GE Healthcare
Life Sciences

Permeate flow control recommended for upstream clarification of target material

Clarification of particulated starting material demands larger lumen diameter fibers (0.75- to 1.0-mm ID). Unlike cell harvesting, or any other simple particulate concentration, clarification of a target molecule from a solution containing particulates requires more attention to process equilibrium. Since protein passage is of paramount concern in clarification processes, open pore size microfiltration membranes, such as 0.2 to 0.65 µm pore size, are typically recommended, especially for larger recombinant proteins and monoclonal antibodies. In general, choose a membrane pore size that is at least 10× larger than the target material to pass through the membrane. Operators must exert more deliberate control of transmembrane pressure and more careful timing of concentration and cell washing to promote protein passage. Even though retentate backpressure is almost certainly reduced or even absent, further steps can also be taken to reduce transmembrane pressure to prevent premature fouling of the membrane. Experience has shown that permeate flow control (Fig 1) can be useful in further reducing inlet transmembrane pressure due to feed pump velocity.

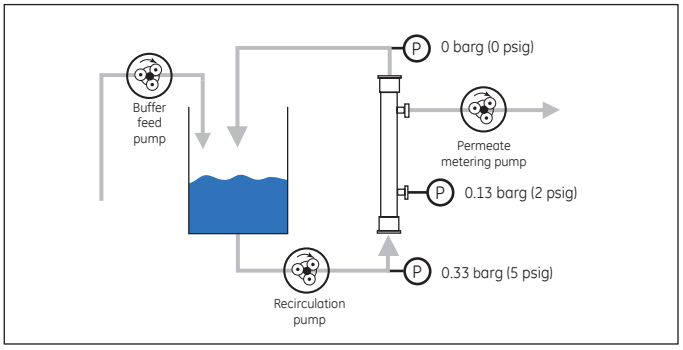

Figure 1. Permeate flow control

When using permeate flow control, the permeation rate is controlled at a lower level than would be achieved initially with an uncontrolled permeate stream. The amount of flow reduction compared to unregulated permeate flow is dependent on the nature of the starting feed stream. Lower flow rates are recommended if the target molecule is very large, if the particulates are variable in size, or if the particulates are very sticky and fouling (Fig 2). A typical monoclonal antibody clarification from hybridoma cell culture with intact cells may be controlled at approximately 30 to 50 lmh, whereas clarification of enzyme from bacterial lysate is almost always at approximately 10 lmh.

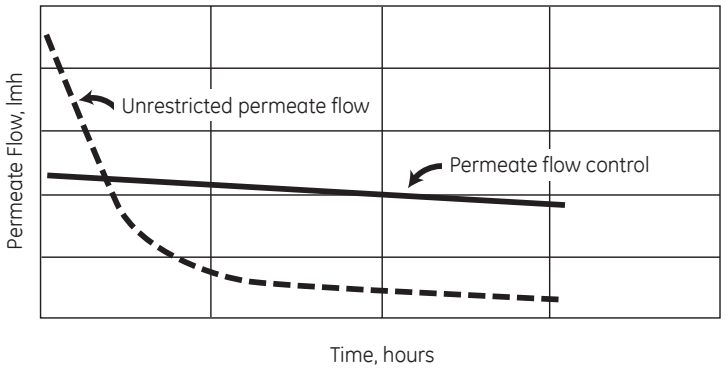

Figure 2. Permeate flow control results in more stable flow, higher protein yields, and often shorter process times.

In clarification tasks, the presence of upstream particulates demands use of shorter path length (30 to 60 cm) cartridges. Pilot scale and large scale processes often employ multiple 30-cm path length cartridges in series with individually controlled permeate streams to manage frictional pressure drop and reduce inlet transmembrane pressures. Contact GE Healthcare membrane separations technical support staff for more details on clarification system design when scaling up.

To promote maximum protein passage, good clarification process design is required (for example, partial concentration followed by brief diafiltration). Particulates can interfere with passage of protein as they become more concentrated. Proper timing of the diafiltration step is therefore essential. It is best to perform a brief diafiltration at a point where protein is still passing freely—that is, not being retained by a secondary rejection layer (gel polarization layer) composed of concentrated particulates.

imagination at work

GE Healthcare Bio-Sciences AB, Björkgatan 30, 751 84 Uppsala, Sweden.

29-0019-01 AB 10/2011