Application Summary

Filtration in Chromatography Systems

Chromatography Systems Operation and Cleaning

Purification processes in biotechnology usually include multiple chromatography columns. Each performs a different function, separating specific components from the base solution. They are an essential part of the biopharmaceutical process, and assuring their efficient operation will maximize their output.

The performance of each column can be enhanced through the use of an optimized filtration system. Both inorganic and organic particles may be in the base solution, buffer solutions, and in the cleaning solutions used to regenerate the columns between batches. Removing those particles helps the columns to perform at the highest level.

Several possible particles can interfere with the peak operation of columns. They can include:

- Bacteria in the base solution or buffer solutions
- Inorganic particles in the base solution, buffer or cleaning solutions
- Organic substances from the fermentation process (lipids, DNA, etc)

Any of these can affect column operation through either mechanical or chemical interference. Filters help assure column performance by removing particles and organisms that clog media, block flow through the column or even cause channeling of the solution through the chromatography media.

This guide shows the functions of filters in chromatography systems with brief explanations of their impact on performance. The filter media and devices commonly used to perform these functions are presented as part of the discussion. There is also a brief overview of the Critical Process Filtration technologies that may be applied in chromatography systems at the end of this guide.

Figure 1 below represents a generic system to feed and clean a single chromatography column. As stated above, a biopharmaceutical process may have many columns, so there may be a series of these systems in a single biotech process.

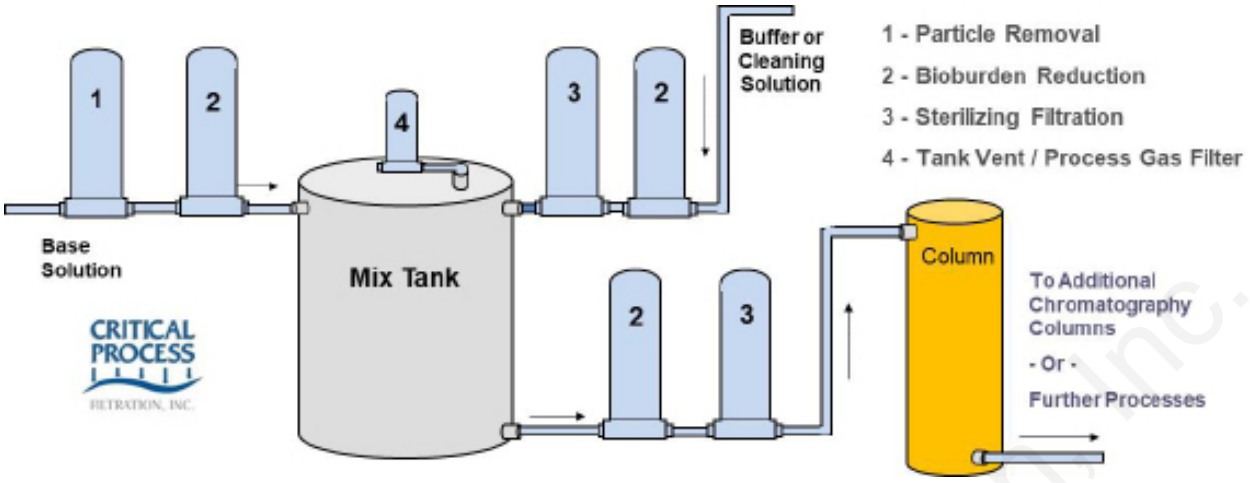

Figure 1: Chromatography Process Protection

The "base solution" noted in Figure 1 has already been processed through initial clarification steps such as centrifugation, but is likely to contain particulates, bacteria or other organic content that could affect column operation. The solution is filtered for particles and to reduce the bioburden (Filters 1 and 2), removing particulates and most bacteria that might interfere with the column before entering a mix tank.

During normal operation, a buffer solution is often added to the base solution to facilitate the chromatography process. After the processing of a batch, an additional buffer may be used to elute the target molecule(s) from the chromatography media. Buffer solutions are mixed in a separate process. See the Application Guide, "Filtration in Preparation of Cell Culture Media and Buffers", for more information on the preparation of buffer solutions. To assure efficient column operation, the final solution is filtered using a bioburden reduction filter and then a sterilizing filter (Filters 2 and 3).

The mixing tank in the figure may not be necessary if the base solution is fed directly to the column or the base solution and buffer can be mixed using an online mixing valve or similar system. If the system includes a mix tank, then the quality of the contents of the tank should also be protected from particles and bacteria in outside air or process gas, usually by a vent or process gas filter (Filter 4).

The filters and their various functions are discussed in more detail below.

System Cleaning

Several cleaning solutions are used to regenerate chromatography columns. The solutions may be acidic (citric acid, guanidine hydrochloride) or alkaline (sodium hydroxide). The columns are regenerated to improve process economics and system efficiency. The filters in the process are commonly subjected to the same cleaning regimen as the column.

Filter compatibility with the cleaning solution is as important as filter compatibility with the base solutions and buffers. The filters may be bypassed during the cleaning process, but doing so risks cross contamination and loss of solution quality. It is best to use filters compatible with both the process solutions and cleaning agents to assure that the entire system is clear of elements that might interfere with efficient operation.

Critical Process Filtration offers multiple filter membrane technologies, so you can find a membrane compatible with your process and cleaning solutions. Contact Critical Process Filtration for more information and for technical support.

Filter Applications

Figure 1 shows "typical" filter applications in a chromatography system. Additional filters may be

required if the solutions contain elements such as gels and colloids. These deformable particles can cause premature filter plugging. Contact the Critical Process Filtration Technical Services Team if you are faced with such a situation.

