TangenX® SC TFF Device

Regulatory Support File

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Abbreviations

AAMI	Association for the Advancement of Medical Instrumentation
APCI	Atmospheric pressure chemical ionization
B/F	Bacteriostasis/Fungistasis
BHT	butylated hydroxytoluene
BPOG	BioPhorum Operations Group
BSA	bovine serum albumin
BSE	bovine spongiform encephalopathy
ccm	cubic centimeter per minute
CFU	colony forming units
cGMP	current Good Manufacturing Practice
CMC	Chemistry, Manufacturing & Controls
Cyt C	cytochrome c
DI	deionized
DI-GC/MS	direct immersion-gas chromatography/mass spectrometry
ea	each
EDPM	Ethylene Propylene Diene Monomer
ESI	electrospray ionization
EU	endotoxin units
FDA	Food and Drug Administration
ft2	Feet squared (square feet)
GC/MS	gas chromatography/mass spectrometry
GDP	Good Documentation Practice
GMP	Good Manufacturing Practice
HPLC/DAD/MS	High-performance liquid chromatography/photodiode-array detection/mass spectrometry
HS-GC/MS	Headspace-gas chromatography/mass spectrometry
ICP/MS	Inductively Coupled Plasma/Mass Spectrometry
IgG	Immunoglobulin G
in	inch
IQ	Installation Qualification
ISO	International Organization for Standardization
kD	kilodalton
kg	kilogram
kGy	kilogray
L	liter

L/min	liter per minute
lb	pound
LMH	liter per square meter per hour
LPB	low protein binding
LPM	liter per minute
m	meter
M	Molar
m2	meter squared (square meter)
min	minute
mL	milliliters
mPES	modified polyethersulfone
MS	mass spectrometry
MWCO	molecular weight cut-off
N-m	Newton-meter
NMP	N-methyl pyrrolidone
NWP	normalized water permeability
OQ	Operation Qualification
PD	Process Development
ppm	parts per million
PPS	polyphenolsulfone
PQ	Process Qualification
PS	polysulfone
psi	pounds per square inch
psig	pounds per square inch gauge
PVP	Polyvinylpyrrolidone
QA	Quality Assurance
QC	Quality Control
RODI	reverse osmosis deionized water
RSF	Regulatory Support File
SAL	Sterility Assurance Level
SOP	Standard Operating Procedure
TFF	tangential flow filtration
TOC	total organic carbon
TSE	transmissible spongiform encephalopathy
UF	ultrafiltration
USP	US Pharmacopeia
VDmax25	Verifcation Dose maximum 25 kGy
WFI	water for injection
ΔP	delta pressure (pressure drop)
µm	micron

1. Introduction

The Regulatory Support File (RSF) for TangenX® SC TFF Devices is intended to be used as:

- A guide for appropriate application use in process development, clinical, and commercial purification processes.
- A guide to validation in manufacturing processes.
- A support reference for CMC submissions for regulatory license approval.
- A guide for supplier audits.
- An alternative to a Drug Master File submission.

Repligen is committed to providing all relevant technical, manufacturing, and quality information; however, only non-confidential information is presented in this document. Confidential details may be made available upon request through a formal confidentiality agreement or as part of a supplier audit.

A copy of the Repligen quality policy can be found at repligen.com.

1.1 Responsible official

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2. Safety precautions

Table 1. Safety precautions for TangenX SC TFF Devices

Symbol	Description
WARNING 	Damage may occur as a result of the following: • Dropping on hard surfaces, or other mechanical shock • Excessive feed pressure • Excessive permeate backpressure or pressurizing the filtrate port • Exposure to harsh chemicals • Freezing • Excessive heat • Drying out - ultrafiltration membrane that is allowed to dry out can permanently damage the pore structure
WARNING 	Membrane devices must remain wet at all times to maintain product integrity and performance. Keep bag sealed until device installation.
WARNING 	All devices are stored in 0.2 M sodium hydroxide. Follow standard safety procedures for handling 0.2 M sodium hydroxide, including the use of gloves, safety goggles, and lab coat.
Information 	It is recommended that you perform a device integrity test (Section 6.4) and meet specifications (Table 9).
Information 	Devices must be equilibrated with an appropriate buffer to ensure neutralization of the 0.2 M sodium hydroxide storage agent in the membrane filter.
Information 	It is important to pre-filter all liquids to avoid fouling the membrane or introducing contaminants into the system that could affect membrane performance and product recovery.
Information 	Select a pump with adequate capacity. Crossflow rate ranges (Table 6) depend upon feed channel type and process fluid type.

3. Product description

TangenX SC TFF Devices are self-contained, closed, single-use, gamma-irradiated tangential flow filtration (TFF) flat sheet devices used for ultrafiltration and diafiltration processes. TangenX SC TFF Devices consist of a membrane housed in a rigid shell containing aseptically sealed inlet and outlet ports. Unlike other flat sheet TFF cassettes and devices, TangenX SC TFF Devices are self-contained, ready-to-use devices that do not require compression in a stainless-steel holder. The complete device is gamma-irradiated for bioburden reduction and carries a sterile claim. TangenX SC TFF Devices provide a sterile, closed system for applications requiring an assembled TFF device that is aseptically isolated from the environment in a plug-and-play holderless format. The TangenX SC TFF Device is constructed using the same membranes as, and delivers equivalent performance to, other TangenX Cassette products for easy cross-scalability.

Figure 1. TangenX SC Devices

Five different sizes of TangenX SC TFF Devices (Figure 1) are available for pilot and process applications with effective membrane surface areas of 0.5, 1.5, 2.5, 5, and 10 m^2 with a range of membrane pore sizes (10, 30, 50, 100, and 300 kD), two (2) membrane chemistries (ProStream and HyStream modified polyether sulfone (mPES)), and one channel configuration (L-screen). The L-screen is ideal for low to medium viscosity streams where high flux and lower recirculation rates are desired. This wide selection of TangenX SC TFF Devices is designed for processing volumes from tens to thousands of liters. In the event a smaller surface area is required, the TangenX SC TFF Device is cross-scalable to other TangenX SIUS® single-use cassettes and PRO reusable cassettes.

The TangenX SC TFF Device represents the latest development in tangential flow filtration device design and membrane performance. Designed to deliver optimal performance and exceptional batch-to-batch reproducibility, each device undergoes rigorous QA lot release testing to verify specifications are met. Devices are tested for both air integrity and hydrodynamic performance. This testing ensures consistency between devices, scalable process development, and reproducible performance.

3.1 Quality standards

To meet the needs of GMP manufacturing, TangenX SC TFF Devices are manufactured in the United States under the following quality standards:

- TangenX SC TFF Devices are manufactured in a facility whose Quality Management System is approved by an accredited registering body to the ISO 9001 2015 Quality System Standard.
- All fluid paths meet USP <88> Biological Reactivity Tests, In Vivo for Class VI plastics criteria

4. Product information

4.1 Device Design

TangenX SC TFF Devices are designed and constructed using FDA-approved materials that have been validated for use in demanding biopharmaceutical applications. Each device is manufactured according to a fully validated and documented manufacturing process

that adheres to ISO 9001:2015 requirements and meets specified release criteria. The TangenX SC TFF Devices are ideally suited for single-use processing in a closed system with optimal performance that is equivalent across the entire TangenX Cassette product line.

4.2 Materials of construction

TangenX SC TFF Devices are constructed of USP Class VI approved materials.

Table 2. Materials of construction

Component	Material
Membrane	Modified polyether sulfone (mPES)
Membrane support	Polypropylene (PP)
Channel configurations: L-screen (feed/retentate channel) Filtrate channel (both L-screen)	 Polypropylene medium grade, woven screen Polypropylene medium grade, woven screen
Encapsulant: Feed/retentate channel Filtrate channel	 Class VI approved polyurethane Class VI approved polyurethane
Gasket	Ethylene propylene diene monomer (EPDM)
Devise Housing	Polyphenylsulfone (PPS)
Device shell ($\geq$2.5 m^2)	Polysulfone (PS)
Tri-clover® clamp	Nylon, glass-filled
Tri-clover gasket	Platinum-cured silicone
Braided tubing	Platinumcured silicone
AseptiQuik® fitting	Polycarbonate
Pinch clamp	Nylon, glass-filled

Table 3. Device specifications

Module characteristics	Surface area				
	0.5 m^2	1.5 m^2	2.5 m^2	5.0 m^2	10 m^2
Channel path length	16 cm				
Hold-up volume Feed/ Retentate	0.17 L	0.44 L	0.65 L	1.17 L	2.34 L
Hold-up volume Permeate	0.16 L	0.40 L	0.58 L	1.03 L	2.07 L
Working volume	1 – 50 L	3 – 150 L	5 – 250 L	10 – 500 L	20 – 1,000 L
Temperature	4 – 40° C				
Maximum forward pressure	60 psi (4 bar)				
Maximum reverse pressure	7 psi (0.48 bar)				
Crossflow @ 10 psi / 0.7 bar	2 – 4 L/min	6 – 12 L/min	10 – 20 L/min	20 – 40 L/min	40 – 80 L/min
Air integrity test pressure	7.3 psi (0.50 bar)				
Max air diffusion rate	323 ccm/m^2				
AseptiQuik connector	G	G	G	G	L
Height	5.9 in (15.0 cm)	7.7 in (19.7 cm)	9.3 in (23.6 cm)	13.1 in (33.3 cm)	20.6 in (52.3 cm)
Diameter	10.0 in (25.4 cm)				
Tube stand-off length	7.5 in	6.75 in	6.75 in	6.75 in	7.25 in
Weight (in box)	20 lb (9 kg)	22 lb (10 kg)	24 lb (11 kg)	32 lb (14.5 kg)	47 lb (21.3 kg)

4.3 Package Contents

TangenX SC TFF Device package contents:

- One (1) TangenX SC Device (0.5, 1.5, 2.5, 5, or 10 m^2)
- Certificate of Conformance
- Safety Data Sheet, 0.2 M sodium hydroxide
- Information Guide

5. Operational Considerations

5.1 Unpacking the Device

TangenX SC TFF Devices should be unpacked from the box and moved to a clean stable surface. Do not lift device using fittings or connectors. Lift from the body of device or the handle if provided (10 m^2 devices only). Failure to do so can result in permanent damage to the device.

The TangenX SC TFF Device is double bagged under a partial vacuum. Depending on environmental conditions, the amount of air in the bags will vary. If vacuum has been lost from outer bag, the device remains sterile. Refer to the User Guide for instructions on how to connect the device to a flow path and conduct process operations.

5.2 Connections

AseptiQuik G aseptic connectors from CPC® (or equivalent) are used on the feed, retentate, and permeate ports of the device for all products up to 5 m^2. The 10 m^2 device is equipped with larger AseptiQuik L aseptic connectors.Refer to the User Guide for information on how to connect the TangenX SC TFF Device.

5.3 Device location

Place the device on a level surface and position it to minimize the tubing length and prevent kinking of tubing. Care must be taken to stabilize the tubing attached to the top of the device. Unsecured tubing may create a device tip hazard and must be addressed through the design of the system being connected to the TangenX SC Device.

5.4 Pump

When using a TangenX SC TFF Device, select a pump with adequate capacity. Crossflow rate ranges are dependent on feed channel type and process fluid. Refer to the User Guide.

5.5 Preparation (water flush) of TangenX SC TFF Devices

Devices can be flushed with water (5 L/m^2 through the retentate and 10 L/m^2 through the permeate) to displace the 0.2 M sodium hydroxide storage agent prior to equilibration.

5.6 Equilibration of TangenX SC TFF Devices

Devices must be equilibrated with an appropriate buffer (e.g., phosphate buffered saline) to ensure the neutralization of the 0.2 M sodium hydroxide storage agent within the device. Verify that the pH of the effluent from the device is neutralized to minimize any possible interaction with your application. For most applications, further sanitization is not required.

5.7 Disposal of used TangenX SC TFF Devices

The fluid path of the TangenX SC TFF Devices should be closed to minimize potential hazards of the process fluid. Disposal will be dependent on the feed stream used and the facility requirements.

5.8 Storage of TangenX SC TFF Devices

Un-used devices should remain sealed in their original packaging prior to use to maintain their characteristics and integrity, and to minimize external contamination. The following temperatures are recommended for storage:

- 15°C – 25°C (optimal)
- 40°C (maximum)
- Do not freeze devices

5.9 Numbering system

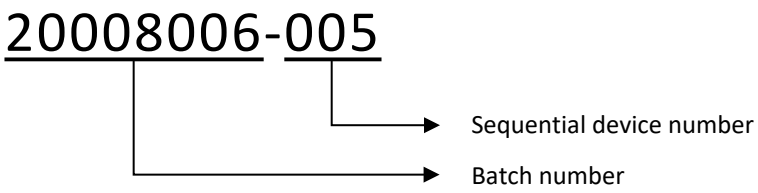
5.9.1 Batch number

Device batch numbers are printed on each device label. The batch number is the eight (8) digit manufacturing process order number assigned by the SAP system. A batch is defined as a group of consecutively serialized devices manufactured on the same day, built from up to six (6) different raw material lots and generated from the same SAP process order. Batch traceability is maintained on the electronic batch record and in the SAP system.

