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TOP TRENDS IN Single-use Biomanufacturing

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A Message from the Editor

Just a few short years ago the idea that single-use/disposable technologies for biopharmaceutical processing would become almost commonplace was almost unthinkable. Yet, here we are. The use of single-use/disposable equipment has exploded during the past several years and the reasons for that monumental growth are clear. Single-use/disposable technologies offer an extremely efficient and cost-effective way to manufacture biopharmaceuticals, while offering significant increases in product quality.

Yet, despite the adoption of these technologies, many questions regarding their use, applicability to processes, and future still remain.

On the following pages we have assembled articles that will give you insight into the current and future uses of single-use/disposable technologies and offer you a look ahead to the market opportunities still ahead for these devices.

Thanks for reading.

Mike Auerbach
Editor In Chief, *American Pharmaceutical Review*

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Trends and Growth in Single-Use System (SUS) Adoption

Eric S. Langer

President and Managing Partner
BioPlan Associates, Inc.

Biopharmaceutical manufacturing directly supports the $240 billion biologics industry, and small improvements in costs of manufacturing can result in very significant savings. However, unlike other industries, where adoption of new technologies are often quickly embraced, in bioprocessing, change evolves slowly. This is due to a few factors, but one of the largest is the intense regulation of the industry. Manufacturers and contract manufacturing organizations (CMOs) must gain regulatory approval for any change in technology or process. Therefore, change is slow to happen because once a manufacturing process has been established and approved, the costs of change are significant, and incentives to change are limited.

Despite the onerous regulatory and testing burdens facing manufacturers who seek innovation and process improvements, change does happen. New technologies are developed, and the industry moves forward, albeit slowly. One such change over the past 20 years has been the adoption of Single-Use System (SUS) and disposables. In recent years, we have seen fewer blockbuster drugs, more biologics having higher potency that require smaller production volumes, advances of biosimilars targeting smaller markets, and ongoing incremental improvements in production yields and efficiencies that create production operations at much smaller scales. This has permitted the use of single-use devices, such as plastic bioreactors, mixing systems, and containers that are now dominating clinical production, and are moving toward commercial operations.

Having adaptable equipment and flexible facilities that can manufacture multiple biopharmaceutical products at once, or in tandem, rather than a single drug that will carry a company for many quarters to come, is now a standard of the bioprocessing industry.

Single-use and disposable devices are being used for a range of applications including upstream production, mixing, filtration, purification, fill-finish, and storage, among others. These systems

provide faster change-overs and reduced times for production. BioPlan Associates has surveyed global biopharmaceutical manufacturers and CMOs to gain insight into current and future trends in the industry. In BioPlan's *14th Annual Report and Survey of Biopharmaceutical Manufacturing Capacity and Production*, we asked 227 bioprocessing decision-makers where they are using SUS and disposables, and the critical factors, trends and hurdles being seen in adoption.

How Common are Single-Use Devices?

The most common single use devices are basic tubing, disposable filter cartridges, and connectors and clamps. Although we do note that these devices must meet exceptionally high standards for quality, and performance, nearly 90% of respondents to our survey indicated they are using these products at some scale. In fact, these devices are reaching market saturation, at least at clinical scale. At the bottom of the 'adoption' list are perfusion devices, membrane adsorbers, and disposable chromatography devices. These are at the lower end of usage ranges because they tend to be newer; given the slow adoption rates in this industry, they are still moving up on the growth curve. While many devices were tracking around a healthy 13% annual growth rate last year, the more saturated devices were showing only single-digit growth. As more facilities use them, growth in adoption of single-use devices necessarily slows as market saturation is reached. For many, probably most of these product classes, slow usage growth rates likely reflect their relatively widespread adoption prior to our collecting these data, particularly the simpler and/or less-expensive products, such as "Sampling systems" and "Media bags, purchased dry". Many of these products achieved relatively high adoption in earlier years. For example, disposable media bags were among the very first single-use products, with single-use filters common even before this.

In contrast, some new(er) single use equipment, such as membrane adsorbers and perfusion/tangential flow filtration devices, simply are newer and continue to have relatively low adoption rates. As we reach a market saturation point for single-use pre-commercial applications, it will take greater regulatory acceptance (commercial product approvals) for plastics usage and/or more approvals of single-use-manufactured commercial biologics to allow this market to capture more significant market shares and growth in sales.

In our study, we evaluated the growth (change) in disposables applications over the past 11 years (See Figure 2), in terms of the difference in percentage facilities actually implementing disposable applications. This year, "Bioreactors" percentages continued to grow rapidly, up from 21% in 2006 to 80.3% adoption; a 59.3% point difference. "Mixing Systems" also saw a large point difference this year, 58.4%, from a point difference of 50.8% in 2016. "Perfusion devices" reported 35.9% growth this year, up from 33.1% in 2016. Other areas have grown at faster rates this year, than reported in 2016, again reflecting either slower and/or earlier adoption and associated higher baseline usage rates (e.g., exemplified by the areas with the least growth – "Media bags (wet)", "Media bags (dry)" both long used in bioprocessing – and "Disposable chromatography" with a high initial baseline).

