Single pass tangential flow filtration: Critical operational variables, fouling, and main current applications

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ABSTRACT

The biopharmaceutical industry has an ongoing interest in improving manufacturing efficiency and productivity while at the same time reducing manufacturing costs by developing intensified, linked and continuous processes. In recent years, upstream productivity of bioprocesses has improved significantly resulting in increased volumes and increased titers, which have put downstream processes under enormous pressure. Single Pass Tangential Flow Filtration (SPTFF) is a technology that offers an opportunity to reduce process volumes efficiently through inline concentration in one single pass through a series of membranes without the need for retentate recirculation. This technology links upstream and downstream operations and has the capability to ensure continuous production. This review seeks to investigate both early papers on SPTFF and recent developments in the field, including single-use SPTFF and 3D printed membrane modules. To our knowledge, this review is the first to address SPTFF and is aimed at gathering all relevant information about the topic, with a special focus on application in bioprocessing from which this technology has evolved. Due to a growing need for intensified processes, interest in SPTFF within the biopharmaceutical industry is increasing, hence the importance of gathering all relevant information on the topic in this review. The review starts with a thorough comparison between normal tangential flow filtration (TFF) and SPTFF, including principles, configuration, and typical features of these technologies. Also addressed are the design equations and how to tackle fouling. The review ends by providing relevant information about the main applications of SPTFF in bioprocessing as well as future perspectives on the operation. The review mainly deals with application in the biopharmaceutical industry but includes examples from the food industry and wastewater treatment.

1. Introduction

Membrane processes are widely used in the separation and purification of biological products [1,2]. Filtration, specifically microfiltration (MF) and ultrafiltration (UF), is a key unit operation in virtually every downstream process of biopharmaceutical production [3,4]. Filtration processes find application where many traditional unit operations are used. For cell harvesting, MF can be used as an alternative operation to centrifugation or in combination with centrifugation [1,3]. When continuous processes in particular are adopted, filtration is preferred over centrifugation or in combination with centrifugation [1,3]. Separation of cells and cell debris can be facilitated by precipitation or flocculation techniques [6]. Expanded bed chromatography is also used for initial harvest of therapeutic proteins [2]. However, all these techniques can potentially be substituted by MF.

UF is mainly used for concentration of proteins and can also be used to remove viruses from the fermentation broth when producing for example, recombinant DNA in mammalian cells. This step is often termed nanofiltration (NF) although a UF membrane with a low pore size is used [3]. Plasmid purification usually relies on precipitation and chromatography that also involve use of harsh solvents, toxic chemicals, or enzymes not recommended for manufacture of pharmaceutical grade plasmid DNA [7]. Here UF can also be shown to be a good substitute [7,8]. For final purification, crystallization and column chromatography can be replaced by UF, and charged UF membranes are of special interest regarding replacing column chromatography [9].

Furthermore, UF and diafiltration (DF) are currently used for the formulation of nearly all biotherapeutics to achieve the final product concentration and buffer composition [10].

Filtration operations are becoming increasingly advanced and are now shifting from conventional tangential flow filtration (TFF) to single pass tangential flow filtration (SPTFF). In contrast to TFF [11] SPTFF...
enables continuous filtration for inline concentration and buffer exchange in downstream bioprocessing of high value biological products [5,12]. In conventional TFF, the fluid mainly flows parallel to the plane of the membrane. A relatively high flow rate prevents concentration polarization and membrane fouling. However, the high speed also leads to only small concentration effects on the stream after a single pass and hence recirculation is required [2]. This increases the energy demand and may result in an unwanted temperature increase [13]. The high flow rate also increases the shear stress on the molecules and can result in denaturation of sensitive biomolecules which can lead to problems with foaming [13,14]. An alternative to TFF is normal flow filtration (NFF) which is also referred to as dead-end or direct flow filtration (DFF) [2].