5.9.2 Serial number

The device serial number is composed of the batch number (SAP process order number) and the three (3) digit sequential device number within the batch. The order number will be between 00000001 and 999999999. The sequential number will be between 001 and 999.

Figure 2. Serial number example



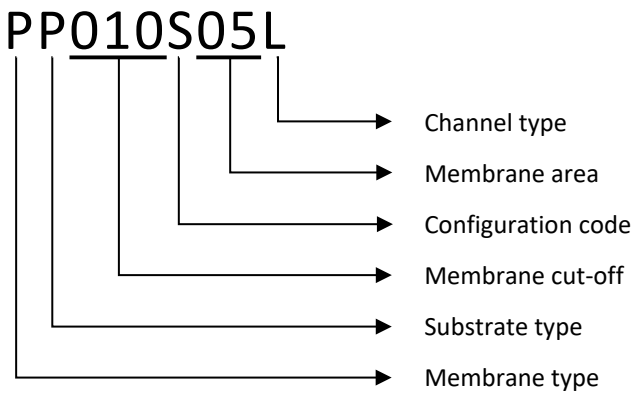
5.9.3 Catalog number

The catalog number is an alphanumeric code that identifies specific membrane, substrate, and channel types, as well as MWCO, surface area, and configuration (Table 4, Figure 3).

Table 4. Catalog part number system

Parameter	Description		Corresponding figure in catalog number
Membrane type	ProStream HyStream	mPES Low protein binding (LPB) mPES Ultra-hydrophilic and LPB	P X
Substrate type	Polypropylene		P
Membrane cut-off	10 kD 30 kD 50 kD 100 kD 300 kD		010 030 050 100 300
Configuration code	TangenX SC TFF Device	Self-Contained	S
Membrane area (m^2 / ft^2)	0.5 / 5.4 1.5 / 16.2 2.5 / 26.9 5 / 53.8 10 / 107	Available: S Available: S Available: S Available: S Available: S	05 15 25 50 99
Channel type	L-screen	Medium woven	L

Figure 3. Catalog part number system



6. Product performance

Designed for use in a wide range of biopharmaceutical applications, especially those that are protein based, TangenX ProStream and HyStream membranes manufactured by Repligen represent the latest in development of modified polyethersulfone (mPES). In contrast to conventional composite mPES ultrafiltration (UF) membranes that are made in multi-step manufacturing processes and often include a post-casting surface modification, TangenX mPES membranes have been developed from state-of-the-art technology, including two unique features that deliver significant user benefits:

Single-cast, uniquely controllable manufacturing process:

- Balanced flux and selectivity are the results of this highly controllable manufacturing process that enables tight control of the macro-porous/UF transition interface, producing a finely controlled continuum. This controlled transition ensures negligible breakthrough of the UF skin, maximizing selectivity performance.
- A reduced number of manufacturing steps delivers excellent consistency and reliability at a lower cost.

Integral cast modification of the membrane chemistry:

- The addition of a second polymer into the pre-casting membrane solution ensures total and consistent surface modification that delivers very low protein binding due to the neutral charge and excellent chemical resistance of the membrane.

The result is an application-focused membrane with a finely balanced performance profile that combines:

- The flux of a highly porous UF membrane substructure with the retention and selectivity of a composite structure.
- Highly desirable low protein binding properties that maximize recovery and have comparable chemical resistance to unmodified polymeric membranes.

6.1 Membrane performance testing

Water flux data was generated using membrane cut to 44.5 mm discs in stirred cells at typcial working conditions (50 psig), using purified water at 20°C to measure the membrane water permeability. TangenX ProStream and HyStream mPES membranes demonstrate comparable water permeability. Purified proteins and molecular weight markers were used to challenge the membranes in the stirred cells. TangenX mPES membranes demonstrate excellent selectivity (Figure 4).

Many membranes are formulated for either retention or flux. TangenX mPES membranes have been designed to be balanced across both. The retention/rejection data for each membrane in the molecular weight cut-off (MWCO) series (Figure 4), when reviewed in conjunction with normalized water permeability (NWP) data (Figure 5), provides the user with a means to select a membrane that best balances flux and retention for their specific application.

Figure 4. Membrane performance: MW vs. % rejection

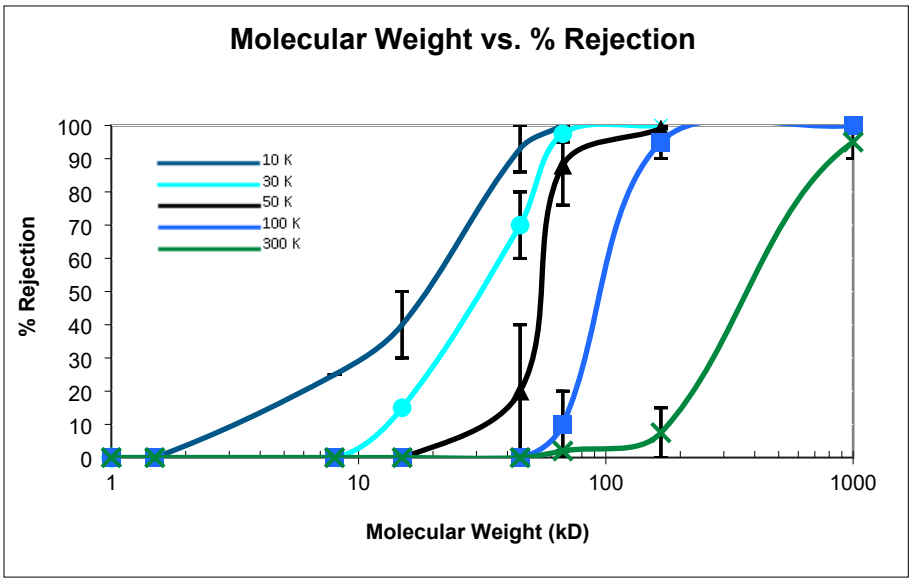
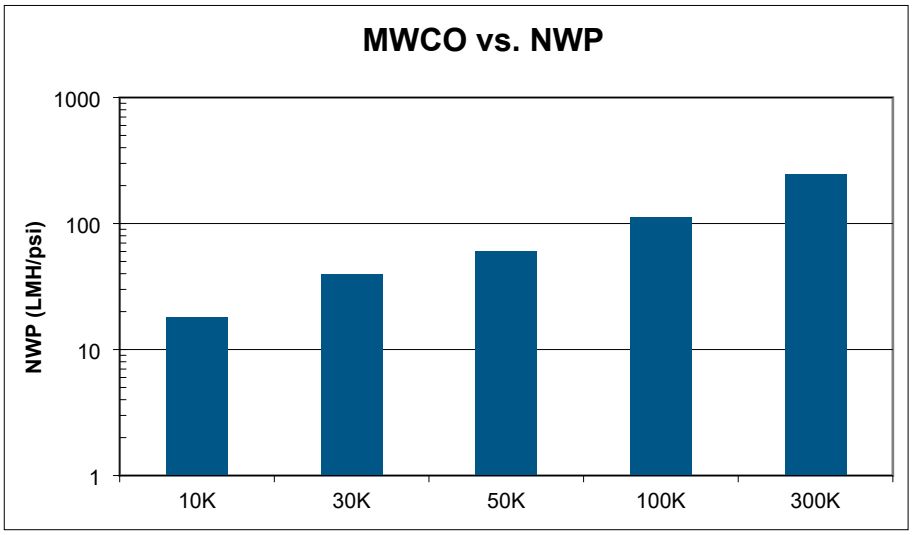


Figure 5. Membrane performance: MWCO vs. NWP



6.2 Non-specific protein binding

Non-specific protein binding is defined as the adsorption of a protein to a surface by one or more modes of attraction (e.g., charge effect, hydrophobic interaction, etc.). Non-specific protein binding is associated with yield loss and membrane fouling; both are undesirable effects.

Non-specific protein binding was quantified for ProStream and HyStream mPES membrane chemistries relative to a PES control membrane. One membrane of each type was chosen since the membrane chemistry is the same for each pore size. Each membrane was challenged with three different types of protein solution: BSA (bovine serum albumin), IgG (immunoglobulin G), and Cyt C (cytochrome c). These proteins were selected because they represent a broad range of molecular weights, structures, and isoelectric points. The amount of protein bound to each membrane was measured by absorbance at 280 nm and then recorded.

ProStream and HyStream mPES membranes bind less BSA, IgG, and Cyt C compared to the PES control membrane (Table 5; average of three data sets).

Table 5. Non-specific protein binding test results

Membrane type	BSA binding	IgG binding	Cyt C binding
PES control	<0.1 µg/cm²	11.34 µg/cm²	36.73 µg/cm²
ProStream**Error! Bookmark not defined.** mPES	<0.1 µg/cm²	2.99 µg/cm²	1.36 µg/cm²
HyStream mPES	<0.1 µg/cm²	3.29 µg/cm²	9.21 µg/cm²

Lower protein binding is associated with higher product recovery and is, therefore, a desirable attribute of mPES membranes. Lower protein binding also decreases the likelihood of a secondary boundary layer forming on the membrane surface. Without this boundary layer, the risk of reduced productivity is reduced.

Non-specific protein binding data support the description of Repligen manufactured ProStream and HyStream modified PES membranes as low protein binding as compared to unmodified PES membranes.

6.3 Device hydraulic performance

Scale-up performance is critical for successful process development and can be demonstrated by evaluating TFF device hydraulic performance using purified water. TangenX SC TFF Devices are manufactured with specific channel geometries and hydrodynamic characteristics that directly impact process performance. Optimized channel geometry is critical for scalable performance. This leaves the end user with two primary factors to consider:

- The effect of operating conditions on the process flux and selectivity profile.
- Linear scaling performance from 0.5 m² to 10 m².

The TangenX SC TFF Devices address these factors, as significant development has been devoted to the channel design. Optimized channel geometry, with enhanced rigidity ensures hydraulic performance is maintained when scaling up through the TangenX SC TFF Device family resulting in optimal and reproducible scaling performance. Additionally, each device undergoes rigorous QA release testing to verify it meets specifications. Devices are tested for both air integrity and hydrodynamic performance. This testing ensures consistent performance from device-to-device. The result is scalable process development and reproducible manufacturing.

To evaluate hydraulic scalability, the pressure drop (ΔP) between the feed and the retentate at 10 psi must be within the specified range of 4 – 8 L/min/m² for the devices with L-screen (Table 6).

Table 6. Recommended crossflow rates

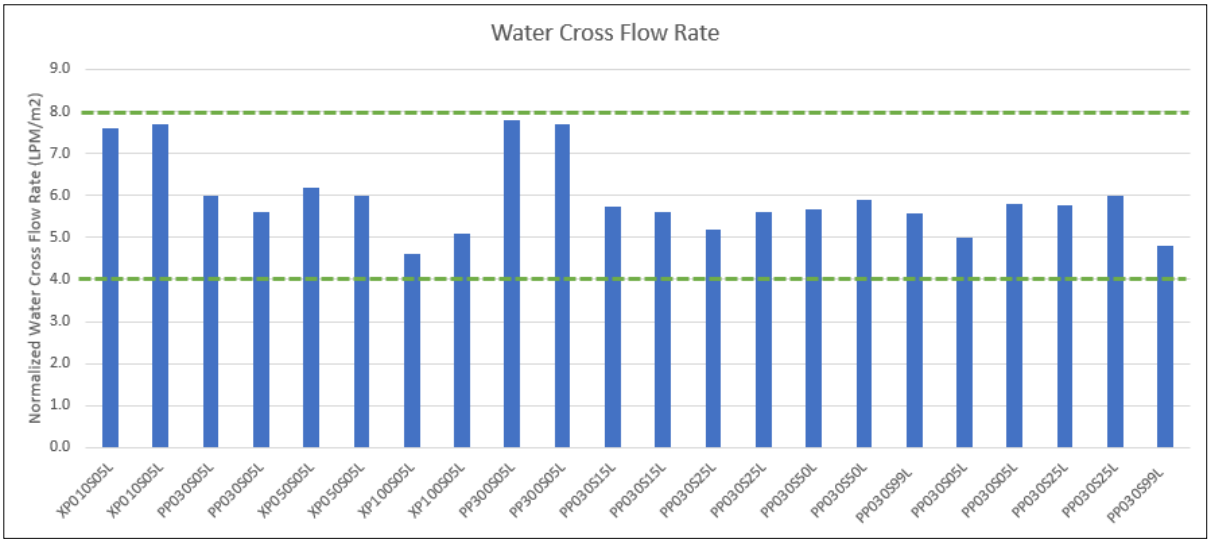
Screen	Crossflow range	Low crossflow	Medium crossflow	High crossflow	ΔP
L-screen	4 – 8 L/min/m²	4 L/min/m²	6 L/min/m²	8 L/min/m²	10 psi (0.7 bar)[1]

[1]Typical ΔP measured with water and permeate closed.

