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Application Highlight

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Faster Diafiltration and Ultrafiltration of Lentiviral Vectors with Vivaflow® SU

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Introduction

Lentiviral vectors (LV) are widely used for gene therapy and genetically modified cell therapies, with many clinical trials in the pipeline[1]. A typical downstream processing workflow for LV comprises clarification, purification, diafiltration, ultrafiltration, and finally sterile filtration. The diafiltration and ultrafiltration steps are usually performed after purification, to formulate the LV into a suitable storage buffer and to concentrate it[2].

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Objectives and Methods

In this study, the new flow path design of Vivaflow® SU cassettes was compared with the previous generation Vivaflow® 50 (Figure 1), to assess diafiltration and ultrafiltration performance for lentiviral vectors.

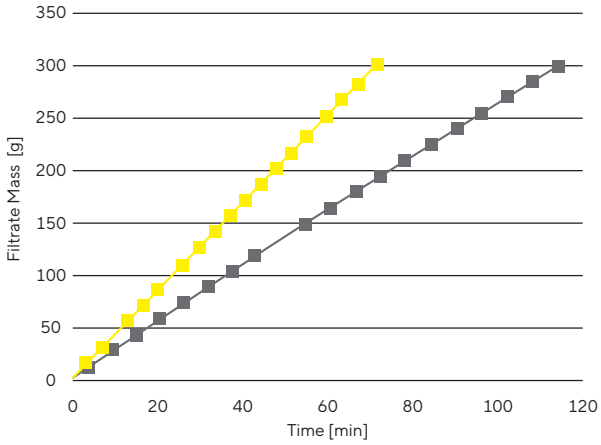
Figure 1: The performance of Vivaflow® SU (foreground) was compared with Vivaflow® 50 for diafiltration and ultrafiltration of an LV sample.

The starting LV material was previously purified by steric exclusion chromatography (SXC)[3]. A PES ultrafiltration membrane with 100 kDa molecular weight cut-off and 50 cm^2 active area was incorporated into both Vivaflow® cassette designs. Each cassette was flushed with purified water (Arium®) and then with PBS before starting the diafiltration. Using a diafiltration reservoir, 175 mL of LV material was exchanged into a formulation buffer (20 mM HEPES, 75 mM NaCl, 2.5% (w/v) sucrose, pH 7) at a constant pressure of 2 bar at the retentate port. Buffer exchange was allowed to proceed until 1.7 diafiltration volumes (300 mL) of formulation buffer had been added to the sample. Finally, the material was volumetrically concentrated 6- to 7-fold.

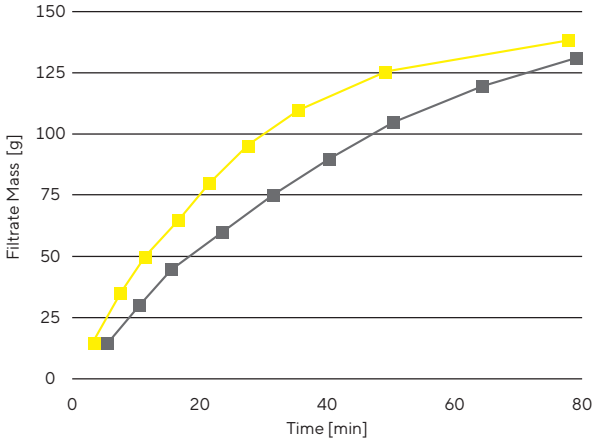
Results and Conclusion

For diafiltration (DF), Vivaflow® SU gave a 36% time saving, when compared with Vivaflow® 50 (Figure 2A). Vivaflow® SU also showed a slight advantage in terms of process speed for ultrafiltration (UF) of the LV sample (Figure 2B). We further demonstrated that infectious LV recoveries were comparable for both cassette designs (Figure 2C).

A. Continuous Diafiltration of LV SXC Eluate



B. Ultrafiltration of LV SXC Eluate



C. Infectious LV Recovery after DF and UF

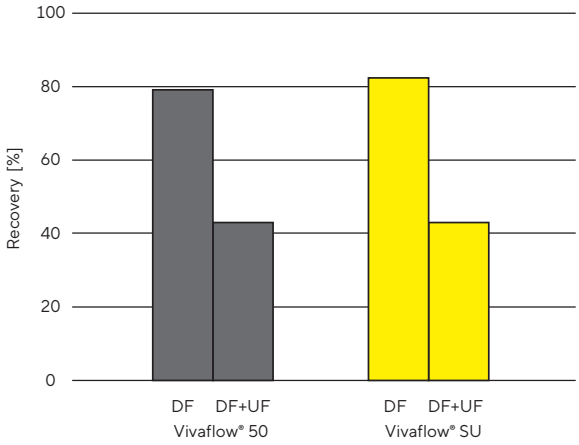


Figure 2: Filtrate mass was measured over time during continuous diafiltration (DF, A) and ultrafiltration (UF, B) of lentiviral vector material obtained after SXC purification. Infectious LV recovery was determined following DF and DF+UF (C). Data were collected for Vivaflow® SU (yellow) and Vivaflow® 50 (gray).

In conclusion, we could show that Vivaflow® is well suited for diafiltration and ultrafiltration of LV. While both cassette designs provided comparable recoveries, the new Vivaflow® SU has the advantage of a substantially shorter processing time.

References

1. Roots Analysis Business Research & Consulting (2021). Viral vectors, non-viral vectors and gene therapy manufacturing market, 2021-2030. Research report, 4th ed. https://www.rootsanalysis.com/reports/view_document/viral-vectors-non-viral-vectors-and-gene-therapy-manufacturing-market-/274.html.
2. Perry, C. & Rayat, A. C. M. E. (2021). Lentiviral vector bioprocessing. Viruses 13.
3. Labisch, J. J., Kassar, M., Bollmann, F., Valentic, A., Hubbuch, J. & Pflanz, K. (2022). Steric exclusion chromatography of lentiviral vectors using hydrophilic cellulose membranes. Journal of Chromatography A 17, 463148.

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