Core Course on Scientific Reasoning and Logic, Fall 2024

Module: Gene Expression and Genetics

Course Outline

Course Faculty

Instructor:	Christopher Vakoc
Invited Experts:	Alexander Gann
	Christopher Hammell
	Adrian Krainer
	Robert Martienssen
	Ullas Pedmale
Tutors:	Deeptiman Chatterjee (chatter@cshl.edu),
	Jason Lynn (lynn@cshl.edu),

Lectures:

August 29, 2024 (10:00-12:00), Delbruck: Pedmale, Vakoc, Hammell

- Primer and overview of genetics and its importance to research and CSHL.

August 29, 2024 (2:00-4:00), Koch, Samet: Hammell

- Intro to genetics (gene, allele, hypomorph, LOF, GOF…),

August 30, 2024 (10:00-12:00), Plimpton, Beckman: Hammell

- Forward Genetics and Epistasis

August 30, 2024 (2:00-4:00), Plimpton, Beckman: Vakoc

- Reverse Genetics-I

Sept. 9, 2024 (2:30-4:00), Plimpton, Beckman: Vakoc

- Reverse Genetics-II

Sept. 10, 2024 (2:00-4:00), James: Gann and Martienssen

- History of Gene Regulation

Sept. 11, 2024 (9:00-11:30), Plimpton, Beckman: Vakoc

- Transcription

Sept. 12, 2024 (2:00-4:00), Delbruck: Pedmale

- Chromatin, polycomb, histone mods, chromatin remodelers

Sept. 18, 2024 (9:00-11:00), Plimpton, Beckman: Vakoc and Hammell

- Paper discussion: TBD (*2 recent papers encompassing forward and reverse genetics*),

Sept. 23, 2024 (2:00-4:30), Plimpton, Beckman: Martienssen

- Transposons/epigenetics

Sept. 30, 2024 (2:00-4:00), Plimpton, Beckman: Wrap-up Session: (Hammell and Vakoc),

- Future of genetics

Oct. 22, 2024 (2:00-4:30), James: Krainer

- Splicing mechanisms and regulation

Student Evaluation:

- Discussions 50%, Class Participation 50%

Learning Objectives

- Genetic organization, mutants, genotype-phenotype relationships
- Forward and reverse genetics
- Genetic interactions and pleiotrophy
- CRISPR
- Transposons and epigenetics
- Cooperativity and specificity in gene regulation
- Splicing and processing of RNAs
- Histone and DNA Modification
- Chromatin

CSHL School of Biological Sciences
Core Course on Scientific Reasoning and Logic, Fall 2024
Course Outline

Module: Gene Regulatory Logic and the Construction of Multicellular Organisms: Insights from humans, flies, and worms

Course Faculty

Lead Instructor: Christopher Hammell (chammell@cshl.edu)
Tutor: Peipei Wu (pwu@cshl.edu; X5210)

Lectures:

Friday, September 20th, 2024, 9 am-11:30 am (Bush Fireplace Room)

- **Cell fate specification and the construction of a rudimentary organ**
- Overview of how intracellular and extracellular signaling defines an array of distinct cell fates using the C. elegans vulva as a model.
- Integrate these general principles of intra- and extra-cellular signaling in the context of disease.

Tuesday, September 24th, 2024, 9 am-11:30 am (James Library)

- **Control of temporal gene expression**
- Overview of *C. elegans* development and the utility of having a hard-wired developmental program versus the spatially-defined one discussed in the last lecture.
- *C. elegans* heterochronic pathway and the emergent themes common to all metazoans.
- Comparison of temporal gene expression strategies: developmental timers, circadian timers, and biological oscillators that construct repeated, spatial elements in development.

Friday, September 27th, 2024, 9 am-11:30 am (Plimpton)

- **Control of shape and size in development**
- Overview of body formation of animals.
- Insights of growth regulation from single cells and model organisms.
- Cell-autonomous and non-autonomous regulatory networks and how to find them genetically.