Normalized crossflow flux data at 10 psi ΔP (Figure 6) show that TangenX SC TFF Devices have average temperature-normalized retentate flow rates within the required specifications (Lower (4 LPM/m² and upper (8 LPM/m²) specification limits, green lines):

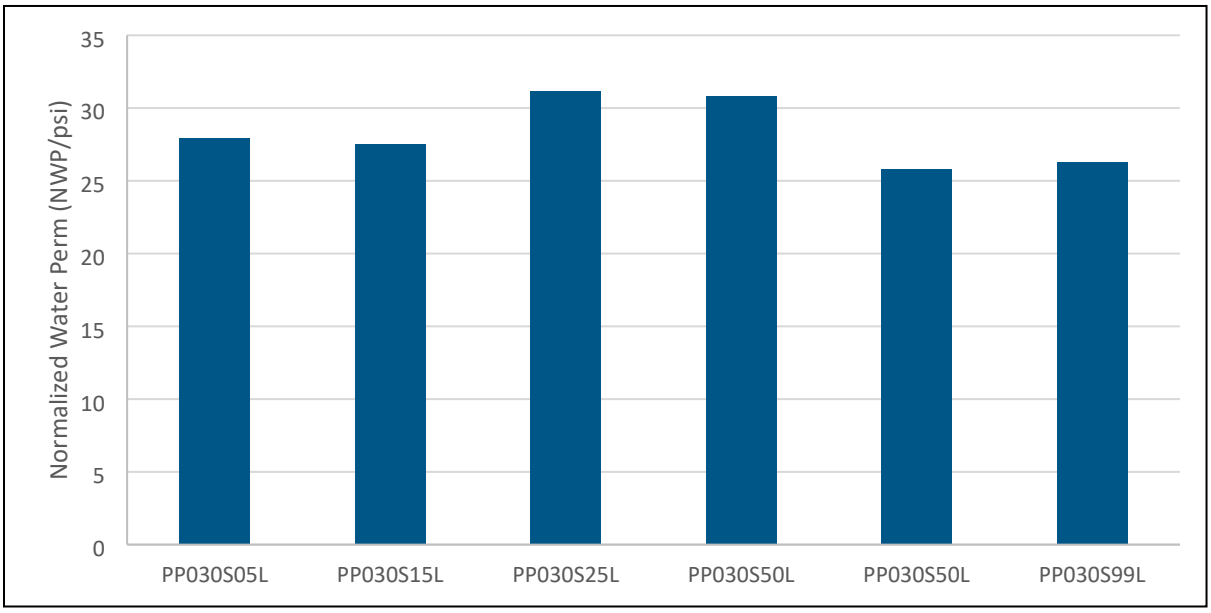
- 2 – 4 LPM for 0.5 m² devices
- 6 – 12 LPM for 1.5 m² devices
- 10 – 20 LPM for 2.5 m² devices
- 20 – 40 LPM for 5 m² devices
- 40 – 80 LPM for 10 m² devices

Figure 6. Crossflow flux at 10 psi ΔP for TangenX SC TFF Devices



The hydraulic scalability of a membrane can also be evaluated using purified water to measure normalized water permeability (NWP). NWP data can be used to further support scalability of the TangenX SC TFF Devices. NWP for the 30 kD ProStream membrane with surface areas from 0.5 m^2 to 10 m^2 was measured. Average NWP was within specification (24 – 41 LMH/psi) across the product range.

Figure 7. NWP for 30 kD TangenX SC TFF Devices



Normalized water permeability (NWP) is a function of the molecular weight cutoff (MWCO) of a membrane. Therefore, there is a range of permeability rates for each membrane of a given MWCO. External influences, such as manifolds, piping, and valves, create restrictions and can affect the measured NWP. Thus, the initial NWP of each device should be measured in its designated system. Typical normalized water permeability (NWP) ranges for a given molecular weight cutoff (MWCO) are provided as a guide for determining if the NWP of a device is within range (Table 7).

Table 7. Typical NWP range for TangenX SC TFF Devices

MWCO	Typical NWP range
10 kD	8.6 – 20 LMH/psi
30 kD	24 – 41 LMH/psi
50 kD	34 – 56 LMH/psi
100 kD	32 – 91 LMH/psi
300 kD	190 – 300 LMH/psi

6.4 Device integrity

Device integrity testing provides a non-destructive method for verifying the integrity of a Tangential Flow Filtration (TFF) device. Each device manufactured by Repligen undergoes strict release testing, including an air integrity test. Release testing at Repligen follows a validated test method for all membrane and device QC testing.

To demonstrate the sensitivity of the integrity testing, both an intact (initial) and a modified (purposely defected) device were evalluated (Table 8). The intact device was tested by pressurizing the upstream side with air. The integral membrane did not allow significant amounts of air to pass through due to the surface tension of liquid in the pores. Next, a pinhole was created in the membrane (Figure 8), and airflow was measured. The pinhole allowed air to pass through the membrane at a rate nearly 100-fold higher than the intact membrane. The difference was specific to the air diffusion rate and not the liquid crossflow rate, as no change in liquid flow rate was detected. The intact device met integrity specifications (Table 9).

Figure 8. Membrane surface with pinhole (100x magnification)

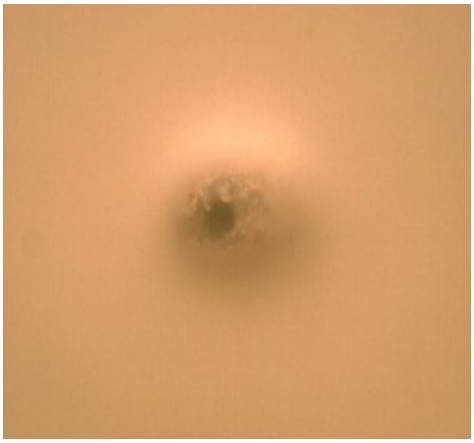


Table 8. Device integrity test results

Device serial #	Device status	Results		Results within spec (Y/N)	Difference observed: air diffusion (Y/N)	Difference observed: flow rate (Y/N)
		Air diffusion rate	Liquid crossflow rate			
17213102	Initial	24 ccm	621 mL/min	Yes	N/A	N/A
	Modified	2196 ccm	620 mL/min	No	Yes	No

Table 9. Device integrity specifications

Device channel type	Membrane type	Specification
L-screen	Ultrafiltration 10 kD – 300 kD	≤323 ccm/m^2 at 0.5 bar (≤30 ccm/ft^2 at 7.3 psi)

6.5 Device pre-flushing

TangenX SC TFF Devices require a purified water flush and a buffer equilibration step prior to use to ensure leachable concentration reduction. The method for flushing the device prior to use was developed to maximize the efficiency of the flushing process and to reduce the total amount of required water and buffer. Storage agents removed by flushing may be considered unwanted leachables by the user if not sufficiently removed by the specified rinse and equilibration procedure recommended by Repligen. The following outlines the recommended flushing and equilibration conditions developed to remove the storage solution and unwanted leachables from the device.

6.5.1 Purified water flush

Flushing the TangenX SC TFF Device with 0.2 µm filtered deionized (DI) water displaces the 0.2 N NaOH storage solution present in the shipped product. Add purified DI water to a vessel. Connect the purified DI water through a pump to the feed port of the device. Direct both the retentate and the permeate flow paths to a waste vessel. Close the permeate. Flush the retentate flow path at the recommended flow rate (Table 6) until 5 L of water per m^2 of surface area is collected from the retentate into the waste vessel (Table 10). The permeate flow path is then flushed by opening the permeate and closing the retentate. Begin pumping at a low flow rate and slowly increase it until the transmembrane pressure is 10 – 15 psi. The permeate is then flushed until 10 L of water per m^2 surface area is collected from the permeate into the waste vessel (Table 10).

Figure 9. Example device flush flow path

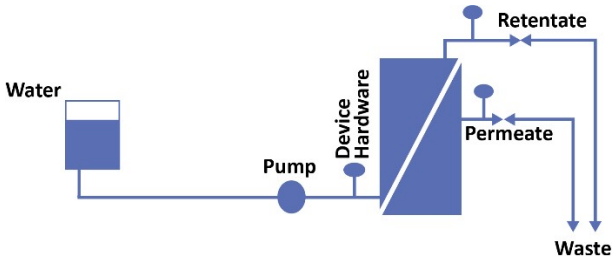


Table 10. Device flush volume (± 10%)

TFF filter surface area	Retentate to waste	Permeate to waste
0.5 m^2	2.5 L	5.0 L
1.5 m^2	7.5 L	15.0 L
2.5 m^2	12.5 L	25.0 L
5 m^2	25 L	50 L
10 m^2	50 L	100 L

6.5.2 Buffer equilibration

The device is then equilibrated with buffer to neutralize residual NaOH. The process is similar to the water flush. Add equilibration buffer to a vessel. Connect the buffer through a pump to the feed port of the device. Direct both the retentate and the permeate flow paths to a waste vessel. Close the permeate. Flush the retentate flow path at the recommended flow rate (Table 6) until 5 L of water per m^2 of surface area is collected from the retentate into the waste vessel (Table 11). The permeate flow path is then flushed

by opening the permeate and closing the retentate. Begin pumping at a low flow rate and slowly increase it until the transmembrane pressure is 10 – 15 psi. The permeate is then flushed until 10 L of water per m^2 surface area is collected from the permeate into the waste vessel (Table 11).

Figure 10. Example buffer equilibration flow path

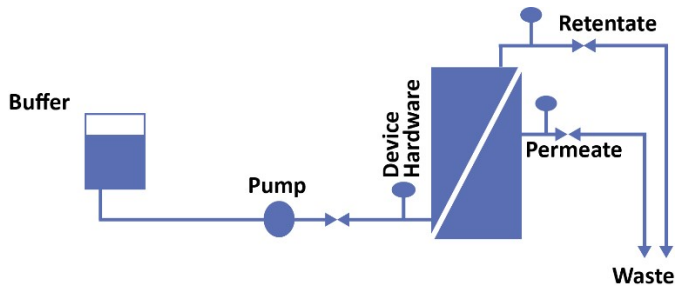


Table 11. Buffer equilibration volume (± 10%)

TFF filter surface area	Retentate to waste	Permeate to waste
0.5 m^2	2.5 L	5.0 L
1.5 m^2	7.5 L	15 L
2.5 m^2	12.5 L	25 L
5 m^2	25 L	50 L
10 m^2	50 L	100 L

A similar pre-use flushing method was used to prepare the devices for the BPOG extractables study, demonstrating the effectiveness of the method (Section 7.2).

6.6 Robustness study

TangenX SC TFF Devices are designed to be single use; however, the device must demonstrate the robustness to withstand use under the full range of recommended conditions. Many different elements contribute to the stress put on TFF membranes: time, temperature, pressure, flow rate, and buffer conditions are several examples. Steps were taken to validate the robustness of the TangenX SC TFF Devices under aggressive operating conditions. The information gathered from this validation study will also be referenced in other supporting documents. Ten (10) 0.5 m^2, two (2) 1.5 m^2, two (2) 2.5 m^2, two (2) 5 m^2, and one (1) 10 m^2 TangenX SC TFF Devices were manufactured, irradiated, then evaluated for robustness. Each device was prepared using current SOPs to reflect the standard manufacturing process at Repligen. The following steps were taken as part of the study:

1. TangenX SC TFF Devices were prepared using the following procedures:
    - SOP-3185 TangenX SC Cassette Assembly
    - SOP-3187 TangenX SC Cassette Trimming and Routing
    - SOP-3188 TangenX SC Cassette QC Testing
    - SOP-3190 TangenX SC Housing Sub-Assembly
    - SOP-3192 TangenX SC Housing Parts Cleaning
    - SOP-3194 TangenX SC Device Assembly
    - SOP-3196 TangenX SC Packaging and Gamma Irradiation
2. Devices were sampled and evaluated following procedure PV-TANGENX-220409.

This study followed the approved method PV-TANGENX-220409 for evaluation of TangenX SC TFF Devices for durability and robustness. Each device was tested and released following the TangenX SC QC test procedures SOP-3188 and SOP-3194. A total of

seventeen different TangenX SC TFF Devices were selected for this study, representative of the entire product line of closed membranes (Table 12). ProStream and HyStream membrane chemistries were included. The molecular weight cut-offs (MWCO) tested ranged from 10 kD to 300 kD, representative of the entire product offering. Each device was tested in triplicate.

In summary, each device was removed from its packaging, installed, and equilibrated with RODI water. Baseline air integrity, normalized water flux (NWP), and pressure drop were measured and recorded. PVP K90 with RODI water (polyvinylpyrrolidone, model solution) was then recirculated through the system for 8 hours at 4°C and 40°C. Once this recirculation was complete, the devices were retested for air integrity, NWP, and pressure drop. All devices met the release specifications (Table 12).

