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# Achieving a Successful Scale-Down Model and Optimized Economics through Parvovirus Filter Validation using Purified TrueSpike™ Viruses

PHILIPPE DE VILMORIN[1], ASHLEY SLOCUM[2], TAREQ JABER[3], OLIVER SCHAEFER[3], HORST RUPPACH[3], and PAUL GENEST[2,*]

*[1]Biogen Idec Inc., Cambridge, MA; [2]EMD Millipore Corporation, Bedford, MA; and [3]Charles River Laboratories International, Inc., Wilmington, MA* 

*ABSTRACT:* This article describes a four virus panel validation of EMD Millipore's (Bedford, MA) small virus–retentive filter, Viresolve® Pro, using TrueSpike™ viruses for a Biogen Idec process intermediate. The study was performed at Charles River Labs in King of Prussia, PA. Greater than 900 L/m$^2$ filter throughput was achieved with the approximately 8 g/L monoclonal antibody feed. No viruses were detected in any filtrate samples. All virus log reduction values were between ≥3.66 and ≥5.60. The use of TrueSpike™ at Charles River Labs allowed Biogen Idec to achieve a more representative scaled-down model and potentially reduce the cost of its virus filtration step and the overall cost of goods. The body of data presented here is an example of the benefits of following the guidance from the PDA Technical Report 47, *The Preparation of Virus Spikes Used for Viral Clearance Studies*.

*LAY ABSTRACT:* The safety of biopharmaceuticals is assured through the use of multiple steps in the purification process that are capable of virus clearance, including filtration with virus-retentive filters. The amount of virus present at the downstream stages in the process is expected to be and is typically low. The viral clearance capability of the filtration step is assessed in a validation study. The study utilizes a small version of the larger manufacturing size filter, and a large, known amount of virus is added to the feed prior to filtration. Viral assay before and after filtration allows the virus log reduction value to be quantified. The representativeness of the small-scale model is supported by comparing large-scale filter performance to small-scale filter performance. The large-scale and small-scale filtration runs are performed using the same operating conditions. If the filter performance at both scales is comparable, it supports the applicability of the virus log reduction value obtained with the small-scale filter to the large-scale manufacturing process. However, the virus preparation used to spike the feed material often contains impurities that contribute adversely to virus filter performance in the small-scale model. The added impurities from the virus spike, which are not present at manufacturing scale, compromise the scale-down model and put into question the direct applicability of the virus clearance results. Another consequence of decreased filter performance due to virus spike impurities is the unnecessary over-sizing of the manufacturing system to match the low filter capacity observed in the scale-down model. This article describes how improvements in mammalian virus spike purity ensure the validity of the log reduction value obtained with the scale-down model and support economically optimized filter usage.

## Introduction

The body of data presented here is an example of the benefits of following the guidance from the Parenteral Drug Association (PDA) Technical Report 47: The Preparation of Virus Spikes Used for Viral Clearance Studies (1). The viral clearance capability of the filtration step is assessed in a validation study. The study utilizes a small version of the larger manufacturing size filter, and a large, known amount of virus is added to the feed prior to filtration. Viral assay before and after filtration allows the virus log reduction value (LRV) to be quantified (2–4).

Manufacturers of biopharmaceuticals are required to validate the virus removal capability of their manufacturing process. Parvovirus (small virus–retentive) filters are utilized as a dedicated virus removal step in the manufacturing process and play an important role

*Corresponding Author: Paul Genest, EMD Millipore Corporation, 900 Middlesex Turnpike Billerica, MA, 01821. e-mail: paul.genest@emdmillipore.com
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in the safety of the drug product. Parvovirus filter membranes remove viruses via a robust size exclusion mechanism (5). To remove greater than 4 $\log_{10}$ of parvovirus (approximately 20 nm in size) while simultaneously passing 100% of the product (monomer monoclonal antibody [MAb] <8 nm), the pore size distribution ideally consists of pores in the narrow 12–18 nm range. The pores may plug if the feed contains species in the same size range.

Typically, process intermediates at the virus filter unit operation are relatively pure, having already been through multiple chromatography steps. Some process intermediates naturally form aggregate as two or more monomers bind together. When aggregates are present, adsorptive pre-filters have proven effective at improving the capacity of the parvovirus filter, providing performance improvements superior to other size based pre-filters (6, 7). Once the optimized filter performance is demonstrated during process development and scaled-up to pilot and manufacturing scales, the expectation is that optimized performance will be achievable during viral clearance validation studies.