The filter applications in chromatography systems are generally for the bacteria control, though inorganic particulates may also be in the fluids. Filters are placed to remove most, if not all, inorganic and bacterial contamination and deliver clean fluid to the columns for effective performance.

Particle Removal Filters

The figure only shows one location for a particle removal filter (housing marked 1) at the point of entry for the base solution. However, depending on the particle load in the solution and in the buffer or cleaning solutions, and the nature of the mixing process, removing particles may be required in multiple locations.

Particle filters, as the term implies, capture particles larger than those the downstream filters will be asked to remove. They usually remove inorganic particles and similar contaminants. Some large organisms, such as molds or yeasts, can also be captured by these filters.

Most operators use depth filters for this step with pleated depth filters used most often. Pleated filtration products, with large media surface area, can remove several times more particles than standard melt-blown or nano-spun depth filters.

Critical Process Filtration offers both standard depth filters in polypropylene and nylon materials, and pleated depth filters using polypropylene or fiberglass flat sheet depth media. The pleated filters are available in “pharmaceutical grade” versions with appropriate documentation and quality certification.

Bioburden Reduction Filters

Filters that do not remove bacteria (bioburden) to the levels required for “sterilizing” are designated as “bioburden reduction” or “bioburden control” filters. The filter micron rating may be 0.22 µm, 0.45 µm or even 0.65 µm. As with sterilizing grade filters, the pore size rating is a secondary consideration. Documentation of the capability to remove the target organisms and validation of results in the application are of primary importance. The goal is to remove most organisms and assure effective system operation.

All bioburden reduction filters (Filters marked “2” in the Figure) are based on microporous membranes. Critical Process Filtration “Biopharmaceutical Grade” filters, using asymmetric polyethersulfone (PES); high capacity PES; and high capacity PVDF membranes. These cartridge and disposable capsule filters are designed to effectively remove most bacteria. They can be validated for performance to assure that the system will consistently deliver solutions that meet or exceed the requirements of your system.

Sterilizing Filters

Typical sterilizing grade filters (housings marked “3”) are rated to remove particles and bacteria that are 0.22 or 0.1 micron or larger in size. In the past, the standard has been 0.22 micron, but a few organisms, such as Acholeplasma laidlawii, have been found to pass through these filters, so some users have moved to using finer, 0.1 micron filters to remove them.

The filters are shown in two locations. Some users may use more locations, but these are the critical ones in the system shown. The buffer must be free from bacteria that would interfere with the chromatography process, so a sterilizing filter will be the last step as the buffer is added to the base solution arriving from upstream processes. After the buffer is added, any bacteria that may still be in the system are removed as the solution is introduced to the column. During the cleaning procedure, the cleaning solution is filtered to prevent potential contamination of the column by bacteria.

Critical Process Filtration has “Pharmaceutical Grade” sterilizing cartridge and disposable capsule filters made using asymmetric PES and nylon 6,6 membranes. These are available with either a 0.1 µm or 0.22 µm pore size rating. Using these filters will assure that the solutions in your chromatography system are free from bacteria that could have an impact on your columns or other downstream purification processes.

Tank Vent Filters

The vent filter (Filter "4" in the Figure) is used to protect solutions in the mix tank from bacteria and particulates. As a tank is filled, the air inside must be allowed to escape. As the tank is emptied, air (or a process gas such as nitrogen) must be allowed to enter the tank to replace lost liquid volume. Filtering the air or gas entering the tank will assure the continued quality of the solution in the tank.

It is critical that the tank vent filter membrane remain dry so that air can pass freely through it. To assure that dryness, hydrophobic membranes such as PTFE or polypropylene membrane are used. Critical Process Filtration's PTM sterilizing grade PTFE membrane cartridges or capsule filters are most often used. If a tank is kept at an elevated temperature, then PTM/HT high temperature PTFE membrane cartridges are used.

NOTE: Stainless steel tanks are susceptible to collapse if they experience a vacuum. It is critical to properly size the filter system to allow the air flow required at the maximum rate of tank outflow with a pressure drop well below the vacuum rating of the tank. Improper sizing can result in permanent tank damage. Tanks should also have rupture discs, vacuum switches or other features to protect against accidental pressure or vacuum excursions that might cause tank failure..

If a large, collapsible bag is used instead of a tank, the bag will expand and collapse without the need to allow air into the container. Vent filters are not required for disposable bag containers.

Filter Options Chromatography Systems

Process Area	Filter Application	Filtration Function	Media **	Typical Pore Sizes
Particle Control	Particle Removal	Reduce particulate load to protect performance of downstream filters and processes	CWPS, PVWL, NC, NM, PS	1 to 5µm
Bioburden control and Sterilizing	Bioburden Reduction	Remove most, but not all, bacteria from the fluid stream	CWPS, PVWL, NM, PS	0.22 to 0.45µm
	Bacteria Removal (Sterilizing)	Remove all bacteria from the fluid stream	PS, NM	0.10 to 0.22µm
	Tank Vent Filtration	Prevent bacteria and particulates from entering tanks when liquids are drawn from the tanks. Protect solution quality	TM, TM/HT	0.22µm

**Media Codes

MB = Melt-Blown Polypropylene Media

NS = Nano-Spun Polypropylene Depth Media

GF = Pleated fiberglass Depth Media

PD = Pleated Polypropylene Depth Media

CWPS = High Capacity PES Membrane

NM = Nylon 6,6 Membrane

PS = Polyethersulfone Membrane

PVWB = High Capacity Hydrophobic PVDF Membrane

PVWL = High Capacity Hydrophilic PVDF MEmbrane

TM = PTFE Membrane

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