3M Is The Innovation Company That Makes Progress Possible

- We create transformational products and solutions that enable customer success and improve people's lives around the world.
- We utilize a collaborative, high-energy approach to solve the toughest problems across industries and markets by:
  - Constantly exchanging and building on each other’s ideas
  - Uncovering new connections between seemingly unrelated markets and more than 50 diverse technology platforms
  - Fostering a culture of intellectual curiosity and creativity that pushes boundaries

At 3M, we are advancing the global biopharmaceutical industry by helping to build better and more efficient manufacturing processes, improving product safety by providing tools for monitoring and tracking, and reducing energy usage with our technologies. The Life Sciences Process Technologies business unit of 3M Purification Inc. provides cutting edge technologies to address clarification, filtration and purification needs of the global biopharmaceutical industry.

Figure 1: The Biopharmaceutical Production and Downstream Purification Train
3M Technologies

At 3M, our technology, products and innovation reflect what we do for our customers every day: advance, enhance and improve their products and processes to enable their success.

3M Purification’s dedicated technical services and laboratory personnel help solve customer’s most arduous separations problems. Our engineers work to provide solutions that reduce the overall cost of ownership. Our researchers are constantly working on breakthroughs that make new separations platforms possible.

Every day our products are used by researchers, process developers and manufacturing personnel for critical filtration, separation and process monitoring steps in the biopharmaceutical industry.

Biopharmaceutical Separations Applications

Biopharmaceutical refers to biologically active therapeutic and diagnostic proteins that are expressed by mammalian, insect, yeast or bacterial cells. Such drugs can be classified into: monoclonal antibodies, growth factors, hormones, cytokines, fusion proteins, and therapeutic enzymes. Viral based therapeutics are poised to grow rapidly and hold great promises for disease treatment in the future. The manufacturing process of diagnostic reagents and sera contains many biopharmaceutical separations applications.

Filtration and purification plays an essential role in manufacturing of biopharmaceutical drugs. 3M offers a range of filtration, purification and process monitoring technologies that can be used in both upstream and downstream steps in every scale of biopharmaceutical manufacturing.
The Bioreactor is at the heart of a biopharmaceutical manufacturing process. For the bioreactor to work at maximal efficiency, filters used for air and media filtration need to completely remove foreign microorganisms.

Bacterial Fermentation e.g. *Escherichia coli*†

Developed in the late 1970s, the bacterial expression system, such as the *E. coli* system, is used to produce many important therapeutic recombinant proteins. The popularity and dominance of the bacterial system has continued until today because of its advantage in speed, simplicity and cost. The challenge of using this system is in downstream purification. A homogenization step after harvest is often needed because most of the expressed proteins are located in inclusion bodies rather than secreted outside the cells. This step increases the level of lipids, host cell proteins, DNA and endotoxins along with the target protein; hence resulting solutions from bacterial harvest are difficult to purify.

Mammalian Fermentation e.g. CHO Cell†

Mammalian cells (e.g., CHO [Chinese Hamster Ovary] Cells) are typically used to produce monoclonal antibodies. Monoclonal antibodies are derived from one single clone and thus are identical in structure. Today over 30 monoclonal antibodies have been approved in the U.S. with indications for a variety of difficult to treat diseases such as autoimmune diseases, cancers, infectious diseases, etc. Clarification involves separation of the cells from bulk media. Cell density (> typical 5 x 10⁶ cells/mL) and cell viability affect the depth filtration step. In general, mammalian cell cultures are easier to filter than bacterial cell culture harvests.