Tuesday, October 1st, 2024, 9 am -11:30 am (James Library)

- **Germline formation**
- Overview of forms of animal/plant growth.
- Examples from model organisms and the genetic analysis of the problem.
- Cell-autonomous and non-autonomous regulatory networks and how to find them genetically.

Wrap-up Session:

Monday, October 7th, 2024, 12:00pm-1:00pm (Urey)

Student Evaluation:

- 50% participation in daily discussions during lectures
- 50% based on paper discussions

Learning Objectives

- To understand the fundamentals of recurrent Gene Regulatory Networks (GRN) that orchestrate various types of cell fate specification.
- To understand what makes a sound “model system” for developmental biology.
- To define a stem cell and how it operates in embryogenesis, post-embryonic development, tissue regeneration, and the germ line.

- To understand the limitations of studying developmental biology from a genetic perspective and to determine the solutions to this problem.
- Integrate large-scale gene expression studies to understand the coordination of gene expression during development.
- Gain a practical understanding of how cell death, developmental timing, cell and organ growth control, germline development, and tissue regeneration contribute to normal developmental processes.

Learning Outcomes

- Elaborate on an understanding of a functional model system for a particular developmental problem.
- Design tractable methods to investigate developmental problems.
- Critically access modern literature focused on developmental biology.
- Gain a fundamental understanding of how high-volume genomic approaches contribute to our understanding of gene expression trajectories and progression of developmental processes.

Reference Material

Textbooks:

- Gilbert, S.F. 2003. *Developmental Biology*, 7th ed. Sinauer Associates, Inc.
- Wolpert, L., R. Beddington, J. Brockes, T. Jessell, P. Lawrence, and E. Meyerowitz. 2002. *Principles of Development*. 2nd ed. Oxford University Press.
- Stern, C. 2004. Gastrulation: From Cells to Embryo. CSHL Press.
- Wilt F. H., Hake, S.C. 2004. *Principles of Development*. W.W. Norton & Company, Inc.
- Alon, Uri. 2006. *An Introduction to Systems Biology: Design Principles of Biological Circuits.* Chapman and Hall. CRC Pr.

Reviews:

- Raj, A. and van Oudenaarden, A. 2009. Single-molecule approaches to stochastic gene expression. *Annu Rev Biophys.* **38**: 255–270.
- Roth, S. and Lynch, J. 2013. Does the bicoid gradient matter? *Cell* **149**: 511–512.
- Tumaneng, K., Russell, R. C. and Guan, K.-L. 2013. Organ size control by Hippo and TOR pathways. *Curr Biol* **22**: R368–79.
- Zhao, B., Tumaneng, K. and Guan, K.-L. 2011. The Hippo pathway in organ size control, tissue regeneration and stem cell self-renewal. *Nat. Cell Biol.* **13**: 877–883.

Problem Set Papers
(Due Wednesday, October 1st, 2024; noon):

- Raj, A., Rifkin, S. A., Andersen, E. and van Oudenaarden, A. 2010. Variability in gene expression underlies incomplete penetrance. *Nature* **463**: 913–918.
- Tursun, B., Patel, T., Kratsios, P., and Hobert, O. 2011. Direct conversion of C. elegans germ cells into specific neuron types. *Science* **331**: 304-308.

Discussion Papers

Tuesday, October 1st, 2024, 6 pm - 8 pm (Plimpton):

- Raj, A., Rifkin, S. A., Andersen, E. and van Oudenaarden, A. 2010. Variability in gene expression underlies incomplete penetrance. *Nature* **463**: 913–918.
- Tursun, B., Patel, T., Kratsios, P., and Hobert, O. 2011. Direct conversion of *C. elegans* germ cells into specific neuron types. *Science* **331**: 304-308.

Thursday, October 3rd, 2024, 6 pm - 8 pm (Plimpton):

- Eorglu, Zocher, McAuley, Webster, Xiao, Yu, Mok, Derry. 2024. Noncanonical inheritance of phenotypic information by protein amyloids, Nature Cell Biology, https://doi.org/10.1038/s41556-024-01494-9
- Böhni, R., Riesgo-Escovar, J., Oldham, S., Brogiolo, W., Stocker, H., Andruss, B.F., Beckingham, K., and Hafen, E. 1999. Autonomous control of cell and organ size by CHICO, a *Drosophila* homolog of vertebrate IRS1-4. *Cell* **97**: 865–875.