Table 12. Robustness testing

Part number	Description	Result
XP010S05L	0.5 m^2 TangenX SC TFF Devices (10 kD ProStream)	Pass
PP030S05L	0.5 m^2 TangenX SC TFF Devices (30 kD ProStream)	Pass
XP050S05L	0.5 m^2 TangenX SC TFF Devices (50 kD HyStream)	Pass
XP100S05L	0.5 m^2 TangenX SC TFF Devices (100 kD HyStream)	Pass
PP300S05L	0.5 m^2 TangenX SC TFF Devices (300 kD ProStream)	Pass
PP030S25L	1.5 m^2 TangenX SC TFF Devices (30 kD ProStream)	Pass
PP030S25L	2.5 m^2 TangenX SC TFF Devices (30 kD ProStream)	Pass
PP030S50L	5 m^2 TangenX SC TFF Devices (30 kD ProStream)	Pass
PP030S99L	10 m^2 TangenX SC TFF Devices (30 kD ProStream)	Pass

Each device type met the post-use integrity and pressure drop specifications. Integrity is one of the best indicators of an issue with the membrane and is critical to its function. The robustness testing demonstrated the design of the membrane stack remained integral following the challenge conditions.

6.7 Shelf life studies

6.7.1 Membranes

The following section describes the conclusion of the shelf life study for ultrafiltration membranes manufactured by Repligen after five years. Ultrafiltration membranes are initially cast and then stored for a period of time prior to being incorporated into a product. The time between when the membrane is manufactured and when it is used in a device may be up to five (5) years.

Several lots of membranes were cast during process validation. Each membrane was prepared using current SOPs and reflected the standard membrane manufacturing process at Repligen. The following steps were taken as part of the study:

- Membranes were prepared using SOP-0448 and SOP-0454
- These membranes were sampled and tested following TX1001-POQ-115

This summary will provide final results for the shelf life study of the ProStream and HyStream membranes manufactured at Repligen. This report will also be used to summarize results of the sampling and testing throughout the study. The membrane storage study procedure (TX1001-POQ115) was applied to both the ProStream and HyStream membranes manufactured at Repligen. One membrane of each type was chosen to represent the product line consisting of all MWCO membranes. These membranes were chosen as they correspond to the device storage study. Each membrane was tested following the standard Repligen QC release procedure SOP-0463.

Membrane acceptance criteria for shelf life results are show in Table 13. The storage study was executed at two different temperatures, one at ambient temperature (Table 14) and the other at 50°C (Table 15). Ambient simulated exposure at a normal or

median temperature. This type of study spanned five (5) years and was the standard shelf life study. Experiments at 50°C simulated exposure to the maximum temperature limit of the product as an accelerated stability study.

Data for each time point was obtained in triplicate. In the event one membrane failed during the study, a failure analysis would have been conducted. Failure mode and product quality impact would have been assessed. If the membrane was deemed to be an anomaly, the study would have continued as planned. The documented failure would have accompanied the final report. Failure of all three membranes would have concluded the study. A detailed analysis of the membranes that did not meet release criteria would have been included in the final report.

Table 13. Membrane acceptance criteria for shelf life studies

Test	Criteria	
Normalized Water Permeability (NWP)	9.5 – 22.0 LMH/psi	Percent deviation: 15
Passing molecular weight (MW) marker (PVP C-15 (~15 kD))	140 – 250 (flux, LMH)	Percent rejection: 30 – 60
Retaining molecular weight (MW) marker (PVP C-30 (~45 kD))	75 – 110 (flux, LMH)	Percent rejection: >85
Integrity test (Air diffusion @ 15 psi; total # of discs with diffusion)	≤6 discs with diffusion	Total of 18 discs

Table 14. Membrane acceptance test results: Ambient temperature

Time point	Normalized water permeability	Passing MW marker	Retaining MW marker	Integrity test
Time initial	Pass	Pass	Pass	Pass
3 months	Pass	Pass	Pass	Pass
6 months	Pass	Pass	Pass	Pass
1 year	Pass	Pass	Pass	Pass
2 years	Pass	Pass	Pass	Pass
3 years	Pass	Pass	Pass	Pass
4 years	Pass	Pass	Pass	Pass
5 years	Pass	Pass	Pass	Pass

Table 15. Membrane acceptance test results: Elevated temperature (50°C)

Time point	Normalized water permeability	Passing molecular weight marker	Retaining molecular weight marker	Integrity test
Time initial	Pass	Pass	Pass	Pass
1 week	Pass	Pass	Pass	Pass
1 month	Pass	Pass	Pass	Pass

6.7.1.1 Conclusions

The results demonstrate that both the ProStream and HyStream membranes meet or exceed all release specifications after five years at ambient conditions and one month at 50°C (Table 14). No significantly measurable change in membrane performance was detected. The membrane storage study successfully reached its 5-year conclusion.

6.7.2 TangenX SC TFF Devices

The following section describes the method used to evaluate the shelf life of the TangenX SC TFF Devices manufactured by Repligen. The TangenX SC TFF Devices are manufactured, packaged, irradiated, and stored for a period of time prior to shipment. Once shipped, the device may then remain in storage for an additional period of time before it is put into use. The TangenX SC single-use devices are stable for up to 3 months at elevated temperature and projected to be stable up to 18 months when stored at ambient temperature based on accelerated aging tests.

Three different sized TangenX SC TFF Devices were manufactured and evaluated in duplicate. Each device was prepared using Repligen SOPs conforming to the Repligen standard manufacturing process. The following steps were taken as part of the study:

1. Devices were prepared using best manufacturing practices.
2. These devices were sampled and studied following this procedure.

Procedure PV-TANGENX-220411 was used to evaluate shelf-life stability, including determination of appropriate time points for sampling and evaluation. Each device was evaluated in accordance with the Repligen standard QC release procedure and acceptance criteria (Table 16).

Table 16. TangenX SC Device acceptance criteria

Acceptance per DID-18159 (R0)	Limit
Pressure drop vs cross flow @ ΔP 10 psi	0.4 – 0.8 LPM
Air diffusion test @ 7.3 psi (0.5 bar)	<300 ccm/m^2
Air diffusion test @ 15 – 60 psi (1 – 4 bar)	Information Only
Visual Inspection	No cracked, broken, or, leaking parts

6.7.2.1 Results

Three different sized devices including 0.5, 2.5, and 10 m^2 were evaluated using an accelerated aging study. Experiments at 50°C provide accelerated aging data designed to simulate exposure at the maximum temperature limit of the product. This portion of the study was concluded within three (3) months and is considered an accelerated study. The 3-month study period of accelerated aging followed the ASTM reference F1980-16 and successfully demonstrated an 18-month shelf life at ambient temperature (Table 17).

Table 17. Device acceptance test results: Elevated temperature (50°C)

Device type	Time initial	3 months at 50C
0.5 m^2 TangenX SC TFF Device	Pass	Pass
2.5 m^2 TangenX SC TFF Device	Pass	Pass
10 m^2 TangenX SC TFF Device	Pass	Pass

The shelf-life study outlined in procedure PV-TANGENX-220411 successfully demonstrated an 18-month shelf life and will continue until its conclusion at three (3) years when a final report will be issued.

6.8 Chemical compatibility

Table 18. ProStream and HyStream membrane chemical compatibility

Reagent	ProStream (pH 1 – 14)	HyStream (pH 1 – 14)
Acetic acid (5%)	✓	✓
Acetic acid (25%)	✓	X
Acetone (≤ 30%)	✓	✓
Acetonitrile (≤ 15%)	✓	✓
Alconox (1%)	✓	✓
Aliphatic and aromatic esters	X	X
Amines	X	X
Ammonium chloride (1%)	✓	✓
Ammonium hydroxide (5%)	X	X
Aromatic and chlorinated hydrocarbons	X	X
Butanol (70%)	✓	✓
Butyl acetate (40%)	✓	X
Butyl cellosolve (10%)	✓	✓
Calcium chloride (5%)	✓	✓
Chloroform (0.8%)	✓	✓
Citric acid (1%)	✓	✓
Dimethyl acetamide (DMAC) (≤ 30%)	✓	X
Dimethyl acetamide (DMAC) (≤ 15%)	✓	✓
Dimethylformamide (≤ 40%)	✓	✓
Dimethyl sulfoxide (≤ 40%)	✓	✓
Disodium salt of EDTA (10%)	✓	✓
Ethanol (70%)	✓	✓
Ethers	X	X
Ethyl acetate (≤30%)	✓	✓
Formaldehyde (1%)	✓	✓
Formic acid (5%)	✓	✓
Glutaraldehyde (0.5%)	✓	✓
Glycerin (50%)	✓	✓
Guanidine HCl (6M)	✓	✓
Hydrochloric acid (0.1N @ 25°C)	✓	✓
Hydrochloric acid (0.1N @ 50°C)	✓	✓
Hydrochloric acid (1.0N @ 50°C)	✓	X
Hydrogen peroxide (1%)	✓	✓

Reagent	ProStream (pH 1 – 14)	HyStream (pH 1 – 14)
Isopropyl acetate (1%)	✓	✓
Isopropyl alcohol (25%)	✓	✓
Ketones	X	X
Lactic acid (5%)	✓	✓
Mercaptoethanol (0.1%)	✓	✓
Methyl alcohol (25%)	✓	✓
Methylene chloride (1%)	✓	X
Methyl ethyl ketone (1%)	✓	X
N-methyl pyrrolidone (1%)	✓	✓
Nitric acid (≤1%)	✓	✓
Oxalic acid (1%)	✓	✓
Phenol (0.5%)	✓	✓
Phosphate buffer (pH: 8.2) (1 M)	✓	✓
Phosphoric acid (1 N)	X	X
Sodium azide (1%)	✓	✓
Sodium chloride (5%) (50°C)	✓	✓
Sodium deoxycholate (5%)	X	X
Sodium dodecyl sulfate (0.01 M)	✓	✓
Sodium hydroxide (0.1 N @ 25°C)	✓	✓
Sodium hydroxide (0.1 N @ 50°C)	✓	✓
Sodium hydroxide (0.5 N @ 25°C)	✓	✓
Sodium hydroxide (0.5 N @ 50°C)	✓	✓
Sodium hydroxide (1.0 N @ 25°C)	✓	X
Sodium hypochlorite (100 ppm)	✓	✓
Sodium hypochlorite (400 ppm)	✓	X
Sodium hypochlorite (1000 ppm)	X	X
Sodium nitrate	✓	✓
Sulfuric acid (1 N)	✓	X
Terg-a-zyme (1%)	✓	✓
Tetrahydrofuran (5%)	X	X
Toluene (1%)	X	X
Tris buffer (pH: 8.2) (1 M)	✓	✓
Triton X - 100 (0.002 M)	✓	✓
Urea (25%)	✓	✓
Ultrasil 11 (1%)	✓	✓

✓ = Compatible, no significant changes in either rejection or flow rate.
X = Not compatible, significant change noticed.

7. Safety information

7.1 USP Class VI

The purpose of UPS Class VI testing is to verify the biological safety of each of the components used in the TangenX SC TFF Device product line. Samples for USP Class VI testing consisted of each of the five components of the TangenX SC Device. Housing components relied upon manufacturers' USP Class VI testing reports to validate USP Class VI requirements. Each component used to construct the membranes, and the sample dimension, sample mass and test regime are provided (Figure 11).

Figure 11. USP testing results

TangenX Sample Matrix **USP Testing** **Vendor: Toxikon**

	Component Description	Composition	Minimum Sample Mass	Sample Dimensions	Tests to be Conducted
1	Cassette Encapsulant	Polyurethane	~ 45 grams from 3 lots	25mm x 25mm x 5mm (1)	A,B,C
2	Screen Spacer	Polyolefin	~ 45 grams from 3 lots	25mm (diameter) x 0.8mm (1)	A,B,C
3	HyStream Membrane	Polyethersulfone	~ 45 grams from 3 lots	25mm (diameter) x 0.2mm (1)	A,B,C
4	ProStream Membrane	Polyethersulfone	~ 45 grams from 3 lots	25mm (diameter) x 0.2mm (1)	A,B,C
5	EPDM Gasket	EPDM	~ 45 grams from 3 lots	25mm (diameter) x 1mm (1)	A,B,C
6	Silicone PSA w/screen	Silicone & Polypro	~ 45 grams from 3 lots	25mm (diameter) x 0.8mm (1)	A,B,C
(1) Must also include 1mm x 1mm x 10mm sample					

Test ID	Test Description	Sample Mass	Sample Dimensions	Total Qty
A	MEM Elution per USP <87>	4 grams	(see above)	7
B	Class VI per USP <88>	16 grams, plus additional pieces ~10g (1)	(see above), plus 12 pieces 1mmx1mmx10mm	7
C	Hemolysis - Indirect with rabbit blood	15 grams	(see above)	7

Samples for both USP and extractables testing required preparation prior to analysis. Each sample was rinsed with WFI, sanitized with 0.5 M NaOH, and then rinsed again with WFI. The purpose of this sample preparation is two-fold:

1. To simulate the sanitization procedure performed on the device prior to release.
2. To sanitize the sample so as not to allow external contamination to interfere with the USP testing.