---

**3M Products**

**Bioreactors**

<table>
<thead>
<tr>
<th>Process Stage</th>
<th>Filter Pore Size</th>
<th>Scope of Application</th>
<th>Retention of bacteria and aerosolized bacteriophage</th>
<th>Short term storage of media, buffers etc.</th>
<th>High UV removal of mycoplasma for media components</th>
<th>Absolute retention of B. diminuta (ATCC 19146) at challenge levels of &gt; a minimum of 10⁷ CFU/cm²</th>
<th>Absolute retention of Acholeplasma laidlawii, ATCC 23206 at challenge levels of &gt; a minimum of 10⁷ CFU/cm²</th>
<th>Portable handheld integrity tester to measure pressure decay</th>
<th>Real-time monitoring of microbial / protein levels from surfaces</th>
</tr>
</thead>
<tbody>
<tr>
<td>Air Filtration</td>
<td>0.2 µm</td>
<td>Not Applicable</td>
<td>LifeASSURE™ PFS</td>
<td>Proprietary Ultrafilter (UPF; sterilization compatible)</td>
<td>LIFEASSURE™ PDA</td>
<td>LifeASSURE™ PDA</td>
<td>MicroCheck™</td>
<td>LIFEASSURE™ PDA</td>
<td>LIFEASSURE™ PDA</td>
</tr>
<tr>
<td>Vent Air Filtration</td>
<td>0.1 µm</td>
<td>Not Applicable</td>
<td>LIFEASSURE™ PFS</td>
<td>PES membrane filter (Sterile and Gamma Compatible)</td>
<td>3M™ Single Use Biocontainer</td>
<td>LIFEASSURE™ PDA</td>
<td>LifeASSURE™ PDA</td>
<td>LIFEASSURE™ PDA</td>
<td>LIFEASSURE™ PDA</td>
</tr>
<tr>
<td>In Process Storage</td>
<td>0.2 µm</td>
<td>Not Applicable</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
</tr>
<tr>
<td>Automated Integrity Tester For Housings And Connections</td>
<td>Not Applicable</td>
<td>Not Applicable</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
</tr>
<tr>
<td>Surface Cleanliness Monitoring</td>
<td>Not Applicable</td>
<td>Not Applicable</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
<td>LIFEASSURE™ PSA</td>
</tr>
</tbody>
</table>

---

**Materials**

<table>
<thead>
<tr>
<th>Media Components</th>
<th>Nutrients e.g. Sugar</th>
<th>Air Filtration</th>
<th>Vent Air Filtration</th>
<th>3M™ Single Use Biocontainer</th>
<th>LIFEASSURE™ PFS</th>
<th>LIFEASSURE™ PSA</th>
<th>LIFEASSURE™ PDA</th>
<th>MicroCheck™</th>
<th>LIFEASSURE™ PDA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Filter Pore Size</td>
<td>0.2 µm</td>
<td>0.1 µm</td>
<td>0.2 µm</td>
<td>Not Applicable</td>
<td>Not Applicable</td>
<td>Not Applicable</td>
<td>Not Applicable</td>
<td>Not Applicable</td>
<td>Not Applicable</td>
</tr>
<tr>
<td>Scope of application</td>
<td>Retention of bacteria and aerosolized bacteriophage</td>
<td>Short term storage of media, buffers etc.</td>
<td>High UV removal of mycoplasma for media components</td>
<td>Absolute retention of B. diminuta (ATCC 19146) at challenge levels of &gt; a minimum of 10⁷ CFU/cm²</td>
<td>Absolute retention of Acholeplasma laidlawii, ATCC 23206 at challenge levels of &gt; a minimum of 10⁷ CFU/cm²</td>
<td>Portable handheld integrity tester to measure pressure decay</td>
<td>Real-time monitoring of microbial / protein levels from surfaces</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

---

**Automation**

**Bioreactors**

- Retention of bacteria and aerosolized bacteriophage
- Short term storage of media, buffers etc.
- High UV removal of mycoplasma for media components
- Absolute retention of *B. diminuta* (ATCC 19146) at challenge levels of > a minimum of 10⁷ CFU/cm²
- Absolute retention of *Acholeplasma laidlawii*, ATCC 23206 at challenge levels of > a minimum of 10⁷ CFU/cm²
- Portable handheld integrity tester to measure pressure decay
- Real-time monitoring of microbial / protein levels from surfaces

---

**Bioreactor**

The Bioreactor is at the heart of a biopharmaceutical manufacturing process. For the bioreactor to work at maximal efficiency, filters used for air and media filtration need to completely remove foreign microorganisms.