In DFF, the flow velocity is perpendicular to the plane of the membrane. NFF prevents high shear stress but quickly results in strong concentration polarization, fouling of the membrane, and low fluxes through the membrane [13]. In TFF, the transmembrane pressure drives the fluid through the membrane but cross-flow velocity provides a force that sweeps molecules away from the membrane surface so that back-diffusion is increased, which minimizes the decline in flux resulting from membrane fouling [15]. To overcome the problems associated with TFF and NFF, SPTFF was developed by Gaston de los Reyes in 2005 [16]. Applications of SPTFF using UF membranes have been reported earlier, for example for blood concentration [17] and production of apple juice [18], but Gaston de los Reyes specifically adapted the technology for concentration of biomolecules with an optimized setup of modules [13].

TFF has traditionally been performed in a batch mode operation with recirculation of the retentate. This approach has some disadvantages for the quality of the system and the overall economics of the process. In this mode of operation, the entire volume of the batch must be handled in a single operation. The fluid also spends time recirculating through the system, which results in a dynamic state that is difficult to monitor. No steady state is ever obtained since the concentration of the feed and the retentate flow changes with time due to the recirculation from retentate to feed [11]. SPTFF is based on the existing technology of TFF by using the latter technique in a different operation mode [11]. For most applications, SPTFF systems can be assembled and operated using existing and well known TFF components that are commercially available. These standard TFF system components include cassettes comprising filtration membranes, cassette holders, tubing and piping for feed, retentate and permeate, valves, gaskets, and pumps [19].

In shifting from TFF to SPTFF, regardless of whether a continuous or a batch process is chosen, the volume of the fluid and the total time it takes to pass through the filter is relatively small compared to TFF. In SPTFF, there is no recirculation of the retentate which means that the fluid moves across the membrane only once to result in a steady state mode of operation [11,20]. Instead of having a concentration process that occurs at phases in time due to recirculation, the SPTFF process occurs throughout the length of the flow path [11,21]. This feature makes it easy to monitor the process constantly by measuring critical quality attributes which will be constant throughout the process. A comparison between TFF and SPTFF is illustrated in Fig. 1. SPTFF is especially suited for concentration of protein solutions [11,15,22], purification of proteins [15], and for buffer exchange through DF to achieve the final targeted buffer formulation [22,23,24]. Common applications include concentration prior to capture chromatography, in-process volume reduction and concentration, in-process dilution and desalting, and as final concentration step [14].

Some important differences between TFF and SPTFF are summarized in Table 1.

The principle behind SPTFF is that the product obtains the desired concentration after the product stream has passed only once through the membrane modules. This is in contrast to the dozens or even hundreds of passes used in conventional TFF where the retentate is recirculated. Since there is no retentate line for the recirculation of retentate, a much simpler setup is possible with SPTFF. In order to make the separation effective enough in one single pass with SPTFF, the residence time in the feed channels has to be increased compared to traditional TFF. The increased residence time can be accomplished by reducing the flow rate in the feed and increasing the path length through a serial configuration of the membrane-holding cassettes [11,15,25]. Serial configuration of SPTFF is illustrated in Fig. 1b. The increased residence time results in an increased conversion, and therefore only a single pass is necessary to obtain the desired product concentration. The products of interest, the biological molecules such as proteins, are retained in the concentrated retentate [20]. The membrane is permeable to the buffer components [25].

SPTFF can also be operated in parallel mode where the feed solution crosses the entire membrane area and the permeate is removed once. Even though parallel SPTFF offers a simpler configuration, installation costs are often higher because SPTFF requires a larger membrane area to achieve the same performance as serial SPTFF [4].

SPTFF is a very important technology with regards to process intensification [26,27], where more product is generated in less time and with smaller equipment, and continuous production methods [24]. During the last few years there has been an upsurge of interest in continuous processing [5,28]. The SPTFF process is itself continuous by nature since no recirculation is needed. The technology helps in maintaining a continuous process by reducing the volumes that need to be handled by subsequent unit operations. These features give SPTFF the potential to play a major role in the shift away from batch processes to continuous processes, for example in the pharmaceutical industry where

