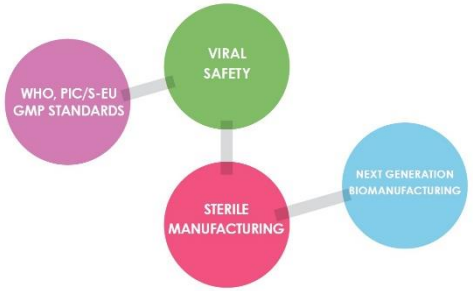

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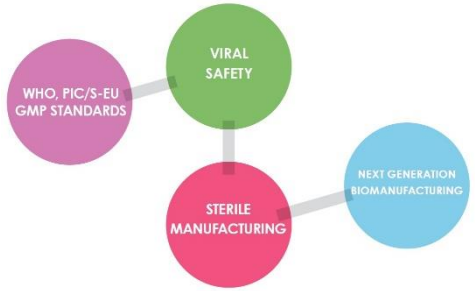
GaBI Educational Workshops

18 December 2019, Hotel Gran Mahakam, Jakarta, Indonesia

2nd ASEAN Educational Workshop on
GMP FOR BIOLOGICALS/BIOSIMILARS



Viral safety of biotechnology-based products: 3 case studies

Elwyn Griffiths, DSc, PhD

18 December 2019

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Viral safety of biotechnology – based products: 3 case studies

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Jakarta, Indonesia, 2019

Outline

- Briefly remind ourselves of the critical manufacturing issues relating to the viral safety of biologicals made using mammalian cells
- Look at three case studies that illustrate importance of the issue
- Outcomes and lessons learned

Viral safety of biological products – critical issue

- Many biologicals produced in mammalian cells
- Measures put in place to ensure absence of adventitious infectious virus agents in a product – **SAFETY issue**
- A contaminating virus MIGHT be devastating to a recipient (patient), might spread in the community.
- Contamination of cell lines, production process intermediates and products also has considerable **ECONOMIC CONSEQUENCES** for manufacturer even if the contaminating virus does not get through to the product
- **Might lead to product supply issues with significant public health impact**

Examples of biologicals produced in mammalian cell lines

- **Live virus vaccines**

Polio (primary monkey kidney cells, diploid cells, Vero cells), MMR (diploid cells, MRC5), Rotavirus (continuous cells, Vero cells)

- **rDNA protein products including biosimilars**

Growth hormone, Factor VIII, t-PA, monoclonal antibodies, cytokines etc. (continuous cells CHO, PER.C6, MDCK)

Viral safety of biological products – source of contamination?

- Cell substrate itself
- Viral seed (for a vaccine)
- Biological materials used in production (other than the cell substrate or viral seed)
- During production processes
- Possible viral contamination **of live viral vaccines** and **rDNA products** produced in **any mammalian cell considered** a **major issue** to be addressed

Detailed Guidelines available
A belts and braces approach

- *WHO Recommendations for the evaluation of animal cell cultures as substrates for the manufacture of biological products and for the characterization of cell banks (2010)* : ICH, national guidance
- Production based on cryopreserved cell bank. Master cell bank, working cell bank exhaustively screened for virus contamination, with documented history.
- **Control of raw material used in production – e.g. growth media, enzymes, virus seed (for vaccine)**
- Closed systems for growth of cell culture
- **Testing of each cell culture lot for viruses**
- **Validation of viral removal / inactivation by downstream processing as appropriate** (possible for rDNA protein products – not live viral vaccines)

Does the system work?

- Generally yes: Testing evolved and updated with time – now includes range of traditional and molecular methods (PCR, PERT assay for reverse transcriptase depending on circumstances)
- However, viral contamination has occasionally occurred but usually prevented from getting into product on the market
- **Seems that not all contaminations are reported publically**; manufacturers concerned about bad publicity in media (see Nature 472, (2011) 389-390)
- Some manufacturers have reported contaminations – MVM, Genentech 1993, 1994: Vesivirus 2117, Boehringer-Ingelheim 2003: Vesivirus 2117, Genzyme (Belgium and USA), 2008: PCV 1 & 2, GSK and Merck, 2010

3 case studies illustrating very different outcomes

- GSK and Merck experience (contaminated Rotavirus vaccine)
- Genentech experience (contaminated product was not released)
- Genzyme experience (contaminated product was not released)

Rotavirus vaccine contamination 2010

- Academic researchers using novel highly sensitive sequencing technologies detected porcine circovirus type 1 DNA (PCV 1) in GSK *Rotarix* (a live attenuated oral rotavirus vaccine), 2010
- Led to intensive investigation by GSK, FDA, EMA, Health Canada, WHO
- Led also, to investigation of Merck's *RotaTeq* rotavirus vaccine by Merck and the other agencies.
- Found *RotaTeq* gave signals for PCV 1 and PCV 2 using range of advanced technologies
- **KEY QUESTION did detection of DNA sequences for these viruses mean there was an adventitious virus present which might impact vaccine safety?**

GSK Rotarix vaccine contamination

- Extensive follow on work showed that GSK Rotarix vaccine (final containers), production intermediates, Working and Master cell banks and the viral seed were all contaminated by PCV1 DNA.
- **PCV 1 DNA found to be infectious at all stages,** including final containers (particle associated)
- GSK expanded investigation and tested all of their live viral vaccines. All were negative for PCV 1 except for Rotarix. In testing IPV made in similar Vero cell line found the harvest tested positive but purified bulks and final containers were negative.
- Seems source of PCV was contaminated **(non irradiated)** trypsin used in mid 1990s to manufacture Vero Cells.

