TangenX® SC Single-Use vs Multi-Use Strategies

Application Note

Introduction

The objective of this application note is to present data supporting the option of both single-use and multi-use strategies with the new TangenX SC Tangential Flow Filtration Device (Figure 1). The following application note outlines several multi-use studies performed at the Repligen manufacturing facility located in Marlborough, Massachusetts. Repligen is a bioprocessing-focused life sciences company bringing expertise and innovation to our customers since 1981. We are inspiring advances in bioprocessing through the development and commercialization of high-value products and flexible solutions that address critical steps in the production of biologic drugs.

The Repligen TangenX line of tangential flow filters is ideally suited to any post-use strategy. The TangenX SIUS product family is a purposefully built line of TFF Cassettes specifically designed for single-use operations and provides many advantages over traditional TFF cassettes. Where conventional reusable cassettes are required, the PRO line of reusable filters is available. Each of the conventional TFF Cassettes require the use of a stainless-steel holder that serves as a clamping mechanism. The holder contributes to higher capital costs and increases the chances of cassette integrity failure if not tightened properly by the end-user.

To address several of these operational challenges, Repligen now offers the TangenX SC, a gamma irradiated, self-contained TFF assembly that eliminates the use of a stainless-steel holder. The self-contained ultrafiltration assembly consists of a membrane cassette housed in a shell with aseptically sealed inlet/outlet ports.

Product performance is comparable to existing TangenX TFF Flat Sheet Membrane Cassettes manufactured by Repligen. TangenX SC TFF Devices are designed to be cross-scalable between the reusable (PRO) and single-use (SIUS) product lines and are available in surface areas from 0.5 m^2 to 10 m^2. The TangenX SC TFF Device can be used both as single-use or in multi-use applications.

TangenX SC TFF Device: Revolutionize TFF Processes

Figure 1. TangenX SC TFF Device Lineup

Background

The two most common post-use strategies for tangential flow filters are single-use and reuse. The single-use strategy uses the device for one discrete process operation before disposal. No time or resources need to be dedicated to the development of a cleaning strategy or to its implementation, and the risk of cross-contamination is minimized. Reuse of the TFF device lowers the filter cost per batch but requires that the filter is cleaned following each batch processed, necessitating the development of a robust and effective cleaning cycle. More recently, multi-use has emerged as a third strategy and fits between single-use and reuse. Multi-use operations use the same filter over the course of a campaign and require fewer cleaning cycles than a traditional reuse strategy. The table below summarizes these three operational strategies.

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Table 1. Operational Strategy Descriptions

Operational Strategy	Number of Uses	Characteristics
Single-use	1 Cycle	Saves time, reduces resources, minimizes cross-contamination
Reuse	> 10 Cycles	Lowers filter cost per batch, requires cleaning cycle
Multi-use	≤ 10 Cycles	Lowers filter cost per campaign, requires cleaning cycle

Several regulatory authorities permit campaign manufacturing for some categories of pharmaceutical products after an appropriate risk assessment has been performed. Standardized, validated procedures are required to ensure quality standards are maintained and cross-contamination risks are minimized. A multi-use operational strategy, a variation on reuse, limits the number of cycles to a single campaign of biological fluid. Repligen has shown that TangenX SC TFF Device can be effectively cleaned and reused for 10 cycles without compromising performance or integrity, and, therefore, supports up to ten (10) multi-use cycles per campaign or where limited production runs are anticipated.

Case Study

The TangenX SC TFF Device is ideally suited for single-use applications; however, because data supporting multi-use applications is desirable, this case study has been performed to subject the TangenX SC TFF Device to the rigors of multiple cleaning cycles. Nine TangenX SC TFF Devices were used to test the performance and robustness of the device following repeated cleaning cycles. Devices ranging from 0.5 m^2 to 10 m^2 were evaluated using the following parameters:

1. Cross flow rate vs. pressure-drop using water: to demonstrate the feed channel is not compressed causing a reduction in crossflow rate.
2. Air integrity testing: to demonstrate the device is integral following repeated cleaning cycles.
3. High pressure operation: to demonstrate the maximum operating pressure is not compromised by the cleaning cycles.

