mAbs 1:5, 443-452; September/October 2009; © 2009 Landes Bioscience

Industrialization of mAb production technology
The bioprocessing industry at a crossroads

Brian Kelley

Bioprocess Development; Genentech; South San Francisco, CA USA

Key words: bioprocessing, cell culture, purification, economics, capacity, manufacturing, production, facility, biopharmaceutical

Manufacturing processes for therapeutic monoclonal antibodies (mAbs) have evolved tremendously since the first licensed mAb product in 1986. The rapid growth in product demand for mAbs triggered parallel efforts to increase production capacity through construction of large bulk manufacturing plants as well as improvements in cell culture processes to raise product titers. This combination has led to an excess of manufacturing capacity, and together with improvements in conventional purification technologies, promises nearly unlimited production capacity in the foreseeable future. The increase in titers has also led to a marked reduction in production costs, which could then become a relatively small fraction of sales price for future products which are sold at prices at or near current levels. The reduction of capacity and cost pressures for current state-of-the-art bulk production processes may shift the focus of process development efforts and have important implications for both plant design and product development strategies for both biopharmaceutical and contract manufacturing companies.

Background

Bioprocessing technology for production of therapeutic monoclonal antibodies (mAbs) has advanced greatly since their introduction into the market in 1986. Early murine mAbs were derived from hybridoma cell lines, using diverse production technology; the first licensed mAb therapeutic, OKT3, was produced in the ascites of mice.[1] The development of recombinant technology based on cloning and expression of the heavy and light chain antibody genes in CHO cells enabled mAb production to take advantage of the common technologies already used for recombinant products like tissue plasminogen activator, erythropoietin, Factor VIII, etc. These recombinant cell culture processes for antibody production initially had low expression levels, with titers typically well below 1 g/L.[2]

The combination of low titers and large market demands for some of the first recombinant mAbs like rituximab (Rituxan), trastuzumab (Herceptin), infliximab (Remicade) and others drove many companies and contract manufacturing organizations (CMOs) to build large production plants containing multiple bioreactors with volumes of 10,000 L or larger. Other products derived from mammalian cell culture in the mid-90s also required large production capacity (Enbrel, while not a mAb, is an Fc-fusion protein which is produced using a similar manufacturing process), driving further expansion. In parallel with the increase in bioreactor production capacity throughout the bioprocessing industry, improvements in the production processes resulted in increased expression levels and higher cell densities, which combined to provide much higher product titers.

Today, the potential of combining high titer process technology with the large installed bioreactor base has resulted in a great excess of production capacity for mAbs, far outstripping the increase in market demands over recent years. This has stimulated discussions of the controversial issues of the best use of current production capacity, the impact of manufacturing cost of goods (COGs), and the choice of the appropriate mAb production technology for emerging product candidates. Should companies choose conventional bioprocessing technologies, or invest in novel technologies which may lead to superior expression levels or lower production costs? Have process development strategies adjusted to this paradigm shift where nearly unlimited capacity and very low COGs are enabled by the current state-of-the-art? If not, how should process development groups respond?

This article will analyze the current mAb production technology, review production capacity and demand estimates, and consider the position of these conventional technologies in the future of commercial mAb production for therapeutic use.

Current State-of-the-Art: Potential for mAb Process Industrialization

The processes for manufacturing recombinant therapeutic mAbs have several common features, and efforts to benchmark the current state-of-the-art draw upon information that is shared at conferences, but often not published. For production of purified bulk drug substance, i.e., the intermediate that is used to produce the final drug product sold to healthcare providers and patients, a consensus process has emerged from the major biopharmaceutical process development groups (**Fig. 1**).

Mammalian cells are used for expression of all commercial therapeutic mAbs, and grown in suspension culture in large bioreactors. The majority of commercial mAbs are derived from just a few cell lines[3] (Chinese Hamster Ovary (CHO), NS0, Sp2/0), with CHO being the dominant choice because of its long history of use since the licensure of tissue plasminogen activator in 1987.