To demonstrate the validity of the scaled-down model and to evaluate the impact of the virus spike addition, it is recommended to perform a concurrent "baseline" run in which the feed material does not contain any virus. Previous published articles (8) and manufacturer technical literature explain the best practices for performing virus filter runs. A significant challenge arises when virus spikes contain impurities that contribute to fouling of the virus filters on their own or via interaction with the process intermediate (9–12). This compromises the scaled-down model and the applicability of the achieved LRV to the large-scale manufacturing process. Filter fouling may affect the filter's viral retention performance, either positively or negatively (1). Validity of the scaled-down model is assured when the same fouling profile is present with and without virus spike.

Virus filters at manufacturing scale are not typically loaded beyond the volumetric filter loading ($L/m^2$) demonstrated with the virus spike present. The loading achieved with the scaled-down model in the validation becomes the maximum loading at manufacturing scale. Given this constraint, increased fouling and decreased capacity due to virus spike addition causes the filter to be oversized in the scaled-up manufacturing process, adding unnecessary cost throughout the life of the drug product. Various mitigation strategies have been proposed to reduce the negative impact of impure virus spikes: diluting the virus spike, substituting high-purity bacteriophage (5, 12–14) for mammalian viruses, and implementing RUNspike (delaying the virus spike addition until representative filter throughput is achieved) (15–17). Higher dilution of the virus spike does help in most cases, but it reduces the claimable LRV. Furthermore, the authors know of no instance where either replacing mammalian virus with bacteriophage or using RUNspike has gained regulatory acceptance.

Another option is to address the root cause of the compromised scale-down model, which is the impurity of the mammalian virus spikes. Purification of virus spikes to minimize their contribution to filter fouling supports the validity of the scaled-down model and optimal process economics. To consolidate efforts toward the goal of improved viral spike purity, a task force was formed with endorsement from the PDA. The task force included biopharmaceutical manufacturers, regulators, viral clearance validation labs, and virus filter manufacturers (1). The result was formal acknowledgement of the issue and a compilation of data regarding existing virus purification methods and the impact of impure virus spikes on virus filter capacity and LRV.

PDA Technical Report 47: The Preparation of Virus Spikes Used for Viral Clearance Studies provides guiding principles to help minimize the impact of viral spike impurities on the performance of the scale-down model. Technical Report 47 prescribes a combination of diluting the virus spike and virus spike quality to achieve this goal for filtration, but it does not specifically define a virus spike purity standard or a standard virus purification method. It recommends minimizing total protein and DNA content, which have been proposed as the main impurities affecting filtration (9).

Technical Report 47 states the following regarding worst-case conditions for virus filter validation: "In virus-retentive filtration experiments it was demonstrated that with some filters the more pure preparation leads to lower LRV compared to crude virus preparations while other virus filter types showed comparable LRV for crude vs purified spikes". However, a higher purity virus spike has an increased propensity for undesirable aggregation of virus. Aggregated parvovirus is specifically not the worst case for the size-based virus filter. Parvovirus filters are not absolute and some breakthrough of parvovirus is observed. Aggre-

