

Lab Filtration Application Note Reference List

Application Support Materials for a Wide Range of Filtration, Concentration and Separation Workflows

Simplifying Progress

SARTURIUS

Lab Filtration Technologies

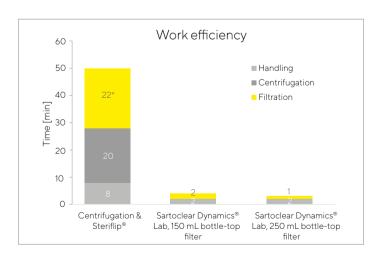
Sartorius Lab Filtration offers a comprehensive range of products and technical guidance for the applications of your choice. This document outlines the areas of application knowledge and resources which are available to you.

Sartorius is constantly improving the quality of its product portfolio and application know-how.

It is often necessary to qualify and optimize products for use in various laboratory applications. To support our customers Sartorius carries out application development in the filtration fields of:

- Clarification
- Sterile filtration
- Concentration
- Molecule separation
- Purification
- Basic filtration

Our application knowledge extends across life science (mAbs, viral vectors, biologics), environmental, clinical diagnostics and food & beverage workflows.



Our Application Notes help to:

- Describe product optimization
- Reduce workflow bottlenecks
- Outline product and application case studies
- Introduce novel techniques
- Provide general tips and tricks
- Improve result accuracy and reliability
- Increase user confidence

If you are interested in working with us on further applications, please feel free to reach out and provide your contact details.



SARTURIUS

Application Note

January 15, 2018

Keywords or phrases:

Cell Culture Supernatant, Protein Purification, Concentration, Diafiltration, Ion Exchange, Desalting, Workflow

Example 1: Vivaflow® and Vivaspin® Workflow in Protein Research Laboratories

Rik McRae¹, Hannes Landmann²,*

1. Sartorius Stedim Lab Ltd, Sperryway, Stonehouse, Gloucestershire, GL10 3UT, UK
2. Sartorius Lab Instruments GmbH & Co. KG, Otto-Brenner-Straße 20, 37079 Goettingen, Germany

E-Mail: john.cashman@sartorius.com

Abstract

In this Application Note, we demonstrate how the Vivaflow® cassettes, Vivapure® Ion Exchange spin columns and Vivaspin® devices can be used to perform a complete protein purification workflow, from concentration and diafiltration of the original protein source, a cell culture supernatant, to final concentration | desalting of the purified protein. The protocol shows in detail the recoveries after each step along with the time needed for every purification and concentration step.

Find out more: www.sartorius.com

^{*} Correspondence:

SARTURIUS

Application Note

April 27, 2018

Keywords or phrases:

CHO Cell Culture, mAb, Clarification, Supernatant, Syringe Filter, Turbidity, Product Recovery, Yield, Throughput, Filter Usage

Example 2:

Minimizing Syringe Filter Usage in Harvesting Monoclonal Antibodies from CHO Cell Culture Supernatants

Michael Grauf^{1*}, Klaus Schöne^{2*}

- 1. Sartorius Stedim Cellca, Erwin-Rentschler-Straße 21, 88471 Laupheim 2. Sartorius Lab Instruments GmbH & Co. KG, Otto-Brenner-Straße 20, 37079 Goettingen
- * Correspondence:

E-Mail: michael.grauf@sartorius.com

Abstract

Clarification of cell culture supernatants with volumes of $< 25 \, \text{mL}$ for harvesting monoclonal antibodies by using syringe filters is often a laborious and, sometimes, an exhausting step. Therefore, selection of a suitable filter type is essential. In this study, we compared the performance characteristics of two suppliers' syringe filter types, each with a similar effective filtration area, for clarification of CHO cell culture supernatant samples. To obtain robust results, we examined ten combinations of cultivation methods and monoclonal antibody products, such as IgG1, IgG2, fc fusion proteins and bispecific antibodies, with regard to turbidity, mAb recovery, relative yield and throughput. As a result, we found that syringe filter type Minisart® High Flow shows an average throughput of 18.0 mL compared with 9.3 mL of Acrodisc® at cell densities between $38.3 \times 105 \, \text{cells/mL}$ and $163.6 \times 105 \, \text{cells/mL}$. For the other parameters, we did not find any significant differences. This finding emphasizes the importance of carefully selecting the syringe filter type to reduce the number of devices needed and thus workload.

