

PRODUCER CELL LINE ENGINEERING FOR LARGE VOLUME MANUFACTURING OF THERAPEUTIC AAV

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Key Words: AAV production, HeLa, PCL

Recombinant adeno-associated viruses (rAAV) remain one of the most promising gene therapy vectors for patients with genetic abnormalities. rAAV can safely deliver long-lasting expression of a therapeutic transgene. To create a rAAV virus the wild-type viral genome is replaced with a functional transgene expression cassette. A constitutive promoter can be utilized to drive strong expression of the transgene once the rAAV vector has infected the target cell. Unlike wtAAV, the recombinant vector avoids the pitfalls of genomic integration by establishing extrachromosomal episomes or concatemers. Multiple studies have shown that rAAV can provide sustained expression of the transgene in cultured cells and pre-clinical models, providing evidence that rAAV could offer a cure for certain diseases. Bioengineering advancements have expanded the viral tropism beyond the constraints of naturally occurring AAV capsids, increasing the types of cells that can be thought of as targets. Taken together, rAAV therapies have attractive qualities to safely address the needs of patients where protein or small molecule therapies would fall short.

One challenge with therapeutic rAAV is the ability to generate enough virus for clinical trials and commercial supply. This is particularly true with neuromuscular or hemophilia patients in which doses can exceed 1×10^{14} viral genomes per patient. Typical yields from a rAAV production are around 10^4 viral genomes per cell, meaning 10^{10} cells will be needed for a single dose. This amount of therapeutic virus is a challenge to deliver with standard gene therapy production platforms, and will require a production platform that can reliably generate sufficient quantities of therapeutic rAAV to meet patient demand. Biogen has selected the producer cell line (PCL) platform to meet the large demand for therapeutic rAAV. Producer cell lines are generated by stably integrating the AAV viral genes along with the ITR flanked therapeutic gene of interest into a host cell line. rAAV production is then triggered by the addition of a 'helper virus' to provide functional genes for AAV replication. Traditional PCL platforms have used HeLa cells as the host and Adenovirus type-5 to deliver helper functions. We have used this platform as a basis for further development. Presented here will be our rationale for selecting the PCL platform, improvements made to the platform for ongoing clinical support, and our vision for the next generation PCL platform.