Correspondence to: Brian Kelley; Email: Kelley.brian@gene.com
Submitted: 06/18/09; Accepted: 07/16/09
Previously published online:
www.landesbioscience.com/journals/mabs/9448

DOCKET ALARM
Find authenticated court documents without watermarks at docketalarm.com.

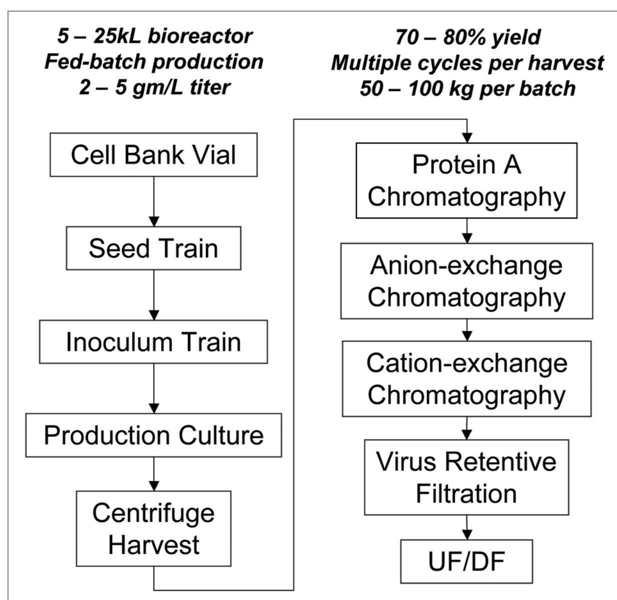

Figure 1. Consensus process flowsheet for mAb Bulk Drug Substance. A consensus process flowsheet has emerged for production of recombinant therapeutic mAbs. Suspension mammalian cell cultures bioreactors operating in fed-batch mode provide high product titers in 10–14 days. Following harvest by centrifugation and depth filtration, Protein A chromatography captures the product, and two additional chromatographic polishing steps complete the purification. Two membrane steps are used to assure viral safety of the product, and concentrate and formulate the drug substance.

CHO cells have attractive process performance attributes such as rapid growth, high expression, and the ability to be adapted for growth in chemically-defined media. Typical production processes will run for 7–14 days with periodic feeds when nutrients are added to the bioreactor. These fed-batch processes will accumulate mAb titers of 1–5 g/L, with some companies reporting 10–13 g/L for extended culture durations. Production bioreactor volumes range from 5,000 L (5 kL) to 25,000 L (25 kL).

The antibody purification process is initiated by harvesting the bioreactor using industrial continuous disc stack centrifuges followed by clarification using depth and membrane filters. The mAb is captured and purified by Protein A chromatography, which includes a low pH elution step that also serves as a viral inactivation step. Two additional chromatographic polishing steps are typically required to meet purity specifications, most commonly anion- and cation-exchange chromatography.[4] A virus retentive filtration step provides additional assurance of viral safety, and a final ultrafiltration step formulates and concentrates the product (the step order of the virus filter and two polishing steps is somewhat flexible, and may vary among company platforms).[5] Overall purification yields from cell cultured fluid range from 70–80%, and the concentrated bulk drug substance is stored frozen or as a liquid, and then shipped to the drug product manufacturing site. While the purification processes developed in the 1990s using the separations media (chromatographic resins and membranes) available at the time were not capable of purifying 2–5 g/L feedstreams, improvements in separations media make it possible today for many facilities to purify up to 5 g/L, which would generate batches of 15–100 kg from 10 kL–25 kL bioreactors.

Large manufacturing plants are designed with multiple bioreactors supplying one (or sometimes two) purification train(s). The individual purification unit operations can be completed in under two days, and often in just one day, and therefore several bioreactors can be matched to the output of a single purification train. The increased capacity of these plants arising from the elevated titers will decrease the drug substance COGs, by virtue of the economies of scale afforded by the large bioreactors. As will be described in more detail below, these plants are capable of producing enormous quantities of mAbs with very attractive costs.