Find out more: www.sartorius.com

SARTURIUS

April 9, 2018

Keywords or phrases:

CHO Cell Culture, mAb, Clarification, Supernatant, mammalian cell culture, clarification, diatomaceous earth, affinity chromatography

Example 3:

Minimizing Syringe Filter Usage in Harvesting Monoclonal Antibodies from CHO Cell Culture Supernatants

Dennis Karthaus*, Sarah Ludwig, Isabel Schuchardt, Stefanie Krame IBA GmbH, Rudolf-Wissell-Str. 28, 37079 Göttingen, Germany www.iba-lifesciences.com

* Correspondence: E-Mail: karthaus@iba-lifesciences.com

Abstract

In this study, the novel Sartoclear Dynamics® Lab V Kit was evaluated for the removal of transiently IgG expressing mammalian MEXi-293E (HEK293) cells from cell cultures. The method was directly compared to the present standard method that required two centrifugation steps. After clarification, recombinant IgG harboring a Twin-Strep-tag® was purified from all samples in parallel by a one-step Strep-Tactin®XT Superflow® high capacity affinity purification process.

Overall, the use of Sartoclear Dynamics® Lab significantly reduced the time for sample clarification by up to 3.6-fold while maintaining total protein yield and quality. Moreover, Sartoclear Dynamics® Lab can be integrated easily into already existing lab processes, substantially decreasing hands-on time and thereby simplifying sample preparation

Find out more: www.sartorius.com/sartoclear-dynamics-lab

Complete List of Application Notes - Page 1/4

The complete list of available Sartorius Lab Filtration application notes and technical guides available to our customers. Please visit www.sartorius.com to download, or contact Sartorius directly to request a free PDF copy

Application notes and technical guides are separated by market workflow category.