**Bioreactor**

- Retention of bacteria and aerosolized bacteriophage
- Short term storage of media, buffers etc.
- High UV removal of mycoplasma for media components
- Absolute retention of *B. diminuta* (ATCC 19146) at challenge levels of > a minimum of 10⁷ CFU/cm²
- Absolute retention of *Acholeplasma laidlawii*, ATCC 23206 at challenge levels of > a minimum of 10⁷ CFU/cm²
- Portable handheld integrity tester to measure pressure decay
- Real-time monitoring of microbial / protein levels from surfaces

---

**Bioreactor**

The Bioreactor is at the heart of a biopharmaceutical manufacturing process. For the bioreactor to work at maximal efficiency, filters used for air and media filtration need to completely remove foreign microorganisms.

**Bioreactor**

- Retention of bacteria and aerosolized bacteriophage
- Short term storage of media, buffers etc.
- High UV removal of mycoplasma for media components
- Absolute retention of *B. diminuta* (ATCC 19146) at challenge levels of > a minimum of 10⁷ CFU/cm²
- Absolute retention of *Acholeplasma laidlawii*, ATCC 23206 at challenge levels of > a minimum of 10⁷ CFU/cm²
- Portable handheld integrity tester to measure pressure decay
- Real-time monitoring of microbial / protein levels from surfaces
3M offers the most comprehensive portfolio of depth filters for cell culture clarification in the biopharmaceutical industry. Zeta Plus™ depth filtration technology, in cartridge systems and sheets, plays an important role in the clarification of cell-derived protein therapeutic products around the world. 3M is recognized as a market leader in depth filter technology.

Table 1: Depth Filter Media Recommendations

<table>
<thead>
<tr>
<th>Feed Composition</th>
<th>Fluid Turbidity</th>
<th>Recommendation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Whole cells, hard particles (density gradient)</td>
<td>&gt; 300 NTU</td>
<td>Centrifuge, TFF, Open pore depth filter (05 or 10 SP)</td>
</tr>
<tr>
<td>Colloidal, Cell Debris</td>
<td>100-300 NTU</td>
<td>Medium Pore Depth filter 30-60 SP</td>
</tr>
<tr>
<td>Colloidal, Small Particles</td>
<td>20-100 NTU</td>
<td>60SP or 60ZA or 90 ZA</td>
</tr>
<tr>
<td>Tiye particles, colloidal (Final Polishing)</td>
<td>&lt; 50 NTU</td>
<td>90 or 120 ZA</td>
</tr>
<tr>
<td>Intra-cellular, requires cell breakage – First Clarification Step</td>
<td>&gt; 300 NTU</td>
<td>Centrifuge</td>
</tr>
</tbody>
</table>

Table 2: Range of Media Adsorption Properties

<table>
<thead>
<tr>
<th>Designation</th>
<th>Media Surface Characteristic</th>
</tr>
</thead>
<tbody>
<tr>
<td>ZA</td>
<td>Strong Anion Exchange</td>
</tr>
<tr>
<td>SP</td>
<td>Medium Anion Exchange</td>
</tr>
<tr>
<td>ZC</td>
<td>Activated Carbon for color / organic adsorption</td>
</tr>
<tr>
<td>DELI</td>
<td>Activated Silica for adsorption of hydrophobic moieties</td>
</tr>
</tbody>
</table>

Figure 4: Zeta Plus™ Depth Filter Family
Traditional Solutions for Cell Culture Clarification

Based on our more than 30 years of technical expertise in filtration and separation products for the biopharmaceutical industry, we offer:

1. The broadest portfolio of lenticular depth filter media in the industry
2. Complex custom engineered systems
3. Single-use storage solutions and customized connector sets
4. Tools for integrity testing of filter systems, handheld pressure decay measurement systems
5. Range of sterile and gamma compatible 0.2 µm PES membrane capsules

3M™ Single-Use Biocontainers

Zeta Plus™ Lenticular Cartridges

Complex Custom Engineered Solutions

Tools for Integrity Testing of Filtering System

LifeASSURE™ PDA 0.2 µm Sterile Capsules

Single-Use Solutions for Cell Culture Clarification

3M offers a complete package of single-use systems for cell culture clarification applications for biopharmaceutical customers. Single-use Zeta Plus™ Encapsulated depth filter clarification solutions are available in scaleable capsule formats from R&D to process development to pilot / clinical production to commercial and large scale production. In addition, we also offer customized solutions and accessories, such as tubing connectors and staging carts to round out a complete package.

