Evolution Of Newborn Screening: Molecular Inversion Probes Targeting Recommended Uniform Screening Panel Genes

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NC Nexus — North Carolina Newborn Exome Sequencing for Universal Screening

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Introduction

Traditional newborn screening (NBS) enables early detection and pre-symptomatic intervention for conditions deemed "medically actionable" that can be detected using biochemical testing. Advancements in next-generation sequencing (NGS) provide opportunities to obtain DNA sequence data and add a vast number of disorders to current NBS.

However, the application of NGS to NBS raises an array of challenges, including ethical, legal, and social issues (ELSI) around screening newborns that the North Carolina Newborn Exome Sequencing for Universal Screening (NC NEXUS) research project at UNC is addressing. Other challenges include the efficacy of sequencing for detection of different conditions, as well as the time and cost of sequencing.

The vast amount of data generated by genome-scale sequencing introduces substantial background noise and complicates the need for informed decision-making. However, a parsimonious alternative would be to increase the usefulness of sequencing as a screening tool by focusing on a selected subset of conditions using a high-throughput assay capable of targeting specific genes. A limited approach could avoid some of the ELSI issues raised by applying NGS to NBS and afford a more cost effective method of screening genes associated with medically actionable disorders.

Acknowledgments & Sources

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[2]Evan A. Boyle, Brian J. O'Roak, Beth K. Martin, Akash Kumar, Jay Shendure (2014). MIPgen: optimized modeling and design of molecular inversion probes for targeted resequencing. Bioinformatics 30 (18): 2670-2672. doi:10.1093/bioinformatics/btu353

[3]HiSeq X Ten System. Illumina, Inc. Illumina, 2016. Web. 30 March 2016.

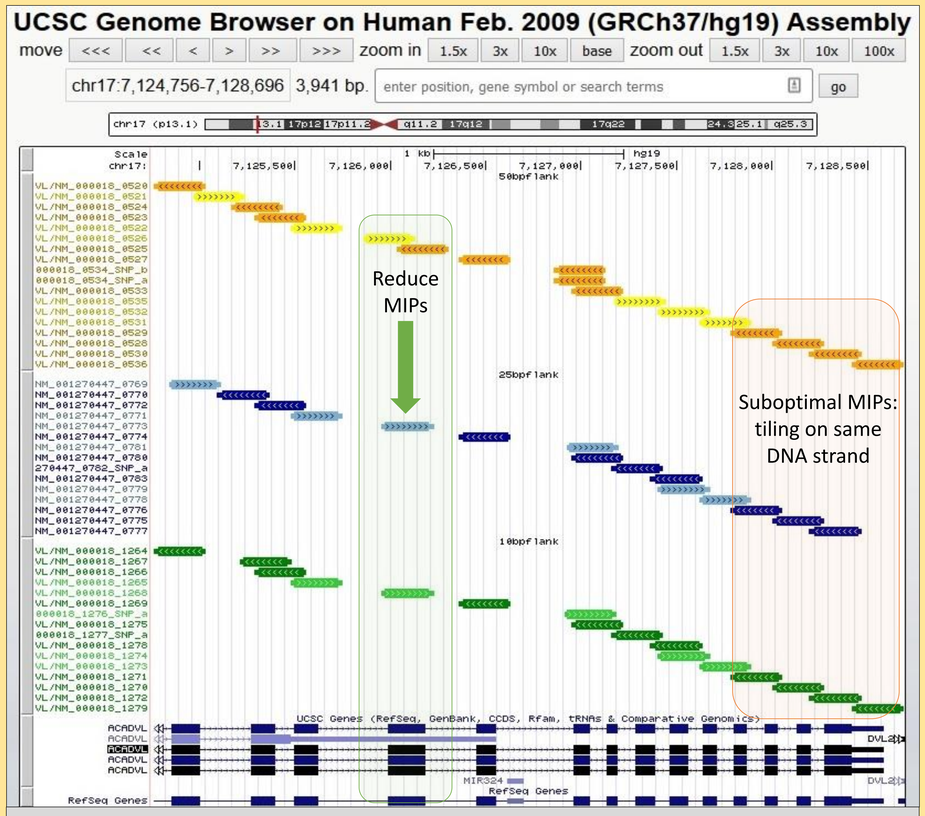

Figure 1. MIP designs with 50bp (yellow/orange), 25bp (blue) and 10bp (green) flanks for the gene *ACADVL* compared to RefSeq Genes on the UCSC Genome Browser. Visualization on the browser aids in identification of tiling probes and confirmation that desired exons are covered.