Proteins at Low Sample Volumes

Title	Brief Description
Centrifuge-Free Clarification of Antibodies from Cell Cultures Using Sartoclear Dynamics® Lab Decimates Working Time: A Comparative Study	This study focuses on the clarification of 50 mL suspension adapted HEK-293-6E cells, transiently expressing human IgG4 by comparing a conventional harvest method and the new Sartoclear Dynamics® Lab method.
From Mammalian Cell Cultures to Pure Proteins: Sartoclear Dynamics® Lab Significantly Reduces Cell Harvest Time	In this study, the novel Sartoclear Dynamics® Lab V Kit was evaluated for the removal of transiently IgG expressing mammalian MEXi-293E (HEK293) cells from cell cultures.
Reducing sample preparation time from <i>Sf</i> 9 insect cultures by using Sartoclear Dynamics® Lab	In this study, the Sartoclear Dynamics® Lab V500 Kit was tested as a novel method for the clarification of cell culture media prior to purification of a recombinant protein expressed in St. 9 cells.
Rapid Mammalian Cell Harvest without Centrifugation for Antibody Purification Using the Sartoclear Dynamics® Lab Filtration System	In this study, we describe our tests of Sartoclear Dynamics® Lab V, a novel system for rapid clarification of cell culture media without the need for centrifugation or any other costly equipment.
Lab-Scale Clarification of Mammalian Suspension Cultures Using Sartoclear Dynamics® Lab V Kits	For clarification of large-volume cell cultures, Sartoclear Dynamics® Lab V Kits are an attractive addition to standard lab instrumentation to increase productivity and throughput prior to protein purification.
How to Increase Recovery at Critical Protein Samples: Impact of Syringe Filter Membrane, Volume and pH	In a design of experiments approach we quantified the recovery of four model proteins under different sample conditions and found that membranes composed of cellulose acetate or polyethersulfone adsorbed on average less than 5% of protein analyte.
Vivaflow® 200: A Critical Sample Preparation Tool for Concentrating Hybridoma Supernatants	This study focusses on the use of Vivaflow® 200 crossflow cassettes to concentrate up to 3 L clear murine hybridoma supernatant 10-fold prior to affinity chromatography.
Scouting Protein Purification Conditions Using Vivapure® Centrifugal Ion Exchange Membrane Absorbers	In this study, a scouting procedure is described where optimal purification conditions for SH2 domain were developed using Vivapure® IEX centrifugal devices.
Purification Polishing of His-tagged Proteins using Vivapure® prior to Crystallization	A method for scouting of ion exchange chromatography (IEC) purification conditions is described, using Vivapure® membrane adsorbers.
Concentration of Low Molecular Weight Peptides with Vivaspin® 500 Ultrafiltration Devices	In this application note, we have used a 3 kDa MWCO PES membrane incorporated into Vivaspin® 500 ultrafiltration devices to concentrate two peptides.
Concentration of an 8 kDa Peptide Fragment with Vivaspin® 6 Ultrafiltration Devices	A process using passivated Vivaspin® 6 devices is described for the concentration of a low molecular weight peptide. This method ensured high recoveries and informed the need for an additional purification step prior to downstream analysis.
Minimizing Syringe Filter Usage in Harvesting Monoclonal Antibodies from CHO Cell Culture Supernatants	In this study, we compared the performance characteristics of two suppliers' syringe filter types, each with a similar effective filtration area, for clarification of CHO cell culture supernatant samples.
Combining Immuno-Affinity Chromatography and Filtration to Improve Specificity and Size Distribution of Exosome-Containing EV Populations	Together with IBA Lifesciences, Sartorius has devised a novel method to isolate and enrich small extracellular vesicles (EVs). This two-step process, utilizing immuno-affinity chromatography and filtration with Minisart® syringe filters, avoids the impurities which typically remain after conventional EV purification techniques.
Vivaflow® and Vivaspin® Workflow in Protein Research Laboratories	In this Application Note, we demonstrate how the Vivaflow® cassettes, Vivapure® Ion Exchange spin columns and Vivaspin® devices can be used to perform a complete protein purification workflow, from concentration and diafiltration of the original protein source, a cell culture supernatant, to final concentration desalting of the purified protein.
Impact of the Claristep® Filtration System on Recovery and Adsorption of Various Therapeutic	We tested the novel filter device Claristep® for the preparation of protein samples containing one of four different target molecules, i.e. an anti-malaria vaccine candidate, aviscuminum, interferent alfa-2B or moneclo

interferon alfa-2B or monoclonal antibody (mAb) P2G12.