Figure 7: Scalable Single-Use Solutions from 0.5 to 25,000 liters

Accessories

Custom Single-use Tubing Connectors for Zeta Plus™ Encapsulated Systems

Staging Carts — For Loading Capsules

<table>
<thead>
<tr>
<th>Capacity</th>
<th>Process Volume</th>
</tr>
</thead>
</table>
| BC25     | 25 cm²  
          | 0.5 - 1 L  |
| E0170    | 170 cm² 
          | 1 - 5 L   |
| E0340    | 340 cm² 
          | 5 - 25 L  |
| E1020    | 1020 cm² 
          | 1 - 5 L   |
| 16EZA    | DL — 1.6 m²
          | SL — 2.5 m² |
| 16EZB    | DL — 11.2 m² 
          | SL — 17.6 m² |
| 16EZC    | DL — 56 m² 
          | SL — 87.5 m² |
| 2000 - 25,000 L |  |
Zeta Plus™ Encapsulated Systems are designed to make cell clarification by depth filtration fast, easy and clean. 3M offers three models of Zeta Plus Encapsulated Holders — 16EZA, 16EZB and 16EZC — as a convenient single-use depth filter system for cell culture clarification. Both the Single Round (Model #16EZB) and Multi-Round (Model #16EZC) can be pivoted between horizontal and vertical positions, allowing for convenient loading and unloading, minimal footprint during filtration, minimal fluid spills during unloading, and full utilization of the filter media. The pilot scale system, 16EZA, uses up to 3.2 m² of depth filter media and is not designed to pivot.

At the heart of the Encapsulated Zeta Plus system is the uniquely designed Depth Filter Capsule.

- Translucent plastic shell allows for easy liquid level detection
- Handles designed for easy loading and unloading
- A cam structure allows reliable capsule-to-capsule connection
- Translucent plastic shell allows for easy liquid level detection
- Upstream hold-up volume is reduced to minimum
- Solid core design eliminates the need for central post and stainless steel band

### Advantages of The Zeta Plus™ Encapsulated system

- Simplifies the operation of depth filtration step
- Full utilization of the filter media, small footprint during filtration
- Avoids spills on the manufacturing floor
- Ergonomic and saves labor

### Table 3: Zeta Plus™ Encapsulated System Specifications

<table>
<thead>
<tr>
<th></th>
<th>16EZB</th>
<th>16EZC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dimensions</td>
<td>1.0 m x 0.5 m x 2.2 m (39.4” x 19.7” x 86.6”)</td>
<td>2.0 m x 2.5 m x 2.2 m (78.7” x 98.4” x 86.6”)</td>
</tr>
<tr>
<td>Rack Capacity</td>
<td>1 rack of capsules</td>
<td>3 or 5 racks of capsules</td>
</tr>
<tr>
<td>Loading per Rack</td>
<td>1 - 7 capsules</td>
<td>7 capsules</td>
</tr>
<tr>
<td>Flexibility</td>
<td>Can plumb for two stage depth filtration in the same system (maximum number of capsules - 4)</td>
<td>Can load just one rack and leave others empty. Can plumb for two stage depth filtration</td>
</tr>
<tr>
<td>Torque Limiter</td>
<td>Manual</td>
<td>Manual (interlocked by PLC to ensure capsules are secure)</td>
</tr>
<tr>
<td>Indexing and Pivoting to Vertical Position</td>
<td>No indexing. Pivot by manual gear</td>
<td>Automated with torsion limited electric motors.</td>
</tr>
<tr>
<td>Control</td>
<td>Manual - All mechanical parts</td>
<td>PLC controlled</td>
</tr>
<tr>
<td>Filter Area</td>
<td>Double Layer Media up to 11 m² Single Layer Media up to 17.5 m²</td>
<td>Double Layer Media up to 55 m² Single Layer Media up to 87.5 m²</td>
</tr>
</tbody>
</table>

### Zeta Plus™ Encapsulated Systems

- **Zeta Plus™ Encapsulated Single-Round System (Model #16EZB)**
  - For up to 5,000 Liters
  - **Load**
  - **Fast**
  - **Pivot**
  - **Easy**
  - **Clean**

- **Zeta Plus™ Encapsulated Multi-Round System (Model #16EZC)**
  - For up to 25,000 Liters
  - **Model 16EZB**
  - **Model 16EZC**

The Zeta Plus™ Encapsulated single-use system has been used to replace a centrifuge and a conventional single layer depth filter in an existing process for CHO cell culture clarification at a major biopharmaceutical manufacturer.