Methods

We are developing a low marginal cost molecular inversion probe (MIP) panel that targets the primary genes in the recommended uniform screening panel (RUSP), which is the current newborn screening standard in the U.S. Using the MIPgen[2] package, molecular inversion probes were designed to target several genes associated with current RUSP conditions. The MIPgen[2] was run three times with different size target regions – 50bp, 25bp, 10bp flanks into intronic regions. From these sets of MIP designs, we chose the fewest number of probes that should appropriately capture most all RUSP gene targets, and which preferably tile on opposite strands of DNA. Modifications are being made to the MIPgen[2] script.

The MIP approach will be validated in 675 samples previously analyzed by whole exome sequencing. This RUSP MIP panel will then be prospectively utilized in 400 participants enrolled in the NC NEXUS project as well as an additional 2000 patients from the Inborn Errors of Metabolism Collaborative with known newborn screen results.

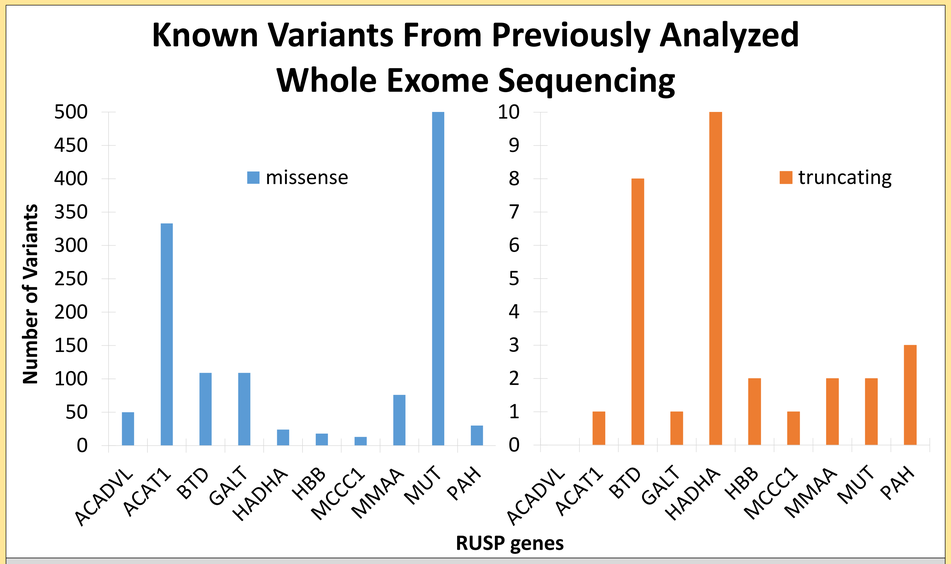

Figure 2. Variants in genes associated with the RUSP conditions detected using whole exome sequencing will be compared to the variants detected using MIPs on the same sample. This will serve as a control to validate performance of MIPs.

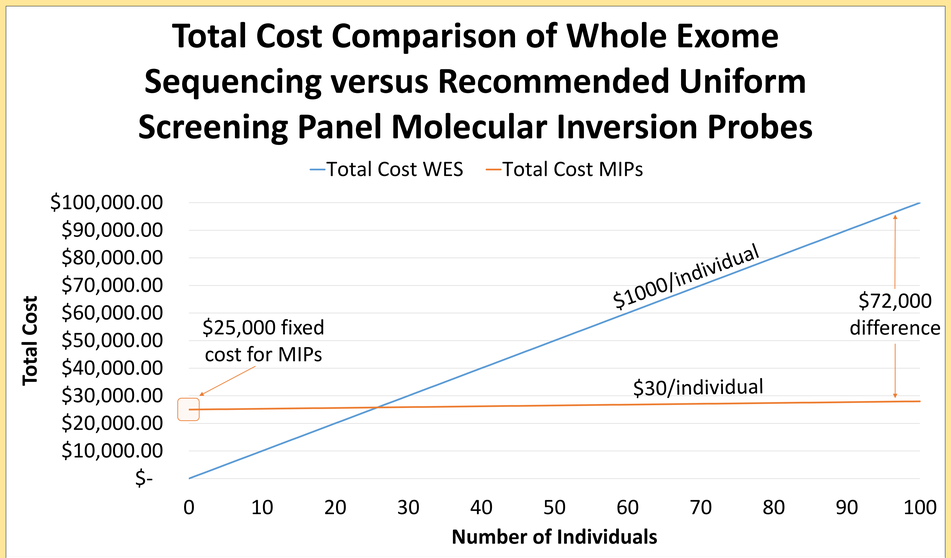

Figure 3. The initial cost of generating the RUSP MIP panel is $25,000 and running the panel is $30 per individual. The cost of whole exome sequencing is $1,000 per individual genome[3].

Future Implications

We anticipate that, if successful, this approach could be translated as an economical secondary screen in clinical care and serve as a proof of concept for adding other medically actionable conditions to the current recommended list for newborn screening.