Further, this consensus manufacturing process is amenable to standardization that establishes a common processing platform for many mAbs. Each company is likely to use a slightly different platform process, but the similarities outweigh the differences when it comes to the process flowsheet (**Fig. 1**) and the typical manufacturing plant design. The use of a platform approach reduces the investment per mAb product candidate, streamlines development efforts, simplifies raw material procurement and warehousing, and reduces scale-up and technology transfer complexities. Several companies have revealed that they have very similar development timelines from the start of cell line development through first-in-human clinical trials, and many are using common tools such as high throughput systems for cell line and purification process development.

This state-of-the-art has the hallmarks of a highly industrialized family of manufacturing processes. Many companies have converged on the use of very similar processes, this common production technology is mature and robust, and the outcomes of product quality, production capacity and costs are predictable. This standardization and maturation of the mAb process technology has emerged relatively recently, since the early years of the 21st century.

Why would companies need to stray from this mature and convergent platform? In some process development groups, continued advances in cell culture technology have driven mAb titers up steadily, putting pressure on purification technology that would eventually limit or bottleneck the plant's production capacity. Concerns have also been raised about the need for increased production capacity, and pressures to reduce COGs further. These factors could drive the development and implementation of novel bioprocess technologies, such as perfusion technology for cell culture, or non-conventional purification methods like precipitation, crystallization, continuous processing or the use of membrane adsorbers.[6]

Assessment of the process fit into a production facility now enables purification bottlenecks to be identified, and process designs can be adapted to enable larger batches to be purified. Often, new technology is not required, but instead simple adjustments of the consensus process will avoid the typical plant limits of product pool tank volume, unit operation cycle time or supply of process solutions. Overall purification yield, if allowed to drop a few percentage points, can often be a key degree of freedom for debottlenecking a plant as well. The use of current separations media combined with a focus on facility has shown that many plants can be debottlenecked to support titers of up to 5 g/L.[7] If

DOCKET ALARM Find authenticated court documents without watermarks at docketalarm.com.

these conventional platform technologies can generate 50–100 kg batches from existing facilities, is there a driver for larger batch sizes? This question can be put in context of product demand in the subsequent sections.

It is valuable to conduct a critical assessment of these drivers for higher production capacity or reduced COGs and determine the validity of the arguments that the bioprocessing status quo is not sufficient. This has fundamental implications for important aspects of process development, facility management, capital investment and broad future trends in mAb production technology. This analysis will focus on commercial operations, as the clinical stages of the product lifecycle have different objectives that could benefit from flexible and lean operations, capital avoidance strategies and minimal upfront investment. The optimization of clinical process development strategies is a separate topic, but the design of clinical processes should reflect the key elements of the eventual commercial process.

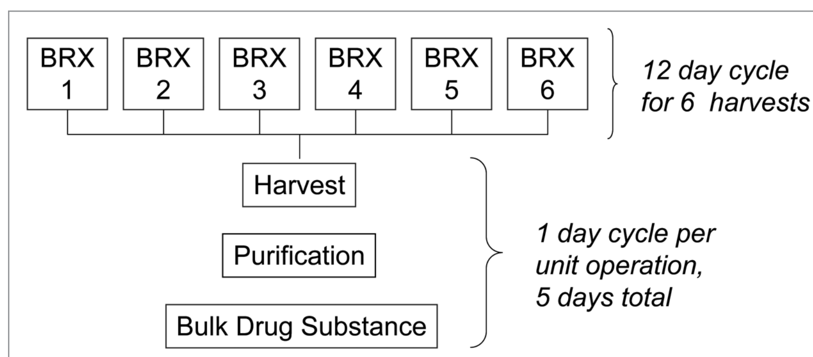

Figure 2. Model mAb production plant design and capabilities. A model large scale mAb production plant employs multiple bioreactors configured to supply a single purification train. A plant having six individual 15 kL bioreactors is potentially capable of supplying 10 tons of purified mAb per year using conventional technologies, or 4–5 products with 1 ton demands. This enormous capacity per plant would result in a marked decrease in drug substance production costs, and results in significant excess capacity throughout the biopharmaceutical industry.