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**Table 1**

<table>
<thead>
<tr>
<th>Equipment</th>
<th>Steady-state</th>
<th>Feed concentration</th>
<th>Concentration profile</th>
</tr>
</thead>
<tbody>
<tr>
<td>TFF</td>
<td>One stage</td>
<td>No</td>
<td>Changes in times</td>
</tr>
<tr>
<td></td>
<td>Recirculation tank and pump needed</td>
<td>Varies due to recirculation</td>
<td></td>
</tr>
<tr>
<td>SPTFF</td>
<td>Multi-stage</td>
<td>Yes</td>
<td>Changes along the flow-path</td>
</tr>
<tr>
<td></td>
<td>No recirculation</td>
<td>Constant</td>
<td></td>
</tr>
</tbody>
</table>

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Fig. 1. Comparison of tangential flow filtration (TFF) and single pass tangential flow filtration (SPTFF). Unlike TFF, SPTFF does not involve recirculation of the retentate. The serial configuration of SPTFF increases the residence time of the fluid and therefore increases the conversion.
continuous production is of increasing interest [29]. A number of cost-reducing elements will be introduced in the shift from batch to continuous processing. These elements include reduction in size and number of hold vessels, reduced size of equipment and hence reduced facility footprint, reduced volume of chromatography sorbents and consumables, reduced buffer requirement, and reduced processing times, all of which will increase productivity [28].

SPTFF has a wide area of application, especially in bioprocesses. The technology can be implemented anywhere where TFF has normally been integrated and anywhere where a volume reduction is required. In bioprocesses, such steps are mostly before or after column chromatography or filtration steps, and as the final concentration and formulation step [22,23,26]. Mild process conditions and low operating costs are some of the major advantages of using membranes for concentration and purification [22,23].

2. Design equations for SPTFF

An SPTFF system is built by using multiple conventional TFF membrane modules set in series and/or in parallel [11]. Most often, UF membranes [11,20,22,23] and flat sheet cassettes are used in processes involving biomolecules [21]. These modules come with a higher retention and yield compared to hollow fiber modules. Hollow fibers have a gentle flow and generate little shear, but at large scale they need a relatively high feed flow rate to generate an acceptable flux and are less efficient than flat sheet cassettes for concentrated feeds [21]. As a rule of thumb the selected membrane should have a molecular weight cut-off that is 3-6 times lower than the molecular weight of the molecules to be retained [30]. UF membranes are often made of regenerated cellulose [11,15,20,29] but polyethersulfone membranes are also a possibility [20,25,31]. A comparison of advantages and challenges of the two polymer materials can be seen in Table 2.

The multiple stages of membrane units in SPTFF are gathered in one single module. The membrane area may be less for the later stages by placing the cassettes in parallel to account for the reduction in volume as the fluid passes through the system and to maintain fluid velocity, control pressure, and avoid fouling [11]. This design is illustrated in Fig. 2 where eleven membranes are gathered in one single unit in a 3-3-2-1 configuration (i.e. the number of parallel cassettes in each stage). Each stage is separated by a diverter plate.

In some configurations, the cassettes are placed only in series with one membrane in each stage each with the same membrane area [20].

The single pass mode ensures that the feed concentration is constant since there is no recirculation. This allows the system to reach steady-state equilibrium. With a constant feed flow rate, \( Q_{\text{feed}} \), retentate flow rate, \( Q_{\text{retentate}} \) and a constant permeate flow rate, \( Q_{\text{permeate}} \) (m³/h), there is the following relationship between the flow rates [11]:

\[
Q_{\text{total}} = Q_{\text{retentate}} + Q_{\text{permeate}}
\] (1)

The conversion, or the volumetric concentration factor, VCF, is also kept constant during the operation and is described by [11]:

\[
VCF = \frac{Q_{\text{retentate}}}{Q_{\text{total}}}
\] (2)

The permeate flux through the membrane, \( J_p \) (L/m²·h), is the ratio of the permeate flow rate (m³/h) and the membrane area \( A \) (m²) [11]:

\[
J_p = \frac{Q_{\text{permeate}}}{A}
\] (3)

As with conventional TFF, concentration polarization is one of the major drawbacks of SPTFF. Concentration polarization reduces the permeate flux compared to the pure water flux, and therefore lowers the retentate concentration [15,22]. After reaching steady state, the flux remains constant, as was shown by Thakur and Rathore [34]. The flux decline caused by the formation of a concentration layer is described by the stagnant film model developed by Michaels [22,23,35,36]:

\[
J_p = k \ln \left( \frac{c_m - c_r}{c_b - c_r} \right) \approx k \ln \left( \frac{c_m}{c_b} \right)
\] (4)