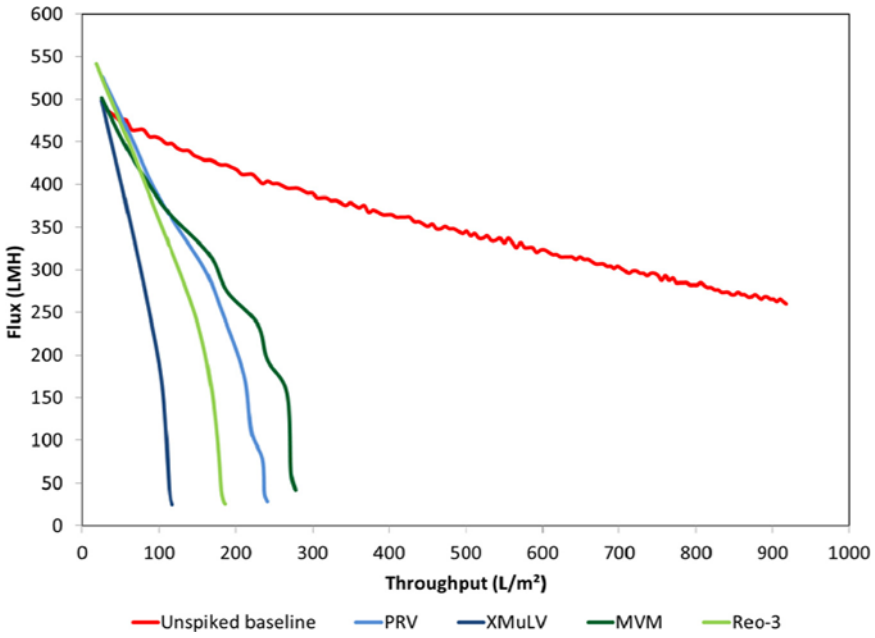


**Figure 1**

**Flux vs throughput for virus filtration runs with minimally purified viral preparations (baseline vs virus-spiked).**

gated parvovirus is more likely to be retained by the filter, resulting in an artificially increased LRV. In contrast, there is significant data showing parvovirus filters exhibiting complete retention of large viruses like Xenotropic murine leukemia virus-related virus (XMuLV) and pseudorabies virus (PRV) (5). Hence, aggregation of these large viruses is less relevant to filter retention performance. Technical Report 47 discusses the use of sonication to break up aggregates, dynamic light scattering to detect aggregates, and microfiltration post–virus spike to assess virus monodispersity. Overall, the advice of Technical Report 47 is to consider all of these factors during the development of virus spike production methods to ensure high purity and minimal aggregation.

Preparation of high-purity virus spikes while avoiding aggregation is not a trivial challenge. Minimizing aggregation is particularly difficult due to the wide range of solution conditions (pH and conductivity) to which the viruses may be exposed when spiked into biopharmaceutical process intermediates. Viruses are produced in mammalian cell culture, and the unpurified virus preparation contains media, host cell protein, DNA, and cellular debris. In the past, the crude virus preparations were minimally purified (centrifugation and/or filtration only). In response to the recommendations of Technical Report 47, many viral clearance validation labs have improved the purity of their virus spikes to support more representative virus filtration.

All of the viruses used in this body of work, called TrueSpike™, have purity specifications established to ensure minimal impact on the performance of EMD Millipore's (Bedford, MA) small virus–retentive filter, Viresolve® Pro (18). Multiple purification steps are

**TABLE I**
**Summary of Virus Filtration Runs with Minimally Purified Virus Preparations**

| | PRV | XMuLV | MVM | Reo-3 |
|---|---|---|---|---|
| Measured Virus Challenge ($TCID_{50}$/mL) | $1.8 \times 10^4$ | $5.8 \times 10^4$ | $5.9 \times 10^6$ | $7.9 \times 10^5$ |
| Log Reduction Value | ≥3.1 | ≥3.6 | ≥5.6 | ≥4.7 |

**TABLE II**
**TrueSpike™ Virus Targets and Characterization Data for Biogen-Idec Study**

| | PRV | XMuLV | MVM | Reo-3 |
|---|---|---|---|---|
| **Virus Titer Target ($\log TCID_{50}$/mL)** | ≥ 7.5 | ≥ 6.8 | ≥ 8.5 | ≥ 8.0 |
| *Actual Virus Titer (log TCID50/mL)* | *6.97±0.25* | *6.97±0.25* | *9.40±0.24* | *7.73±0.29* |
| **Protein Target (μg/mL)** | ≤ 100 | ≤ 100 | ≤ 150 | ≤ 100 |
| *Actual Protein (μg/mL)* | *2.5* | *63.1* | *50* | *2.5* |
| **DNA Target (μg/mL)** | ≤ 2 | ≤ 2 | ≤ 10 | ≤ 2 |
| *Actual DNA (μg/mL)* | *0.35* | *0.30* | *7* | *0.11* |

used for each virus to deliver purity within specification. The purified virus preparations are characterized for virus titer, total protein content, and total DNA content. The virus stock impurity profile may also be generated using sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) as an additional confirmation of purity. Characterization data from several analytical methods provide the biopharmaceutical manufacturer with a high level of assurance that a virus preparation will better support a representative scaled-down virus filtration model.

This case study presented some unique challenges: a large change in the potential of the MAb to aggregate post freeze/thaw, increased turbidity upon transfer between containers, a high level of cell based assay cytotoxicity and interference, and a relatively large amount of XMuLV aggregation in the presence of the Biogen process intermediate. Most of these challenges are artifacts, like freezing and thawing, which are only present in the validation environment and do not exist in the manufacturing process.