Approved procedures were followed during preparation of samples and used for USP and Class VI testing. The procedures were used to provide a record of the samples to be prepared, as well as the method of preparation. Experimental deviations were recorded in a laboratory notebook and a copy attached to the final report. Appendicies were created to document deviations, to identify ways to rectify them, and to record whether or not they would significantly affect the result of the experiment.

7.1.1 Results and discussion

The results of the studies show that all component materials meet:

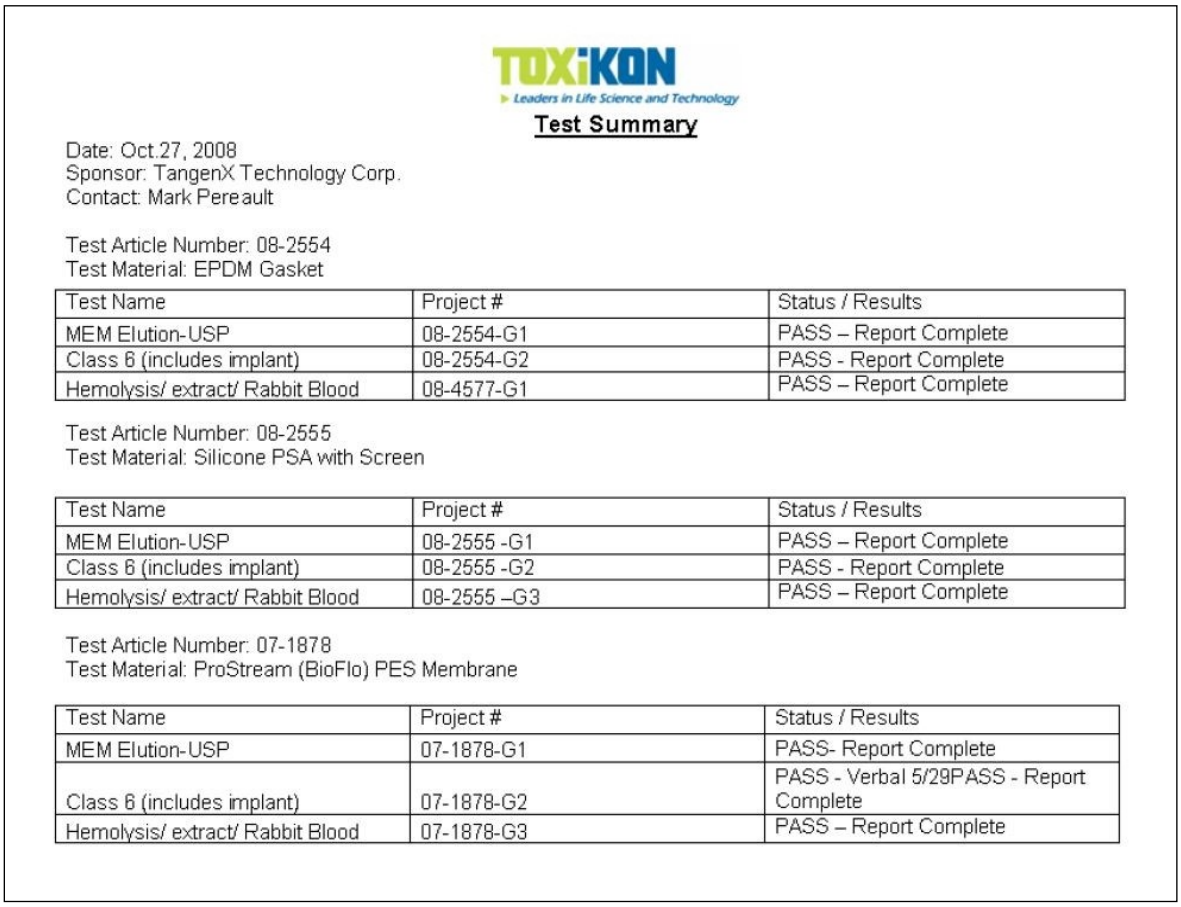
- Current requirements for USP Class VI biological testing for plastics
- The test articles meet the test requirements as defined in the USP guidelines: USP 30, NF 25, 2007, <788> Particulate Matter in Injections

All component materials used in membranes manufactured by Repligen have been independently tested for USP safety and were shown to be safe according to:

- L929 MEM Elution per USP <87>
- Class VI per USP <88>
- Hemolysis - Indirect with Rabbit Blood

The study proposal for the USP testing conducted with Toxikon is found in Toxikon laboratory proposals #07-2-26TF7757 and #08-5-8TF9874. The study results generated by Toxikon are found in the complete USP report and can be provided by Repligen on request. Summaries of the test results is below (Figure 12 and Figure 13).

Figure 12. Summary of USP testing results (A)



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Test Summary

Date: Oct.27, 2008
Sponsor: TangenX Technology Corp.
Contact: Mark Pereault

Test Article Number: 08-2554
Test Material: EPDM Gasket

Test Name	Project #	Status / Results
MEM Elution-USP	08-2554-G1	PASS – Report Complete
Class 6 (includes implant)	08-2554-G2	PASS - Report Complete
Hemolysis/ extract/ Rabbit Blood	08-4577-G1	PASS – Report Complete

Test Article Number: 08-2555
Test Material: Silicone PSA with Screen

Test Name	Project #	Status / Results
MEM Elution-USP	08-2555 -G1	PASS – Report Complete
Class 6 (includes implant)	08-2555 -G2	PASS - Report Complete
Hemolysis/ extract/ Rabbit Blood	08-2555 –G3	PASS – Report Complete

Test Article Number: 07-1878
Test Material: ProStream (BioFlo) PES Membrane

Test Name	Project #	Status / Results
MEM Elution-USP	07-1878-G1	PASS- Report Complete
Class 6 (includes implant)	07-1878-G2	PASS - Verbal 5/29PASS - Report Complete
Hemolysis/ extract/ Rabbit Blood	07-1878-G3	PASS – Report Complete

Figure 13. Summary of USP testing results (B)

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Test Article Number: 07-1880
Test Material: Screen Spacer

Test Name	Project #	Status / Results
MEM Elution-USP	07-1880-G1	PASS – Report Complete
Class 6 (includes implant)	07-1880-G2	PASS - Report Complete
Hemolysis/ extract/ Rabbit Blood	07-1880-G3	PASS – Report Complete

Test Article Number: 07-1881
Test Material: Channel Spacer

Test Name	Project #	Status / Results
MEM Elution-USP	07-1881-G1	PASS – Report Complete
Class 6 (includes implant)	07-1881-G2	PASS - Report Complete
Hemolysis/ extract/ Rabbit Blood	07-1881-G3	PASS – Report Complete

Test Article Number: 07-1882
Test Material: Cassette Encapsulent

Test Name	Project #	Status / Results
MEM Elution-USP	07-1882-G1	PASS – Report Complete
Class 6 (includes implant)	07-1882-G2	PASS - Report Complete
Hemolysis/ extract/ Rabbit Blood	07-1882-G3	PASS – Report Complete

Test Article Number: 07-1885
Test Material: HyStream (HyFlo) PES Membrane

Test Name	Project #	Status / Results
MEM Elution-USP	07-1885-G1	PASS – Report Complete
Class 6 (includes implant)	07-1885-G2	PASS - Report Complete
Hemolysis/ extract/ Rabbit Blood	07-1885-G3	PASS – Report Complete

Note: *Test article identified as BioFlo is ProStream. Test article identified as HyFlo is HyStream.*

7.2 TangenX SC TFF Device Extractables per BPOG Guidance

A controlled extraction study was performed on the TangenX SC TFF Devices (and tubing components) using solvents and extraction techniques across a broad range of polarities. The methodologies utilized were described in the study plan M-TangenX-221006 and results generated are summarized in Study Report 11862.329 v1. BPOG extractables testing was performed by an accredited analytical laboratory. The results from this study were generated under the 2014 BPOG recommended study conditions. They present a worst-case scenario, since neither the temperature nor dissolution properties of the solvents used during this investigation are more aggressive compared to the solvents used during routine component exposure.

Several different TangenX SC TFF Devices were manufactured and evaluated for extractables following the latest guidance outlined by BPOG. Each device was prepared using current SOPs and reflected the standard manufacturing process at Repligen. TangenX SC TFF Devices were prepared and met release criteria following methods established by Repligen.

Test samples were initially received by the contracted laboratory, flushed with purified water to remove the storage solution, then equilibrated with the extraction solution. Extraction of the test samples was performed using 50% ethanol in USP purified water, 0.5 M sodium hydroxide, 0.1 M phosphoric acid, and purified water (WFI). Samples were extracted for 24 hours and 21 days at 40°C. Each device sample was composed of three different lots of membrane forming a composite sample. The test articles were agitated using a rocking table for the entire duration of the extraction. Once the sample time point was reached, the extraction fluid was drained from the device and analyzed for extractables. The following is a summary of the testing performed.

HPLC/DAD/MS was performed on selected component extracts according to the conditions as described in the study plan. All sample extracts were analyzed for antioxidants and additives by HPLC-DAD/MS with the DAD operating at 220 nm wavelength, and the MS operating in ESI (±) and APCI (±) modes. A number of unknown and tentatively identified extractable peaks were identified in all sample extracts. No extractable peaks were identified in the APCI- extracts. Concentrations for all analytes were determined using the response factor for the internal standard for each sample injection. All peaks greater than 0.1 µg/mL that were detected in the sample extracts at levels 3x higher than in the associated method control are reported as extractables. Results are available in the study report 11862.329 v1; summarized in Tables 3 – 12 of that report.

GC/MS was performed on selected component extracts according to the conditions as described in the study plan. All sample extracts were assayed for semi-volatiles by DI-GC/MS. A number of analytes were detected in all sample extracts. Concentrations of BHT were determined using an authentic reference standard. Concentrations of all other analytes were determined using the response factor for the internal standard for each sample injection. Results are available in the study report 11862.329 v1; summarized in Tables 13 – 22 of that report.

Headspace GC/MS was performed on selected component extracts according to the conditions as described in the study plan. All sample extracts were assayed for volatiles by HS-GC/MS. Concentrations of 1,3-di-tert-butylbenzene were quantified using the response from an authentic reference standard. Concentrations of all other analytes were determined using the response factor for the internal standard for each sample injection. Results are available in the study report 1082719-3528; summarized in Tables 23 – 32 of that report.

Induction Coupled Plasma/MS was performed on selected component extracts according to the conditions as described in the study plan. The 21-day 0.1 M phosphoric acid and USP purified water sample extracts were outsourced to SGS North America Inc. for metals analysis by ICP/MS. Results are available in the study report 11862.329 v1; summarized in Tables 33 – 37 of that report.

TOC, pH, and Non-Volatile Residue analysis were performed on selected component extracts according to the conditions as described in the study plan. Results for total organic carbon, pH, and non-volatile residue are provided in Tables 38 – 47 of the study report 11862.329 v1. A summary of the TOC (Table 19), pH (Table 20), and Non-Volatile Residue analysis (Table 21) are provided.

Table 19. TOC results summary

Sample description	Extraction solvent	Results			
		1 day		21 days	
TangenX SC TFF Device	WFI	220 µg C/mL	11.8 µg C/cm^2	687 µg C/mL	36.7 µg C/cm^2
	0.5 N NaOH	263 µg C/mL	14.0 µg C/cm^2	767 µg C/mL	40.9 µg C/cm^2
	0.1 M H_3PO_4	198 µg C/mL	10.6 µg C/cm^2	793 µg C/mL	42.3 µg C/cm^2

Table 20. pH results summary

Sample description	Extraction solvent	Results	
		1 day	21 days
TangenX SC TFF Device	WFI	10.9	10.2
	0.5 N NaOH	13.1	13.1
	0.1 M H_3PO_4	2.0	2.1

Table 21. Non-volatile residue results summary

Sample description	Extraction solvent	Results			
		1 day		21 days	
TangenX SC TFF Device	WFI	494 µg C/mL	25.8 µg C/cm^2	1.59E+03 µg C/mL	84.8 µg C/cm^2
	50% EtOH	1.10 x 10^3 µg C/mL	58.4 µg C/cm^2	3.23 x 10^3 µg C/mL	172 µg C/cm^2

7.2.1 BPOG Acceptance criteria

The extractables testing is compliant when the study has reached its 21-day conclusion. Information gathered will be presented in a report format and reviewed to ensure study protocols were followed. Failure to follow protocols as written will require a deviation to be written in order to justify the validity of the results of the extractables testing.

- Operators must follow approved protocols
- All other test components must perform their function as described in the protocol
- Instrument control test results must be valid

7.3 Endotoxin Test

TangenX SC TFF Devices produced by Repligen are flushed, packaged, stored in 0.2 M sodium hydroxide, and gamma irradiated prior to shipment. The careful preparation of these devices allows them to be used in a biopharmaceutical process following a brief buffer equilibration step. No additional sanitization of the device is required. The following study was conducted to verify that TangenX SC TFF Devices do not contain endotoxin that could potentially contaminate a process stream. This study quantifies the amount of endotoxin flushed from a set of ten (10) TangenX SC TFF Devices ranging from 0.5 – 10 m^2. A specified volume of purified water was used for the flush and then evaluated for endotoxin count by an approved contract lab using the following methods.