Table 2. TangenX SC TFF Devices

Quantity	Part Number	Area	Description
3	PP010S05L	0.5 m^2	10 kD, TangenX SC TFF Device
3	XP030S25L	2.5 m^2	30 kD, TangenX SC TFF Device
3	XP030S99L	10 m^2	30 kD, TangenX SC TFF Device

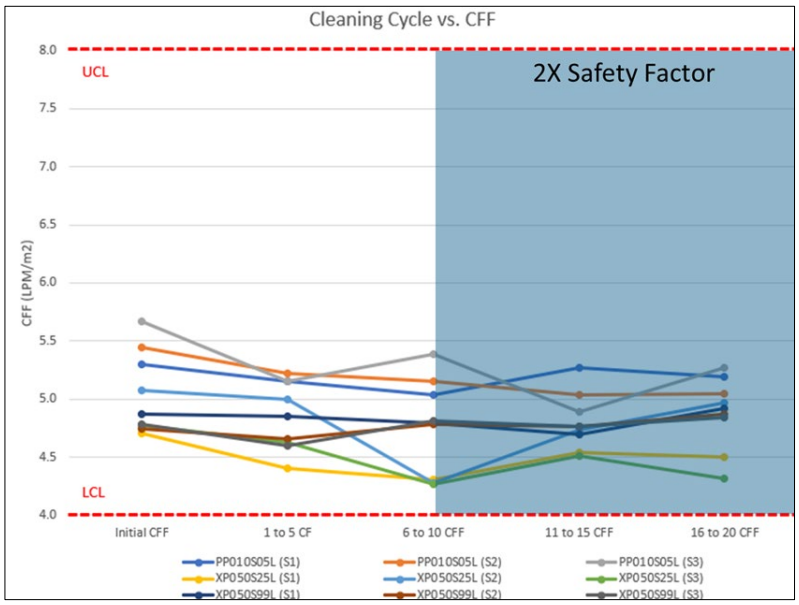
Following the repetitive cleaning study, simulated processes with model proteins were used to demonstrate the TangenX SC TFF Device continued to perform as intended. Devices were evaluated for the following parameters:

1. Flux rate: to confirm the cleaning cycles were effective at recovering the membrane permeability.
2. Molecular weight marker retention: to demonstrate the membrane retention did not shift following repeated cleaning cycles.
3. A 10-fold concentration experiment with bovine serum albumin: to compare a new device with a device that had been subjected to multiple cleaning cycles.

Initially, each device was connected to a TFF system, flushed with purified water, and evaluated for baseline performance using crossflow flux (CFF) and air integrity testing. These tests are also release tests for the product, demonstrating different aspects of performance. Next, each device was subjected to repeated and subsequent two-hour cleaning cycles with 0.5M NaOH at 40°C. The device was flushed with purified water following each cleaning cycle and evaluated for performance using water. Crossflow rate vs. pressure-drop was measured to demonstrate the feed channel was not compressed, which would cause a reduction in crossflow rate. A significant reduction in crossflow rate creates higher inlet pressure when operating at a specified flow rate. If the pressure

drop becomes too great, the predetermined process parameters for the operation will not be maintained. The pressure drop over 20 cleaning cycles is shown in Figure 2.

Figure 2. Crossflow Rate vs Pressure Drop



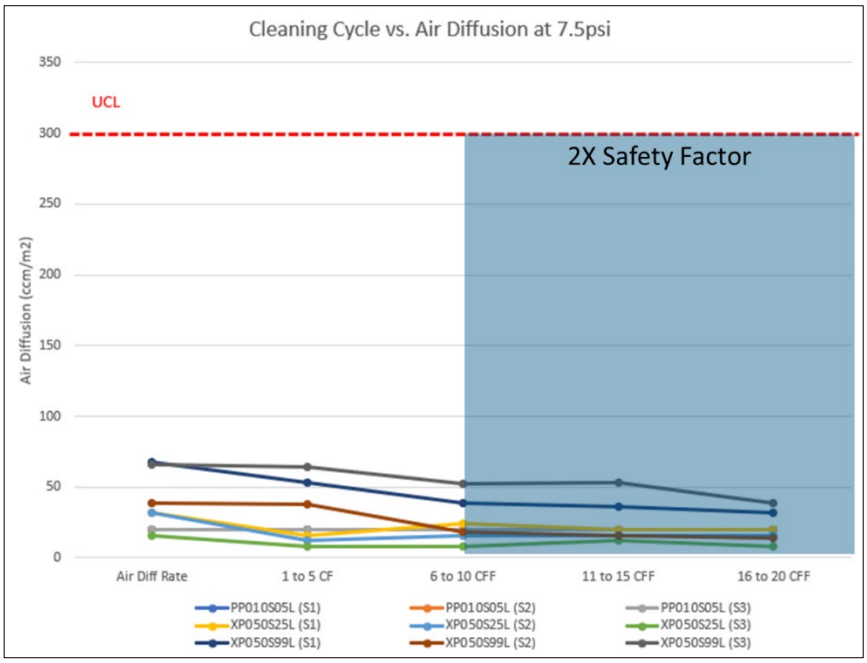
Analysis of the data shows the average pressure drop averaged less than 5% over the entire study (Table 3). Data points remained above the lower control limit of 4.0 LPM/m^2 and stabilized after the tenth cleaning cycle. The study shows the crossflow rate at a ΔP of 10 psi remained within specification with a minimal change in the channel height that would not impact performance.