Previous attempts by Biogen Idec to model the virus filtration process with minimally purified virus preparations resulted in a maximum volumetric throughput of approximately 300 L/m$^2$ and greater than 90% flow decay, as shown in Figure 1. The virus preparations used in this study were purified with non-chromatographic methods and characterized only for viral titer. Table I lists the viral challenges and LRVs achieved. Although effective removal of virus was demonstrated, the volumetric throughput did not meet the study target.

As capacities exceeded 900 L/m$^2$ in unspiked baseline runs, it was hypothesized that impurities in the viral preparations contributed to the low volumetric throughput. The subsequent use of TrueSpike™ at Charles River Labs allowed Biogen Idec to achieve a more representative scaled-down model and potentially reduce the cost of its virus filtration step and the overall cost of goods.

## Materials and Methods

### *Viruses*

**TrueSpike™ Virus:** The production and purification processes for the TrueSpike™ viruses are described in European Patent Application number 11162488.8. Each virus is produced in cell culture and processed using multiple unit operations to purify and/or concentrate the virus (18). The goals are to reduce impurities to the lowest possible level, maintain the highest possible virus titer, and minimize virus aggregation. Table II summarizes the target titer, protein, and DNA characterization data for the TrueSpike™ virus panel as well as the actual values for the lots used in this

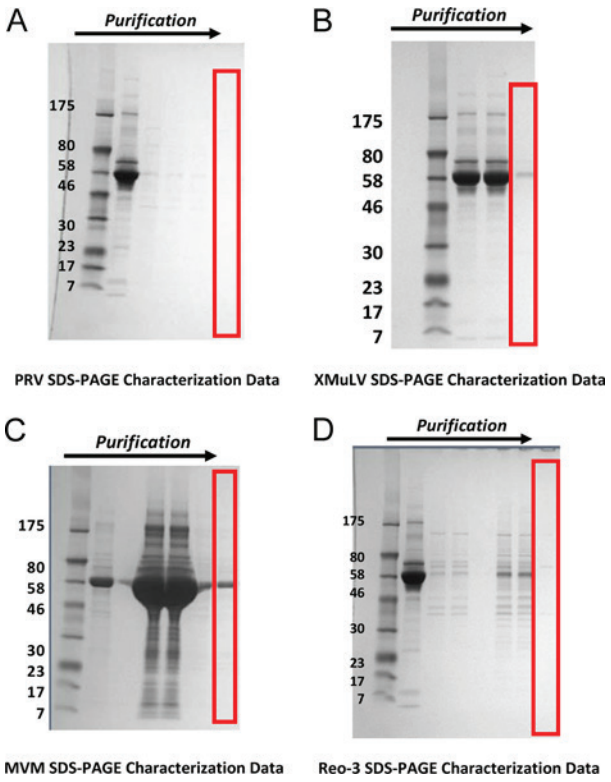


**Figure 2**

**SDS-PAGE images for TrueSpike™ viruses (panels: A = PRV upper left, B = XMuLV upper right, C = MVM lower left, D = Reo-3 lower right).**

work. Shown in Figure 2 (panels A–D) are silver-stained SDS-PAGE gels with samples taken throughout the purification processes of the virus lots used. Red boxes indicate the lane with the final, purified virus preparation. The target levels (μg/mL) for the protein and DNA impurities are lower for the minute virus of mice (MVM) virus as compared to the other viruses due to the higher achievable titer of the MVM virus. The critical attribute when it comes to virus preparation purity is the amount of impurity per virus particle challenged, as the total virus particles challenged is targeted to demonstrate a particular level of LRV.

**Virus Titer and LRV Determination:** To determine the virus titer of a sample, serial 3-fold dilutions were made with cell culture medium. One hundred microliter aliquots of each dilution were added to eight wells of a 96-well microtiter plate with cells (in 100 μL cell culture medium per well). The filtrate samples were additionally analyzed in a large-volume plating analysis by taking a defined number of wells already containing the indicator cells in 100 μL of cell culture medium and adding 200 μL of filtrate sample, already diluted for cytotoxicity/interference, into each well. The cells were cultivated for a specified incubation period, then inspected microscopically for virus-induced changes in cell morphology. The indicator cells and period of incubation used for PRV, XMuLV, MVM, and Reovirus type 3 (Reo-3) were the following: Vero76 after 5–8 days, PG4 after 6–8 days, 324K after 11–15 days, and LLC-MK2 after 11–15 days, respectively. The virus titer (median tissue culture infective dose [$TCID_{50}$]) was calculated according to the Spearman-Kaerber formula (19, 20) or the Poisson distribution. The reduction factor was calculated by dividing the total amount of virus in the spiked test item by the total amount of virus in the filtrate. Due to the cytotoxicity and interference of the process intermediate, the required sample dilutions for PRV, XMuLV, MVM, and Reo-3 are 1:162, 1:162, 1:1458, and 1:486, respectively.

