Validation Guide

PVDF Hydrophilic Filter Cartridges 0.45 µm

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1.Introduction

1.1 Product Description

Our hydrophilic PVDF 0.45μm filter cartridges are sterilize grade cartridge filters manufactured from hydrophilic PVDF membrane and polypropylene hardware providing high flow rates and throughputs, low extractable, broad chemical compatibility. These cartridge filters are 100%integrity tested during manufacturing to assure sterilizing-grade performance.

The hydrophilic PVDF 0.45μm filter cartridges are designed for removal of particles and the sterile filtration of aqueous solutions.

The hydrophilic PVDF 0.45μm filter cartridges are designed,developed and manufactured in accordance with an ISO 9001:2015 certified quality management system. These filters are manufactured in a controlled environment that meets the air quality standards of an ISO class 8 room with respect to viable and nonviable particulate and positive pressure.

1.2 Materials of Construction

1.2.1Component materials used in this product meets the FDA Indirect Food Additive requirements cited in 21 CFR 177-182.

Items	Material
Filter media	hydrophilic PVDF membrane
Support	polypropylene
Cage	polypropylene
Core	polypropylene
End Caps	polypropylene
O-ring	silicone

1.2.2Component materials used in this product meets the regulation(EC)No.1935/2004.

Items	Material
Filter media	hydrophilic PVDF membrane
Support	polypropylene
Cage	polypropylene
Core	polypropylene
End Caps	Polypropylene

1.3 Ordering Information

Application	Type	Material	Micron	Length	Adaptor	Sealing	R
D- Dorsan Biopharma Food&Beverag e	PVDFPHIL Pleated Filter	PVDF	0.45μm	5″ 10″ 20″ 30″ 40″	E2-222/Flat with SS insert E3-222/Fin with SS insert E7-226/Fin with SS insert E6-226/Flat with SS insert	S S-Silicone E-EPDM N-NBR V-Viton F-PTFE F-Encapsul Viton K-Encapsul Silicone	

The Catalogue Number is shown as below:

Adaptor No.	Description	Top	Bottom
BLANK SPACE	DOE		
E7	226/Fin with SS insert		
E6	226/Flat with SS insert		
E2	222/Flat with SS insert		
E3	222/Fin with SS insert		

2. Validation Item

Retention	3.1 BCT (ASTM F838-20)	3.2 Integrity Test
Flow Rate	3.3 Flow Characteristic	
Durability	3.4 Maximum Operating Conditions	3.5 Steam Sterilization
Cleanness	3.6.1 Gravimetric Extractables	3.6.2 Non-Fiber Releasing
Biological	3.7.1 Biological Safety	3.7.2 Bacterial Endotoxin
Chemical	3.8 Chemical Compatibility	

3.Test Methods and Results

3.1Correlation of non-destructive integrity testing to liquid bacterial challenge with *Serratia marcescens*(ATCC 14756) for sterilize grade filters

3.1.1 Introduction

The FDA guidelines on Sterile Products Produced by Aseptic Processing (2004) state, "A sterilizing filter is one which, when challenged with the micro-organism *Serratia marcescens*, at a minimum concentration of 10^7 organisms per cm^2 of filter surface, will produce a sterile effluent".