General procedure for bacterial endotoxin test (BET)

1. The contents of one (1) container were assayed in duplicate at the neat liquid concentration.
2. An endotoxin standard curve was prepared in duplicate, to a lysate sensitivity of 0.06 EU/mL.
3. Duplicate product controls were prepared containing 0.1 mL of liquid test article mixed with endotoxing standard at twice the indicated sensitivity of the lysate.
4. Water for BET served as the negative control.
5. Lysate (0.1 mL) was added to 0.1 mL of liquid in each tube.
6. All tubes were incubated in a 37 ± 1°C heat block for 60 ± 2 minutes.
7. After incubation, all tubes were examined for agglutination.

Note: *The study and its design employed methodology to minimize uncertainty of measurement and control of bias for data collection and analysis.*

References for BET

1. USP-NF 2023 <85> Bacterial Endotoxins Test
2. ISO 10993-12, 2021 Biological Evaluation of Medical Devices — Part 12: Sample Preparation and Reference Materials
3. ANSI/AAMI ST72: 2019 Bacterial Endotoxins — Test Methods, Routine Monitoring, and Alternatives to Batch Testing

Each size of TangenX SC TFF Devices was manufactured and evaluated in duplicate. Each device was prepared using current SOPs and reflected the standard cassette manufacturing process at Repligen. The device was connected to a purified water outlet source and the retentate and permeate tubing were directed to a sterile media bottle. The device was flushed with purified water through the retentate into the vessel to collect a minimum volume with the permeate valve closed. Flushing continued, opening the permeate valve while closing the retentate valve, and collecting a minimum volume of water through the filtrate. Once the specified volume of purified water was flushed through the device, a pipette was used to transfer the contents to sample vials. The samples were sent to a contract lab for endotoxin testing. The samples were analyzed, and the results reported in Table 22 below. The results of the endotoxin count study show that the level of endotoxin was below the detection limit and below acceptable limits when compared to industry standards (Table 22). Samples were shown to contain less than 0.06 EU/mL of endotoxin. The devices met the acceptance criteria for endotoxin (Table 23).

Table 22. Results of bioburden and endotoxin count study

Sample ID	Area	Collection Volume		EU/ml
		Retentate	Filtrate	
99912348-009	0.5 m^2	500 mL	500 mL	< 0.06
99912348-010	0.5 m^2	500 mL	500 mL	< 0.06
99912349-007	1.5 m^2	750 mL	750 mL	< 0.06
99912349-008	1.5 m^2	750 mL	750 mL	< 0.06
99912350-005	2.5 m^2	1250 mL	1250 mL	< 0.06
99912350-006	2.5 m^2	1250 mL	1250 mL	< 0.06
99912351-003	5 m^2	2500 mL	2500 mL	< 0.06
99912351-004	5 m^2	2500 mL	2500 mL	< 0.06
99945678-001	10 m^2	3250 mL	3250 mL	< 0.06
99945678-002	10 m^2	3250 mL	3250 mL	< 0.06

Table 23. Acceptance criteria

Type	Specification limit
Endotoxin	Result <0.25 EU/mL

7.4 Sterility

The TangenX SC TFF Device was validated for sterility following TIR35 methods of product adoption. A technical review showed that the candidate product and gamma irradiated devices in the TangenX family of products are similar. Therefore, the VDmax25 method (11137: 2006) used to validate sterility of the TangenX product line applies to the TangenX SC TFF Device. This method determines the lowest sterilization dose necessary for the determined bioburden population. Sterility testing was carried out by qualified microbiological experts to verify the effectiveness of a sterilization of the wetted fluid path and to validate removal of microorganisms from the product. Sterility testing was performed to confirm requirements for sterility following exposure to gamma irradiation at 25 – 40 kGy. Furthermore, ANSI/AAMI/ISO 11137-1: 2006 and -2: 2006 address the issue of validation and Quarterly Dose Audits for product validated using the VDmax25 method. Once the sterilization dose is established, periodic audits are performed at a defined and documented frequency. The audits are performed to determine the continued validity of the sterilization dose. The audit will be performed at three-month intervals to detect any changes in the bioburden that could require an augmentation in the sterilization dose.

ANSI/AAMI/ISO 11137: 2006 sterilization dose auditing consists of three major steps.

- Dose Setting - Bacteriostasis/Fungistasis (B/F) & Recovery Efficiency (RE)
- Bioburden testing
- Verification dose experiment

7.5 Dose Setting

The bacteriostasis fungistasis test was performed with selected microorganisms to demonstrate the presence of substances that inhibit the multiplication of microorganisms. This was done prior to a sterility test to assure that the readings of the sterility test are true. This test determined that the product is not leaching anything in the test media during the sterility test resulting in false negatives. Three irradiated samples were required for the B/F test. A validation of recovery efficiency was performed prior to conducting bioburden testing, providing an assessment of the efficiency of the specified extraction technique to remove microorganisms from the device. A correction factor was derived based on the recovery efficiency, and extraction efficiency was determined by using inoculation recovery. Five irradiated samples were required for this method. This was a one-time test for a

product unless a change is made in materials, supplier, product configuration, or other factors that may impact bioburden or dose absorption.

7.5.1 Bioburden Testing

The bioburden testing is the process of determining the population of viable microorganisms on a given sample (product). In this step of the dose audit, 10 samples are taken from a production batch (lot) to determine bioburden count, also known as the number of colony forming units (CFU). The results are then used for comparison with bioburden counts that were determined at the time of the initial validation. If for any reason the bioburden count is significantly higher than the initial bioburden, passing a sterilization dose audit may be unlikely. It is also recommended that a gram stain be performed at the time of the bioburden testing. This performance is helpful in identifying if the microorganisms have changed in type as well as in number.

7.5.2 Verification Dose Experiment

The verification dose experiment was performed to determine whether or not a change in the sterilization dose was needed. The verification dose experiment was performed at the dose determined at the time of validation. The verification dose audits were performed as follows:

- Randomly select 20 samples from a production batch prior to the sterilization phase of production
- Ten of these samples are used for bioburden testing. The bioburden testing of these samples was used for trending purposes only
- The remaining 10 samples were used for the verification dose experiment. The samples were irradiated at the verification dose established at the time of initial validation or the verification dose from the last sterilization dose audit
- The sterility testing was performed on the irradiated samples to determine if viable microorganisms were still present
- A bioburden assessment was conducted for the candidate component(s) and listed in the adoption report M-TANGENX-230701.

7.5.3 Acceptance Criteria

If after results of the sterility test show that one or no positive sterility samples are obtained, the original sterilization dose is considered acceptable, and no action is required. The positive sterility tests are sterility test samples, which exhibit detectable microbial growth after incubation. If after completion of the sterility test two or more positive sterility tests are obtained, the original sterilization dose is not acceptable and further action is required.

A full report for the VDmax25 protocol (21-061-TT) is available upon request and provides additional details of the sterility validation for the SC device. A summary of the results follows in Table 24 through Table 27.

Table 24. Bacteriostasis/Fungistasis(B/F)

Bacteriostasis/Fungistasis (B/F) Testing Summary	
Lot Tested	20023159
Delivered Dose Range (kGy)	27.91 – 31.88
Certificate of Processing RRID#	48771
Laboratory Test Report #	M21083085

No bacteriostasis or fungistasis was observed from the product under the conditions of the test.

Table 25. Recovery Efficiency (RE)

Bioburden Recovery Efficiency (RE) Testing Summary	
Lot Tested	20023159
Delivered Dose Range (kGy)	27.91 – 31.88
Recovery Efficiency (%)	152
Correction Factor	1.0
Certificate of Processing RRID#	48771
Laboratory Test Report #	R21083084

Table 26. Bioburden Determination Summary

Bacteriostasis/Fungistasis (B/F) Testing Summary	
Lots Tested	20023159, 20034247, & 20034246
Average Bioburden (CFU)	<4.0
Verification Dose (kGy)	6.1 ± 10%
Laboratory Test Report #	B21092072, B21101247-1, & B21101247-2

Table 27. Test of Sterility Summary

Bacteriostasis/Fungistasis (B/F) Testing Summary	
Lot Tested	20034246
Verification (Testing) Dose (kGy)	6.1 ± 10%
Delivered Dose Range (kGy)	5.63 – 6.52
Certificate of Processing RRID#	62601
Laboratory Test Report #	S21111886

7.5.4 Conclusion

The fluid path of the gamma irradiated TangenX product line has been validated following ANSI/AAMI/ISO 11137 guidelines for VDmax25 to provide a minimum Sterility Assurance Level (SAL) of 10^{-6} for an established irradiation dose. Sterility testing was carried out by qualified microbiological experts to verify the effectiveness of sterilization within the wetted fluid path and to validate removal of microorganisms. A minimum dose of 25 kGy identified for routine sterilization confirmed a sterility assurance level of 10^{-6} was achieved for the TangenX product family. Quarterly dose audit testing will continue to be conducted on samples and be irradiated at the sterility testing verification dose.

7.6 BSE free materials

Raw materials used in the manufacture of these products have been accepted for use in accordance with standard operating procedures and meet all incoming release criteria. Repligen certifies that the components used in the production of both membranes and filtration cassettes are BSE free. Statements from manufacturers were used to verify new components used in TangenX SC TFF Devices are TSE/BSE free.

The raw materials used in the manufacture of Repligen TangenX membrane and filtration cassettes do contain traces of animal derived material. Process stabilizers required for the production of several of the polymer-based materials are made using stearic acid. This originates from tallow, a rendered form of beef lard.

However, risk is minimized using this tallow-based stabilizer. Tallow derivatives for industrial, cosmetic, or pharmaceutical uses are considered safe with regard to the risk of contracting TSE/BSE when certain inactivation conditions are met. The reasons are as follows:

- The beef tallow used is TSE/BSE free, as the beef tallow is supplied together with a certificate from the authorities responsible, which states that the tallow originates from healthy animals (ante and postmortem).
- The processing conditions meet the requirements of the "Note for Guidance on Minimizing the Risk of Transmitting Animal Spongiform Encephalopathy Agents via Human and Veterinary Medicinal Products" EMEA/410/01 Rev. 3, effective July 1, 2011.
- The above document(s) define an inactivation method and a hydrolysis process of at least 200°C under an approximate pressure for 20 minutes. These conditions are far exceeded in the production of stabilizer as the tallow is hydrolyzed at about 230°C under 30 bars for at least six (6) hours.
- The stearic acid does not come from high-risk countries.

7.7 Particulates

A study was conducted to quantify the particulate count from an initial flush of the TangenX SC Device. The following report outlines steps that were taken to determine the ideal conditions under which to remove the storage solution. Several devices were manufactured and evaluated for particulates following USP NF 2023, <788> Particulate Matter in Injections. The following steps were taken as part of the study:

Three sizes of tubing, corresonding to device size, were selected as controls and were cleaned and prepared by flushing the same volume of water as the specified device. One control was prepared for each device size (0.5 m^2, 2.5 m^2, and 10 m^2). Next, each device was removed from its bag and placed on the table, and the tube sets were connected. The feed tube set was connected directly from the RODI line to the device feed port, and samples were collected from the retentate and permeate outlets. Each device was rinsed by flushing 5 L/m^2 through the retentate and 10 L/m^2 through the permeate. After rinsing the device, a sample of 1 L/m^2 was collected per device (0.5 L/m^2 retentate and 0.5 L/m^2 permeate). Samples were collected in a sterile container and pooled. All flush samples were then labeled for shipment to external vendor for analysis. The particulate count analysis was conducted under USP NF 2023, <788> Particulate Matter in Injections.

Table 28. Acceptance criteria

Type	Particle Size	Specification
Particulate	Particles 10-25 microns	25.0000 per/mL
	Particles >25 microns	3.0000 per/mL

Upon the conclusion of the testing, all the devices passed the particulate testing. A summary of the test data is summarized below.

Table 29. Data summary

Part Number	Sample Number	Particles 10 – 25 microns	Particles >25 microns	Pass/Fail
PP010S05L	20102971-004	6.0500 per/mL	0.3000 per/mL	Pass
PP010S05L	20102971-007	19.0000 per/mL	0.3000 per/mL	Pass
XP300S05L	20102972-002	12.7500 per/mL	0.2000 per/mL	Pass
XP300S05L	20102972-007	12.3500 per/mL	0.8000 per/mL	Pass
PP030S25L	99912346-001	11.5000 per/mL	1.2500 per/mL	Pass
PP030S25L	99912346-002	5.8500 per/mL	0.4500 per/mL	Pass
PP030S99L	99912347-001	7.2000 per/mL	0.7000 per/mL	Pass

In conclusion, the particulate count study showed the test articles meet the test requirements as defined in the USP General Chapter <788> Particulate Matter in Injections.

