

Massively parallel sequencing (MP-Seq) a new tool for adventitious agent detection and virus discovery

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Recent contaminations of manufacturing processes by porcine circovirus and vesiviruses have highlighted the need for broadly based and rapid methods to detect adventitious agents in cell banks, virus seeds and bulk product (drug substance). Massively parallel sequencing is a powerful new method for the identification of viruses and other adventitious agents, without prior knowledge of the nature of the agent. BioReliance have developed MP-Seq methods to detect free viruses in raw materials and fermenter samples. Our application of this technology has resulted in the discovery of a new parvovirus in bovine serum capable of infecting human cells and we have used this technology in the investigation of fermenter contaminations.

In some cells, the genomes of latent or transforming viruses may be present in a cell but no virus particles are produced. However, latency associated or transforming gene mRNAs are expressed. We have developed a method to identify these latent viruses by sequencing the total transcriptome of the cell and applying an algorithm to identify the viral specific transcripts. Enormous amounts of data (~400Mb) are generated in this process and a robust algorithmic process is required to analyse the data. Using this method we have been able to identify a new retrovirus expressed in Vero cells and identify nodavirus and errantivirus contamination of insect cells. The definitive nature of the methodology provides considerable reassurance that cell banks are free of unexpected contaminating agents.

We are also developing MP-Seq to provide rapid end points in *in vitro* virus detection assays. We have shown that we can detect virus infection as early as day 4 post infection with MP-Seq while conventional methods may require 14 or 28 days to reveal infection