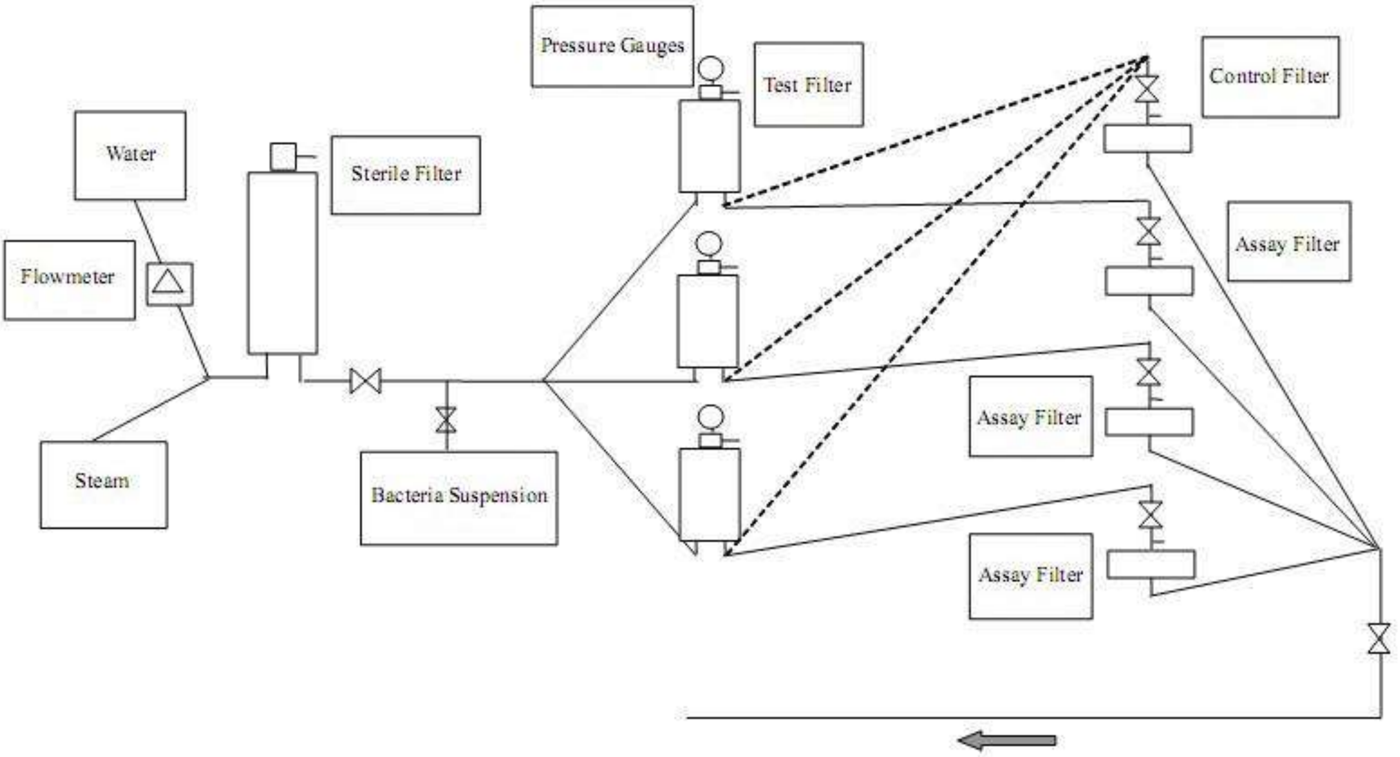
In order to meet the requirements of this guideline, liquid challenge tests using *Serratia marcescens* (ATCC 14756) were performed with hydrophilic PVDF 0.45μm filter cartridges using a minimum of $1x10^7$ colony forming units (CFU)/cm^2 of effective filtration area.

The correlation between microbial retention and a non-destructive integrity test is also an important aspect of the validation of sterilizing grade filters. The FDA guideline further states, "After a filtration process is properly validated for a given product, process and filter, it is important to assure that identical filter replacements (membrane or cartridge) used in production runs will perform in the same manner. One way of achieving this is to correlate filter performance data with filter integrity testing data". The integrity tests used during this validation study were the Forward Flow and Bubble Point tests.

The Forward Flow Integrity Test
In the Forward Flow test, a filter is wetted with a suitable test liquid and a pre-determined gas pressure is applied to the upstream side of the filter assembly. After a stabilization period, the gas flow through the wetted membrane can be measured manually on the downstream side or on the upstream side, using sensitive flow measurement equipment such as the integrity test devices.

The Bubble Point Integrity Test
In the Bubble Point test, a filter is wetted with a suitable test liquid. After a stabilization period, increasing the gas pressure to the upstream side of the filter assembly, using sensitive flow measurement equipment such as the integrity test devices to test the change point of the gas flow rate.

3.1.2 Summary of Methods



Hydrophilic PVDF0.45μm filter with different batch lots were subjected to microbial challenge tests using an aqueous suspension of *Serratia marcescens*(ATCC 14756).

