

Cold Spring Harbor Laboratory Course on:
Expression, Purification & Analysis of Proteins & Protein Complexes
March 30- April 11, 2022

INSTRUCTORS:

Albert Courey, University of California, Los Angeles, Los Angeles, CA

Michael Marr, Brandeis University, Waltham, MA

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COURSE TEACHING ASSISTANTS:

Will Dahl, Brandeis University, Waltham, MA

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William Sander, University of California, Los Angeles, CA

Tom Yau, University of California, Los Angeles, CA

SUMMARY:

This course was for scientists, including graduate students, postdoctoral scholars, staff scientists, and principal investigators, who wanted a rigorous introduction to expression and purification of proteins as well as analysis of protein structure and function.

Through hands-on experience in the lab as well as extensive lecture and discussion, each student became familiar with key approaches in expression, purification, and analysis of soluble and membrane proteins and protein complexes from both natural sources and overexpression systems. The emphasis of the course was on the following:

- 1) Approaches in protein expression: Choosing the best bacterial or eukaryotic expression system tailored for the particular protein and experimental problem; determining how to optimize expression; understanding protein tagging: the advantages and pitfalls of various affinity and solubility tags.
- 2) Approaches in protein purification: Choosing the best strategy for a given protein including solubilization; bulk fractionation; liquid chromatography: including conventional methods (ion exchange, size exclusion, reverse phase, etc.) and affinity methods (e.g., MAC, DNA affinity, immunoaffinity, etc.), as well as FPLC/HPLC.
- 3) Approaches in protein analysis: introduction to common approaches for characterization of proteins including binding assays; activity assays; mass spectroscopy to identify protein interaction partners and post-translational modifications.

In addition to purification, students also gained exposure to fundamental analytical approaches such as mass spectroscopy and protein structure determination (e.g., X-ray crystallography, cryo-EM, etc.).

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PARTICIPANTS:

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Cassandra Taber, Graduate Student, University of Toronto, Laboratory Medicine and Pathobiology, Toronto, Canada. Lab Head: Dr. Michael Ohh

Bonnie Tillotson, Lab Head, C4 Therapeutics, Inc., Biochemistry, Biophysics and Crystallography, Watertown, MA. Lab Head: Dr. Joe Patel

12 Participants (6 Male, 6 Female, 1 URM)

* **NIH scholarship support**

SEMINARS:

Andrew Quigley, Diamond Light Source, Oxfordshire, United Kingdom
"Membrane Proteins"

Albert Courey, University of California, Los Angeles, Los Angeles, CA
"Regulation of protein function by SUMO"

Leemor Joshua-Tor, Cold Spring Harbor Laboratory
"A shape shifting nuclease unravels structure RNA"

Michael Marr, Brandeis University, Waltham, MA
"Introduction to Protein Purification"
"Gene regulation in response to stress"

Sergei Nechaev, University of North Dakota School of Medicine, Grand Forks, ND
"RNA Polymerase II Pausing"

Darryl Pappin, Cold Spring Harbor Laboratory, Cold Spring Harbor, NY
"Introduction to Mass Spectrometry of Proteins"
"Quantitative Approaches to Mass Spectrometry of Proteins"

Jan Witkowski, Cold Spring Harbor Laboratory, NY
"Ethics, Rigor & Reproducibility"