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WHITE PAPER

Assessing Animal-Derived Raw Materials for Viral Risk

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TABLE OF CONTENTS

01

Why are Viral Risk Assessments Performed for Animal-Derived Raw Materials?

In the realm of biologic manufacturing, the risk of introducing an infectious virus into the drug substance bulk harvest is one of the fundamental concerns articulated within the International Council for the Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) Q5A(R2) Viral Safety Evaluation of Biotechnology Products Derived From Cell Lines of Human or Animal Origin.[1] Why the concern? Have there been known viral contamination events leading to lot rejection during biologic manufacture? The answer is yes, though rarely.[2] These events often have been attributed to use of animal-derived raw materials (ADM), such as fetal bovine serum or porcine trypsin. Have such viral contaminants ever made it into commercial drug products administered to humans? Yes, two instances come to mind. The first was the contamination of inactivated polio vaccine with infectious simian virus 40 (SV40) between 1953 and 1963 as a result of use of an infected production cell[3] . The second instance was the more recent finding that a liveattenuated rotavirus vaccine was contaminated with porcine circovirus attributed to the porcine trypsin used during culture of the production cell[4]. Hence the concern over viral contamination of a biologic. To help prevent such viral contaminants of a biologic from making their way into drug products and potentially harming recipients of the therapeutics, there is a so-called safety testing triangle5 that has been articulated within regulatory guidance documents from the late 1980s to the present, including ICH Q5A (R2) (see Box 1).

Box 1. The Biologics Safety Testing Triangle [from ICH Q5A (R2)]

"Three principal, complementary approaches are applied to control the potential viral contamination of biotechnology products:complementary approaches are applied to control the potential viral contamination of biotechnology products:

- **Selecting and testing cell lines and other raw materials, including media components, for the absence of undesirable infectious viruses**
- **Assessing the capacity of the production processes to clear adventitious and endogenous viruses**
- **Testing the product at appropriate steps of production for demonstrating the absence of contaminating infectious viruses"**

> The first bullet point of Box 1 articulates the concern over introduction of a viral contaminant through use of an ADM. As is implied in this bullet, ADM include the production cell line, as well as any other ADM that may be used in the upstream (cell culture through bulk harvest) or downstream (purification through final formulation and fill) manufacturing processes. For the purpose of this white paper, ADM are considered to include raw materials of animal or human origin. If raw materials are sourced from prokaryotes, yeast, or protists, they typically are not assessed unless it is known that there are ADM (such as animal peptone) used in the fermentation processes. Mitigation of viral risk associated with the biologic production cell itself (again, excluding bacterial or yeast production cells) is accomplished through following[6] ICH Q5D Derivation and Characterization of Cell Substrates Used for Production of Biotechnological/Biological Products[7] which mandates the viral testing required to assure freedom from viral contaminants such as the SV40 which contaminated the early inactivated polio vaccines. Other than the production cell, the reduction or elimination of use of ADM, where possible, is the most straightforward approach for mitigating viral risk associated with these materials. Where ADM must be used, the viral risk associated with the specific ADM is expected to be assessed and mitigated to the extent possible [i.e., to bring the viral risk to a level as low as reasonably achievable (ALARA)[8]], recognizing that zero risk is not achievable.

The assessed viral risk associated with any ADM used during biologic manufacturing is information which is expected to be conveyed to regulatory authorities in the 3.2.A.2 (Adventitious Agents Safety Evaluation) section of the Common Technical Document (CTD) used for filing new drug applications and biologic licensure applications. In fact, provided that a proper assessment of each ADM used in the biologic manufacturing process (including the production cell) has been performed, the conclusions of those assessments may be used to populate the 3.2.A.2 section. The remainder of this white paper provides guidance on how to conduct such an ADM risk assessment. As mentioned above, characterization of the production cell itself for viral risk is handled through a separate process6,7 and is not in scope for this white paper. In addition, the second and third bullets in Box 1 address viral clearance validation and viral testing of in-process samples, respectively, and these topics also are considered out of scope for this white paper.