Prior to the challenge tests the filters were installed in an appropriate housing, flushed with DI water at a flow rate of 4 L/min for 15~20 minutes, and then autoclaved at 121 °C for 30 minutes. The filter assembly was then aseptically connected to a pre-sterilized challenge apparatus.

A Forward Flow integrity test was then performed using an integrity test instrument with an air test pressure of 0.12Mpa (17.4 psi).

An aqueous suspension of *Serratia marcescens* was passed through the filter to achieve a challenge level of > 1 x 10^7 colony forming units (CFU) per cm^2 of effective filtration area.

During the challenge test, the entire filter effluent was passed through a 0.45 μm-rated analysis disc on the downstream side of the test filter assembly. The filter disc was incubated on TSA and following incubation, the disc was examined to determine if any colonies had grown, indicating whether or not bacteria had passed through the test filter during the challenge.

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3.1.3 Test Results

Table listed below indicates hydrophilic PVDF 0.45μm that are forward flow tested before and after bacterial challenge. The bacterial challenge was conducted using ASTM F838-20 to provide the necessary correlation between a bacterial challenge and a non-destructive integrity test. The table also indicates that a hydrophilic PVDF 0.45μm filter with Forward Flow ≤16.0 mL/min/10"@ 1.2bar at 20℃ has sterile filtration efficiency when challenged with > 1 x 10^7 CFU per cm^2 of filtration area using *Serratia marcescens*.

Filter type: Hydrophilic PVDF 10", 0.45μm
Challenge organism: *Serratia marcescens*(ATCC 14756)

Filter Serial Number	Forward Flow mL/min	Challenging bacteria(CFU/10inch)	Sterile Effluent	LRV/ cm^2
402200511001	8.5	$1.65*10^{11}$	Yes	7.43
403060211009	8.8	$2.05*10^{11}$	Yes	7.53
402200511003	8.9	$2.49*10^{11}$	Yes	7.61
402200511002	9.2	$2.05*10^{11}$	Yes	7.53
403120111004	9.3	$1.53*10^{11}$	Yes	7.40
403120111005	9.3	$1.53*10^{11}$	Yes	7.40
403060211004	9.6	$1.65*10^{11}$	Yes	7.43
403060211003	10.1	$1.95*10^{11}$	Yes	7.51
403120111008	10.2	$1.85*10^{11}$	Yes	7.48
403120111001	10.2	$2.49*10^{11}$	Yes	7.61
402200511004	10.5	$1.95*10^{11}$	Yes	7.51
402200511005	10.6	$2.49*10^{11}$	Yes	7.61
403060211005	10.8	$1.77*10^{11}$	Yes	7.46
403060211001	11.2	$2.31*10^{11}$	Yes	7.58
403120111003	11.4	$2.12*10^{11}$	Yes	7.54
403120111009	11.8	$1.65*10^{11}$	Yes	7.43
402200511007	12.1	$1.85*10^{11}$	Yes	7.48
403060211002	12.5	$2.05*10^{11}$	Yes	7.53
403060211006	12.6	$2.31*10^{11}$	Yes	7.58
402200511008	13.1	$2.82*10^{11}$	Yes	7.67
403120111002	13.4	$1.19*10^{11}$	Yes	7.29
403120111006	13.5	$1.65*10^{11}$	Yes	7.43
403060211008	13.7	$2.05*10^{11}$	Yes	7.53
403060211012	14.8	$2.82*10^{11}$	Yes	7.67
402200511009	16.2	$2.28*10^{11}$	Yes	7.57
402200511006	16.8	$1.65*10^{11}$	Yes	7.43
402200511011	17.1	$2.28*10^{11}$	Yes	7.57
403120111010	18.9	$2.82*10^{11}$	No	<7
403060211010	20.5	$2.28*10^{11}$	No	<7
403120111012	22.4	$2.05*10^{11}$	No	<7

3.1.4 Conclusions

A typical hydrophilic PVDF Series 0.45μm filters from production, 27pcs were found to pass the forward flow integrity test. The table also indicates that hydrophilic PVDF 0.45μm filter with Forward Flow ≤17.1 mL/min/10"@ 1.2bar at 20℃ has sterile filtration efficiency when challenged with > 1 x 10^7 CFU per cm^2 of filtration area using *Serratia marcescens*.