Analysis of Drug Substance Production Capacity for mAb Products

An analysis of the production capacity for mAb drug substance is relatively straightforward, as much of the information on plant capabilities is available to the public. Both internal and external databases[8] were used to develop estimates of mammalian cell capacity and demand. While the number of bioreactors and their volumes are known, details of the purification train capacities are generally not. It has been reported that some facilities can purify up to 5 g/L titers and potentially generate a 100 kg batch from a 25 kL bioreactor, yet it should not be assumed that all facilities could purify such large batches. It would be safe to assume that a 2 g/L titer should be easily supported, however, and that a 5 g/L titer would fit in some, but possibly not all, facilities.

It is useful to examine the production capacity of a single plant, which could be described as a model plant for the purposes of this article. The model plant would have six 15 kL bioreactors, for an installed base of 90 kL capacity (the largest plant in operation today has 200 kL of capacity), and be supported by a single purification train (**Fig. 2**). If this plant ran a cell culture process with a titer of 5 g/L and had no purification limitations, it would offer a capacity of 10 tons of mAb drug substance per year. The design basis for this model plant has been described in the literature,[9] and would use conventional purification technologies that are available today.

In 2007, the installed capacity for mammalian cell processes was 2.3 million liters, and is projected to rise to 4 million liters in 2013 based on current plans for capacity expansion for both CMOs and biopharmaceutical companies (**Table 1**). There will be at least 25 plants with the same or greater capacity of the model plant described above by 2013, with many other smaller plants in operation as well. A conservative estimate can be taken, such that each bioreactor generates 20–24 batches per year, which is

Table 1. Production capacity estimates for mammalian cell-derived mAbs[a]

Year	CMO	Product company	Total	Capacity at 2 g/L	Capacity at 5 g/L
2007	500 kL	1,800 kL	2,300 kL	70 tons/yr	170 tons/yr
2010	700 kL	2,700 kL	3,400 kL	100 tons/yr	255 tons/yr
2013	1,000 kL	3,000 kL	4,000 kL	120 tons/yr	300 tons/yr

[a]Capacity estimates from ref. 8.

consistent with a 12–14 day production culture cycle and a short plant shutdown. When combined with a purification yield of 75%, this equates to 300 tons/yr if the process titer averaged 5 g/L, or 120 tons/yr for a titer of just 2 g/L (**Table 1**). These theoretical capacities need to be compared to current and projected market demands to provide the appropriate context for implications to facility utilization.

While the estimates for drug substance production capacity should be corrected for overage required for drug product manufacturing, the losses in mixing vessels, filling lines, filters and ancillary equipment decrease with filling volumes, and diminish at very large production scales that require large filling volumes. Stability and testing requirements will also impact overall yields. Because these losses are a function of scale, facility and configuration, they are not accounted for in this analysis, but typical losses could be 10–30%, and are not large enough to change the primary conclusions of the capacity analysis.

Analysis of Drug Substance Demand for mAb and Fc-Fusion Products

The estimates of the drug substance market demand rely on a combination of several publically disclosed factors, and cannot be considered a precise value. By using the annual product revenue

DOCKET ALARM Find authenticated court documents without watermarks at docketalarm.com.