02

Assessing Animal-Derived Raw Materials for Viral Risk

The process involved in assessing ADM for viral risk has been described previously 9 (although in less detail). Parts of the assessment process consist merely of collecting the relevant information from the manufacturer/supplier of the ADM, and this information may best be obtained by the biologic manufacturer's supplier quality or purchasing groups. The information that typically is used in the assessment is displayed in Table 1. Note that not all of this information may be available from the manufacturer/supplier, so the assessment necessarily will be based upon the information that is available to the risk assessor. In particular, the ADM manufacturing process information may, in some cases, be considered proprietary and therefore withheld. Where the manufacturing process for the ADM contains multiple viral inactivation or removal steps with the same mechanism of action, only the most stringent of the similar steps is considered during the viral risk assessment, in alignment with the manner in which viral clearance validation is performed and interpreted 10 . It is relatively atypical for ADM manufacturers/suppliers to conduct viral clearance validation studies, despite the fact that certain regulatory jurisdictions (Japan is an example) may expect to see such validation results.

Note that a separate viral risk assessment needs to be done for each manufacturer/ supplier of an ADM and, in some cases, for different catalog numbers of a given ADM from a given manufacturer/supplier, since manufacturers sometimes supply a given ADM in different stock keeping units (SKU), each having differing testing specifications and/ or administered barrier treatments (e.g., gamma irradiation) (see also Table 1).

ADM Information Source	Information contained	Look for
Product Certificate of Analysis	Product name, product catalog number, lot number, limitations on use, release testing assays and results	Is the certificate for a recently released lot? Is any viral safety release testing performed?
Product Certificate or Statement of Origin	Animal species and tissue type(s), geographical source(s)	Lower risk geographical regions are preferred.
ADM manufacturingprocess description	Possible viral inactivation or removal steps (certain of these, such as gamma or UV irradiation,are often termed barrier treatments)	Heating, low pH, gamma irradiation, UV irradiation, etc. (need to know worst-case conditions of temperatures, durations, pH, fluences, etc.)
Viral clearance validation summary	Validation of efficacy of viral inactivation or removal steps	Log 10 reduction of relevant challenge viruses under worst-case conditions

Table 1. Information Needed from ADM Supplier for Conducting Viral Risk Assessments

The availability (or non-availability) of the information listed in Table 1 might represent a consideration for selecting potential suppliers for a given ADM. This is a good segue for another recommendation, which is to consider selection of ADM sources relatively early in the biologic development process with a view to the viral risk associated with a given ADM and differences in viral risk mitigation approaches taken by different suppliers of the ADM.

Once the information in Table 1 has been obtained, it is time to conduct the viral risk assessment proper. At this point, the help of a virology subject matter expert from the biologic manufacturing organization or an external virology consultant may be required.

The viral risk assessment itself must take into account:

1. The species of origin from which the ADM was derived and the viruses of concern for that animal species. Sources for this information may include A) for human origin materials, Requirements for Blood and Blood Components Intended for Transfusion or for Further Manufacturing Use[11] ; and B for animal-origin materials, Detection of Extraneous Viruses by the Fluorescent Antibody Technique[12]. For animal species not covered by the[9] CFR reference[12] , a literature search must be conducted to determine the viruses of concern.

2. The expected (estimated) efficacy of the ADM manufacturing process steps for inactivating the viruses of concern identified in bullet 1 above. This assessment of viral inactivation efficacy for the manufacturing steps (e.g., low pH treatment, heat treatment, etc.) or barrier treatments (gamma, electron beam, or UV treatment) is facilitated by searching the published results of empirical viral inactivation studies[e.g.,13-17] as well as by using modeling techniques[15-18] allowing extrapolation of $\log_{10}$ reduction in virus titer from empirically tested treatment conditions (°C for x minutes; kGy for gamma or electron beam, mJ/cm^2 for UV) to the conditions actually used by the ADM manufacturer. The modeling is made possible by the first-order relationship between log 10 reduction of virus and the applied dose of heat or radiation, such that (for example) the time required to inactivate[2] log[10] of virus by 56°C is approximately twice that required to inactivate[1] log[10] of the virus. Of course, the estimates of viral inactivation efficacy obtained by such modeling are to be regarded as estimates only, yet the modeling results are useful for estimating viral risk reduction by such treatments and for assessing remaining viral risk post treatment.