Complete List of Application Notes - Page 2/4

Application Area: Life Science Research (
Title	Brief Description
High Recovery of Cationized Protein using Vivaspin® Filtrate	In comparison to conventional techniques such as lyophilization, ultrafiltration shows increased processing speeds and high cationized protein recovery.
Lab-Scale Harvest and Clarification of Lentiviral Vectors Using Vacuum Filtration Units	In this study, Sartolab® RF50 and Sartoclear Dynamics® Lab V50 (Sartorius), as well as other manufacturers' filters, named products A, B, and C, were evaluated for clarification of lentiviral vectors produced by transient transfection of HEK293T/17 SF suspension cells.
Lab-Scale Harvest and Clarification of Lentiviral Vectors Using Sartoclear Dynamics® Lab Filtration Systems	In this study, the use of Sartoclear Dynamics® Lab V50 (Sartorius) comprising the diatomaceous earth (DE) filter aid was evaluated for clarification of lentiviral vectors (LV) produced by HEK293 suspension cells.
Using Ultrafiltration with Vivaspin® 500 as an Alternative to Sample Volume Reduction by Lyophilization	The recombinant mutated lectin purified through Ni-NTA (nickel nitrilotriacetic acid) column was concentrated using Vivaspin® 500 in parallel to lyophilization.
Removal of Endotoxin from Monoclonal Antibodies using Vivapure® Q Membrane Adsorbers	Using anion exchange devices in optimal conditions for the removal of endotoxins from research grade monoclonal antibodies.
Efficient Purification of Adult Rat Brain and Chick Embryo Membrane Proteins Using Centrifugal Vivapure® D and Vivaspin® Devices	A workflow featuring Vivapure® ion exchange spin columns and Vivaspin® centrifugal concentrators is presented for the purification of two membrane proteins.
Protein Complex Concentration with Vivaspin® Prior to Fourier Transform Infrared (FTIR) Spectroscopy	The ability to generate high quality strucutral data is typically dependent on using a protein sample which is high in both concentration and purity. Vivaspin® offers both fast processing speeds and high concentration factors, while also enabling the removal of sample components which would be detrimental to FTIR.
Vivaspin® Ultrafiltration Devices: Tools for Concentrating His-Tagged Proteins Eluted in High Imidazole Following Affinity Chromatography	The high levels of imidazole used for elution during purification of his-tag proteins by metal affinity chromatography can present challenges for concentration and the accuracy of quantitative measurements. Vivaspin® devices were evaluated to show their effectiveness for concentrating and re-buffering such samples.
Vivapure® S Ion Exchange Membrane Adsorbers For the Rapid Fractionation of Peroxidases from Plant Calluses	The spin column format and high binding capacities of Vivapure® ion exchange devices are ideal for rapid screening, since multiple low-volume samples can be purified in parallel, with minimal equipment requirements.
Rapid DNA Removal from Mammalian Cell Lysates by Ion Exchange Purification with Vivapure®	$\label{thm:purity} Vivapure ^{@}\ ion\ exchange\ spin\ columns\ can\ efficiently\ remove\ DNA\ without\ compromising\ the\ purity\ or\ integrity\ of\ cell\ lysate\ samples.$
Concentration of Soluble and Membrane Proteins by Pressurized Ultrafiltration with Vivaspin® 100	Vivaspin® 100 is presented for its unique ability to process sensitive samples - such as membrane proteins - under positive pressure, and capacity to handle sample volumes in the range of $20 - 100 \text{ mL}$.
Vivaspin® 20 Diafiltration Cups: A Rapid Alternative to Buffer Exchange by Dialysis	To assess the effectiveness and performance of diafiltration in comparison to the traditional dialysis approach, Vivaspin® 20 products were used in parallel to conventional laboratory dialysis frames.
Concentration of Mammalian Cell Culture Supernatants with Vivaspin® Turbo 15 PES.	Here we benchmark two different membranes and MWCO devices for the concentration of a molecule of interest, TFF1, to demonstrate the resulting differences in yield percentage recovery.
Does Syringe Filtration Impact Recovery of Cannabinoids Prior to Potency Testing?	The objective of this study was to determine if using syringe filters (Sartorius Minisart® RC, SRP, NY and NY Plus, all 0.2 µm) either released interfering compounds or retained cannabinoids during sample preparation for HPLC testing.