Here, \( k \) is the mass transfer coefficient, \( c_m \) is the concentration at the membrane surface, and \( c_b \) is the bulk phase concentration. \( c_r \) is the concentration of protein in the permeate, and the approximation in Eq. (4) is valid for \( c_r \approx 0 \) [35]. The mass transfer coefficient \( k \) can be experimentally determined from Eq. (4). The mass transfer coefficient increases with increasing feed flow rate [35]. The permeate flux decreases with an increasing degree of fouling at a constant transmembrane pressure [37].

A method for sizing SPTFF UF systems is given by Eq. (5) [35]. The maximum achievable conversion \( \varphi_{\text{max}} \) in the pressure-independent region is based on Eq. (4) and is given by:

\[
\varphi_{\text{max}} = \frac{J_p A}{Q_{\text{feed}}} = \frac{A}{Q_{\text{feed}}} \cdot k \ln \left( \frac{c_m}{c_b} \right)
\] (5)

The conversion is a function of the membrane area \( A \), which is determined from a specified desired conversion \( \varphi_{\text{max}} \). By increasing the membrane area, the permeate flow rate increases and hence an increased volume reduction is obtained [35]. The ability to determine the membrane area needed for a given separation remains one of the challenges in the design phase. A recent approach is that of Jabra et al. [12] who modelled the membrane area as a function of channel width for an SPTFF module for a desired protein concentration at a given flow rate. Future studies will examine the design of a staged SPTFF module to take into account the specific configuration of the channels i.e., the number of parallel channels in each stage of the module.

The structure of the channels in SPTFF is specified by two quantitative parameters. The first is the specific membrane area of the channel \( \alpha_c \) (m⁻¹), which is expressed as the ratio of the membrane area to the void volume of the channel [38]:

\[
\alpha_c = \frac{A_c}{V_c}
\] (6)

<table>
<thead>
<tr>
<th>Advantages</th>
<th>Challenges</th>
</tr>
</thead>
<tbody>
<tr>
<td>Regenerated cellulose</td>
<td>High flow rates and high durability. High thermal stability and chemical resistance. Limited stability in oxidizing agents.</td>
</tr>
<tr>
<td>Polyethersulfone</td>
<td>High thermal stability and chemical resistance. Hydrophilic nature but can be modified by use of co-polymers. Higher hydrophobicity.</td>
</tr>
</tbody>
</table>

Table 2: Advantages and challenges associated with the two polymer materials used in SPTFF and TFF ultrafiltration membranes [1,2,32,33].
A_{fc} is the area of the flow channel (m²) and V_{fc} is the void volume of the flow channel (m³). The second parameter is the dimensionless length \( \lambda \) which is the product of the specific membrane area \( \sigma \) and the length of the channel in either one stage or the whole system [38]:

\[
\lambda_{\text{stage}} = \sigma \cdot L_{\text{stage}}
\]

\[
\lambda_{\text{system}} = \sigma \cdot L_{\text{system}}
\]

\( \lambda_{\text{stage}} \) is the dimensionless length of one stage, and \( L_{\text{stage}} \) is the channel length of one stage, while \( \lambda_{\text{system}} \) is the dimensionless length of the whole system and \( L_{\text{system}} \) is the channel length of the whole system. These equations are used to compare the attributes of different SPTFF systems [38].

Extensive models are being developed to account for concentration polarization, pressure drop, and mass transfer through the membrane in SPTFF systems [12,22]. If the properties of the membrane modules are known from models, an SPTFF system can be designed and integrated in almost any process sequence with less experimental effort and increased understanding of the process [23]. Just recently, a hybrid model has been developed by Krippel et al. [4] to predict the concentration performance of SPTFF at various pressures, feed flows and protein concentrations for up to three membranes. The hybrid model consists of a data-driven, statistical part and a mechanistic, knowledge-based part, and requires only a minimal number of experiments for model training. The hybrid model can be used as a Digital Twin to perform process simulations, which facilitates faster process transfer from batch to continuous mode and predicts how SPTFF performs under varying process conditions. Another recent modelling approach has been presented by Thakur and Rathore [34]. This model leverages the gel polarization model of protein UF to develop a model for the permeate flux as a function of time of a single membrane inside an SPTFF module. The flux is predicted based on three resistances: boundary layer resistance, resistance of the deposited protein layer over time, and intrinsic membrane resistance. This single membrane model is then used to model complex SPTFF configurations based on operating targets. The model is able to predict the flux vs. time profile in various SPTFF configurations within 10% accuracy.