*Filtration*

Prior to virus spiking, the Biogen process intermediate was filtered with 23 $cm^2$ Millistak+® A1HC

**TABLE III**
**Process Intermediate Lot Summary**

| | Run ID | Feed Lot |
|---|---|---|
| PRV Scoping | Baseline A | |
| | PRV 1 | A |
| | PRV 2 | |
| PRV Official | Baseline 1 | C |
| | PRV Run 1 | A |
| | PRV Run 2 | C |
| XMuLV Scoping | Baseline B | |
| | XMuLV 1 | B |
| | XMuLV 2 | |
| XMuLV Official | Baseline 2 | A |
| | XMuLV Run 1 | A |
| | XMuLV Run 2 | B |
| MVM Scoping | Baseline C | |
| | MVM 1 | C |
| | MVM 2 | |
| MVM Official | Baseline 3 | C |
| | MVM Run 1 | C |
| | MVM Run 2 | B |
| Reo-3 Scoping | Baseline D | |
| | Reo-3 1 | B |
| | Reo-3 2 | |
| Reo-3 Official | Baseline 4 | B |
| | Reo-3 Run 1 | B |
| | Reo-3 Run 2 | C |

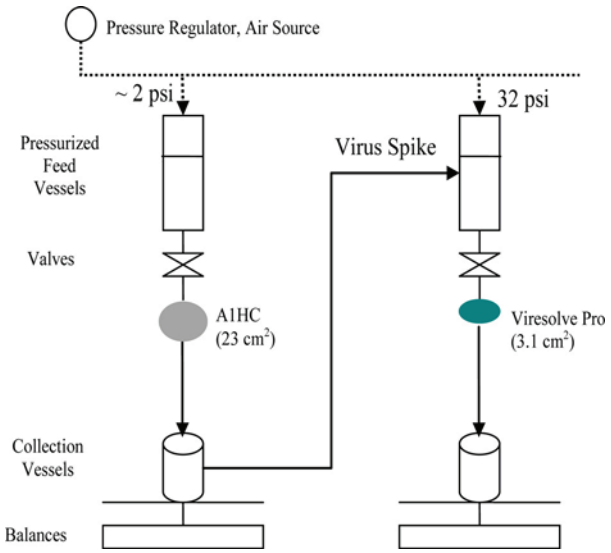


**Figure 3.**

**Schematic of experimental setup.**

Micro Pod adsorptive depth filters (catalog number MA1HC23CL3). Scaled-down virus filtration was performed with two different lots of 3.1 $cm^2$ Viresolve® Pro Micro devices (catalog number VPMCVALNB9).

Three different process intermediate lots, each with a MAb concentration of approximately 8 mg/mL, were used as feed material for this work: A, B, and C. Two different lots of feed were used for each official virus spiked run, including an additional baseline run. Scoping runs were completed prior to the virus spiked runs to determine the optimal level of virus spike needed to ensure the target throughput. Table III summarizes the feed lots used in each run.

The process intermediate feed was stored frozen in 1 L containers prior to this work. On the first day of the study, all feed samples were thawed in a water bath at approximately 25 °C. After thawing, the process intermediate was stored at 2–4 °C. Prior to prefiltration, the feed was warmed to room temperature in the water bath. The buffer (20 mM acetate, 350 mM ammonium sulfate, pH 5.8) was stored at room temperature. Milli-Q purified water was used to perform an initial flushing of the filter devices. Figure 3 is a schematic of the experimental setup.

The scoping runs for each virus consisted of three total runs: baseline, high-level spike, and low-level spike. The higher virus spike level was used for the official spiking runs if the virus filter throughput was minimally affected; otherwise the lower spike level was used. The official spiking runs also consisted of three total runs. Two lots of feed material were spiked with the same amount of virus, determined from the results of the scoping runs (data not shown). One baseline run, using at least one of the same feed lots, was run at the same time as the spiked runs.