Merck RotaTeq vaccine contamination

- Low levels of both PCV 1 and PCV 2 DNA fragments found in Merck RotaTeq vaccine final containers using highly sensitive assays but below detection level in Vero Cell bank and viral seed.
- **No infectious viruses found**
- Investigations showed porcine derived trypsin used in production to be the likely source of the PCV DNA fragments.
- Residual fragments of PCV DNA are now thought to come from porcine viral contaminants present in porcine pancreas used in manufacture of the trypsin: effectively destroyed by methods used to inactive adventitious agents, leaving only the remains behind.

Outcomes and lessons learned

- **Detection of signal for an adventitious agent in a marketed vaccine triggered extensive actions and risk-benefit discussions** between manufacturers, major regulatory agencies (FDA, EMA, Health Canada) and WHO because rotavirus vaccine is of global importance
- PCV1 and PCV 2 infect many mammalian cells but cause no major cytopathic effects. Not easily detected
- PCV 1 is not pathogenic for humans
- PCV 2 causes disease in post-weaning pigs but existing evidence suggests not pathogenic to humans

Outcomes and lessons learned

- **REGULATORY** decision was **NOT** to remove either vaccine from the market (although Rotarix was temporarily suspended in the USA during the investigations)
- No evidence of pathogenic role of PCV in humans.
- Post market surveillance of rotavirus vaccines had raised no issue.
- Conclusion of FDA/EMA/Health Canada, WHO) based on a risk-benefit assessment:- benefits of rotavirus vaccine substantial in preventing death and hospitalization in children and outweighed the risks which were theoretical

Outcomes and lessons learned

- **MANUFACTURERS** have focussed on improving the quality of trypsin used in the production of cell based live viral vaccines
- Irradiated trypsin being used to prepare new master cell banks and in production of rotavirus vaccines
- **New EMA Guideline on the quality of porcine trypsin used in the production of human biologicals was developed**

Outcomes and lessons learned overall

- Recognised that discovery of a previously undetected contaminant in a licensed biological using advance technologies is now a possibility
- Emergence of new technologies will inevitably result in new challenges in safety assessment of vaccine
- However, a thorough scientific investigation of the signal by manufacturers, independent laboratories and regulators is essential in supporting a scientifically based regulatory decision on what to do about the situation

Outcomes and lessons learned overall

- Also it was recognised that the discovery of a signal for a potential adventitious agent in a vaccine already on the market raised concerns that were not well addressed in available guidelines.
- **WHO has developed Guidelines on carrying out national regulatory risk evaluation strategies**

Genentech experience with contamination in cell culture 1993

- Contamination of large scale cell culture by Minute Virus of Mouse (MVM) detected during routine production control process
- Traditional testing takes time and product already well on way through downstream purification processes by time detected
- Production of the contaminated lot promptly stopped, reported to US FDA and clean up started
- Investigation of source instigated

Genentech experience with contamination in cell culture 1993

- No definitive source of contamination identified; consistent with media used in production as source. Feral mice from land surrounding plant examined but no MVM found
- Clean up process expensive
- **At no time was a contaminated product let through the system and the regulator was aware of all developments**
- New PCR and infectivity assay developed to speed up early testing and introduced routinely

Genentech experience with contamination in cell culture 1994

- New PCR and infectivity assays used and nothing found for 12 months
- Then another MVM positive signal but this time contamination detected before any downstream processing started. **Downstream protected**
- Source again highly likely to be contaminated cell culture media but not shown directly
- New heat treatment of medium developed, approved by FDA and installed
- **No viral contamination detected since 1994**

The Genzyme Experience

- Several bioreactor runs (Belgium and USA, 2008–2009) terminated early due to poor growth of cells – **suspected contamination**. Seem not to have dealt with problem promptly. Eventually informed FDA.
- US FDA warning letter and re-inspection
- Virus identified as Vesivirus 2117 using PCR in 2009: not known to be a human health risk but interferes with growth of CHO cells.
- Likely introduced by contaminated media
- USA plant shut down for major clean up and re-organization. Virus had spread into manufacturing facility – bioreactors and expensive chromatography columns. Clean up very costly.

The Genzyme Experience

- Global supply of two rDNA derived orphan drugs, Cerezyme (Gaucher's disease) and Fabrazyme (Fabry's disease), were seriously compromised and the products rationed. No alternative to Fabryzyme.
- Cause of concern to regulators (e.g. Health Canada) as to how to handle the situation.
- Overall Genzyme needed lot of GMP actions, stock prices dived and together with sales shortfall left the company vulnerable to takeover – acquired by Sanofi in 2011

Outcomes and lesson learned

- **Virus contamination is a serious business.**
- Manufacturers need to deal **promptly** with contamination or suspected contamination (compare Genentech & Genzyme)
- As new inexperienced manufacturers come into operation it is essential that they understand the need for great care and attention regarding development and production of biological products.
- Role of NRA in overseeing these developments is critical and GMP inspection is a key aspect of this oversight
- **Continued vigilance essential. Don’t be complacent.**

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THANK YOU FOR YOUR ATTENTION