A recent study by Jabra and Zydney [39] has interestingly revealed the influence of cassette configuration on concentration factor and pressure drop. By placing six cassettes with the same membrane area all in series, the highest concentration factor was obtained compared to placing the cassettes in various parallel configurations. Placing all the six cassettes in parallel, however, led to the lowest pressure drop at all feed flow rates, but also the lowest concentration factor. When aiming for a particular concentration factor, the lowest membrane area was obtained when placing the cassettes in series only, but this configuration also caused the highest pressure drop. This pressure drop could be significantly reduced by various parallel configurations but with a corresponding increase in total membrane area. For example, a module with a 3–2–1–1 configuration provided a two-fold reduction in pressure drop with less than a 35% increase in membrane area when a two-fold concentration factor was desired. The optimal trade-off between membrane area and pressure drop is determined by the overall process economics.

3. Fouling and cleaning

Several studies have evaluated the effect of flux and protein concentration on fouling. The results, however, are unclear because the degree of fouling depends on both the nature of the proteins and the membrane material [27]. For SPTFF systems with membranes of regenerated cellulose, a static cleaning procedure has been proposed after protein concentration to avoid fouling [32]. Polyethersulfone membranes are in general more susceptible to fouling and may require more extensive cleaning than regenerated cellulose membranes [32]. In TFF and SPTFF, the effect of concentration polarization is not as problematic as in dead-end or direct flow filtration [15].

The retained solutes that accumulate at the surface of the membrane form a polarized boundary layer. This phenomenon is what is called concentration polarization, and the concentration of solute at the membrane surface \( c_m \) is related to the concentration of solute in the bulk solution \( c_b \). By isolating \( c_m \) from Eq. (4) the expression in Eq. (9) is obtained [25].

\[
c_m = c_b \left( S_o + (1 - S_o) \exp \left( \frac{L}{L_{\text{system}}} \right) \right)
\]

\( S_o \) is the sieving coefficient determined by Eq. (10) [25].

\[
S_o = \frac{c_f}{c_b}
\]

In SPTFF the sieving coefficient of partially retained proteins is not constant because protein concentration changes throughout the length of the module. The hydraulic also change at each point in SPTFF, which makes it difficult to measure \( S_o \). An overall mass balance can be used, however, to calculate an overall average sieving coefficient of the system. If the protein is completely retained, the sieving coefficient is set to \( S_o = 0 \) [25].

Turbulence promoters, or feed screens, can be incorporated into the feed channel to reduce concentration polarization. The screen, which is a net-like element present between the membrane sheets, generates a complex secondary flow within the channel. The screen provides directional changes of the flowing fluid, which increases pressure drop and the channel shear, and creates eddies ensuring mixing within the channel. In this way complete pore-plugging and the extent of concentration polarization is reduced [37].

UF membranes are commonly used several times, which makes it important to develop cleaning procedures for thoroughly cleaning after each use. This requires both expensive raw materials, such as cleaning
buffers and pure water, and is also very time-consuming because a cleaning procedure can take several hours to run [40]. The cleanliness of a membrane after a cleaning procedure is determined by the normalized water permeability (NWP). The NWP method involves measuring the passage of clean water through the membrane under standard pressure and temperature conditions. The water flux through the membrane (L/m²/h) is divided by the transmembrane pressure (bar) to obtain NWP (L/m²/h/ bar). The values of NWP after cleaning are compared to initial levels and can be analyzed for trends over time. Fouled membranes typically have NWP values of less than 50% of the original NWP [1]. A substantial reduction in NWP has also been reported by Thakur et al. [41]. After concentration of monoclonal antibodies with SPTFF, NWP dropped to 50–60% of the initial value but recovered to more than 95% after cleaning with NaOH [41]. A cleaning procedure involving NaOH followed by flushing with buffer or water is often used for cleaning the SPTFF system after concentration of proteins [15,20,32,41]. To avoid cleaning the membranes, single-use membranes that are replaced after each filtration process can be used [40]. This will also avoid the risk of cross-contamination [9]. Whether to use single-use or multi-use membranes depends highly on the membrane costs. For multi-use membranes, the cost for cleaning agents dominates the operating cost. The operation cost due to cleaning procedures needed in membrane separations is astonishingly high and seems to be the main objective for further improvements in this area [40].