The Millistak+® A1HC devices were flushed with approximately 400 mL of water for injection (WFI) at 20 psi, followed by equilibration with 200 mL of buffer at 20 psi. To prepare the baseline feed and the feed for the spiked runs, approximately 320 mL of Biogen process intermediate was prefiltered through the Millistak+® A1HC (139 $L/m^2$) at a constant flow rate of approximately 10 mL/min (~2 psi). As soon as the prefiltration was completed, the feed was spiked with virus.

Previous attempts to microfilter the Biogen process intermediate post–virus spike affected the material such that the minimum target throughput could not be achieved. Based on those results and the handling sensitivity of the feed material, bulk microfiltration post–virus spike was not performed for any virus during this study. Instead, samples of approximately 5

**TABLE IV**
**Measured Virus Challenge**

| Run ID | Feed Lot | Measured Virus Challenge ($TCID_{50}$/mL) |
|---|---|---|
| Baseline 1 | C | N/A |
| PRV Run 1 | A | $4.7x10^5$ |
| PRV Run 2 | C | $1.4x10^5$ |
| Baseline 2 | A | N/A |
| XMuLV Run 1 | A | $1.4x10^4$ |
| XMuLV Run 2 | B | $1.8x10^4$ |
| Baseline 3 | C | N/A |
| MVM Run 1 | C | $3.8x10^6$ |
| MVM Run 2 | B | $1.9x10^6$ |
| Baseline 4 | B | N/A |
| Reo-3 Run 1 | B | $4.7x10^4$ |
| Reo-3 Run 2 | C | $3.1x10^4$ |

**TABLE V**
**Summary of Filtration Capacity Results**

| Run ID | Measured Virus Challenge ($TCID_{50}$/mL) | Filtration Time (min) | Achieved Product Throughput Before Buffer Flush ($L/m^2$) | Achieved Throughput After Buffer Flush ($L/m^2$) | % Flow Decay |
|---|---|---|---|---|---|
| Baseline 1 | N/A | 114 | 1004.2 | N/A | 12% |
| PRV Run 1 | $4.7x10^5$ | 107 | 991.9 | 1013.2 | 7% |
| PRV Run 2 | $1.4x10^5$ | 114 | 991.3 | 1013.5 | 16% |
| Baseline 2 | N/A | 131 | 999.0 | N/A | 18% |
| XMuLV Run 1 | $1.4x10^4$ | 136 | 997.7 | 1018.4 | 28% |
| XMuLV Run 2 | $1.8x10^4$ | 136 | 978.7 | 1001.0 | 23% |
| Baseline 3 | N/A | 127 | 1047.1 | N/A | 4% |
| MVM Run 1 | $3.8x10^6$ | 141 | 986.8 | 1007.7 | 28% |
| MVM Run 2 | $1.9x10^6$ | 128 | 990.3 | 1011.0 | 24% |
| Baseline 4 | N/A | 144 | 994.5 | N/A | 36% |
| Reo-3 Run 1 | $4.7x10^4$ | 113 | 998.4 | 1019.7 | 11% |
| Reo-3 Run 2 | $3.1x10^4$ | 140 | 1008.1 | 1029.7 | 34% |

mL of virus spiked process intermediate were microfiltered with vacuum filters to assess virus monodispersity. A 0.1 μm vacuum filter was used for MVM, a 0.2 μm vacuum filter was used for Reo-3, and 0.45 μm vacuum filters were used for both XMuLV and PRV.

Viresolve® Pro devices were purged of air and then wet with Milli-Q water for 5 min at 50 psi and then for 5 additional minutes at 32 psi. Approximately 30 mL of buffer was subsequently flushed through the Viresolve® Pro devices at a constant pressure of 32 psi. After the Viresolve® Pro devices were prepared, the virus-spiked process intermediate was sampled for viral titer and then added to the viral filtration feed vessel. Table IV contains information on the measured virus challenge.

Filtrate weight was measured as a function of time to determine changes in flux with time. Flow decay is determined by expressing the final flux as a percentage of the initial buffer flux and subtracting this result from 100%. Processing was stopped when the feed was depleted. After loading was complete, each filter was flushed with 20 $L/m^2$ (6.2 mL) of buffer. The pooled filtrate was sampled for virus titer. According to the filter manufacturer's recommendations, a post-use integrity test at 50 psi was completed for all virus challenged devices; no breach of integrity was observed in any of the devices.