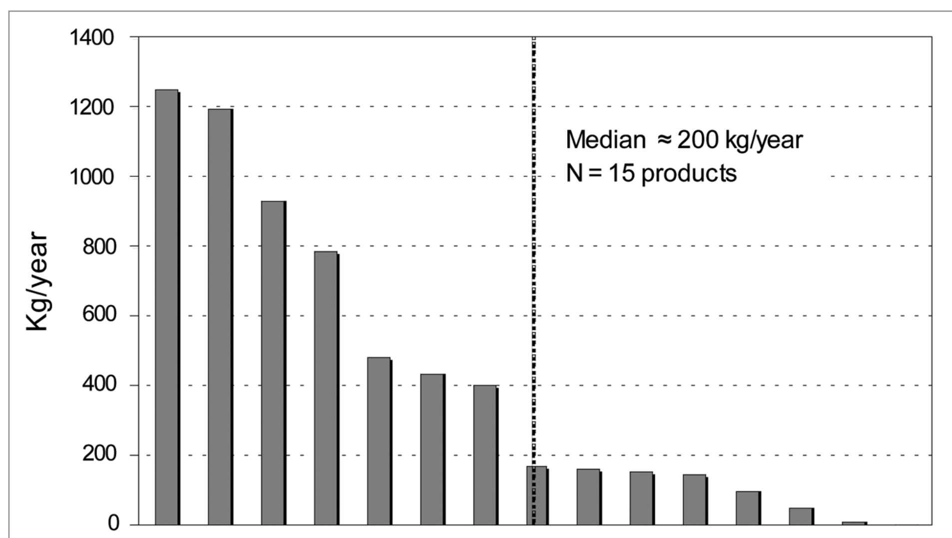

Figure 3. Estimated demand for therapeutic mAbs and Fc-fusion products in 2009. The total demand for the top 15 mAbs and Fc-fusions in 2009 is estimated to be approximately 7 tons, with the four largest volume products requiring approximately one ton per year. More than half of the products were estimated to require less than 200 kg per year (reviewed in ref. 8).

provided in annual reports, and an average wholesale price from public and private databases,[8,10] combined with a modest process yield loss and fill overage upon drug product manufacturing, a rough estimate of the annual drug substance demand can be generated. Both mAb and Fc-fusion proteins such as Enbrel are included in this analysis, as they would share the production capacity given their use of similar production technology. Some Fc-fusion proteins do not accumulate to titers as high as mAbs, and therefore would require proportionately more production capacity. In addition, other recombinant proteins not included in this analysis will also require mammalian cell culture production capacity. A survey of these products is beyond the scope of this review, and their total mass and volumetric demands are much lower than mAbs.

Again, both internal and external databases[8] were used as sources of information. The total estimated demand for therapeutic mAbs and Fc-fusion proteins in 2009 will be 7 tons. The median demand for the 15 licensed products in the database was approximately 200 kg/yr (**Fig. 3**). It is useful to note that this median product demand would be satisfied by just four batches from the model plant described above if the titer was 5 g/L, and only nine batches if the titer was just 2 g/L. It is not uncommon for some companies to have Phase III mAb processes today with titers as high as 4–5 g/L. Even titers of 2 g/L for very late stage products that reflect older cell culture processes will provide sufficient supply for nearly all pipeline products, given access to the large excess capacity in the industry.

Future demand estimates are even less certain, and are a complex combination of the factors that drive mAb clinical development: the probability of clinical success, competition from other pipeline or commercial products, development and regulatory review timelines. Several consultants provide estimates of the demands, which could increase to as much as 25 tons per year in 2013 (data not shown). This value would correspond to an annualized growth rate of 37%, which seems an aggressive value for growth of this sector, which has shown a revenue growth rate of 11%.[11] This demand is still small compared to the production capacity of the industry as a whole, even at modest titers of 2 g/L.

These analyses of production capacity and demand strongly suggest that there will be a significant amount of excess mAb production capacity throughout the biopharmaceutical industry in the foreseeable future. Even if several blockbuster products are licensed which far exceed the current maximum demands of approximately 1 ton per year, they will not give rise to a production challenge if multiple plants can be accessed for production, which has been the pattern of the production lifecycles for bevacizumab (Avastin), etanercept (Enbrel), rituximab and trastuzumab, or if their titers are sufficiently high (2–5 g/L). Access to large production facilities can be assured through the contracts with CMOs, or by partnering with the biopharmaceutical companies that hold the majority of mammalian cell production capacity.

Often, arguments which state that a new technology is required to meet growing therapeutic mAb demands assume that many products will reach blockbuster status and the highest peak product demands in industry are likely to grow in future years. Still, the forces of competition from other biologics or small molecules for common indications, and improved mAb characteristics such as selection for extended pharmacokinetic profiles or lower dose will likely combine to cap demands below 2–4 tons per year for all but the most unusual products. It is important to note that even in the case where a landmark product commands 10 tons per year, a single one of the model plants could cover this demand. Further, as cell culture titers increase in concert with movement of today's molecules through the pipeline and on to becoming commercial products, a smaller number of batches will be required to satisfy the demand.