7.8 Residual solvents

Impurities related to residual solvents are specified under ICH Topic Q3C (R4) Impurities: Guideline for Residual Solvents which outlines acceptable amounts for residual solvents in pharmaceuticals for the safety of the patient. The guideline recommends use of less toxic solvents and describes levels considered to be toxicologically acceptable for some residual solvents.

Residual solvents in pharmaceuticals are defined as organic volatile chemicals that are used or produced in the manufacture of drug substances or excipients, or in the preparation of drug products. The solvents are not completely removed by practical manufacturing techniques. Since there is no therapeutic benefit from residual solvents, residual solvents should be removed to the extent possible to meet product specifications, good manufacturing practices, or other quality-based requirements. Drug products should contain no higher levels of residual solvents than can be supported by safety data.

It is only necessary to test for solvents that are used or produced in the manufacture or purification of drug substances, excipients, or drug product. Although manufacturers may choose to test the drug product, a cumulative method may be used to calculate the residual solvent levels in the drug product from the levels in the ingredients used to produce the drug product. If the calculation results in a level equal to or below that recommended in this guideline, no testing of the drug product for residual solvents need be considered.

Results from the BPOG extractables study were used to determine residual solvent levels in the TangenX SC TFF Device. Both HPLC/MS and GC/MS data detected residual quantities of N-methyl pyrrolidone used in the manufacture of the membrane. The highest levels were detected using the HPLC method after 21 days of extraction (Table 28).

Table 30. Results of residual solvents: HPLC-MS

Device[1]	Result
Solvent detected	1-methyl-2-pyrrolidinone
Highest level detected	14.1 ug/cm^2
	141 mg/m^2
Process loading	200 L/m^2
NMP max concentration	0.71 mg/L
	0.71 ppm
ICH guidelines for NMP	<530 ppm

[1]Highest value reported, 21-day time point in 50% EtOH

N-methyl pyrrolidone (NMP) is a Class 2 solvent and should be limited to 530 ppm in pharmaceutical products. A typical process loading of 200 L/m^2 would result in a maximum concentration of approximately 1 ppm in the batch of drug processed assuming the NMP is not concentrated, diluted, or removed throughout the purification process. This level is below the <530 ppm limit established in the ICH guidelines.

8. Qualification

An IQ and OQ were performed for each piece of critical equipment utilized in the production of the membrane and device assembly. Initial qualifications were performed when a new process or equipment were introduced that was unique in application. Repeat or supplementary qualification activities are performed when a significant change in process, equipment or system are introduced.

8.1 Installation Qualification (IQ)

Verification that the item qualified was installed correctly. The following criteria was considered in each IQ:

- Equipment design features (i.e. materials of construction, cleanability, etc.)
- Installation conditions (wiring, utilities, functionality, etc.)
- Calibration, preventative maintenance, cleaning schedules

- Safety features
- Supplier documentation, prints, drawings, and manuals
- Software documentation
- Spare parts list and inventory
- Environmental conditions (such as clean room requirements, temperature, humidity)

8.2 Operation Qualification (OQ)

Parameters were challenged to assure that outputs met all defined requirements under all anticipated conditions of operations. To establish process control limits, critical parameters and or product characteristics were challenged. These control limits were established and documented to determine the robustness of the process through the spectrum of potential ranges. The following criteria were considered in each OQ:

- Process control limits (time, temperature, pressure, line-speed, setup conditions, etc.)
- Documented procedures and work instructions
- Software parameters
- Raw material specifications
- Process operating procedures
- Material handling requirements
- Potential failure modes, action levels and worst-case conditions (failure mode and effects analysis, fault tree analysis)

Qualifications were performed with a risk-based approach, addressing potential risks associated with the processes, system, analytical method, and/or equipment that were addressed with appropriate risk level and mitigation. Any risk that is identified as major or critical was challenged during qualification and/or validation activities.

8.3 Responsibilities

System Owner: Ensures that the validation parameters, requirements, and the acceptance criteria are properly determined.

Operator/Executor: Ensures protocol is performed as per the approved document and executes the qualification/validation in accordance with the protocol and good documentation practices (GDP).

Technical Lead: Generates draft protocols which are circulated to subject matter experts within the organization for review and approval. Reviews and approves the final validation report to confirm that the process, system, analytical method, and/or equipment are suitable for its intended use.

Quality Assurance: Reviews and approves the validation protocol and approves the final report to confirm that the protocol was properly executed and that any deviations/discrepancies have been addressed and documented.

9. Manufacturing process validation

The objective of process validation is the collection and evaluation of data, from the process design stage throughout commercial production, which establishes scientific evidence that a process is capable of consistently delivering quality product. This involves a series of activities taking place over the manufacturing process. Validation of the membrane manufacturing process was carried out independently as the membrane is used throughout several different product lines at Repligen. The TangenX SC Device process validation was conducted following the Repligen validation program specified in the procedures PV-TANGENX-220910 and PV-TANGENX-230710.

The process validation consists of an evaluation of a defined, documented procedure to consistently deliver an expected result. Process Validation considerations include:

- Actual product and process parameters and procedures established in OQ
- Acceptability of the product
- Assurance of process capability and control as established in OQ
- Process repeatability, long term process stability

Final reports summarize the results of an executed qualification and/or validation protocol and document the supported conclusion based on the data obtained. The report summarizes of all data collection and analysis as specified in the protocol. It also summarizes and discusses all non-conformances, deviations, observations, and data acquired during the execution of the approved protocol for the validation and/or qualification.

9.1 Membrane process validation

The membrane process validation activities were conducted for the specific process of the flat-sheet membrane manufacturing operation at the Repligen Marlborough facility. The process validation applied to the membrane manufacturing process using equipment specified in the master equipment list. The validation report (R-Tangenx-190902) includes a summary of results for the casting solution preparation, membrane casting procedure, post-treatment, drying, slitting, membrane QC testing procedure and their corresponding forms.

The membrane manufacturing process referenced in this validation report consisted of six major parts. The casting solution preparation required individual components of the membrane be dissolved in a solvent creating a casting solution that will be coated onto a non-woven substrate. The membrane casting process used the casting machine to coat the casting solution and form the membrane as it moved through the machine. The extraction post-treatment system flushed the membrane with purified water and removed residual solvents. The glycerin post-treatment system treated the membrane with a preservative. The membrane drying operation dried the membrane, removing water from the pores. During the validation of the membranes, samples were taken once the membrane rolls were processed according to the membrane manufacturing casting SOP-0564. The membrane was cut into 33-inch sheets and retain samples were collected as described in the procedure SOP-0565.

The casting solution data collected for the first three batches of membrane processed during the membrane manufacturing process validation were HyStream 100 kD (XP100) membrane. Each of these membranes met the critical parameters specified in the approved procedure (SOP-0448). Figure 14 summarizes the critical conditions captured for the casting solution used for each lot of the XP100 membranes cast. The table is formatted in a comparable manner to the casting solution specification guide FORM-0449.

The casting solution data collected for the next three batches of ProStream 10 kD (PP010) membrane processed during the membrane manufacturing process validation met the critical parameters specified in the approved procedure SOP-0448. Figure 14 summarizes the critical conditions captured for the casting solution used for each lot of the PP010 membranes cast.

Membranes produced as part of the process validation were tested following a validated test method as instructed in the procedure SOP-0463. Membranes were sampled and tested using 44.5 mm stirred cells. Data collected were compared to release criteria for the following:

- Normalized Water Permeability (NWP)
- Percent deviation of NWP
- Passing solute flux
- Passing solute % rejection
- Retaining solute flux
- Retaining solute % rejection

The test data collected for the membrane processed during the membrane manufacturing process validation was used to confirm the process validation was successful. Figure 14 summarizes and compares the test data generated for each lot of membranes produced. The first row of the table identifies the contents listed in the columns below, release specifications for the HyStream 100 kD membrane are shown in red colored text on the second row of the table. Release specifications for the ProStream 10 kD membrane are shown in red colored text on the sixth row of the table below.

Figure 14. Membrane validation: Data summary

LOT NUMBER	MEMBRANE TYPE	MEMBRA MWCO	PART NUMBER	CAST LENGTH (lin foot)	AVERAGE NWP		PASSING SOLUTE		RETAINING SOLUTE	
					LMH / psi	PERCENT DEVIATION	FLUX	REJECTION	FLUX	REJECTION
					50-140	≤15	305-550	≤20	50-90	≥90
F19220A	Hystream	100	XP100-S2	393.0	55.0	4.7	449.2	3.2	54.8	96.8
F19220B	Hystream	100	XP100-S2	430.0	52.1	3.9	432.5	3.1	58.9	97.1
F19220C	Hystream	100	XP100-S2	404.0	53.8	5.4	470.3	3.9	57.8	96.9
					9.5-22	≤15	140-250	30-60	75-110	≥85
F19220D	Prostream	10	PP010-S2	270.0	14.8	4.3	142.3	34.6	81.2	90.4
F19220E	Prostream	10	PP010-S2	341.0	13.3	9.7	170.4	39.4	79.9	89.7
F19220F	Prostream	10	PP010-S2	312.0	11.8	9.8	163.3	41.4	81.6	91.1

The membrane lot number shown in the first row of the table corresponds with the data set generated for each membrane during testing. The HyStream 100 kD membrane met the critical parameters specified in the approved procedure SOP-0463 and specification guide FORM-0467. The observed NWP was at the lower end of the accepted range but is consistent with 100 KD membranes currently produced using the original membrane equipment being replaced. The passing solute flux rate and percent rejection are within the accepted range and ideally fell into the middle of the specification. The retaining solute flux rate and percent rejection were within the accepted range and showed each membrane batch met the specifications for a 100 kD membrane. Each of the ProStream 10 kD membranes met the critical parameters specified. The NWP was within the accepted range and ideally fell into the middle of the specification. The passing solute flux rate and percent rejection were also within the accepted range for a 10 kD membrane. The retaining solute flux rate and percent rejection were within the accepted range and showed each membrane batch met the specification for a 10 kD membrane.

Three consecutive lots of two different membrane types were manufactured as part of the membrane process validation. The validation was considered successful as three lots of each membrane type were in conformance with the defined specification. Two membrane types, HyStream 100 kD and ProStream 10 kD were each validated and were identified to represent the TangenX membrane product line. Each membrane batch was manufactured in accordance with approved procedures. Each of the membrane lots was shown to meet product specifications following procedure SOP-0463 and found to be within compliance. The membrane validation is complete and the membrane manufacturing process at the Repligen Marlborough site is considered validated.

9.2 TangenX SC Device process validation

This section of the document describes the process validation report for the TangenX SC TFF Device manufacturing operation at the Repligen Marlborough facility. The validation included approved procedures for device assembly, device testing, part assembly, part washing, device assembly, integrity testing, bagging, packaging, and gamma-irradiation with their corresponding forms. The device assembly process has met the acceptance criteria (Table 33) and is currently a validated process.

This summary defines the validation within the Repligen Marlborough facility for the TangenX SC TFF Device manufacturing process. Six (6) batches consisting of a total of fifty-four (54) devices were produced during the process validation. The matrix includes sets of 0.5 – 10 m^2 representing the full TangenX SC TFF Device product line. The membrane type was varied but is not specific as the device assembly process is the same. Each membrane chemistry was evaluated (ProStream and HyStream); tight and open pore sizes were used (10, 50, and 300 kD) to represent the full range. This process validation is per DHF0044, and the data generated shows conformance to the process validation protocols, PV-TANGENX-220910 AND PV-TANGENX-230710.

Responsibilities for the validation fall upon product engineering to write the report and ensure that the report meets the requirements stated the Repligen process validation protocols. Engineering is also responsible for ensuring that the validation is performed in compliance with the protocol and with any designated SOPs. Drafting, reviewing, and approving validation protocols and/or reports and all associated data also falls on the engineering group. Manufacturing supports validation activities, generating and providing supporting data. Manufacturing also ensures access to necessary raw materials, utilities, and resources for execution. Quality is responsible for reviewing and approving validation protocols and/or reports and all associated data. Quality is also

responsible for maintaining records of executed and completed validation protocols and/or reports and all supporting documentation.

Prerequisites include evidence that all raw materials used in the validation were approved and released by quality assurance. All specifications must be written and approved. All the equipment used during the process validation must be qualified. The critical equipment must be calibrated and recorded in their corresponding logbooks. Protocols, process parameters to be controlled/calibrated can be referenced in approved IQ/OQ test protocols.

The individual procedures were combined and executed as two validation groups where six (6) consecutive serialized batches of devices were manufactured. Each of the membranes used in the devices were individually tested according to the approved TangenX SC Cassette QC Testing SOP-3188. Each device was tested in the device QC test area for liquid volume flow rate and air mass flow rate. Completed assemblies were tested for leaks and a final visual inspection per the assembly procedure SOP-3194.

Following the validation, Quality Assurance conducted a review of the test data, verifying the adherence to set specifications. Quality Assurance was responsible for the final review of the executed validation procedures and test results. The test results for each device are found below in Table 29.