**Justification:**

During harvest runs, the continuously stacked disc centrifuge broke down periodically, causing maintenance and operational bottleneck. Going from two-unit operations to one and converting a hard plumbed system into a single-use process results in significant savings.

**Solution:**

- 3M’s Zeta Plus EXT media grade 60SPO2A in an encapsulated format

**Details:**

- 11 m² of Zeta Plus Encapsulated module processed 1,000 liter batch and differential pressure across the downstream 0.2 μm membrane was < 0.05 psi. (1.0 bar) throughout the run

**Operator Feedback:**

- Minimal residual liquid was observed during system break-down.
- Zeta Plus Encapsulated system was easy and convenient to set up.
- Single-use depth filtration significantly reduced cleaning time for equipment and the process suite.
- No CIP or cleaning validation studies were required.
- Further, the cycle times were shortened thus reducing manufacturing costs.
Exploiting The Charge Effect Of Depth Filters

Depth filters are made of cellulose fibers, a filter aid (e.g. Perlite) and binding resins that impart a charge to the filtration matrix. 3M offers a range of depth filter matrices that have strong anion exchange (ZA grade media) to weak anion exchange (SP grade media) characteristics.

Monoclonal antibodies (mAbs) typically have isoelectric points (IEP or pI) that vary from pH of 4.5 to 8.5. If the pI of the mAb is greater than the pH of the HCP/DNA contaminant these contaminants can be removed by exploiting the operating pH and choosing the optimal depth filter resin system. It is important that the feed stream be relatively free of colloidal particles as they are also adsorbed by the charged depth filter.

Graph 1 shows charge capacity of strong anion exchange and weak anion exchange resin as a function of pH. The graph shows Zeta Plus ZA (strong anion exchange [AEX]) maintains the charge at higher pH, while Zeta Plus SP (weak AEX) has reduced charge capacity as pH increases. DNA removal at a pH 7.4 and 9.0 for Zeta Plus SP grade filters is shown in Table 4.

Zeta Plus SP grade depth filters have lower AEX capacity at pH 9 compared to pH 7.4.

Figure 8: Host Cell Protein Removal

Host Cell Proteins (HCPs) are undesirable components in downstream protein purification processes. HCPs are known to foul Protein A resins, thereby impacting the operation of this important purification step. In addition, the presence of high levels of HCP may lead to protein precipitation, or aggregation, either before or after the Protein A column. Low pH viral inactivation is particularly prone to protein precipitation. Protein aggregates can sporadically plug the sterile filters.

Figure 9 shows the flow schematic and levels of HCP removal using Zeta Plus™ 90ZA depth filter media.

Endotoxin Removal

Endotoxins are cell wall components of gram-negative bacteria consisting of lipopolysaccharides that can cause a pyrogenic response in a parenteral formulation. When a protein therapeutic is made in a bacterial expression system, such as E. coli, downstream processes are required to remove these endotoxins below the threshold level. Fortunately, because of its high negative charge, endotoxin can be removed using media that have anion-exchange characteristics. Graph 2 shows, endotoxin removal of various Zeta Plus grades filters.

DNA Removal

Downstream processes are required to reduce the residual host cell DNA to levels less than 100 pg / dose of the final protein therapeutic formulation per safety requirements of US FDA. Relatively high levels of DNA are seen in perfusion harvest and large amount of DNA co-elutes with the protein from the Protein-A column. The challenge for the downstream purification process is to consistently reduce DNA levels in the final product. DNA can bind nonspecifically to the backbone of Protein A column. Due to its phosphate groups, DNA is highly negatively charged at physiological pH and thus is well suited to be removed by binding to AEX ligands.

Zeta Plus™ depth filters can be used for DNA removal. Graph 3 shows Zeta Plus ZA media’s DNA removal properties.

