specific aims

Regulation of Quiescence in Eukaryotic Cells

• **Aim 1. Define the conserved quiescence program.** Using multiplexed analysis of genome-wide libraries of mutants in budding and fission yeast and high throughput sequencing, we will quantitatively define the role of each gene in quiescence initiated by carbon, nitrogen or phosphorous starvation (nutrients essential for cell growth that are also depleted in some tumor microenvironments). We will study physiological hallmarks of quiescence in homologous mutants defective in quiescence to define their functional roles.

• **Aim 2. Determine how signaling pathways coordinately regulate initiation of quiescence.** We hypothesize that diverse signals are integrated by a network of interactions between signaling pathways. To test this hypothesis, we will identify the targets and interactions of conserved protein kinases that regulate quiescence using quantitative genome-wide genetic interaction mapping in both species in different quiescence-inducing conditions. In addition, we will test the role of subcellular localization of signaling pathway components in regulating quiescence using protein mislocalization experiments.

• **Aim 3. Test the role of regulated variation in mRNA stability as cells enter quiescence.** We will quantify variation in the rate of mRNA synthesis and degradation for each transcript in both species as cells enter quiescence using *in vivo* metabolic labeling of mRNAs and RNA-Seq in chemostat cultures. We will identify factors that contribute to variation in mRNA degradation rates using computational analyses. In addition, we will test the functional role of mRNA stabilization and storage in P bodies in quiescent cells using imaging studies.
**NIH RO1 subcontract**

- **Alexander Statnikov (NYU Med School)**

- **Sub-aim 3:** Assess pathway discovery accuracy and experimental efficiency of both new and existing state-of-the-art methods in real biological data using an experimentally-derived gene regulatory network of *S. cerevisiae* and a unique database of gene knockout experiments covering a significant part of the yeast genome.

- Perform experimental validation of the predicted local causal pathway of TOR1, a conserved regulator of cell growth that is frequently dysregulated in human tumors.
2015

• Lab composition
• Grants
• Papers
• Meetings
Current Lab composition

Lab Manager
  Nathan Brandt (Joined December 2009)

Postdocs
  Niki Athanasiadou (joined May 2011)

Faculty Fellows
  Sam Diaz-Munoz (joined August 2014)

PhD Students
  Benjy Neymotin (joined April 2010)
  Naomi Ziv (joined April 2010)
  Jungeui Hong (joined December 2010)
  Darach Miller (joined April 2013)
  Steff Lauer (joined April 2014)

Masters Students
  None

Undergraduate Students
  Christina Nunez (January 2014)

High School Students
  Wei Wu (joined May 2013)
  Anise Ru (joined May 2013)

Other
  Jill Trivedi (joined June 2014)
Lab composition

Anticipated departures in 2015

Following PhD thesis defense
  Benjy Neymotin (joined April 2010)
  Naomi Ziv (joined April 2010; thesis defense March 10)
  Jungeui Hong (joined December 2010)

Graduating from undergrad
  Christina Nunez (January 2014)
  Jill Trivedi (joined June 2014)

Graduating from High School
  Wei Wu (joined May 2013)
  Anise Rau (joined May 2013)
Declined grants

• none
Grants submitted by lab members

- Steff -> NSF GRFP
- Sam -> K99/R00 (NIH)
  -> MCB (NSF)
2015 Grants

• mRNA decay RO1 – June 5\textsuperscript{th}
• NSF Center Grant
Papers for 2015
Airoldi et al

- Nitrogen regulated gene expression
- Reviewed
- Addition of new experiments by Darach
- Resubmission due by Feb 12, 2015.
Neymotin, Ettore and Gresham

• Determinants of mRNA degradation
• Draft complete
Hong and Gresham

- Customized sequencing adaptors for unique molecular identification
- In draft format
- Additional analysis underway
Hong and Gresham

• Transcription factor evolution
• In draft format
• Additional Experiments
  – Additional fitness assays
  – Functional assay of GAT1 mutants
    – ChIP-PCR, ChIP-seq, in vitro
Growth rate regulation of gene expression

In draft format

Additional Experiments

- Replicate of RATE-seq
- Resequencing of steady-state samples
- Single-cell RNA and protein content (using flow)?
- Batch culture RNA quantification?
Ziv, Siegal, Gresham

• Growth rate QTL
• In draft format
Athanasiadou, Rau, Wu, Gresham

- Mechanisms and Evolution of 5-FU resistance
- Written report
- Additional experiments
  - Mutation identification using NGS
Where we publish

