A Neuronal Atlas of RNA-Binding Protein Expression at Single-Cell Resolution

John Laver¹, Eviatar Yemini², Oliver Hobert², Mihail Sarov³, and John Calarco¹

¹Department of Cell and Systems Biology, University of Toronto, Toronto, Canada; ²Department of Biological Sciences, Howard Hughes Medical Institute, Columbia University, New York; ³Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, Germany

Background

The nervous system consists of diverse neuronal subtypes

Distinct neuronal subtypes differ in:
- function
- morphology
- susceptibility to diseases

RNA-binding proteins are important regulators of neuronal gene expression

RNA-binding proteins (RBPs) bind specific sequence or structural elements in mRNAs to control their expression

C. elegans as a model for studying the nervous system

C. elegans has a uniquely well-defined nervous system:
- 302 neurons divided into 118 classes
- stereotypical neuron positions and morphologies
- completely mapped developmental cell lineage and neuronal connectome

Objectives

1. Determine the expression and localization of RNA-binding proteins in neurons with single-cell resolution
2. Understand the roles of RNA-binding proteins in specification and function of neurons and neuronal subtypes

Results

Half of the ~650 C. elegans RNA-binding proteins are enriched in neurons

Tissue-specific gene expression in C. elegans has been measured by:

1. Translating ribosome affinity purification coupled with RNA-seq (TRAP-seq) (Gracida et al., 2017, Cell Rep 21:3089-101)
2. Whole animal single cell transcriptional profiling (Cao et al., 2017, Science 357:661-7)

A preliminary microscopy-based survey of 40 neuronally-enriched RNA-binding proteins reveals diverse patterns of neuronal expression and subcellular localization

Expression of 40 RNA-binding proteins was assessed in transgenic animals

Examples of RNA-binding protein expression patterns in the head of C. elegans:

ptb-1/PTBP
mec-8/RBMS
larp-5/LARP4
asd-1/RBFOX

Future Directions

Annotation of neuronal RNA-binding protein expression patterns at single-cell resolution using NeuroPAL

Assessment of phenotypic consequences of neuron-specific RNA-binding protein depletion

Identification of neuron-specific mRNA targets of RNA-binding proteins

Acknowledgments

We thank Wesley Hung, Mei Zhen, Arneet Saltzman, and members of the Calarco and Saltzman labs for help and advice.

This work is supported by funding from the Canadian Institutes of Health Research, the Natural Sciences and Engineering Research Council of Canada, the Canada First Research Excellence Fund, and the University of Toronto.