Core Course on Scientific Reasoning and Logic
Module: Macromolecular Structure and Function
Course Syllabus

Course Faculty
Organizer: Leemor Joshua-Tor
Module Tutor: Natalie Jones (njones@cshl.edu

Lecture 1, Fri., Nov. 1, 9-11:30am: Joshua-Tor
- Basic principles

Lecture 2, Tue., Nov. 5, 9-11:30am: Garg
- Pymol Tutorial

Lecture 3, Wed., Nov. 6, 9-11:30am: Joshua-Tor
- A structural perspective of RNA interference

Lecture 4, Fri., Nov. 8, 9-11:30am: Joshua-Tor
- X-ray crystallography

Lecture 5, Tues., Nov. 12, 9-11:30am: Joshua-Tor
- CryoEM and other methods in structural biology

Wrap-up Session:
- TBD

Student Evaluation:
Presentation and written portion of protein tales: 60%
Lecture participation: 20%
Problem Set: 20%

Learning Objectives
- Elements of macromolecular structure
- Hydrophobic vs. ionic interactions
- Protein-nucleic acid and protein-protein interactions
- RNA folding/recognition
- Crystallography in a nutshell – what you need to know in reading structure papers critically
- Single particle negative stain and cryoEM
- Principles of CD, SAXS, NMR spectroscopy

Learning Outcomes
- Understand the principles of RNA interference pathways
- Demonstrate understanding of protein and nucleic acid structure and their utility in understanding biology
- Have the ability to download structures, visualize and interrogate them.
- Demonstrate understanding of protein-nucleic acid and protein-protein interactions
- Design methods to distinguish between direct and indirect protein interactions
- Discuss strategies for obtaining macromolecular structure and learn to decide which approach to use and what information one can obtain from each method
- Learn how to read structural biology papers and critically assess them

Reference Material
Textbooks:
- Watson, J.D. *et al., Molecular Biology of the Gene,* 2013
- Liljas, et al., Textbook on Structural Biology
  Alberts, B. *et al., Molecular Biology of the Cell,* 2008, pp. 329-400 and 411-454.
- Rupp, *Biomolecular Crystallography*
- McPherson, *Introduction to Macromolecular Crystallography*

Reviews:

- Ipsaro, J.J. and Joshua-Tor, L. 2015. From guide to target: molecular insights into eukaryotic RNA-interference machinery. *Nat Struc Mol Biol*. **22**: 20-28.
- Ozata, DM et al and Zamore, PD. 2019. PIWI-interacting RNAs: small RNAs with big functions. *Nat Rev Genet*, **20**, 89-108.
- Gutbrod, MJ and Martienssen, RA, 2020. Conserved chromosomal functions of RNA interference. *Nat Rev Genet*, **21**, 311-331.
- Crowther, R.A. 2016. *Methods in Enzymology* **579**: 2-445.

Discussion Session 1 Nov. 18, 2pm-4:30pm: Protein Tales
Discussion Session 2 Nov. 19, 2pm-4:30pm: Protein Tales

Scientific Reasoning and Logic
Study Section Module Guidelines
Fall 2024

Tuesday	11/5/24	5:00 p.m.	Receive grant abstracts
Monday	11/11/24	5:00 p.m.	Submit grant rankings
Thursday	11/14/24	12:00 p.m. – 1:00 p.m.	Module Overview
Wednesday	11/27/24	12:00 p.m.	Written Critiques due
Monday	12/02/24	2:00 p.m. – 4:30 p.m.	Study Section I
Tuesday	12/03/24	9:00 a.m. – 11:30 a.m.	Study Section II
Tuesday	12/03/24	2:00 p.m. – 4:30 p.m.	Study Section III
Wednesday	12/04/24	9:00 a.m. – 11:30 a.m.	Study Section IV
Wednesday	12/04/24	2:00 p.m. – 4:30 p.m.	Study Section V (if needed)

In the Study Section Module, you will read and critique grants, much as an NIH Study Section reviewing applications would do (although we will have much more time per grant for presentation and discussion). We have pre-selected real grants for review. Every student is expected to read every grant and participate in the discussion of every grant. In addition to this, each student will be assigned as PRIMARY on one grant as SECONDARY on another grant and as a READER on a third grant. Read the abstracts and rank your top 3, send via email to Razan Alnahhas (alnahhas@cshl.edu) by **5:00pm** on **Monday November 11th**.