Table 3. Pressure Average Pressure Drop Change

	Initial CFF	1 to 5 CF	6 to 10 CFF	11 to 15 CFF	16 to 20 CFF
Average	5.0	4.9	4.8	4.8	4.9
% Change	---	3.8%	5.7%	4.8%	3.2%

Air integrity testing was performed to demonstrate the device was integral following repeated cleaning (Figure 3). Air integrity testing is a non-destructive method used to assure the user that the device will meet the criteria of removal under targeted process conditions. Integrity testing for ultrafiltration cassettes can identify both membrane defects as well as structural cassette defects. Air diffusion is a quantitative test that measures the rate of air diffusing through the wetted membrane at 7.5 psi (0.5 bar). The TangenX SC TFF Device was flushed with purified water. A mass flow meter was used to measure the rate of the air diffusing through the wetted membrane. The study shows the air integrity at 7.5 psi (0.5 bar) met specification throughout the 20-cycle cleaning study. Meeting the air integrity specification ensures the device is free of defects and will function as intended, not allowing bypass of the feed to the permeate. Figure 3 demonstrates the air diffusion rate remains below the upper control limit following each of the cleaning cycles.

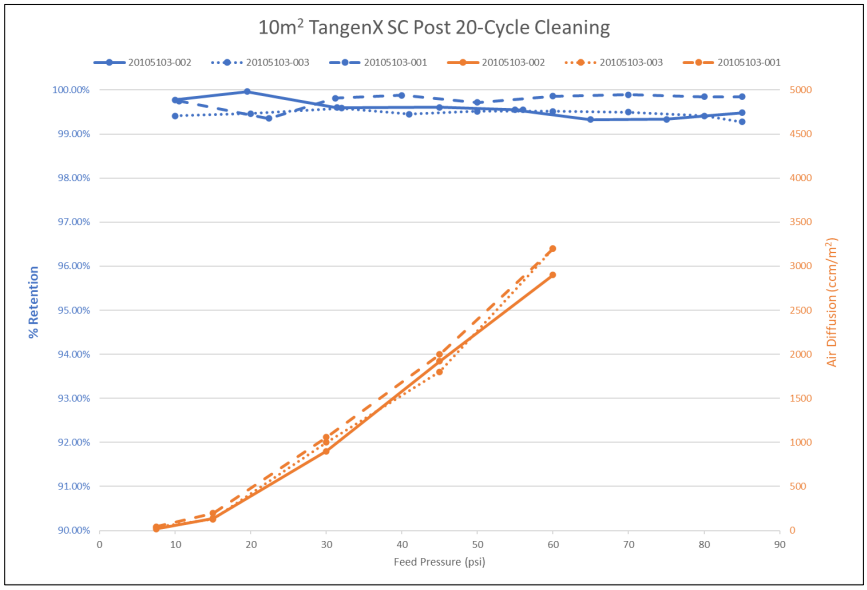
Figure 3. Air Integrity Testing



A high-pressure test was performed to demonstrate that the maximum operating pressure of the device was not compromised. The 10 m^2 devices are impacted the most by cassette compression and will experience the greatest reduction in compressive force due to stack height. Without adequate compression within the device, the feed will be allowed to bypass to the permeate and the product retention rate will drop. A demonstration of marker retention after 20 cleaning cycles was performed. This testing challenged the device with a retaining test marker up to 90 psi (6 bar), 50% higher than the maximum specified operating pressure of 60 psi (4 bar). Additionally, the air diffusion rate was measured up to 60 psi (4 bar) to confirm the device was integral.