Analysis in base of results of forward flow, tested filter ≤16mL/min gave sterile effluent when challenged with LRV>7 of *Serratia marcescens*.

3.2Integrity Test Standard(20±5℃)

In base of test results of validation test, forward flow is approved as suitable test methods for hydrophilic PVDF 0.45μm filter cartridges. We also test the bubble point before and after bacterial challenge. The hydrophilic PVDF 0.45μm filter with bubble point ≥1.6bar at 20℃ has sterile filtration efficiency.

The Integrity test value standard is as below:

Wetting Liquid	DI Water	
Temperature	20±5℃	
Test Gas	Air	
Allowable Integrity Value limit	Forward Flow:	≤16mL/min@1.2bar
	Bubble Point:	≥1.6bar

3.3 Flow Characteristic(20±5℃)

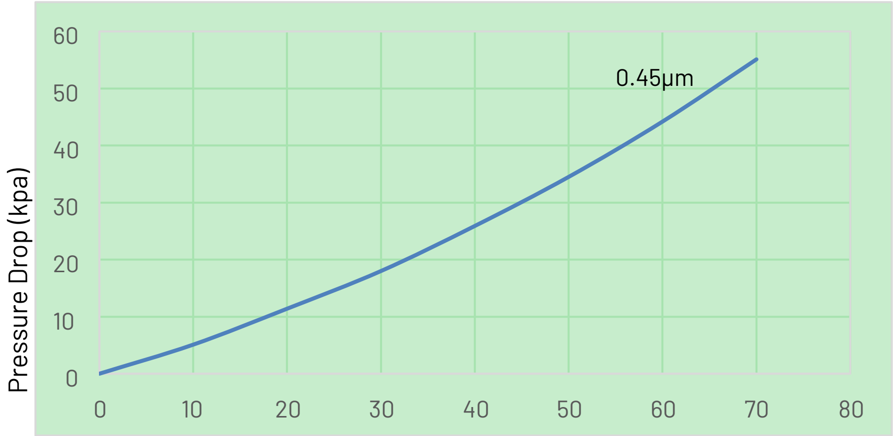
3.3.1 Summary of Methods

Typical hydrophilic PVDF 0.45μm filter Cartridge from production were used for the tests. The filters were flushed with DI water@1.0bar for15-20mins. Then passed forward flow in base of 1.2bar air test pressure.

Standard production hydrophilic PVDF 0.45μm filter cartridge (10", EFA 0.6m^2) were installed with flow rate test system device, which adjust the flow rate and pressure drop by auto-valve. Then take the continuous records with flow rate, upstream & downstream pressure value, temperature etc.

3.3.2 Test Results

Here is the flow chart of filter cartridge.



Flow Rate@20℃（L/min）

3.3.3 Conclusions

These data can be used to assist users in sizing filter systems.

3.4 Maximum Operating Conditions

3.4.1Summary of Methods

Typical hydrophilic PVDF 0.45μm filter Cartridge from production were used for the tests. The filters were flushed with DI water@1.0bar for 15~20mins. Then passed forward flow in base of 1.2bar air test pressure.

Standard production hydrophilic PVDF 0.45μm filter cartridge(10", EFA 0.6m^2) were installed with pressure drop test system device, which adjust the upstream and downstream pressure by auto-valve.

Use ISO Standard powers to increase the pressure drop till 5.0bar ,temperature 25℃and keep the pressure for 30mins, total 4 cycles.

Use Standard test dust to increase the pressure drop till 2.0bar,temperature 80℃ and keep the pressure for 30mins, total 3 cycles. Then test the integrity.