• Open Access
• We should try:
  – Biorxiv (http://biorxiv.org/)
  – And pay attention to Haldane’s Sieve (http://haldanessieve.org/)
2015 Meetings

1. Microbial Population Biology (July 19-24, NH)
2. Molecular Mechanisms in Evolution (June 28-July 3, MA)
3. SMBE (July 12-16, Vienna)
4. RNA 2015 (May 26-31, Madison)
5. Berlin Summer Meeting (June 4-6, Berlin)
6. The CRISPR/Cas Revolution (Sept 24-27, CSHL)
Gresham Lab Conference Policy

If you want to go to a conference you must:
• Submit an abstract and request an oral presentation
• Identify all possible sources of support (the meeting, department, university, GSA, microphone runner)

The lab can support with prior approval:
• Travel (flight, train, bus)
• Registration
• Accommodation

The lab cannot support:
• meals that are not part of the conference registration
• taxi fares

Before registering for a conference talk to David about why you want to go, who will be there, what you plan to do, how it will be funded and what the expected cost for the lab will be.

https://wikis.nyu.edu/display/greshamlab/Lab+Information
Support for conferences

- Delill Nasser (GSA)
- Conference-specific
- GSAS
- Biology Department
Welcome to the Gresham Lab

We are located in The Center for Genomics and Systems Biology in the Department of Biology at New York University.

Our lab studies adaptive evolution, regulation of cell growth and mRNA degradation.

Genetic interactions and adaptive evolution

One of the central challenges in genetics is to understand how genes interact to result in phenotypic variation. We are studying how interactions between alleles at different loci affect fitness and the role of environmental variation in determining the outcome of genetic interactions. We study the role of genetic interactions in adaptive evolution using experimental evolution to identify the contribution of individual mutations, and their combinations, to fitness. We are determining whether loci interact in additive or non-additive ways and how these interactions affect fitness landscapes. We also study how genetic interactions differ in different environments using synthetic genetic array (SGA) technology to study double mutant phenotypes in different environments.

Regulation of cell growth and quiescence

In order to regulate their rate of growth cells must sense the environment and integrate complex signals. The rate at which cells grow can vary from minutes to hours depending...
Lab wiki

Gresham Lab
Created by Nathan Brandt, last modified on Sep 30, 2014

Click Here to Access the Gresham Lab NYU Homepage.
To access the labgear website for booking machines in the core Click Here.
To access the 2nd Floor Conference Room Calendar Click Here.
To access the 3rd Floor Conference Room Calendar Click Here.
To view a google doc of the status of known issues in the CGSB Click Here.

Home
Calender
Materials
Lab Data Entry
Lab Information
Protocols
Code

Like Be the first to like this
Data management

- Strain database
- Plasmid database
- Oligo database
- Microarray database

- Next-gen seq database?
Gresham Lab S. cerevisiae Strains

Date: 10/21/2014
Entered By: Derakh Miller
DGYS: 1320
ALIAS: SEC63-GFP

Genotype: MATa His3_1 Leu2_0 me15_0 ura3_0 SEC63::GFP::His3MX6

Strain: BY4741
Source: Invitrogen Yeast GFP Clone

Notes: His(+); qdp not verified yet

Plate Images:
Add Image

Date Modified: 10/30/2014
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Gresham Lab Plasmids

Plasmid Name: pCM188-GFP229
Plasmid Number: 231
Base Vector: pCM188
Gene Cloned: GFP229
Promoter: tetO2/pcYC
Size (kb): 8.8
Description: GFP227 (from DGP 143) cloned into pCM188 (DGP138)

Notes:

Plasmid Map:

Plasmid Stock Location:
Bacterial Stock Location:
Sequencing and Other Data:

Date Modified: 11/4/2014
Lab Books

• Record of all experiments
• One page per date
• Pen
• No removed pages
• Legible by everyone (including yourself)
• Property of the lab
Data sharing

• Let’s use Google Drive -> unlimited storage space
Lab meetings

• Weekly lab meeting
  – Lab business
  – Research update from one researcher
  – Paper discussion

• One on one meetings
  – Weekly
  – Bring lab book, print outs (preferably no computer)
  – Brief (<30 minutes)

All meetings start on time
Career development

• Personal career development statement
Other opportunities

- Travel to spend time in other labs
- Visit Max Delbruck Center in Germany
Beyond lab

• David
  – Teaching Human Genetics Spring 2015 (Darach TA’ing)
  – Teaching Applied Genomics Spring 2015

– Director of Bioinformatics
  • Hired fulltime bioinformatician who starts Feb 17.
Thanks