Complete List of Application Notes - Page 3/4

Application Area: Food & Beverage				
Analyzing Drinking Water by ICP-MS for Heavy Metal Detection	A basic step in drinking water analysis is preparation of samples during which undissolved constituents are removed from each sample by filtration through a 0.45 μm filter.			
Ergonomic and Efficient Filtration for Food Analytics	Fourteen commercially available food samples – among them very particle-rich dispersions were filtered at various levels of dilution with the syringeless Claristep® 0.45 μm and 0.2 μm pore size filters and the manual Claristep® base.			
Application Area: Medical Research & Di	agnostics			
Title	Brief Description			
Concentration and Purification of Viruses by using Ultrafiltration, Incl. Coronavirus – a Short Review	Investigations into general Coronavirus virology, Coronavirus spike proteins, capture of Coronavirus virons and free RNA from water streams, are three core examples of focus point for study in the scientific community, as such we provide some examples of concentration and purification steps in these Coronavirus research workflows also.			
Sartorius Ultrafiltration Products in the Preparation of Biological Nanoparticles and Medical Nanocarriers - a Short Review	Here, ultrafiltration is used to purify, concentrate and separate nanoparticles from substrate			
An Improved Method to Wash Graphene Prior to Use as a Drug Delivery Vehicle	An improved method for washing graphene oxide dispersions to rapidly neutralize pH levels is described using tangential flow filtration (TFF) with the Vivaflow® 50, 100 kDa MWCO.			
Free Label Removal with Vivaspin® 2 Prior to Radioimmunoassay	We used Vivaspin® 2 ultrafiltration devices with a 50 kDa MWCO PES membrane to separate an antigen-antibody complex from unbound tracer.			
Ethylene Oxide Treatment as a Method for Introducing PCR Grade Vivacon® Centrifugal Concentrators	This study shows that an EtO treatment regime is capable of reducing DNA contamination, by preventing amplification of contaminant extraneous DNA, such that database loadable contaminant DNA profiles should not be possible after gas sterilization.			
Urine Protein Concentration with Vivaspin®	Procedure for the concentration and separation of protein disease markers from urine, using centrifugal ultrafiltration devices.			
Vivaspin® Turbo 4 PES: An Economic Approach to Separate Disease Metabolites and Proteins	Here we demonstrate ultrafiltration with Vivaspin® Turbo 4 PES centrifugal devices as a high effective method for the rapid and precise separation of biomolecules from blood serum, using NMR and off-gel fractionation to evaluate.			
De-lipidation and De-proteinization of Hyperlipidaemic Serum Samples using Vivaspin®	Serum lipids and lipoproteins are important clinically, because hyperlipidemia is a risk factor for disease. To permit the study of low molecular weight species in serum samples, a two-ste ultrafiltration process using Vivaspin® to remove lipids and larger proteins is presented.			
Application Area: Hints and Tips				
Desalting and Buffer Exchange with Vivaspin® Centrifugal Concentrators	This short application note highlights the ability to reduce protein sample salt concentration by up to 99%, or to exchange the buffer sample entirely, using Vivaspin® 20 and Vivaspin® 6 centrifugal ultrafiltration devices.			
Treatment of Vivaspin® Concentrators for Improved Recovery of Proteins at Low Initial Concentration	Here we describe passivation techniques that demonstrate increased recovery when used with low starting concentration samples.			
Concentration to a Defined Final Volume with Vivaspin® Turbo 15 PES, Vivaspin® Turbo 4 PES and Vivaspin® 500 PES	This short Application Note describes how you can use Vivaspin® Turbo 15 PES, Vivaspin® Turbo 4 PES and Vivaspin® 500 PES to concentrate samples to defined final volumes.			
Comparative Study of Resin- and Membrane- Based Ion Exchange Technologies for Protein Purification	The performance of conventional DEAE resins is compared with Vivapure® D spin columns. Vivapure® benefits from lower cost and higher yields.			
Application Area: Environmental				
In-line Filtration of Groundwater Samples for Heavy Metal Analysis with Sartolab® P20 Plus	The Sartolab® P20 Plus in-line filter presented in this paper was compared with syringe filter with regard to its suitability based on throughput and particle reduction.			

