Testing of raw materials is an essential step in the production cycle of biological therapeutics and vaccines. The implementation of Quality by Design (QbD) in manufacturing processes is required across the pharmaceutical industry to ensure the consistent production of a product to the required level of quality. The holistic mapping of extraneous agents in raw materials is an essential step in the QbD process, and requires molecular techniques capable of detection of known and novel contaminants.

Results from the evaluation of testing technologies are presented, highlighting appropriate technologies for evaluation of raw materials, and the drawbacks of comparable technologies, specifically for the identification of extraneous viruses.

Case studies regarding the discovery of several viruses in animal sera and cell lines will be presented, and will be considered in the context of current regulatory recommendation and guidelines for testing. Strategies for routine testing to mitigate risk of extraneous agents in raw materials will also be presented.

Quality by Design in Raw Materials Testing: Considerations, Strategies and Experience of a Testing Laboratory

Reginald Clayton1, Donna McMurtrie2, Anne Ilichmann1, David Onions1, Audrey Chang3, Colette Cote1, John Kolman1, Alison Armstrong4

1BioReliance, West of Scotland Science Park, Todd Campus, Glasgow G20 OXA, 2BioReliance, 14920 Brosschat Road, Rockville MD 20850, USA

Abstract

Testing of raw materials is an essential step in the production cycle of biological therapeutics and vaccines. The implementation of Quality by Design (QbD) in manufacturing processes is required across the pharmaceutical industry to ensure the consistent production of a product to the required level of quality. The holistic mapping of extraneous agents in raw materials is an essential step in the QbD process, and requires molecular techniques capable of detection of known and novel contaminants.

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Introduction

In evaluating the viral risk of biological products several factors have to be considered. These include the risks associated with individual viruses, the probability of the viruses being present in the material and the procedures used to inactivate or clear contaminating viruses.

New methods of virus detection based on massively parallel sequencing (MP-Seq™) in molecular based technologies i.e., degenerate PCR have initiated a new era of virus discovery. Recently, the number of human polyomaviruses has increased from 2 to 5. Scientists at BioReliance have identified a new bovine parvovirus in bovine serum using MP-Seq™ (Onions and Kolman 2010) and new porcine viruses in the Bovacrus and Hokovirus genera have been reported (Lau et al. 2008, Cheung et al. 2010). Testing regimes have often lagged behind this recent phase of virus discovery. (see adjacent tables). For instance for assays for porcine parvovirus 1 are always required, but until recently the other porcine parvoviruses were rarely considered. This is now changing with tests for porcine hokovirus being requested by regulatory authorities evaluating the safety of porcine parvoviruses, and assays for anellovirus being required for veterinary vaccines.

Global regulatory requirements

Table 1. Regulatory update: bovine and porcine contaminants

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New enabling technologies: Massively parallel sequencing: MP-Seq™

- Screening of raw materials using molecular technologies has highlighted the detection of novel viruses (see case studies).
- MP-Seq™ is performed using the Roche/454 GS FLX “Next Generation Sequencing” approach to generate high throughput sequencing data.
- The system can generate as many as 1,000,000 sequences at 35–50 bp read length. The length of “coverage” of the sequencing run, which is the average number of times that a base is sequenced, is often several thousands (see case studies for MP-Seq™).
- If a positive MP-Seq product is analyzed, the depth can be large, possibly 1000-10,000 times.
- MP-Seq™ detects sequences that are not detectable from a sample of extracts of viruses or agents detectable is not limited by degenerate primer or PCR, resulting in an holistic analysis of the raw material.

- MP-Seq™ enables us to ask, and answer the question: what agents and sequences are present in these raw materials?

MP-Seq™ enables detection of novel viruses

- RNA viruses
- DNA viruses

Conclusions

- There are numerous routes through which adventitious viruses may be introduced into the Biologic manufacturing process. Most are associated with the use of animal-derived raw materials.
- Materials such as serum, hormones, insulin, plasma proteins, attachment factors, tissue extracts and established cell lines, plant explants and raw materials such as cold liver and oil protein extracts can all potentially harbor viral contaminants.
- Risk can be minimized by testing raw materials by MP-Seq™ to enable identification of the contaminants present in the batch of raw materials.
- Once identified, the use of screening processes using PCR-directed methods or Molecular based methods for detection of specific contaminants are necessary.
- Massively parallel sequencing enables the holistic and detailed analysis of raw materials, the detection of viruses specific to raw materials, and the implementation of screening programmes for the assessment of raw material safety.

References