Sponsored Seminar: Eclipse Bioinnovations

08:00 - 09:00 Friday, 29th May, 2020
Talk

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971 Part 1: Introduction - The eCLIP platform: A powerful technology to study RNA binding protein interactions, microRNA targets and RNA modifications

Peter Chu
Eclipse Bioinnovations, San Diego, CA, USA

Abstract

Eclipse Bioinnovations RNA genomics technology can take your research to the next level. Easier and Faster.

Eclipse Bio has quickly become a leader in the new frontier of RNA genomics. Our eCLIP technology platform enables all scientists to understand post-transcriptional regulation by mapping targets for *RNA binding proteins (RBPs) or RNA modifications on mRNAs and non-coding RNAs such as miRNAs and lncRNAs.*

Why is this important? RNA processing regulates all aspects of human and model organism biology. This has been shown by the ever-increasing number of RNA regulatory events linked to key steps in development, and more importantly, the initiation and progression of both inherited and infectious diseases. RNA binding proteins (RBPs) control the fate and function of RNA by regulating the post-transcriptional machinery responsible for RNA splicing, polyadenylation, stability, localization, translation and degradation.

Eclipse products and services enable researchers in all research fields (including cancer, CNS, genetic disease and virology), to understand RNA regulation in their diseases, cells and model organisms by robust and precise mapping of RNA binding protein, m6A methylation and miRNA-mRNA targets.

In this seminar, we will have 3 main presentations: First, Dr. Sergei Manakou (*Eclipse Bio Principal Scientist Computation Biology)* will provide a scientific overview of Eclipse's RNA post-trancriptional regulation genomics and NGS methods. Second, our guest speaker Prof. Stephanie Ceman (*University of Illionois Urbana Champaign*) will present her Fragile X Syndrome research and how she used Eclipse technologies to advance her studies. Finally, we will showcase Eclipse's products and services and how scientists can take advantage of these products to simplify and advance their own research.

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972 Part 2: RNA regulation by RNA-binding proteins, m6A and ncRNAs

Sergei Manakov
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Abstract

RNA binding proteins play key roles in regulating RNA processing by interacting with RNA primary sequence and secondary structures to control splicing, RNA stability, translation initiation and elongation, and localization. With the discovery of more and more RNA binding proteins associated with disease or other biological processes, it is often essential to be able to directly map which RNAs are bound by the protein of interest. Crosslinking and immunoprecipitation (CLIP) methods enable this by immunoprecipitating the protein of interest along with crosslinked RNA, followed by library preparation and high-throughput sequencing to identify regions that show significantly enriched signal.

Recently, improved CLIP-seq methods have been described that dramatically improve these experiments by increasing the conversion of RNA into high-throughput sequencing libraries. In particular, eCLIP (developed in the Yeo laboratory at UCSD and described in Van Nostrand et al., Nature Methods 2016 and Van Nostrand et al., Nature 2020 in press) has been shown to be highly robust, with over 150 RNA binding proteins profiled to date. In this presentation, we will describe the eCLIP method and show how eCLIP can be used to profile the targets of an RNA binding protein of interest in an unbiased, transcriptome-wide manner. We will also discuss how the use of proper controls (input, IgG-only, and wild-type versus tagged protein) can provide significantly improved signal to noise in detecting true signal and removing abundant RNAs. Finally, we will show how the basic eCLIP method can be modified to more deeply probe individual aspects of RNA biology, by showing how adapted eCLIP methods have been used to directly profile miRNA targets (through chimeric ligation of miRNA with their mRNA targets) and RNA modifications (using an m6A-specific approach).

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970 Part 3: Fragile X Mental Retardation Protein FMRP regulates Argonaute 2 (AGO2) association with cobound mRNAs through binding MOV10

Stephanie Ceman
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Abstract

The Fragile X Mental Retardation Protein (FMRP) is an RNA binding protein that regulates translation and is required for normal cognition. FMRP both facilitates and blocks microRNA (miRNA)-mediated silencing in the 3'UTR of a subset of mRNAs through its interaction with the RNA helicase MOV10. This bi-functional role is modulated through RNA secondary structures known as G-Quadruplexes. We mapped the interacting domains of FMRP, MOV10 and Argonaute (AGO) and found that AGO associates with the N-terminus of FMRP and the N-terminal domain of MOV10 binds the KH1 domain of FMRP. The RGG box of FMRP has been shown to bind RNA G-Quadruplexes and the RGG box is necessary to block AGO association with cobound mRNAs. The N-terminus of MOV10 is required for this protection: its over-expression leads to increased levels of the endogenous proteins encoded by this co-bound subset of mRNAs and increased RGG box dependent binding to a model G-Quadruplex. Finally, FMRP has a global role in miRNA-mediated translational regulation by recruiting AGO2 to a large subset of RNAs in mouse brain.

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973 Part 4: Eclipse Bioinnovations products and services

Ines Rabano
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Abstract

In this presentation we will provide specific examples of how eCLIP can be adapted into individual projects through the use of Eclipse products. Eclipse's kits and services can help you identify RNA binding proteins, directly profile miRNA targets (through chimeric ligation of miRNA with their mRNA targets) and RNA modifications (using an m6A-specific approach).

For expert users, we will discuss details of how different sample types can be incorporated into eCLIP workflows, what reagents and optimizations need to be performed before beginning full eCLIP experiments, and what controls provide useful comparisons for analysis. We will further discuss options for non-expert users, including Eclipse-provided service options for both eCLIP experiments as well computational data analysis support.

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