## Results and Discussion

### *Viresolve® Pro Throughput Results*

Table V and Figure 4 (panels A–D) show that spiking with high-purity virus had minimal impact to the virus filter throughput. Filter performance in the virus-spiked runs was comparable to the baseline runs with no virus. All runs achieved greater than 900 $L/m^2$ throughput with less than 40% flow decay. The results were a significant improvement over previous viral clearance studies, in which only 300 $L/m^2$ throughput was achieved. The biopharmaceutical manufacturer's validation procedure requires that the spike runs were performed with two different lots of process intermediate. In both baseline and spiked runs, the B lot was observed to promote greater flow decay relative to the A and C lots. For example, the Reo-3 virus–spiked run using the C lot showed less flow decay compared to the other Reo-3 virus spiked run and the baseline, both

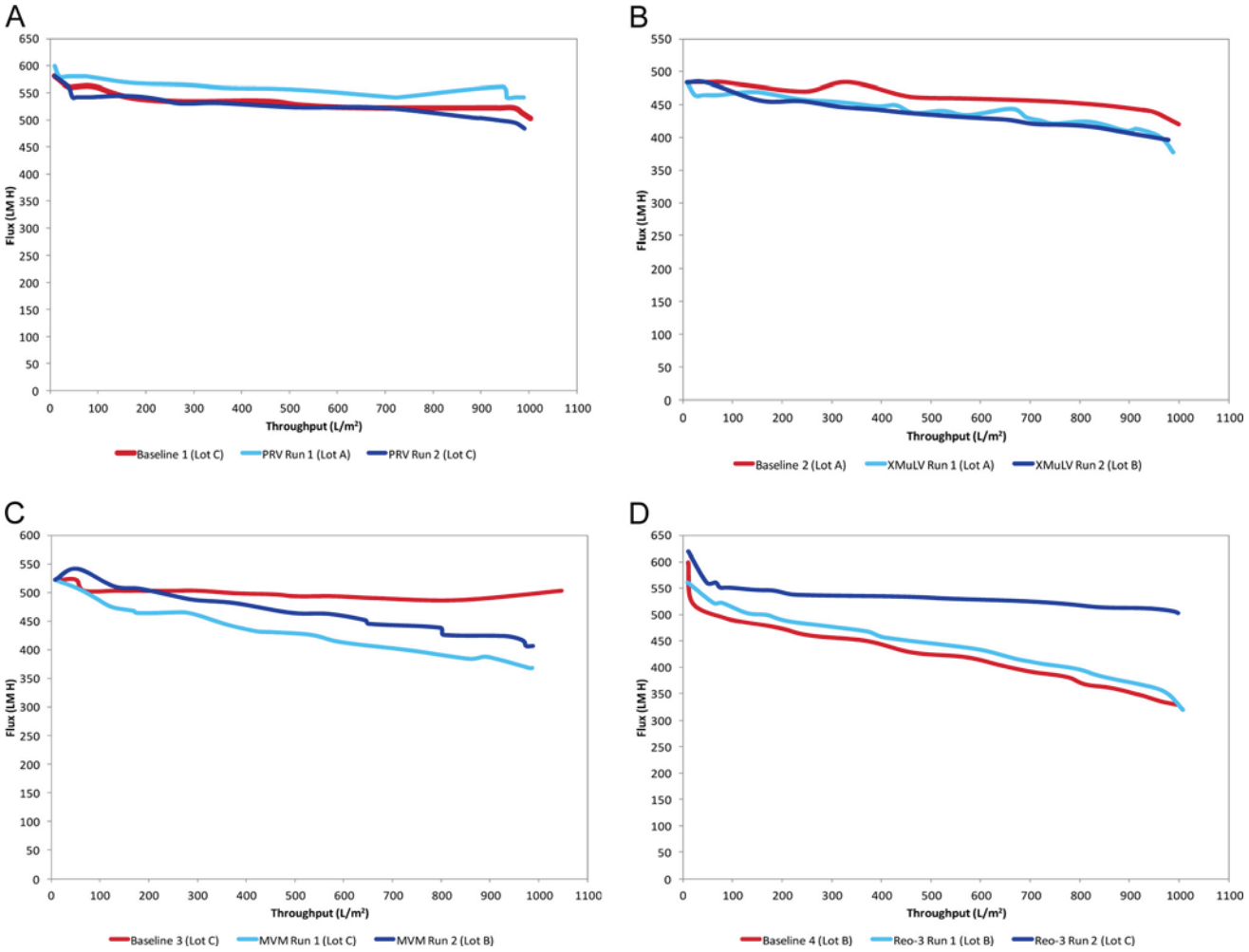


**Figure 4**

**Flux vs throughput for all virus filtration runs (baseline vs virus-spiked) (panels: A = PRV upper left, B = XMuLV upper right, C = MVM lower left, D = Reo-3 lower right).**

of which used the B lot (Figure 4, panel D). However, the presence of the virus spike showed no impact on filter performance, as evidenced by the comparable performance observed in the baseline runs and the virus-spiked runs using the same process intermediate lot.