Devices are integral up to the maximum operating pressure of 60 psi (4 bar), with a 1.5X safety factor demonstrating robust performance with no bypass of the feed to the filtrate. Similarly, 2.5 m^2 and 0.5 m^2 devices were evaluated in a similar manner with the same result showing no sign of bypass up to 90 psi. Figure 4 shows percent retention of the molecular weight marker as the operating pressure was increased in 20 psi increments. Retention remained constant demonstrating the devices remained integral beyond the maximum recommended operating pressure of 60 psi after being subjected to 20 repeated cleaning cycles.

Figure 4. High Pressure Testing

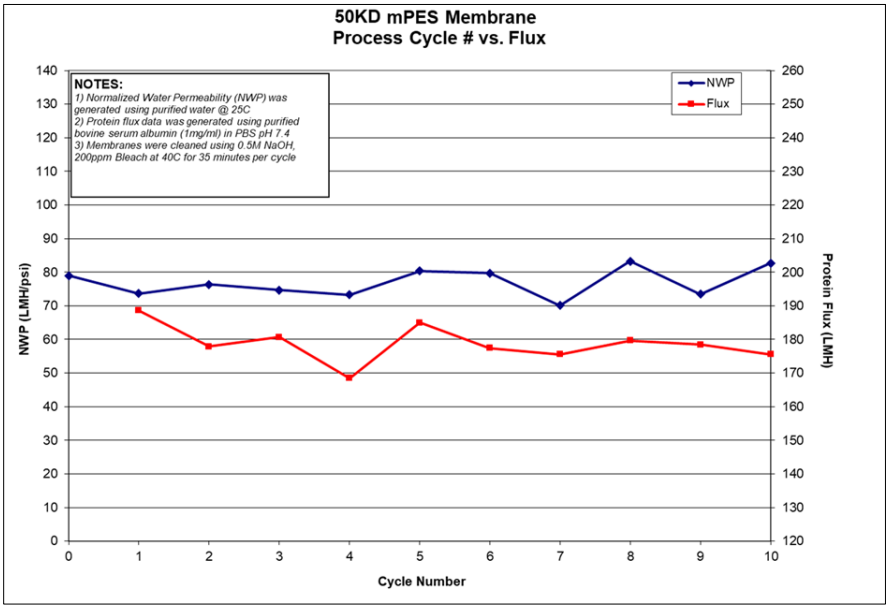


The air diffusion rate confirmed the devices were integral at 7.5 psi (0.5 bar) and increased proportionately as the pressure was increased up to 60 psi (4 bar). In the event of a seal bypass or integrity failure, the slope of the curve for air diffusion would increase sharply at the point of failure. No such change in slope was noted, and data confirm the devices were integral following 20 cleaning cycles. Based on these data, the number reuse cycles should be limited to ten (10) cycles, accounting for a conservative safety factor.

The innovative modified polyether sulfone (mPES) membranes used in the TangenX SC TFF Device can withstand multiple cleaning cycles without a shift in performance characteristics. To be of practical use following each reuse cycle, the performance of the membrane in the TangenX SC TFF Device must be recovered both in terms of water permeability, process flux, and retention. Flux rates measured following simulated processing operations confirmed the cleaning cycles were effective at recovering the membrane permeability.