The cost is not just due to the cleaning agent itself but also due to disposing of the used agents and the time the cleaning procedure takes. In principle, there is no difference in cleaning procedure between conventional TFF and SPTFF since the membranes used are the same.

4. Applications of SPTFF in bioprocesses

Bioprocesses have progressed significantly in the recent years. Unless stated otherwise the examples in this section deal with application in the biopharmaceutical industry. The upstream productivity of bioprocesses has improved [40,9,25], which has led to an increased demand on the downstream processes because the volumes that must be handled have increased [42]. In many existing facilities, the downstream capacity has become the bottleneck [11]. The increased volumes [42] and titers [25,27] put an enormous pressure on the downstream operations. Increased titers mean that the downstream processing should be able to handle more biomass and product [25]. One solution to this is SPTFF that provides an effective though simple way of reducing process volumes through inline concentration and thus a debottlenecking of downstream capacity. SPTFF can be used to concentrate process intermediates with the result that smaller holding tanks are required and loading times for subsequent unit operations are reduced [27]. SPTFF has the potential to reduce the volumes from upstream production by a factor of 15 to 25 [42], meaning that downstream capacity could potentially be reduced to handle only 1/25 of the volume of the upstream production. Hence, implementation of SPTFF will reduce the tank size of some of the subsequent unit operations and lead to better process economics since only smaller volumes would need to be handled. This is beneficial especially for the capital cost of downstream equipment, which can be smaller [29]. Since SPTFF is designed from conventional TFF stages in a single unit, the SPTFF unit itself requires minimal capital investment and footprint area [11]. Another major reason for moving from batch production to a continuous process is the stability of the products. Biomolecules are very labile and are progressively denatured if not continuously recovered [3]. Furthermore, by moving from TFF to SPTFF the risk of damaging the sensitive molecules in the recirculation loop is precluded. Product recoveries for SPTFF are typically above 99% under optimized conditions [14]. One example of a commercial application of SPTFF was reported by C. A. Teske [35] for the production of a monoclonal antibody. Improved cell culture titers placed increased demands on the downstream processes, and to address this challenge inline UF for volume reduction in the already existing manufacturing facility was implemented with great success and minimal cost.

UF membranes are used extensively in the dairy industry for the fractionation of protein, lactose and minerals [43,44]. This is also an industry where SPTFF has been widely used for whey manufacturing [32]. Wastewater treatment is also one of the areas where SPTFF has been utilized for many years [45]. Just recently, SPTFF has been used in biopharmaceutical processes where the technology is now gaining a foothold [32].

Some of the many applications of SPTFF are summarized in Table 3 where the components in the permeate and the retentate are listed together with the stream in which the product of interest is found. Since most of the literature dealing with these applications refers to laboratory and pilot scale only, membrane areas and flow rates will vary considerably and are not indicative of industrial application, and therefore this information is not included in the table.

As early as in 2006, a biopharmaceutical industry survey revealed that most companies at that time considered downstream processing to be more important than the upstream production phase for productivity and process economy. The same survey also concluded that disposable membrane technology was one of the most promising ways to address this challenge, for instance for flow-through operations where the objective is to capture impurities rather than the product [9]. Now, technology is also moving towards single-use SPTFF units [29,48] and this development is highly relevant for the pharmaceutical industry. The single-use SPTFF units are sterilized beforehand and are fully scalable [29]. After use, the units are disposed of and there is no risk of cross-contamination [14] or any need for handling fouling [48]. Down-time for cleaning is thus avoided, increasing the viability of the process. Combining principles of single-use technology with continuous UF systems is of increasing interest in the biopharmaceutical industry [48].