Graph 3: Product Recommendations For Pre-Purification Applications

<table>
<thead>
<tr>
<th>Choice of Media</th>
<th>Antigen Exchange Functionality</th>
</tr>
</thead>
<tbody>
<tr>
<td>ZA, LA, LA, SP Grades</td>
<td>Quaternary Amine</td>
</tr>
<tr>
<td>ZA Grade</td>
<td>Tertiary Amine</td>
</tr>
</tbody>
</table>

Table 4: Percent Capture of DNA by Charged Depth Filter Media

<table>
<thead>
<tr>
<th>Zeta Plus™ Grade</th>
<th>Lot Number</th>
<th>% Removal of DNA</th>
<th>pH 7.4</th>
<th>pH 8.0</th>
</tr>
</thead>
<tbody>
<tr>
<td>ZA</td>
<td>20127</td>
<td>99.9</td>
<td>29</td>
<td></td>
</tr>
<tr>
<td>ZA</td>
<td>23443</td>
<td>99.8</td>
<td></td>
<td>68</td>
</tr>
</tbody>
</table>

Graph 1: Pre-Purification HCP, DNA, Endotoxin Removal Applications
Zeta Plus™ DEL series filters combine high lipid removal with cellulose-based depth filtration and can remove hydrophobic components and particulates such as lipids, protein aggregates and cell debris in a single step prior to a HIC unit operation. Table 5 shows the effect of pretreatment with Zeta Plus DEL series filter prior to an HIC column and the resultant improvement in performance.

### Table 5: HIC Performance with and without Zeta Plus™ DEL Series Media

<table>
<thead>
<tr>
<th>Pre-treatment</th>
<th>Lipid in the Feed</th>
<th>Feed to HIC Column Pretreatment</th>
<th>Zeta Plus™ DEL Series Filters</th>
</tr>
</thead>
<tbody>
<tr>
<td>No Filter</td>
<td>100%</td>
<td>Loading: 35%</td>
<td>80%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Elution: 90%</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Yeast: 35%</td>
<td>72%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>40th Cycle:</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Loading: 62%</td>
<td>87%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Elution: 29%</td>
<td>43%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Yeast: 18%</td>
<td>37%</td>
</tr>
</tbody>
</table>


**Zeta Plus™ Media and Selection Recommendations**

- Zeta Plus™ depth filters are used to remove particulates prior to chromatography columns
- Zeta Plus DEL series filter media contains activated silica that selectively adsorbs lipids, detergents, anti-foams that foul chromatography resins ultrafiltration (UF) systems and sterile membrane filters

**Zeta Plus Media Choices and Selection Recommendations**

Two different types of Zeta Plus DEL series filter media are available to suit varying application requirements. The chart below serves as a guideline for selecting the appropriate media type:

<table>
<thead>
<tr>
<th>Type</th>
<th>Lipid Reduction Capacity</th>
<th>Optimized for Low Aluminum Extractable Levels</th>
<th>Optimized for Sensitive LAL* Test Procedures</th>
</tr>
</thead>
<tbody>
<tr>
<td>DEL</td>
<td>Intermediate</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>DELP</td>
<td>High</td>
<td>Yes</td>
<td>Yes</td>
</tr>
</tbody>
</table>

*Limulus Amoebocyte Lysate
PES — The Preferred Membrane For Biopharmaceutical Applications

- Polyethersulfone (PES) membranes offer higher flow rates
- Broad chemical compatibility
- Wide pH range: Same filter for acid and basic buffers
- Low protein binding and adsorption

Sterilizing grade filters are widely used in many downstream processing steps. In addition to the sterile filtration prior to the final filling step, sterilizing filters are used widely in many intermediate steps to reduce cross-contamination risk from in-process liquids such as buffers. Cell culture growth media are filtered using 0.1 µm filters to mitigate the risk of mycoplasma contamination. Since biopharmaceutical unit operations are discontinuous in nature, elution pools (e.g. Protein A pool) need to be filtered with 0.2 µm filters prior to storage in order to manage bioburden loads. To prevent growth of microorganisms in downstream chromatography steps, post depth filter permeate is filtered using 0.2 µm sterilizing or bioburden reduction grade filters. Performance characteristics of LifeASSURE™ PDA PES membranes in various biopharmaceutical fluids are shown in graphs 6, 7 and 8.