You will be assessed on your presentation; your written critique; pre-prepared questions on your secondary grant; and participation in all of the discussions.

PRIMARY reviewers will: (A) Prepare a 30 minute-presentation to serve as the basis of discussion of the grant. 15 minutes will be on the scientific Background and to introduce Specific Aims (Background presentation). 15 minutes will be devoted to summarize Preliminary Results and to evaluate the Experimental Design (Grant presentation) and 5 minutes will be devoted to the PI, environment, etc. (B) Each primary reviewer will also prepare a written critique of the grant, along the lines of the NIH Center for Scientific Review guide (http://www.csr.nih.gov/guidelines/R01.htm).

SECONDARY reviewers will read the grant in detail and in advance of the meeting. They should prepare specific commentary and questions and will have ~ 10 minutes to present them before the general discussion period.

READERS will read the grant in detail and in advance of the meeting. They should prepare specific questions for the general discussion period.

For most of the module, you will be on your own to do your literature search, develop your critiques, and produce your oral presentations and written reviews. For general questions, you should also feel free to contact the Study Section instructor (Linda Van Aelst, vanaelst@cshl.edu) But you shouldn't use the instructor to assess the specifics of the grant

themselves-- that's your job!

Written critiques should be short and succinct: no longer than three pages long (no font smaller than Arial 11 point or Times 12point, 0.75inch margins), to be e-mailed to Razan Alnahhas (alnahhas@cshl.edu) by **12:00pm** on **Wednesday November 27th**. The written critiques do not have to provide introductory information. They should begin by summarizing the overall goal of the proposed research in context with the importance of the questions to be addressed within the field of proposed study. The strengths and weaknesses of each approach should be outlined, with an aim-by-aim critique being generally the easiest to present and understand. Alternative and perhaps better ways to approach each question should be presented if they exist. In exemplary cases, it would be good to postulate why such approaches might not have been proposed. The review should finish with a brief discussion of the P.I. and their qualifications, the environment in which the research is to be performed and the appropriateness of the available facilities and budget/personnel. Any concerns about regulatory issues with vertebrate animals, human subjects, data sharing and human embryonic stem cell research should be noted. An overall score should then be suggested based on the standard NIH priority score rating scale (see below).

The oral presentations for the primary referee should proceed similarly except that they should include an introduction to the field and the work proposed. This should be sufficient to bring a non-expert up to speed with the topic of the grant. Secondary referees should follow the primary referee with comments and concerns on the science proposed and other areas (P.I. etc.) outlined above. The reader will not be responsible for a formal presentation but is expected to support the discussion.

(1) Scientific background. We recommend that you spend the first few days of this module searching for, and reading, review articles on your primary and secondary grants' research topics. One of the key issues to consider is whether a grant is asking important and novel questions. This can only be done by placing the grant in the context of the research going on around it. Journals specializing in reviews will be particularly useful: for example, the Annual Review suite (http://www.annualreviews.org/), or the Trends In ... suite (http://www.trends.com/), or the Current Opinion suite (http://www.current-opinion.com/) or the Nature Reviews series (http://www.nature.com/reviews/index.html), but reviews can also be found in regular journals. Happy literature search!

Covering this background will be particularly important in the oral presentations; we expect you might spend up to 12 minutes of the presentation introducing your audience to the scientific context of the work. You should describe the global background of the field, and identify and justify important unanswered questions that are relevant to the proposed studies. (In the written critiques, it is not necessary for you to lay out the background. But you could, if you thought it appropriate and based on what you know, criticize the grant in your written critiques for failing to consider the relevant background.)