Table 31. Device validation: Data summary

Catalog #	Membrane chemistry	Area	MWCO	Batch	Qty tested	Qty passed	Test yield	Target Yield	Result
PP010S05L	ProStream	0.5 m^2	100 kD	20102971	10	8	80%	>75%	Pass
XP300S05L	HyStream	0.5 m^2	300 kD	20102972	10	8	80%	>75%	Pass
XP050S15L	ProStream	1.5 m^2	50 kD	20102973	10	10	100%	>75%	Pass
XP050S25L	ProStream	2.5 m^2	50 kD	20104359	9	7	78%	>75%	Pass
XP300S50L	ProStream	5.0 m^2	300 kD	20104360	9	9	100%	>75%	Pass
XP050S99L	ProStream	10 m^2	50 kD	20105103	6	5	83%	>75%	Pass

All devices passed testing per criteria per Table 33 and final inspection defined in in the procedure SOP-3196. All validation documentation was successfully completed with a process yield >75%, per the validation protocol PV-TANGENX-220910 and PV-TANGENX-230710. The TangenX SC Device assembly process was considered validated and released for production usage with the completion of the Design History File, DHF0044 requirements.

10. Release testing

10.1 Analytical method validation

Selective analytical methods for the quantitative evaluation of membrane and membrane-based products are necessary for the QC release of these devices. Analytical method qualification includes all the procedures that demonstrate that a particular method used for quantitative measurement of samples in a given matrix is reliable and reproducible for the intended use. The fundamental parameters for qualification include specificity, linearity, accuracy, precision, and robustness.
Method validation involved documenting that the performance characteristics of the methods were suitable and reliable for the intended applications. The acceptability of analytical data corresponds directly to the criteria used to qualify the method. Specific, detailed descriptions of the analytical methods were written in the form of a standard operating procedure for both membrane and device QC testing. Each step in these methods was investigated to determine the extent to which environmental, matrix, or procedural variables can affect the estimation of material in the matrix.

In the case of sensitive quantitative procedures such as these, appropriate steps were taken to ensure the lack of matrix effects throughout the application of the method. These analytical methods were validated for the intended use of membrane characterization and release. All experiments used to make claims or draw conclusions about the validity of the method are presented in a method qualification report. In-process test methods include both membrane and device QC methods.

10.2 Membrane QC method validation

The purpose of the membrane QC testing method validation was to validate the membrane QC testing procedure. This procedure refers to ultrafiltration and microfiltration membranes manufactured by Repligen. Membranes are initially manufactured and then tested for performance prior to being incorporated into a product. A report summarizing the verification of specificity, linearity, accuracy, precision, and robustness of the membrane QC test procedure was written. Minimum requirements, including acceptance specifications for the methods, were set during the method development and validation cycle. The acceptance criteria are found in each of the data sheets found in the body of the report.

The principles followed for the membrane QC method validation were based on cGMP guidelines and helped Repligen ensure the test method was acceptable for use. The membrane QC procedure is used to verify membrane water permeability and protein rejection. This information is then used to accept or reject the membranes manufactured at Repligen. At the conclusion of the validation, it was proven that membrane QC method meets requirements set by Repligen for specificity, linearity, accuracy, precision, and robustness. Minimum requirements, which were essentially acceptance specifications for the methods, were met during the method development and validation cycle and the QC membrane test procedure considered validated.

10.3 Cassette QC method validation

TangenX SC Devices incorperate a TFF cassette within the device and are individually QC tested prior to device assembly. The purpose of the cassette QC testing method validation was to validate the cassette QC testing procedure. This procedure refers to ultrafiltration and microfiltration cassettes manufactured by Repligen. The cassettes are initially manufactured and then tested for performance prior to being released as final product. A written report summarizes the verification of specificity, linearity, accuracy, precision, and robustness of the cassette QC test procedure. Minimum requirements including acceptance specifications for the methods, were set during the method development and validation cycle. The acceptance criteria are found in each of the data sheets found in the body of the report. The procedure used for the method validation was described in the validation protocol listed the steps that were followed during the validation.

The principles followed for the validation were based on cGMP guidelines and helped Repligen ensure the cassette QC test method was acceptable for use. The cassette QC procedure was used to verify each cassette’s air diffusion and cross flow rate. This information is then used to accept or reject the cassettes manufactured at Repligen. At the conclusion of the validation, it was proven that cassette QC method meets requirements set by Repligen for specificity, linearity, accuracy, precision, and robustness. Minimum requirements including acceptance specifications for the methods were met during the method development and validation cycle and validation cycle and the QC membrane test procedure considered validated.

10.4 Release specifications

Table 33 presents the release specifications for both membrane chemistries, as taken from document FORM-0467. A complete set of TangenX SC Device release specifications are listed in Table 30 through Table 33, as taken from document SOP-3194.

- Membrane QC Release Specifications: FORM-0467
- Device Release Specifications: SOP-3194
- Part Number Suffix: S1 = 12.5" width, S2 = 18.0" width

Table 32. Membrane QC release specifications: NWP (10 kD – 300 kD)

Part Number	MWCO (Daltons)	Normalized Water Permeability[1]		
		Water flux (LMH/psi)	Deviation %	Pressure
PP010 / XP010	10 K	9.50 – 22.0	≤15%	50 psi
PP030 / XP030	30 K	30.0 – 50.0	≤15%	50 psi
PP050 / XP050	50 K	45.0 – 75.0	≤15%	50 psi
PP100 / XP100	100 K	50.0 – 140	≤15%	10 psi
PP300 / XP300	300 K	190 – 300	≤15%	10 psi

[1]NWP test measurements are performed at the specified applied air pressure. Latex bead solutions are tested with no applied air pressure; head pressure is approximately 5.5 in H_2O (140 mm H_2O)

Table 33. Membrane QC release specifications: Specificity (10 kD – 300 kD)

Part Number	MWCO	Selectivity									
		12 18 kD C15		45 kD C30		67 kD BSA		167 kD IgG		2000 kD Blue Dextran	
		Flux	%Rej.	Flux	%Rej.	Flux	%Rej.	Flux	%Rej.	Flux	%Rej.
PP010 XP010	10 K	140 – 250 LHM	30 – 60	75 – 110 LMH	≥85						
PP030 XP030	30 K			80 – 110 LMH	60 – 80	120 – 170 LMH	≥95				
PP050 XP050	50 K			95 – 150 LMH	30 – 60	140 – 200 LMH	≥75				
PP100 XP100	100 K					305 – 550 LMH	≤20	50 – 90 LMH	≥90		
PP300 XP300	300 K							≥530 LHM	≤15	65 – 110	≥90
Test Solution Concentration		1.0 g/L		1.0 g/L		1.0 g/L					
Test Solution Buffer[1,2,3]		DI water		DI water		PBS pH 7.4					
Spectrophotometer Measurement Technique											
Mode / Wavelength		Absorbance @ 214 nm		Absorbance @ 214 nm		Absorbance @ 280 nm					
Optical Path		NanoDrop(R) Pedestal		NanoDrop Pedestal		10 mm cuvette					

Table 34. Membrane QC release specifications: Integrity testing (10 kD – 300 kD)

Part Number (4)	MWCO (Daltons)	Integrity Total # of discs positive with air diffusion 1K – 300 K @ 15 psi; 0.1 µm – 0.65 µm @ 3 psi
PP010 / XP010	10 K	≤6 (of 18 Discs)
PP030 / XP030	30 K	≤6 (of 18 Discs)
PP050 / XP050	50 K	≤6 (of 18 Discs)
PP100 / XP100	100 K	≤6 (of 18 Discs)
PP300 / XP300	300 K	≤6 (of 18 Discs)

Table 35. Device QC Release Specifications

Device Format	Area	Pressure Hold	Pressure Drop	Air Integrity Pressure	Air Flow Rate
S05	0.5 m^2	25 psi	0.05 psi/min	7.5 psi	<150 ccm
S15	1.5 m^2	25 psi	0.05 psi/min	7.5 psi	<450 ccm
S25	2.5 m^2	25 psi	0.05 psi/min	7.5 psi	<750 ccm
S50	5.0 m^2	25 psi	0.05 psi/min	7.5 psi	<1,500 ccm
S99	10.0 m^2	25 psi	0.05 psi/min	7.5 psi	<3,000 ccm

10.5 Certificate of conformance

Figure 15 shows an example of the standard Quality Assurance Certificate provided with each TangenX SC TFF Device manufactured by Repligen. A specific product part number, serial number, and description will be included on the label attached in the upper left corner of the certificate.

Figure 15. QA Certificate of conformance for TangenX SC TFF Device

REPLIGEN
INSPIRING ADVANCES IN BIOPROCESSING

Repligen Corporation
111 Locke Drive
Marlborough, MA 01752
Phone: 508-845-6400
Fax: 508-845-3050

Quality Assurance Certificate

This is to certify that the TangenX® SC Device as indicated by the affixed label complies with the following descriptions and specifications:

Product Quality – TangenX® SC Device

This product has been manufactured in a fully validated and documented manufacturing process under an ISO 9001:2015 quality management system.

This product was manufactured and tested according to standard operating procedures and was found to meet all release criteria. This article will perform according to the manufacturer's published specifications when used according to the manufacturer's recommendations.

REPLIGEN
TangenX® SC Device
xxStream
www.repligen.com
Marlborough, MA USA

Use By: 09-Mar-2024
DOM: 09-Sep-2022
PN: xx kD
LN: 5678 L Screen
SN: 5678-001 5.0 m

MED-18 EXPOSED

100% Release Testing

Each membrane lot is inspected prior to incorporation into a cassette. Before assembly, the membrane used in each cassette is tested for flow rate, retention, and physical specifications.

Each cassette has been flushed with D.I. water, sanitized with 0.2M NaOH, and individually tested against the following performance specifications:

1. Hydraulic performance - a measure of the cross-flow rate at a specified pressure drop.
2. Integrity - a measure of the rate of air diffusion through the cassette at a specified pressure differential.

The minimum requirements for each test was set by our Quality Assurance Department.
Each cassette has been gamma irradiated to 25.0 – 40.0 kGy and verified following procedure SOP-0568.

Validation

All component materials were shown to meet:

1. USP Class VI biological test for plastics.
2. Certifications for BSE/TSE free components.

All component materials used in cassettes manufactured by Repligen were tested for USP safety and were shown to be safe according to:

1. Class VI per USP <88>.

All finished component materials were tested under controlled conditions for extractables following BPOG guidelines.

1. 4 Model Solvents
2. 21 Day Extraction
3. 40°C Extraction Temperature

A representative of population TangenX® SC Devices were tested for:

1. Endotoxin testing following references the limulus amebocyte lysate (LAL) test as an end product. Acceptance criteria is specified as < 0.25 EU/ml as determined by the LAL test method.

Signature Required:
Reviewed and approved for accuracy and completeness.
Paul Wallace, Director of Quality
Signature and Title

Document Number: QS-4431 Revision: 01 Effective Date: DD/MM/YYYY Legacy Document #: N/A
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11. List of TangenX Cassette and Device studies

1. TX1001-POQ-117-R Protein Binding Study
2. 10827-19-3528 SC Gamma Cassette & Tubing Assembly Extractables Assessment
3. TX1001-POQ-159-R SC Gamma Cassette Robustness Study
4. TX1001-POQ-125-R Membrane QC Testing Method Validation
5. TX1001-POQ-132-R Cassette QC Testing Method Validation
6. TX1001-POQ-164 SC Gamma Cassette Shelf Life Study
7. R-TANGENX-190902 Membrane Validation
8. PV-TANGENX-220910 Process Validation Report — SC Gamma Manufacturing (0.5 – 5 m^2)
9. PV-TANGENX-230710 Process Validation Report — SC Gamma Manufacturing (10 m^2)

12. References

1. Class VI Test per USP <88> Includes: Systemic Injection, Intracutaneous Injection, and 7-Day Muscle Implantation.
2. ANSI/AAMI/ISO 11137-1. 2006/(R) 2010 & A1:2013 Sterilization of health care products — Radiation — Part 1: Requirements for development, validation, and routine control of a sterilization process for medical devices.
3. ISO/IEC 17025, 2017, General requirements for the competence of testing and calibration laboratories.
4. USP 42, NF 37, 2019 <85> Bacterial Endotoxin Test, USP current revision, <161> Medical Devices — Bacterial Endotoxin and Pyrogen Tests.
5. BPOG - Best Practices Guide for Evaluating Leachables Risk from Polymeric Single Use Systems Used in Biopharmaceutical Manufacturing: 2017; Sexton, Aidan W., et.al.
6. Standardized Extractables Testing Protocol for Single-Use Systems in Biomanufacturing: 2014; Weibing Ding, Gary Madsen, Ekta Mahajan, Seamus O'Connor, Ken Wong.
7. EMA Note for guidance on minimising the risk of transmitting animal spongiform encephalopathy agents via human and veterinary medicinal products (EMA/410/01 rev.3).
8. ICH Topic Q3C (R4) Impurities: Guideline for Residual Solvents.

13. Index

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