Table 6 Comparative Choice Of Membranes

<table>
<thead>
<tr>
<th>Strengths</th>
<th>Nylon</th>
<th>PES</th>
<th>PTFE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Good wettability</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Positive charge</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Lower rates of false integrity test (FT) failure</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>High flow rates</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Greater throughput</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Low and high pH compatibility</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Gamma stable</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Low protein adsorption</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Weaknesses</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lower flow rates</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>Low pH compatibility</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Nonspecific adsorption</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Immersible</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Need to autoclave/steam vent</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>Metallurgical</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Difficult conducting integrity tests, such as the need to wet the membrane with alcohol</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Lead applications</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Many cytotoxic drugs</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>UHP applications</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Relative flow rates</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Critical wettability</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Buffer / media filtration</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Critical wettability</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Solvent / sterile AP applications</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
</tbody>
</table>

Table 7: 3M Membrane Choices For Sterile Filtration Applications

<table>
<thead>
<tr>
<th>Membrane</th>
<th>3M Product Type</th>
<th>Pore Size</th>
<th>Applications</th>
<th>Additional Features</th>
</tr>
</thead>
<tbody>
<tr>
<td>Polyethersulfone (PES)</td>
<td>LifeASSURE™ PDA Series Filters</td>
<td>0.2 µm</td>
<td>In-process filtration, Buffer filtration</td>
<td>Sterilizing grade filter, Gamma stable, Solvent stable, Sterilizing performance in wet or dry conditions</td>
</tr>
<tr>
<td>Nylon 66</td>
<td>LifeASSURE™ PSA Series Filters</td>
<td>0.2 µm</td>
<td>In-process filtration, Buffer filtration</td>
<td>Sterilizing grade filter, Gamma stable, Solvent stable, Sterilizing performance in wet or dry conditions</td>
</tr>
</tbody>
</table>

Figure 13: Biopharmaceutical — Sterile Final Filtration and Filling Process

Graph 6: Water Flow

Relative water flow of 10 inch LifeASSURE™ PDA and commercially available PES cartridge filters. PES A, B, C cartridges water flow from product literature.

Graph 7: Bovine IgG

Relative throughput of bovine IgG (50 mg/ml) with equal area polyethersulfone membrane. Relative filtrate volume after 10 minutes at 15 psi feed pressure was measured.

Graph 8: CHO Cell Media

Relative throughput of CHO cell media feed with equal area polyethersulfone membrane. Tests were conducted at a constant flow and throughput volume was measured at a terminal pressure of 25 psi.
3M makes innovative products for biopharmaceutical monitoring applications. 3M Comply™ chemical indicators can provide a visual 'Accept' / 'Reject' for steam sterilization. Our innovative Attest™ Rapid readout biological indicators can provide rapid results when using biological indicators while simultaneously avoiding aseptic transfers, and the need to prepare media. CleanTrace™ technologies use ATP monitoring to test for biological activity on surfaces and to validated the cleaning process. 3M Petrifilm Aqua Plates are suitable for efficient monitoring of plant’s water.

3M™ Attest™ 1292-S Rapid Readout Biological Indicators and 3M™ Attest™ Auto-readers

- Provide final readout in 3 hours to validate steam sterilization applications
- Self-contained biological indicator design reduces risk of contamination during transfer
- Recognized by the US Food and Drug Administration as a biological indicator
- Auto-reader has either 12 or 36 vial capacity and provides automatic calibration on every biological indicator

Chemical Integrators For Monitoring Steam Sterilization

- “Accept” or “Reject” at a glance
- For use in all 118-138°C (245-280°F) steam sterilization cycles
- Conform to ANSI/AAMI/ISO 11140-1:2005 and EN ISO 11140-1:2005 Class 5 Integrating Indicators
- Identify non-sterile instruments before they enter the sterile field

3M™ Petrifilm™ Aqua Plates for Water Testing

3M™ Petrifilm™ Aqua Plates are sample-ready media that replace conventional agar, petri dishes, media pads and disposable filter funnels used in the microbial testing of water. Each plate contains a water-soluble gelling agent, nutrients and indicators in a dry, shelf-stable format. We offer four 3M Petrifilm Aqua Plates to cover your unique testing needs:

- Heterotrophic Count
- Coliform Count
- Enterobacteriacea Count
- Yeast and Mold Count

Advantages:

- 80% increase in productivity
- Ease of use
- Compatible with membrane filtration
- 85% savings in storage space (50 3M Petrifilm Aqua Plates vs. 50 agar dishes and 50 disposable filter funnels)
- Confirmed coliform results in just 24 hours
- Longer shelf life vs premade agar plates

Why wait 24 hours for biological results?