(2) Questions asked. Are the goals of the grant important? Why? Will answering the questions

resolve important scientific issues? Or would they merely lead to small increments to our knowledge? Are there better questions that could be feasibly asked? Is sufficient preliminary data presented to support the proposed research plan? What is the quality of the preliminary data, particularly that which supports key or novel methods?

(3) Experiments planned. Are the proposed experimental methods the most appropriate to answer the questions posed? Are they a novel approach to solving the questions asked? Will the experiments lead to definitive, clearly interpretable conclusions? Or, even if successful, would the experimental results be likely to produce results of ambiguous interpretation? Does the PI foresee the potential problems and offer alternatives? Are there better experiments that could be carried out to address the goals of the grant?

(4) Is the timeline of proposal reasonable or is the PI claiming that she/he will do more than is humanly possible? Are the preliminary data convincing in terms of establishing the feasibility of the proposed studies and clarity of ensuing interpretations? Are the resources planned for these experiments (personnel, equipment, time, etc.) too few or too many?

Overall, remember that the goal is not simply to present the grant, but also to evaluate it critically.

Real referees of NIH grants do not confer before the study section meets. Their opinions are formed independently as are their critiques. We therefore expect that you will not discuss your critiques of each grant before the study section meets.

One thing that you must remember is that you are to judge the grants based upon the state of their respective fields on the day that the grant was submitted. Investigators should not be rewarded or penalized because some of their hypotheses might have been addressed either by them or by others subsequent to the grant's submission. Therefore, you must judge these in their historical context rather than in the present.

Scoring

Each reviewer assigned to an application will give a separate score for each of five review criteria: Significance, Investigator(s), Innovation, Approach, and Environment.
The scoring system utilizes a 9-point rating scale (1 = exceptional; 9 = poor). The final overall impact/priority score for each discussed application is determined by calculating the mean score from all the eligible members' impact/priority scores, and multiplying the average by 10; the final overall impact/priority score is reported on the summary statement. Thus, the final overall impact/priority scores range from 10 (high impact) through 90 (low impact). The following guidance has been given to reviewers to determine individual review criterion and overall impact/priority scores:

Impact	Score	Descriptor	Additional Guidance on Strengths/Weaknesses
High	1	Exceptional	Exceptionally strong with essentially no

			weaknesses
	2	Outstanding	Extremely strong with negligible weaknesses
	3	Excellent	Very strong with only some minor weaknesses
Medium	4	Very Good	Strong but with numerous minor weaknesses
	5	Good	Strong but with at least one moderate weakness
	6	Satisfactory	Some strengths but also some moderate weaknesses
Low	7	Fair	Some strengths but with at least one major weakness
	8	Marginal	A few strengths and a few major weaknesses
	9	Poor	Very few strengths and numerous major weaknesses
Minor Weakness: An easily addressable weakness that does not substantially lessen impact **Moderate Weakness:** A weakness that lessens impact **Major Weakness:** A weakness that severely limits impact			

Resources

There are a number of sources to assist you in this assignment.

The NIH has produced a video of a mock study section:
https://www.youtube.com/watch?v=Vx6qO8z9swQ&t=2067s

You can see sample grants and summary statements on the NIH NIAID website:
https://www.niaid.nih.gov/grants-contracts/sample-applications

You can read NIH Reviewer Guidelines:
https://grants.nih.gov/grants/peer/guidelines_general/reviewer_orientation.pdf

Some terms you may come across (from the Center for Scientific Review):

Percentile: represents the relative position or rank of each priority score (along a 100.0 percentile band) among the scores assigned by a particular study section.

Priority score: A numerical rating that reflects the scientific merit of the proposed research relative to the "state of the science."

Study section: panel of experts established according to scientific disciplines or current research areas for the primary purpose of evaluating the scientific and technical merit of grant applications. Also called scientific review groups (SRGs).

Summary statement: a combination of the reviewers' written comments and the SRA's summary of the members' discussion during the study section meeting. It includes the recommendations of the study section, a recommended budget, and administrative notes of special consideration.