Several single-use SPTFF modules have already been developed in

Table 3
Examples of applications of SPTFF.

<table>
<thead>
<tr>
<th>Application of SPTFF</th>
<th>Product of interest in</th>
<th>Components in permeate</th>
<th>Components in retentate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Purification of plasmid DNA [7, 8, 25]</td>
<td>Retentate</td>
<td>RNA (or short oligonucleotides hereof)</td>
<td>DNA</td>
</tr>
<tr>
<td>Concentration of monoclonal antibodies [11,15, 20, 25, 31]</td>
<td>Retentate</td>
<td>Proteins remaining after lysis</td>
<td>Water</td>
</tr>
<tr>
<td>Production of apple juice [18]</td>
<td>Permeate</td>
<td>Clarified apple juice</td>
<td>Suspended solids, partly retained sugars</td>
</tr>
<tr>
<td>Wastewater treatment [32, 45, 46]</td>
<td>Permeate</td>
<td>Purified water</td>
<td>Organics, suspended solids, bacteria</td>
</tr>
<tr>
<td>Whey manufacturing [32, 47]</td>
<td>Retentate</td>
<td>Water, small molecules such as lactose, non-protein nitrogen, minerals, riboflavin, cheese color</td>
<td>Whey protein</td>
</tr>
<tr>
<td>Blood concentration [17]</td>
<td>Retentate</td>
<td>Plasma water</td>
<td>White blood cells, hemoglobin, platelets, albumin, fibrinogen</td>
</tr>
</tbody>
</table>
the past few years. The membranes can be rapidly employed in the downstream processing because they are all pre-sanitized. Pre-sanitization also eliminates the need for developing and validating a cleaning schedule for the membranes [48]. Until recently, single-use SPTFF systems have been quite expensive. However, by utilizing existing cheap hollow-fiber membrane modules, a low-cost format of SPTFF appropriate for single-use applications can be achieved. This type of single-use SPTFF system has been reported to provide a more than ten-fold concentration of Immunoglobulin G. The membranes could also be operated for more than 120 h without the need for chemical or physical cleaning and with only minimal levels of fouling. This resulted in a total membrane module cost of less than 0.004 $ per g of protein for the UF process. This cost could perhaps be further reduced by extending the operating time, which might be possible due to the absence of any significant fouling during the 120 h run [48].

Some disadvantages of single-use technology count ongoing replacement of the disposables and thus an increased cost of solid waste disposal [49]. The disposal must be compared with waste from traditional cleaning [50], and an economic and environmental evaluation needs to be done to compare the viability of single-use versus traditional membrane systems, taking end-of-life processes into consideration [51]. DF is the industrial-scale unit operation for buffer exchange [3] and can also be run continuously, but requires a more complex setup than continuous filtration, as multiple filter units and intermittent dilution steps need to be included [52,53,21]. Rucker-Pezzini et al. [54] reported a single-pass DF process using three consecutive commercial SPTFF modules with inter-retenate vessels into which the DF buffer was added. A buffer exchange rate of 99.75% was achieved in a monoclonal antibody case study. Although the buffer demand is higher, single-pass DF could be beneficial in fully continuous processes. Nambari et al. [55] presented a countercurrent two-stage configuration with a recirculation of the permeate buffer to reduce the buffer demand, which provides 99% buffer exchange. A buffer exchange of 99.9%, which is the target for most bioprocessing applications, could however only be achieved with a three-stage design [56]. In general, advantages of single-pass DF are continuous operation, fewer pump passes for the product, and potentially lower buffer requirements [55]. However, this comes at the cost of a higher system complexity and the use of a larger membrane area. As described by Jabra et al. [56], more studies will be needed to demonstrate the effectiveness of single-pass DF in commercial manufacturing processes, which currently makes conventional UF/DF the preferred solution in industrial applications [21].