*Filter LRV Results*

All filtrate samples showed no detectable virus, therefore all LRVs are expressed as "greater than or equal to" (≥) values. Most of the LRVs met the study target of ≥4 $\log_{10}$ removal; see Table VI. For XMuLV, the samples were diluted due to the high cytotoxicity of the process intermediate, which limited the claimable LRV values. In the absence of cytotoxicity, reported LRV values would most likely have been higher. One option to avoid sample dilution is to use buffer exchange by size exclusion chromatography or dialysis to remove cytotoxic components from samples prior to assay. However, this approach presents the risk that the buffer exchange process will change the amount of virus in the sample. Separate validation is required to prove that no infectivity is removed during the buffer exchange process. Addressing cytotoxicity through di-

**TABLE VI**
**Summary of LRV Results for Duplicate Virus-Spiked Runs**

| | PRV | XMuLV | MVM | Reo-3 |
|---|---|---|---|---|
| Run 1 | ≥4.62 | ≥3.66 | ≥5.60 | ≥4.18 |
| Run 2 | ≥4.09 | ≥3.79 | ≥5.30 | ≥4.00 |

lution alone is the most reliable method to ensure full retention of infectivity, and therefore this study relied solely on dilution and large-volume assays.

Virus removal by microfiltration of MVM-, Reo-3-, and PRV-spiked process intermediate was $\leq 0.5$ $\log_{10}$. This removal of virus is insignificant and supports the monodispersity of the virus preparations. The XMuLV-spiked material, however, showed a reduction of 1.2 $\log_{10}$ across the 0.45 μm microfilter, suggesting significant aggregation of virus. This is in contrast to the same XMuLV TrueSpike™ virus prep which, when 0.45 μm–filtered in its final purified formulation, showed minimal virus loss. The amount of virus removed by microfiltration is virus- and product-specific. The high cytotoxicity and interference of the filtrate and the apparent tendency of XMuLV to aggregate in or interact with this process intermediate limited the capability to demonstrate reduction of monodispersed XMuLV. Based on the high level of monodispersity demonstrated for the other viruses, specifically the smallest virus MVM, all LRVs were calculated based on the feed titer measured before microfiltration.

Although monodispersity was not demonstrated for XMuLV, there is an expectation that in a real process challenged with XMuLV, the viruses would aggregate in or interact with this process intermediate as observed in this study. This is the rationale for considering aggregated or interacting XMuLV in the calculation of the reduction factor. The aggregation of XMuLV induced by this process intermediate provides additional assurance that this type of contamination will effectively be removed during virus filtration. Unlike the smallest viruses such as MVM, it is well established that even monomeric XMuLV virus is too large to pass through small virus–retentive, parvovirus filters (5).

## Conclusion

The case study presented here shows the positive outcome of utilizing highly purified and well characterized virus preparations, as recommended in the PDA Technical Report 47. The study met the goal of demonstrating robust virus clearance for all viruses tested. Filter fouling was minimal and comparable to the results of baseline runs, supporting the representativeness of the scaled-down model. The higher throughput achieved with the high purity virus spike presents an opportunity for the biopharmaceutical manufacturer to reduce filter usage at manufacturing scale. In summary, the use of high-purity virus in viral clearance validation studies enables improved compliance, reduced production costs, and a more reliable assessment of the viral safety of the biopharmaceutical product.

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## Declarations

The contents of this publication do not represent a Biogen endorsement of any products described herein and are not meant to imply that Biogen uses any of these products for clinical or commercial manufacturing. The authors declare that they have no competing interests related to this manuscript, financial or otherwise.

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Bethesda Towers
4350 East West Highway
Suite 150
Bethesda, MD 20814 USA
Tel: +1 (301) 656-5900
Fax: +1 (301) 986-0296
E-mail: info@pda.org
Web site: www.pda.org

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