Spotlight on Science

Transcriptional profiling of pain-transmitting neurons in the spinal dorsal horn

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What is the main focus of your research?

The general focus of my research is to understand the causes and mechanisms of chronic pain. To that end, I have been using RNA-seq to identify different cell populations in the spinal cord and to uncover the genes that govern the function of these cells under normal and pathological conditions.

What are the implications of accurate gene detection in neuronal development and specifically the spinal dorsal horn?

The spinal dorsal horn is where pain and other sensory signals make their first connection to the Central Nervous System. Accordingly, this is a prime location to intervene to try to thwart the pain from getting to the brain. However, one would like to only interfere with pain signals and not all sensation. It turns out that some neurons in the spinal dorsal horn mediate pain while others are used for non-pain sensations like touch or warm. If we can identify gene targets on those pain-transmitting neurons, this opens avenues for the specific inhibition of pain. Thus, accurate identification of genes in those pain-transmitting neurons is critical.

What are the advantages of low input RNA-seq for your study?

Rather than look at all the RNA of the spinal cord, as people had done previously, I wanted to investigate the genes expressed by a specific subpopulation of pain-transmitting neuron. So, instead of having copious RNA from a whole piece of tissue, I could only obtain RNA on the level of nanograms. To make matters even more challenging, it's difficult to isolate intact neurons from adult spinal cord, so I had to resort to using neuronal nuclei, which are easier to isolate but have ~1/10th the RNA content of a whole cell. So even thousands of nuclei got me only low nanogram quantities of RNA. So the tradeoff was specificity of signal vs. amount of material, and for my purposes, only looking at the transcriptome of a single neuronal population was key. Low input RNA-seq was the only way to achieve this goal.

How has using Ovation® SoLo RNA-Seq better enabled your research?

Nuclear RNA is less abundant than whole-cell RNA,and much of it is comprised of nascent transcripts that still retain intronic regions. Thus, I needed a library preparation kit that could take the lowest of inputs and that would faithfully and comprehensively represent the transcriptome of the nuclei I was using. I tried many kits, but only SoLo was able to give me what I needed. I was able to take pure nuclear RNA in the sub-nanogram range and get ready-to-sequence libraries that gave excellent data.

Why is a whole transcriptome approach important for your studies?

My work aimed to discover novel, unkown genes that govern the identity and function of pain-transmitting spinal neurons. Many genes have already been discovered, but I didn't want to rehash old work. I was

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looking for new targets. A whole transcriptome approach provided an unbiased and comprehensive view of all the genes expressed in my population of interest. In this way, we uncovered hundreds of enriched genes that I would have never thought to look for. That's the power of the WTA approach.

What are the broad implications of your work?

Our study using the Ovation SoLo RNA-seq kit identified several novel genes in pain-transmitting neurons in the spinal cord, many of which have features that are desirable for therapeutic candidates. In future work, I hope to test the contribution of these genes in preclinical pain models, with the ultimate goal of developing better and safer analgesic drugs than the current offerings.

Learn more about this work in this recent publication:

Transcriptional Profiling of Somatostatin Interneurons in the Spinal Dorsal Horn
Alexander Chamessian, Michael Young, Yawar Qadri, Temugin Berta, Ru-Rong Ji, and Thomas Van De Ven

https://www.biorxiv.org/content/biorxiv/early/2017/11/08/215657.full.pdf

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