5. Future perspectives

A recent advance, which will probably be more frequent in the future, is 3D printed membrane modules [13,24]. This technology has been specifically applied to produce 3D printed UF/DF modules for continuous and simultaneous concentration of retained biomolecules and exchange of buffer salt [13]. All parts of the membrane module, except for the membrane itself, can be 3D printed. The 3D printed parts provide mechanical support for the membrane and form the required liquid distribution system. By 3D printing the modules, the liquid distribution system can be customized to the exact purpose of the module [13].

In contrast to the multi-stage DF setups described above, only a single membrane module is used for buffer exchange in the studies by Tan et al. [13,24]. The modules consist of two membranes with a continuous in and out flow perpendicular to the flow direction in the feed channel. To reduce the emergence of a concentration polarization layer, the buffer flow direction is alternated, yielding a buffer exchange of 99.9% [24].

If 3D printed modules for liquid distribution and mechanical strength are really to evolve, there will be a need for more research since only small-scale systems have been tested to date [13,24].

Complete control of SPTFF is very important for continuous production. Recently, Process Analytical Technology (PAT) has been introduced to control the output concentration of a continuous SPTFF regardless of variations in volume or feed concentration. PAT is used to measure critical process parameters and critical quality attributes so as to provide a robust process control in biopharmaceutical processes. However, PAT approaches are very limited in SPTFF technology today but will most probably be introduced to a greater extent over the coming years due to the crucial role played by PAT in controlling continuous manufacturing processes [41].

Some recent patents utilize the SPTFF technology for the production of stable protein compositions, injectable pharmaceuticals, and generally as a process for purifying target substances. However, only a few patents deal with the invention of new SPTFF systems. One was published in 2019 by Gaston de los Reyes [57], the inventor of modern SPTFF, who holds several patents in membrane technology and SPTFF systems. In this patent, a continuous SPTFF device is disclosed for providing a constant concentration output. The process is said to provide a high conversion concentration at relatively low feed flows. The system should also be able to provide constant output for DF.

A patent published in 2019 by Abhiram Arunkumar [58] describes a hybrid method configuration for enhancing the concentration of macromolecule solutions. The system uses at least two semi-permeable membranes with different molecular weight cutoffs in a SPTFF unit at a high feed flow rate for an efficient concentration of biological macromolecules. This system is claimed to reach final product concentrations up to 150-fold higher than the feed concentration.

A patent by Nathan Landry [59] published in 2020 discloses a method of manufacturing a spiral wound membrane module which might be suitable for single-use. This underlines the interest in moving towards cheap membrane modules that can be discarded after a short operating time.

With the increasing interest in moving towards continuous processing and process intensification it is expected that more patents and articles on SPTFF will be seen in the coming years. As SPTFF is still a relatively new technology, more research is desirable to increase the current knowledge and understanding of the technology. Both more theoretical experience, for example through mechanistic modelling, and more practical experience, for example from systematic experimental testing, could help to use SPTFF under optimal conditions i.e., optimal operating conditions and filter configurations, to maximize efficiency and boost the application of SPTFF in many bioprocesses.

6. Conclusion

SPTFF is a technology that has been of increasing interest during the last decade in the biopharmaceutical industry. As a connecting link between upstream and downstream processing, SPTFF can be utilized to reduce the volumes to be handled by holding tanks and downstream processing. In contrast to conventional tangential flow filtration, in SPTFF the fluid moves across the membrane only once because there is no recirculation of the retentate. A high concentration of the retentate is reached by increasing the residence time, by reducing the flow rate in the feed, and by increasing the path length through the membrane. The introduction of SPTFF can ensure a continuous process that may be used both as a volume reduction procedure and as a final purification step.

Single-use SPTFF modules are being developed as part of process intensification efforts to decrease both capital and operating costs by introducing cheap membrane modules and eliminating the need for cleaning procedures. 3D printing introduces user-designed membrane modules tailored to the exact process of interest.

Even though the literature on SPTFF is still limited, most papers have been published in the last few years and more patents are being filed. This field may therefore evolve rapidly during the next decade in step with increasing interest in continuous manufacturing of biopharmaceuticals and the need to reduce volumes from upstream to downstream processes to bring down equipment and operational costs.
Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References