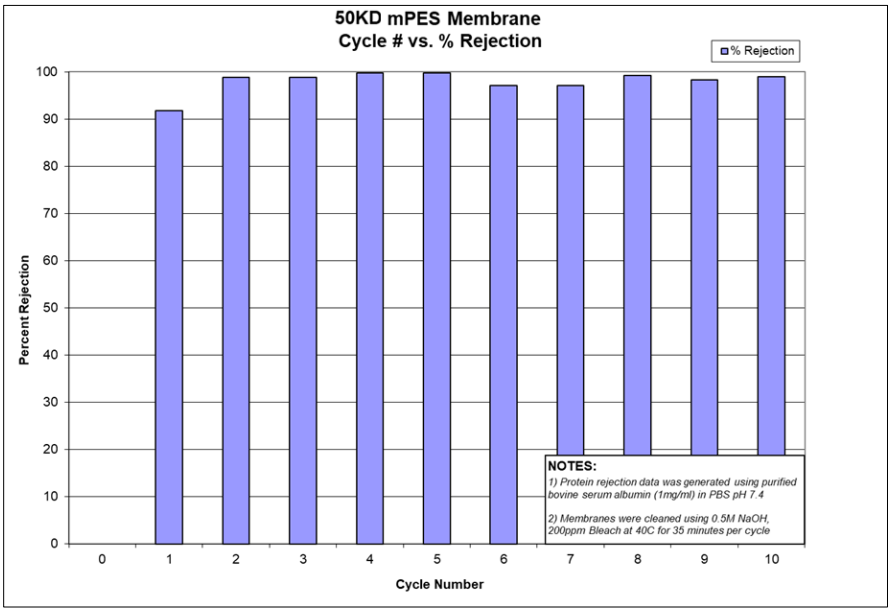
Membrane permeability is a key indicator of the effectiveness of the cleaning cycle used on a TFF device. To demonstrate this, a TangenX 50 kD mPES membrane was challenged with 1 mg/ml bovine serum albumin (BSA) solution for a concentration operation. The clean water flux was evaluated and recorded as a baseline measurement. Next, the membrane was used to concentrate 1mg/ml BSA and cleaned using 0.5N NaOH, 200 ppm Bleach at 40°C for 35 minutes. Following cleaning, the water flux was measured again and compared to the initial water flux, where recovery of greater than 90% was established as a target. Following this cycle, the membrane was challenged and cleaned nine more times, for a total of ten cycles. Figure 5 demonstrates that after 10 cycles (or 6 hours of exposure) with the cleaning solution, the membrane consistently shows greater than 90 percent water flux recovery. Additionally, the graph shows that protein flux remains consistent from cycle to cycle as well. The data support that the mPES membranes used in TangenX SC TFF Devices can maintain initial performance following each cleaning cycle and can be consistently recovered without a change in water permeability or process flux.

Figure 5. Process vs Flux Rates



Molecular weight marker retention for the membrane was also measured to confirm the pore size had not shifted following repeated cleaning cycles. A demonstration of membrane retention of BSA over 10 cleaning cycles was performed. Figure 6 shows the rejection of the membrane is equal to (or greater than) the initial rejection of a new membrane. The data support that the TangenX membrane has good chemical resistance to aggressive cleaning processes and can be consistently recovered without a significant change in retention.

Figure 6. Molecular Weight Marker Retention



As a final test, a 10-fold concentration experiment with bovine serum albumin was conducted on both a new SIUS Cassette and a TangenX SC TFF Device that had been subjected to multiple cleaning cycles. A 5 g/L BSA solution was concentrated ten-fold using a

new 0.1 m^2, 10 kD SIUS Cassette and a cleaned 0.5 m^2, 10 kD TangenX SC TFF Device. The SIUS Cassette was evaluated using a KrosFlo KR2i TFF System and the TangenX SC TFF Device was evaluated using the KrosFlo FS-15 System (Figure 7).

Figure 7. KrosFlo FS 15 System

Each cassette was flushed with purified water and equilibrated in PBS buffer. The buffer was drained from the system and replaced with the BSA solution. Total loading was 200 g/m^2 for each cassette. The solution was concentrated ten-fold while maintaining a crossflow flux of 6 LPM/m^2 and a transmembrane pressure of 15 psi. Critical process parameters such as pressure, flow rate, and volume were recorded throughout the process. Figure 8 shows the relationship between concentration factor and flux for both cassettes. The flux rate, instantaneous retention, and percent recovery of the BSA solution were comparable. The average percent retention and final percent recovery were also measured using UV/Vis at 280 nm. The results (Table 4) demonstrate that both retention and recovery of the devices are comparable. These data also show that repeated cleaning cycles do not impact the performance or recovery of the SC Device when compared to a new SIUS Cassette.