Thus, it seems that production capacity and cell culture titer will not be drivers for process design targets for almost all pipeline mAb products. Arguments that improved process technologies are needed to debottleneck today's mAb production to satisfy market demand appear to be largely unfounded, but for very exceptional circumstances.

mAb Drug Substance COGs Evaluation and Sales Prices

Data on the production COGs for pharmaceutical products are not typically available in the public domain, but there are

DOCKET ALARM
Find authenticated court documents without watermarks at docketalarm.com.

indications that mAb drug substance costs have dropped significantly in the last decade as larger plants came on line and process improvements increased titers. Published estimates for COGs have shifted from approximately $300/gm[12,13] to $100/gm[14,15] with a potential minimum as low as $20/gm for the model plant producing 10 tons per year.[9] Note that these are only projections, not actual costs, and may reflect the ideal situation where a plant is operated at full capacity. The cell culture titers increased from <1 g/L, to 1–2 g/L, and then 5 g/L for these estimates. Several other COGs estimates from conference presentations and publications range from $50–100/gm for current processes with titers ≥2 g/L, as economies of scale serve to reduce costs.

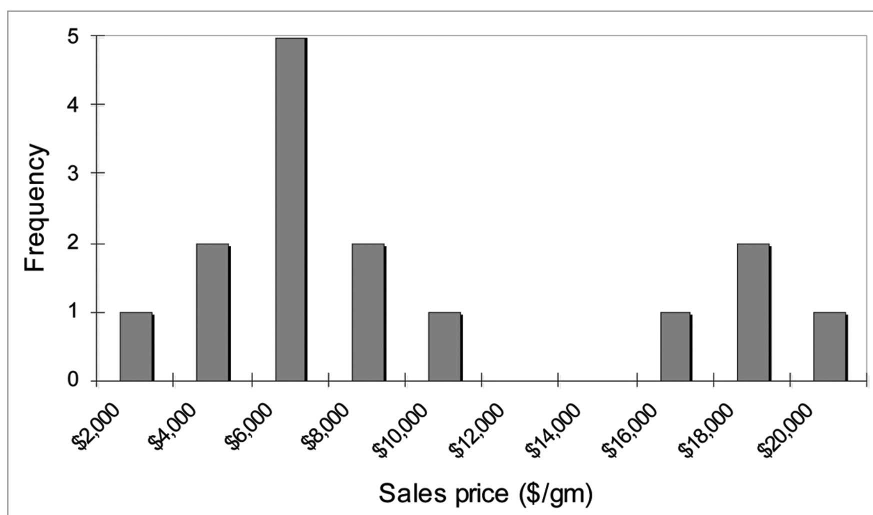

Figure 4. Distribution of average wholesale prices for mAb and Fc-fusions in 2008. The average U.S. wholesale prices per gram for 15 commercial mAbs and Fc-fusions are shown. The minimum is approximately $2,000 per gram, and the median is approximately $8,000 per gram. Note that a significant price erosion (50% of the minimum shown here) for a product with modest demand (100 kg/yr) could result in an unprofitable market, as revenues for the therapeutic product ($100 million/yr) may never provide a positive return on investment.

Raw material costs are estimated to be less than $8/L for cell culture media (with an 75% purification yield, this becomes a small cost element for high titer processes, as it may be only $2/gm for a process with a 5 g/L titer) and approximately $4/gm for the purification process. It appears that COGs reduction provided by reducing raw material costs further will only be a significant benefit for very large products with very large production scales. For the median mAb, a savings of 25% of raw material costs (25% of $6/gm x 200 kg/yr) would only result in a $0.3 M savings per year, and likely not recover the investment necessary to develop the improved process using cheaper raw materials, considering the fully burdened labor cost for development staff of $0.3–0.5 M per year.