Complete List of Application Notes - Page 4/4

Application Area: Environmental (continued)		
The Measurement of Soluble (< 10 kDa) Trace Metals in seawater by the Vivaflow® 50	This preliminary data suggest that the Vivaflow® 50 is a fast, convenient and accurate way to separate soluble and colloidal metal species in seawater.	
Application Area: Applied Research		
Filter Aided Sample Preparation (FASP) with Vivacon® 500	The effectiveness of different filtration devices for analysis of proteomes and glycoproteome was compared.	
Using Vivacon® 500 for Primer Removal after PCR	Here, we show in an experiment the effective removal of primers using Vivacon® 500 ultrafiltration devices and show that the 30 kDa Hydrosart® membrane is effective at retaining 300 bp DNA fragments, while removing the 25 bp primers.	
Using Vivacon® 2 for Primer Removal after PCR	Using a 30 kDa Vivacon® 2 device, primers and PCR reaction components can effectively be removed from a PCR sample containing 300 bp and larger DNA fragments for subsequent applications.	
Depyrogenation of Vivaspin® Turbo 15 PES in Comparison to Ultrafiltration Devices With a Regenerated Cellulose Membrane	Here we demonstrate low endotoxin concentrations within the Vivaspin® Turbo 15 PES devices and show further successful depyrogenation using sodium hydroxide.	
Application Area: Technical Guides		
Title	Brief Description	
Laboratory Ultrafiltration How to Choose the Optimal Device & Method	Selection Guide for proteins, viruses, DNA, polymers, nanoparticles, exosomes, etc.	
Laboratory Ultrafiltration FAQ	Questions about Ultrafiltration Devices and various applications are answered in this FAQ.	
Syringe Filters — Sartorius Minisart® Selection Guide: How to Choose the Optimal Housing and Membrane Material for Your Application	How to Choose the Optimal Housing and Membrane Material for Your Application.	
Effectively Use Picus® Pipettes with Vivaspin® Concentrators	In this practical guide, we provide guidance on the efficient use of Picus® pipettes and extended length pipette tips with centrifugal concentrators.	
How to Choose: Sartoclear® Dynamics Lab	An eBook to help you identify and choose the best combination of vacuum filtration unit and diatomaceous earth filter aid for your sample.	
Sartolab® Multistation: How to Filter Multiple Samples in Parallel	A guide to combining vacuum filtration units with a vacuum manifold for handsfree, rapid filtration of multiple samples.	
Total Cost of Ownership (TCO) Analysis: Sartoclear Dynamics® Lab	Case study analysis of the ownership and running cost comparisons of filter aid versus centrifugation for cell culture clarification.	
Lab Ultrafiltration Tips and Tricks	Build knowledge on desalting samples, concentrating samples and recovery of molecule targets with a selection of dedicated application guides.	
Technical Cleanliness Guide	Complete guide on the analysis of particles from clean manufacturing parts and component using filtration and gravimetric sample prep and testing.	

Application Partnerships - Simplify Progress With Us

Despite the wide-ranging application support, there are many more new and emerging applications at the forefront of science to investigate.

Sartorius is interested in cooperating with scientists on the front line of science, to optimize protocols and develop application knowledge.

There are many ways to partner and exchange with us:

1	Development Partnerships	Influencing and supporting the development of new products and technologies
2	Application Partnerships	Sharing working protocols and data to optimize and improve procedures
3	Customer Interviews	Exchange of thoughts and ideas on how to improve processes and products
4	Market Surveys	Complete simple surveys to provide your input into key areas of importance for Sartorius, often with prize draws!
5	Application Specialist Meetings	Onsite or virtual meetings to discuss application optimization with existing products

If interested in any of the above or you would like more information, please contact Sartorius Lab Filtration Product Management at LabFiltrationPM@sartorius.com and/or reach out to your local Sartorius representative office with a request to contact Sartorius Lab Filtration Product Management.



Germany

Sartorius Lab Instruments GmbH & Co. KG Otto-Brenner-Strasse 20 37079 Goettingen Phone +49 551 308 0

USA

Sartorius Corporation 565 Johnson Avenue Bohemia, NY 11716 Phone +1 631 254 4249 Toll-free +1 800 635 2906