Figure 8. Flux vs CF Results

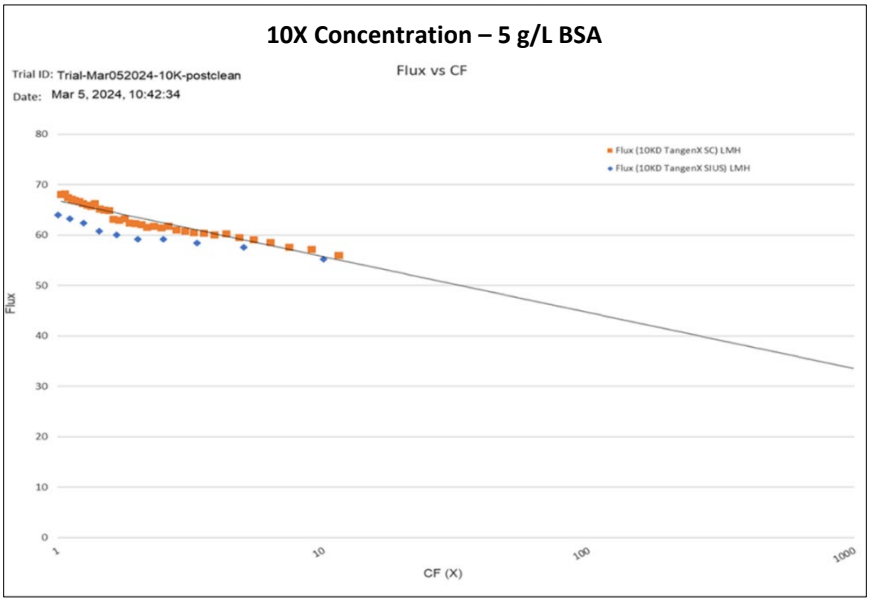


Table 4. Device Flux, Retention and Recovery

Description	Average Flux	% Retention	% Recovery
SIUS	60 LMH	97.8 %	105 %
TangenX SC TFF Device	62 LMH	99.2 %	106 %

These data not only confirm the TangenX SC TFF Device performance has not been changed, but also demonstrate the cross-scalability between all TangenX TFF Cassette products. These attributes allow processes to be developed on any one of the TangenX Cassette products and scaled up to a larger surface area or to another format that suits the application.

Developing a Cleaning Strategy

Cleaning and cleaning validation is a key priority when using conventional reusable TFF cassettes. A rigorous CIP (clean-in-place) cycle is used to remove residual contaminants from the TFF cassettes following each use. A validated membrane CIP cycle is used to maximize:

- Product safety
- Product recovery
- Batch to batch consistency
- Membrane NWP recovery
- Membrane productivity

Cleaning is important because it affects the safety of the drug product with regard to contamination risk. The goal is to maintain a clean and sanitary system by removing residual proteins and microorganisms. Cleaning can also impact the consistency and performance of the TFF process where the objective is to maintain efficient productivity by counteracting the effects of membrane fouling. Cleaning validation is not only good business practice, but also prevents batch failures, improves product consistency, improves patient safety, and supports regulatory agency requirements. Cleaning validation studies provide documented evidence that the cleaning methods used are effective and will not impact product identity, safety, purity, or potency.

Effective and consistent cleaning procedures require users to identify an effective cleaning agent for the mode of cleaning. Hydrolysis, oxidation, solubilization, emulsification, and digestion are chemical methods used for effective membrane cleaning. The ideal mode of cleaning is dependent on the composition of the foulants on the membrane surface and varies with the type of feed streams processed. Process conditions including contact time, temperature, volume, crossflow rate, and water quality impact the effectiveness of the cleaning cycle. Likewise, operational procedures such as pre-flushing of the foulant and post-flushing of the CIP agent also need to be considered in the validation process.

Physical tubing connections to the TangenX SC Device must also be considered as part of the clean-in-place (CIP) strategy. The TangenX SC Device is supplied with AseptiQuik® fittings from CPC (Colder Products Company) and are designed to be permanently connected to a mating fitting. This ensures the flow path remains closed to the environment and sterility is maintained. In the case where the SC Device will be re-used, there are two viable options that are available once the process is finished, and the CIP is complete.

1. The SC device remains connected to the flow path where it is cleaned in place and the flow path is also reused.
2. Sanitary genderless AseptiQuik adapters (*sold separately*) are used between the TangenX SC Device and the flow path. The Tri-Clover® connection serves as a point where the connection to the TangenX SC Device can be broken (Figure 9).

Figure 9. Sanitary Genderless Adapter

Conclusion

The objective of this application note was to present data supporting the option of both single-use and multi-use strategies with the new TangenX SC TFF Device. The data presented support multi-use strategies for TangenX SC TFF Device, demonstrating the device is robust and can be effectively cleaned without a loss in performance. Repeated cleaning cycles show the SC device can withstand at least ten (10) cleaning cycles without a significant impact to its crossflow flux or integrity up to its maximum operating pressure of 60 psi (4 bar). Based on these supporting data, Repligen endorses up to and including ten (10) cleaning cycles for the TangenX SC TFF Device in multi-use applications.

References

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