The 2008 average sales prices for the top 15 mAbs and Fc fusions range from $2,000–20,000/gm, and the median sales price is $8,000 (**Fig. 4**). The fraction of the sales price associated with the drug substance COGs for a process with a titer of 2 or 5 g/L would be very small (approximately 1–5% at most). It may not be widely recognized or reported that because of these increase in titers and economies of scale, mAbs will be a class of biological products with relatively low production costs, although this calculation does not account for many other expenses, such as royalties incurred for accessing either the necessary process technology, or for the antibody sequence or target, in addition to the burdens of the cost of research, sales and failed projects in the research pipeline. This will have critical implications for process development, manufacturing and product lifecycle strategies.

Thus, it appears that drug substance COGs will not be a significant driver for process technology decisions for pipeline products as long as reasonable titers (>2 g/L) can be achieved; titers greater than 5 g/L are very unlikely to have a meaningful impact on either capacity or COGs, and even higher titers could have no impact on costs as the bioreactor output would exceed the purification process capacity. For nearly all mAb products, with the exception of blockbusters with a very low sales price, there will be no direct link between mAb drug substance production costs and sales prices in the future, as companies are able to take advantage of the economies of scale provided by large production capacities and increasing titers. However, the current slate of mAb products may have very different cost bases given that their process titers are likely to be much lower, as a consequence of earlier technologies used to establish their cell lines, media formulations and bioreactor management strategies.

A summary of COGs components for the final product vial is shown in **Table 2**. Cell culture titer is a strong influence on COGs, but the difference between 0.5 and 2 g/L is much larger than between 2 and 5 g/L. The rough cost of the upstream process is inversely proportional to titer, while the downstream costs are in direct proportion to the product mass purified. As the titer increases from 0.5 to 5 g/L, the majority of the drug substance COGs shifts from upstream to downstream unit operations, as has been described by other models.[16] The clear benefit in increasing product titers for these large-scale production facilities is evident, as the 10-fold increase in titer decreases the drug substance COGs by over 85% ($124/gm to $16/gm). The cost of manufacturing the drug product is estimated at $10 per vial, which represents a reasonable average for a parenteral product, but will depend upon many factors including configuration, batch volume and testing requirements. The fill-finish costs could become a larger component of final product costs than drug substance COGs in some cases, although this is largely dose and product titer dependent. When drug product device or delivery technologies are employed, the proportion of costs associated with drug substance production will be reduced even further, sometimes dramatically. Recognizing that drug product manufacturing costs may exceed drug substance costs for some high titer mAb processes emphasizes the diminishing returns of increasing titer further.

DOCKET ALARM Find authenticated court documents without watermarks at docketalarm.com.

DOCKET ALARM

Explore Litigation Insights

Docket Alarm provides insights to develop a more informed litigation strategy and the peace of mind of knowing you're on top of things.

Real-Time Litigation Alerts

Keep your litigation team up-to-date with **real-time alerts** and advanced team management tools built for the enterprise, all while greatly reducing PACER spend.

Our comprehensive service means we can handle Federal, State, and Administrative courts across the country.

Advanced Docket Research

With over 230 million records, Docket Alarm's cloud-native docket research platform finds what other services can't. Coverage includes Federal, State, plus PTAB, TTAB, ITC and NLRB decisions, all in one place.

Identify arguments that have been successful in the past with full text, pinpoint searching. Link to case law cited within any court document via Fastcase.

Analytics At Your Fingertips

Learn what happened the last time a particular judge, opposing counsel or company faced cases similar to yours.

Advanced out-of-the-box PTAB and TTAB analytics are always at your fingertips.

API

Docket Alarm offers a powerful API (application programming interface) to developers that want to integrate case filings into their apps.

LAW FIRMS

Build custom dashboards for your attorneys and clients with live data direct from the court.

Automate many repetitive legal tasks like conflict checks, document management, and marketing.

FINANCIAL INSTITUTIONS

Litigation and bankruptcy checks for companies and debtors.

E-DISCOVERY AND LEGAL VENDORS

Sync your system to PACER to automate legal marketing.

WHAT WILL YOU BUILD? | sales@docketalarm.com | 1-866-77-FASTCASE

fastcase®
Smarter legal research.