3.4.2 Test Results

Filter Serial Number	Integrity Test（Before the pressure test）		Integrity Test（After the pressure test）	
	Forward Flow(mL/min@1.2bar)	Bubble Point(bar)	Forward Flow(mL/min@1.2bar)	Bubble Point(bar)
403060211011	11.9	1.846	11.7	1.890
402200511012	12.2	1.925	12.5	2.005
402200511015	13.7	1.887	14.1	1.827
403120111007	10.8	2.012	11.2	1.989
403060211016	11.2	1.968	10.9	1.924

The Integrity test value standard is forward flow≤16mL/min@1.2bar,bubble point≥1.6bar,test temperature 20±5℃.

3.4.3 Conclusions

Here is the Max. pressure drop and temperature of filter cartridge

Maximum Differential Pressure (Forward)	Maximum Operating Temperature
5.0bar @ 25 ℃ 2.0bar @ 80 ℃	80°C

3.5 Steam Sterilization(Thermal Stability)

3.5.1 Summary of Methods

The purpose of these tests was to determine the effects of repeated exposure to in-line steam or autoclave cycles on filter integrity using standard hydrophilic PVDF 0.45μm filters from production. The filters were flushed with DI water @1.0bar for 15~20mins. Then passed forward flow in base of 1.2bar air test pressure.

During this autoclave study, filters were steamed using high initial differential pressures (1.0bar (14.5 psi)) at 125°C (257°F). The tests were performed in 30 minutes cycles in the forward (out to in) direction.

During the on-line steam sterilization,These filter cartridges were installed in stainless steel housing and steamed in place in the forward (out to in) direction using saturated steam at constant pressure and flow while ensuring effective condensate drainage. After each steam-in-place cycle the filters were cooled by passing dry compressed air through them.

Integrity tests maintained integrity after Sterilized in place and Autoclave to determine the ability of the filter to provide a sterile filtration.

3.5.2 Test Results

Forward Flow (mL/min) after the following Number of 30 minutes Steam Cycles.

Serial No.	0Cycles	10cycles	30cycles	50cyecles	100cycles	150cycles
402200511016	10.5	10.7	10.3	11.1	11.2	11.3
402200511017	12.7	12.8	12.4	13.0	12.9	12.6
402200511018	10.7	11.1	10.8	10.9	11.5	11.2
403060211013	11.6	11.4	12.2	12.4	11.8	12.2
403060211014	13.9	13.1	13.8	13.9	13.2	13.1
403060211015	11.9	11.7	12.1	12.6	11.9	12.1
403120111011	13.5	13.7	13.6	13.2	13.1	13.8
403120111013	12.6	12.8	12.9	11.5	12.9	13.1
403120111014	12.4	12.2	12.1	12.8	12.1	12.8
403120111015	12.9	12.7	12.8	13.1	12.8	12.6

The Integrity test value standard is forward flow≤16mL/min@1.2bar,test temperature 20±5℃.

Forward Flow (mL/min) after the following number of 30 minutes on-line steam cycles

Serial No.	0Cycles	10cycles	20cycles	30cyecles	40cycles	50cyecles
403060211016	12.9	13.3	12.5	13.2	13.5	13.1
403060211017	13.6	13.7	13.5	13.8	13.5	13.9

403120111017	11.8	11.7	11.1	11.9	11.1	11.9
403120111018	14.1	14.0	14.5	14.6	14.5	14.6
402200511019	12.2	12.9	12.3	12.6	12.3	12.6

The Integrity test value standard is forward flow≤16mL/min@1.2bar,test temperature 20±5℃.

3.5.3 Conclusions

Hydrophilic PVDF0.45μm filters have been demonstrated to be capable of withstanding multiple in-line steam/autoclave sterilization cycles.

The data presented in this section support the following product claims for in-line steaming/autoclaving hydrophilic PVDF0.45μm filter cartridges:

Sterilized in Place				**Autoclave**		
Temp.	Time	ΔP	Cycles	Temp.	Time	Cycles
135℃	30min	30kPa	50	125℃	30min	150

3.6 Cleanness

3.6.1Gravimetric Extractable

3.6.1.1Summary of Methods

Typical hydrophilic PVDF0.45μm filter Cartridge from production were used for the tests.