The difference between Conventional Biological Indicators and 3M™ Attest™ Rapid Readout Biological Indicators

- Nutrient-enriched broth allows microorganisms to grow and multiply.
- Nutrients allow microorganisms to grow and multiply.
- Nutrient-enriched broth allows microorganisms to grow and multiply.
- Spore-associated enzyme breaks down nutrients attached to a fluorescent dye. During breakdown of nutrients, fluorescence is released.
- Spore-associated enzyme breaks down nutrients attached to a fluorescent dye. During breakdown of nutrients, fluorescence is released.
- Fluorescence is detected by Auto-reader.
- Fluorescence is detected by Auto-reader.

Same-day results can be up to 95% faster than conventional BIs!
Scientific Applications Support Services (SASS)

3M has a global team of market-focused scientists and engineers who excel in supporting, collaborative efforts between our customers and 3M. Our technical team is skilled in performing on-site bench-scale tests and relating their results to full scale manufacturing filter operations. When unique processing problems are encountered, our expert product and application specialists are equipped to identify issue solutions using either 3M’s broad array of existing products or work directly with our customers to design a custom solution for the job.

Validation Services

3M provides the following validation and scientific services for the biopharmaceutical industry from its various regional global technical service centers.

Post-use Integrity Test
- GMP Guidelines
- Process Fluid
- Safety Margin
- Correlation

Bacterial Challenge Test
- ASTM B. Dimunuta ATCC 19046
- HIMA

Compatibility
- Sterilization
- CFIR
- USP
- Integrity
- Hardware

Adsortion
- GMP Guidelines
- Binding
- Preservatives
- Equilibrium
- Saturation
- Recovery

Extractables
- Post-use Integrity Test
- Biological Safety
- USP
- Toxicity
- Identity
- Permitted materials
- GMP Guidelines
- Quantity
- Active Materials

End Notes


† The information presented here contains data and conclusions from studies where process conditions of filtration and separation technology was optimized by the respective researchers. Each application of filtration and separation technology may have significant process differences and it is important for the user to conduct similar process and scale-up studies to validate performance in a particular application. Please contact 3M Purification’s global SASS team for help with specific applications.
## Important Notice

The information described in this literature is accurate to the best of our knowledge. A variety of factors, however, can affect the performance of the Product(s) in a particular application, some of which are uniquely within your knowledge and control. **INFORMATION IS SUPPLIED UPON THE CONDITION THAT THE PERSONS RECEIVING THE SAME WILL MAKE THEIR OWN DETERMINATION AS TO ITS SUITABILITY FOR THEIR USE. IN NO EVENT WILL 3M PURIFICATION INC. BE RESPONSIBLE FOR DAMAGES OF ANY NATURE WHATSOEVER RESULTING FROM THE USE OF OR RELIANCE UPON INFORMATION.**

It is your responsibility to determine if additional testing or information is required and if this product is fit for a particular purpose and suitable in your specific application.

**3M PURIFICATION INC. MAKES NO REPRESENTATIONS OR WARRANTIES, EITHER EXPRESS OR IMPLIED INCLUDING WITHOUT LIMITATION ANY WARRANTIES OF MERCHANTABILITY, FITNESS FOR A PARTICULAR PURPOSE OR OF ANY OTHER NATURE HEREUNDER WITH RESPECT TO INFORMATION OR THE PRODUCT TO WHICH INFORMATION REFERS.**

## Limitation of Liability

3M Purification Inc. will not be liable for any loss or damage arising from the use of the Product(s), whether direct, indirect, special, incidental, or consequential, regardless of the legal theory asserted, including warranty, contract, negligence or strict liability. Some states do not allow the exclusion or limitation of incidental or consequential damages, so the above limitation may not apply to you.