3. As mentioned above, zero viral risk is unachievable, so in a practical sense the results of the above viral risk assessments are best couched in terms such as minimal viral risk, some viral risk, or considerable viral risk. If the manufacturer's process includes steps which are capable of inactivating ≥6 $\log_{10}$ of all of the identified viruses of concern, the ADM may be assessed as conferring minimal viral risk. If, on the other hand, one or more of the identified viruses of concern are inactivated less than[3] log 10 by the manufacturing steps, and no testing of the ADM for those particular viruses is performed per the ADM Certificate of Analysis or by the biologic manufacturer itself, the ADM may be assessed as conferring some viral risk. If no viral inactivation or removal steps are performed during manufacture of an ADM, and no viral testing of that ADM is performed by the manufacturer during lot release or by the biologic manufacturer, that ADM might be assessed as conferring considerable viral risk. As implied by the abbreviated decision tree above, mitigation of viral risk can take the form of additional viral safety testing, commissioned either by the ADM manufacturer or by the biologics manufacturer. Risk mitigation involving viral inactivation during ADM manufacturing is always preferable to risk mitigation involving additional viral testing of the ADM, due to the limitations associated with sampling of the ADM (typically one sample tested per ADM lot) and by the limitations of the viral detection assays used (in terms of volume of sample tested and the sensitivity for detection of specific viruses of concern). biologics manufacturer. Risk mitigation involving viral inactivation during ADM manufacturing is always preferable to risk mitigation involving additional viral testing of the ADM, due to the limitations associated with sampling of the ADM (typically one sample tested per ADM lot) and by the limitations of the viral detection assays used (in terms of volume of sample tested and the sensitivity for detection of specific viruses of concern).

4. The stage of the biologic manufacturing process at which the ADM is to be used may be considered during the viral risk assessment process. An ADM used only during the upstream portion of the manufacturing process may confer less viral risk than one used, for example, as an excipient during final formulation. In the former case, the biologic manufacturing process may include downstream purification steps capable of removing or inactivating viruses introduced during the upstream steps. How much virus may be removed or inactivated by the purification processes is determined during viral clearance validation studies that are required for all biologics prior to administration to humans[10]. On the other hand, any virus introduced by an ADM used as a formulation excipient will have no opportunity of being cleared by downstream purification processes and will make its way to the drug product, remaining undetected and potentially harming recipients of the drug product. It should be noted that if a biologic manufacturing process must be considered in order to allay concern of viral risk for a given ADM as described in this bullet, that ADM viral risk assessment must be considered a process-specific viral risk assessment. If the same ADM is then to be used in the manufacturing process for some other biologic, the viral risk assessment would need to be performed anew for that second manufacturing process.

Documentation of Viral Risk Assessments

It is preferable to document the conduct of a viral risk assessment as one would document any Technical Report. That is to say, the assessment optimally would be performed per a Standard Operating Procedure, documented on an approved template[9] , and archived in an organization's document storage and retrieval system, with signoff by appropriate individuals within the organization.

Those individuals might include the risk assessment author (i.e., the subject matter expert) and representatives from manufacturing, procurement (purchasing), and Quality. The assessments need to be easily retrievable, as they should be considered fair game for request during regulatory agency inspection.

Conclusions

It is a regulatory expectation that ADM be assessed for viral risk [1,8,9,19] , and the information obtained as a result of the conduct of the viral risk assessments may be used in completing Section 3.2.A.2 of regulatory filings for a biologic. The information provided in this white paper should demystify the processes involved in the conduct and documentation of such viral risk assessments.

How Syner-G Can Support Viral Risk Assessments for ADM

Syner-G BioPharma Group stands at the forefront of regulatory and scientific consulting for biologic development, with particular strength in viral risk assessment and mitigation for animal- derived materials (ADM). Given the complexity, regulatory scrutiny, and scientific rigor required to assess and document viral safety in biologics manufacturing, partnering with a knowledgeable and experienced organization is critical.

Syner-G offers deep expertise in the interpretation and application of regulatory guidance such as ICH Q5A(R2), ICH Q5D, and regional expectations such as those in Japan and Europe.

Our team specializes in:

Designing and executing ADM risk assessments tailored to the specific material, supplier, and intended use stage in the biologics process.

Identifying viral risks associated with specific animal species and processing conditions using the latest literature and empirical modeling.

Providing comprehensive documentation packages aligned with Section 3.2.A.2 of the Common Technical Document (CTD), which are inspection-ready for global regulatory agencies.

Advising on ADM supplier selection and qualification strategies that balance manufacturing practicality with viral safety compliance.

Additionally, Syner-G brings a holistic, phase-appropriate approach to biologics development, integrating ADM viral risk assessments with broader CMC strategy and regulatory submission planning. This integrated view ensures that viral safety evaluations are not isolated exercises but are embedded into the overall quality and risk management frameworks of our clients' development programs.

In an environment where viral safety is both a scientific imperative and a regulatory requirement, Syner-G delivers the insight, precision, and compliance assurance needed to de-risk your supply chain and accelerate your product to market with confidence.

To learn more, visit: www.synergbiopharma.com

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