Preparation of Filter Samples

Extractables tests were performed on typical production filter cartridges (10inch, 0.6m^2), which had been autoclaved in order to maximize the quantity of any extractable material present. The filters were wrapped in aluminium foil and autoclaved for half hour at 121°C, using a slow exhaust cycle. Visible droplets of water remaining on the filter elements were allowed to evaporate at room temperature before the extraction was performed.

Extraction Procedure

Dynamic extraction tests were performed. The test filters were immersed in 1800 ml of extraction fluid in a clean measuring cylinder for 24 hours. For four hours the filter was gently moved up and down. This movement created flow through the filter membrane as a result of the pressure head that was created each time the element was partially lifted out of the liquid.

Analysis of Material Extracted

After the extraction, 1500mL of the extraction liquid was evaporated to dryness and the non-volatile extractable were determined gravimetrically.

3.6.1.2 Test Results

Extraction Fluid	Filter serial number	Gravimetric Extractable (milligrams per Filter)	Average(milligrams)
DI Water	402200511021	13.1	14.4
	402200511022	15.4	
	403060211018	14.8	

3.6.1.3 Conclusions

The extractable determined of hydrophilic PVDF0.45μm filter cartridge were depended by different solvent. The extractable levels under different solvents, different solubility, different temperature and different contact time are not consistent, so it is recommended to test under actual process conditions.

3.6.2 Fiber Releasing

3.6.2.1 Summary of Methods

Typical hydrophilic PVDF 0.45μm filter Cartridge from production were used for the tests.

Filters were autoclaved for one cycle of 30 minutes, 121℃, and then flushed with a total of 10 liters of 0.1 μm filtered water at a flow rate of 1 L per minute. The filtrate was passed through a 0.65μm black gridded disc filter to collect any fibers removed from the filter. Filters were then integrity tested to verify that only integral filters were used in the test.

3.6.2.2 Test Results

hydrophilic PVDF0.45μm Filter – Fiber Shedding Results		
Filter serial number	Number of Fibers in filtrate	Forward Flow(mL/min/10"@ 1.2bar,20 ℃)
403120111019	0	12.1
403120111020	0	13.5
403060211019	0	12.6

The Integrity test value standard is forward flow≤16mL/min@1.2bar,test temperature 20±5℃.

3.6.2.3 Conclusions

The hydrophilic PVDF0.45μm filter cartridge from production don't have Fiber releasing and meet the request of FDA 21 CFR 210.3(b)(6).

3.7 Biological Safety

3.7.1 Biological Tests

3.7.1.1 Summary of Methods

These filters are non-toxic per USP Class VI Biological Tests forPlastics.
Systemic Injection Test, Intracutaneous Test as well as Implantation Test were performed to determine the toxicity of this filters. This testing was performed by an independent laboratory.

3.7.1.2 Conclusions

The materials used in hydrophilic PVDF0.45μm filter cartridges from production met the specifications for Biological Reactivity Tests, in vivo, listed in the current revision of the United States Pharmacopeia (USP) for Class VI –121 °C Plastics.

3.7.2Bacterial Endotoxin: LAL Test

3.7.2.1Summary of Methods

The test filter was flushed with each of endotoxin-free normal saline .The filtration amount of per filter area is not more than 1mL/cm^2. Ensure normal saline flow through the entire filter. The aqueous extract was tested with an LAL reagent and all tubes were incubated at 37±1℃ for 60±2min.

3.7.2.2 Test Results

Extracts from filters contain <0.25EU/mL endotoxin units per milliliter per the USP Bacterial Endotoxins Test. The results are shown in the following table.

hydrophilic PVDF0.45μm **filter--Bacterial Endotoxin: LAL Test per USP(+Clotted;-Not Clotted)**					
Filter series number	Positive Control	Negative Control	Positive control of test solution	Test solution	Test results （0.25EU/mL）
403060211020	+ +	- -	+ +	- -	<0.25
403060211022			+ +	- -	<0.25
402200511023			+ +	- -	<0.25
402200511024			+ +	- -	<0.25
403120111022			+ +	- -	<0.25
403120111024			+ +	- -	<0.25

3.7.2.3 Conclusions

The hydrophilic PVDF0.45μm filter cartridges from production met the specifications for USP Bacterial Endotoxins Test.

