Spring 5-12-2016

# Biotherapeutic development in the 'Omics Age: The CHO genome and beyond

Chapman Wright
*Biogen*, chapman.wright@biogen.com

Joost Groot
*Biogen*

Scott Estes
*Biogen*

# BIOTHERAPEUTIC DEVELOPMENT IN THE 'OMICS AGE: The CHO Genome and Beyond

Chapman Wright, Cell Culture Development, Biogen, Cambridge, MA
chapman.wright@biogen.com
Joost Groot, Computational Biology & Genomics, Biogen, Cambridge, MA
Scott Estes, Cell Culture Development, Biogen, Cambridge, MA

Key Words: CHO genome, transcriptomics, sequence variant detection, contaminant detection

The Next-Generation Sequencing (NGS) revolution has had a profound effect on the manner in which we approach the fields of biology and medicine, but the effects are not limited to these fields alone. With the process of collecting and analyzing large datasets becoming more seamless, a growing number of disciplines are incorporating NGS and other 'Omics technologies into their workflow. Biotherapeutic development is one such field that is benefitting from the comprehensive and rich datasets that NGS can provide. At Biogen, we are using NGS in two ways: to provide a deeper understanding CHO cell biology during bioprocessing and to improve our cell line development workflows. A straightforward use of NGS technology is to verify that the biotherapeutic mRNA sequence is free of mutations during clone screening. We developed a method that takes advantage the multiplexing and high-throughput capability of NGS to sequence confirm the mRNA from multiple antibody producing clones in one experiment. Results from these NGS experiments are combined with other product quality and productivity data points to inform clone decisions. A single mutated clone has been identified and subsequently discarded in three separate programs to date. As an additional QC step to minimize the possibility of sequence variants, we have also implemented NGS plasmid confirmation prior to transfection. These two NGS methods are now an integral part of our cell line development workflow and have added quality to the clones moving forwarded for more rigorous screening.

Biogen has also made generating, characterizing and annotating the genomic sequence of our CHO host cell lines a priority. To this end, we have partnered with a number of vendor and academic labs to move this initiative forward and arrive at what we believe are industry-leading CHO genomes. Our collaborators utilized a combination of the shorter, more accurate Illumina reads with SMRT technology, commonly called PacBio reads. This work has filled in gaps within our CHO genomes and reduced the number of contigs and scaffolds reported for public genome resources. While the updating and curation of our CHO genomes will continue into the future, the state of these genomes allowed us to explore other 'Omics techniques we deemed important for characterization of our production clones. Transcriptomics has become a central characterization technique of production clones, giving us a deeper understanding of the CHO cell during production. By comparing RNAseq datasets at various time points during our bioprocess, we have identified changes in cellular pathways as they relate to a particular growth stage. As such, we are aiming to exploit these cellular changes and use them to our advantage in expressing our biotherapeutics. The development of targeted integration strategies has also been a primary goal for us in the 'Omics space. The ability to specifically target GOIs to multiple safe harbor areas within the CHO genome can offer flexibility for screening expression modifiers as well as accelerate the cell line development process. To identify the integration sites in our production clones, we are using a 'pull-down' method to enrich for genome sequences that are abutting the expression vector. Results of these experiments will be discussed.

Taken together, Biogen has made 'Omics a priority to gain a deeper understand of our CHO cell biology at the cell line development stage and during the production bioreactor cultures. We are now taking meaningful steps forward, and believe that 'Omics will continue to shape how we engineering, select and culture CHO cells expressing our biotherapeutics.