3.8 Chemical Compatibility

The chemical compatibility of hydrophilic PVDF0.45μm filters is shown in the chart below. Recommendations are based upon static soak for 72 hours at 25℃ and 1.0 atmosphere (14.5 psi,1.01 bar absolute) pressure. Dynamic (operating) conditions at moderate temperatures (±10% fluctuation) will not change the recommendations, but high liquid temperature may do so in some cases.

NOTE: This data is intended to provide expected results when filtration device are exposed to chemicals under static conditions for 48hours at 25℃, unless otherwise noted, membrane integrity was tested by bubble point.

This chart is intended only as a guide. User should verify chemical compatibility with a specificfilter under actual use condition, such as various temperatures, pressure, and concentration.

R = Resistant.
L = Limited resistance
N = Not resistant
- = No data

Chemicals	**PVDF Membrane filter**	**PP Plastic Parts**	**Silicone O-ring**	**EPDM O-ring**	**Viton O-ring**
Acetic Acid, glacial	R	R	L	L	N
Acetic Acid, 90%	R	R	L	L	N
Acetic Acid, 30%	R	R	R	L	L
Acetic Acid, 10%	R	R	R	L	L
Hydrochloric acid, conc. 35%	R	R	N	N	R
Hydrochloric acid, 20%	R	R	N	N	R
Hydrochloric acid, 3.3%	R	R	-	N	R
Nitric Acid， conc .67%	R	R	N	-	R
Nitric Acid， 27%	R	R	L	L	R
Sulfuric Acid， conc. 96%	R	N	N	-	R
Sulfuric Acid， 16%	R	R	N	-	R
Ammonium Hydroxide 3N,5.7%	R	R	R	-	R
Ammonium Hydroxide 6N,11.4%	R	R	R	-	R
Potassium Hydroxide， 15%	R	R	N	R	R
Sodium Hydroxide3N， 11%	N	R	R	R	R
Sodium Hydroxide， 22%	N	R	R	R	R
Amyl Alcohol	R	R	N	R	R
Benzyl Alcohol	R	R	L	-	R
Butanol	R	R	L	-	R
Isopropanol	R	R	R	-	R
Methanol	R	R	R	R	N
Ethylene glycol	R	R	R	-	R
Glycerol	R	R	R	R	R
Propylene glycol	R	R	R	-	R
Ethyl ether	R	R	N	N	N
Tetrahydrofuran	N	R	N	N	N
Tetrahydrofuran， 50% v-v	N	R	-	N	N
Acetone	R	R	R	R	N

Cyclohexanone	R	R	L	L	N
Methyl Ethyl Ketone (MEK)	R	R	N	R	N
Methyl Isobutyl Ketone (MIBK)	N	R	N	R	N
Amyl acetate	R	R	N	R	N
Butyl Acetate	R	R	R	-	N
Cellusolve Acetate	R	R	R	-	N
Ethyl Acetate	R	R	L	N	N
Isopropyl acetate	R	R	L	R	N
Methyl acetate	R	R	N	R	N
Carbon Tetrachloride	R	R	N	N	R
Chloroform	R	R	N	N	R
Ethylene dichloride	N	R	N	-	R
Methylene Chloride	R	R	N	N	L
Tetrachloroethylene	R	R	N	-	R
Trichloroethane	N	R	N	N	-
Benzene	R	N	N	N	R
Toluene	R	N	N	N	R
Xylene	R	N	N	N	R
Cottonseed	R	R	R	-	R
Peanut	R	R	R	-	R
Formaldehyde 37%	R	L	L	R	R
Formaldehyde 4%	R	R	R	R	R
Hexan	R	N	N	-	R
Acetonitrile	N	N	N	R	R
Dimethyl Formamide(DMF)	R	R	R	N	L
Dimethylsulfoxide(DMSO)	R	R	N	N	N
Kerosene	R	–	N	N	R
Pyridine	R	L	N	N	N
Petroleum spirits	R	N	N	N	-
Hydrogen Peroxide	N	R	R	R	-
Ozone	N	R	N	L	-
Phenol	-	R	-	-	-

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