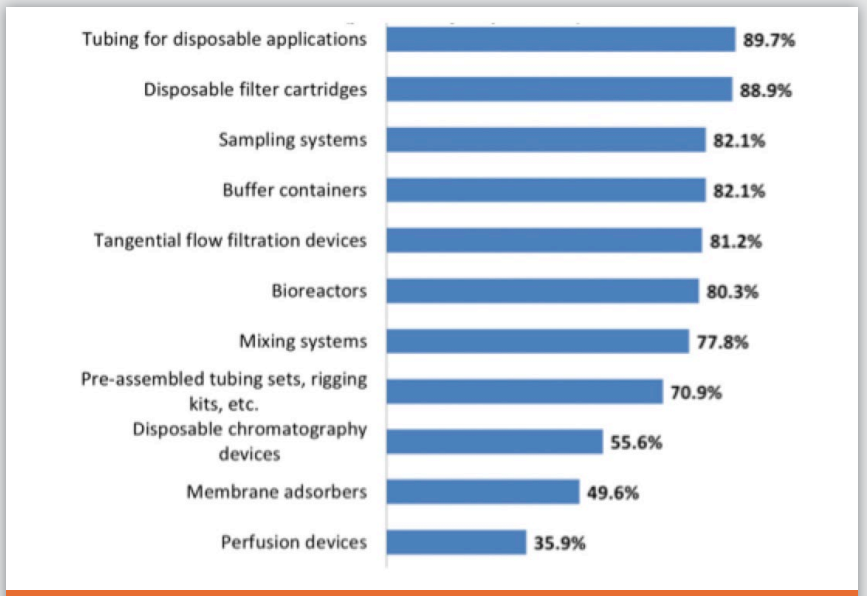

Figure 1. Usage of Disposables in Biopharmaceutical manufacturing, any Stage of R&D or Manufacture (Selected Data)

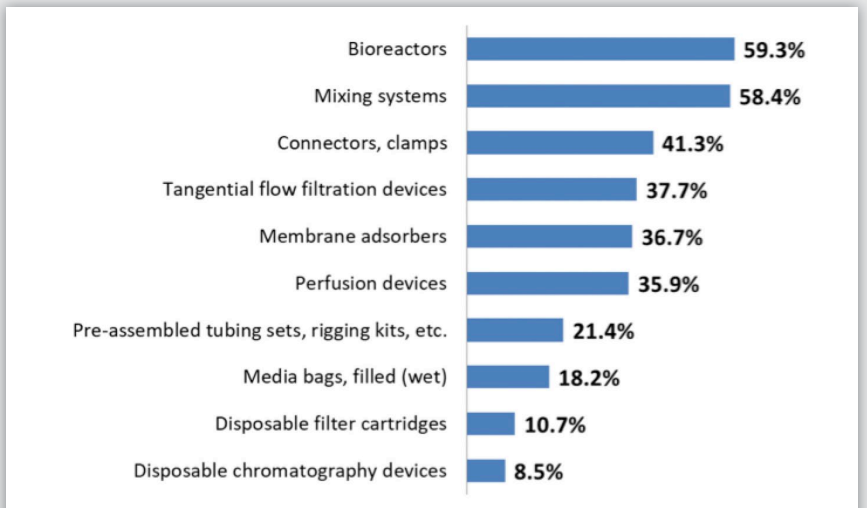

Figure 2. Selected Devices-11-Year Percentage-Point Change in First-Usage of Disposables, 2006-2017

The average annual growth rate (CAGR) for some of these devices between 2006 and 2017 has been relatively high, around 13% for mixing systems, membrane adsorbers, and bioreactors. Other more common devices that have seen less *average* growth due to the fact that these were already in steady use when BioPlan began collecting data in 2006. In addition, some other single-use equipment may be showing slower growth today, such as perfusion/tangential flow filtration devices, because there is more regulatory approval required to allow these devices to reach market saturation.

Process Scales Where SUS and Disposables are Being Used

We looked at commercial production, scale-up/clinical production, process development, and early R&D. By far, disposables are being more widely used in scale-up/clinical production and process development than commercial production. In scale-up/clinical production-scale, adoption rates for nearly every type of single-use product is over 70%, with several areas over 80%. In contrast, disposable chromatography, for example, is used by only 20% at commercial scale. This is not unexpected, since devices like larger scale SUS chromatography are not (yet) available, and membrane adsorbers have not yet entered mainstream commercial markets.

Reasons for Adopting Single-Use Technology And Disposables

Study participants cited reducing capital investment in facilities and equipment as the most critical reason for increasing disposable use. This was cited by 27.7% of respondents, an almost 50% increase over that response in 2016. This is likely due to the fact that manufacturers are continuing to focus on productivity, efficiency, and short-term cost savings and therefore see a decrease in facility costs as a good way to accomplish these goals. The next most critical reasons cited for adopting SUS were to eliminate cleaning requirements (15.2%), faster campaign turnaround time (8.9%), decrease risk of cross-contamination (8%), and flexibility of a modular approach (7.1%).

We also asked the most critical reason for not increasing disposable use. The number one listed reason was the high cost of disposables, cited by 23.4% of respondents. The fact that cost issues have risen to the top is indicative of how SUS device manufacturers have generally begun to resolve concerns of the past, including breakage, and leachables and extractables, both of which have taken the top spots in prior years.

When Will Facilities Be Using 100% Fully Disposable Technology?

A majority of industry experts, 64.9%, either said they 'strongly agree' or 'agree' that there

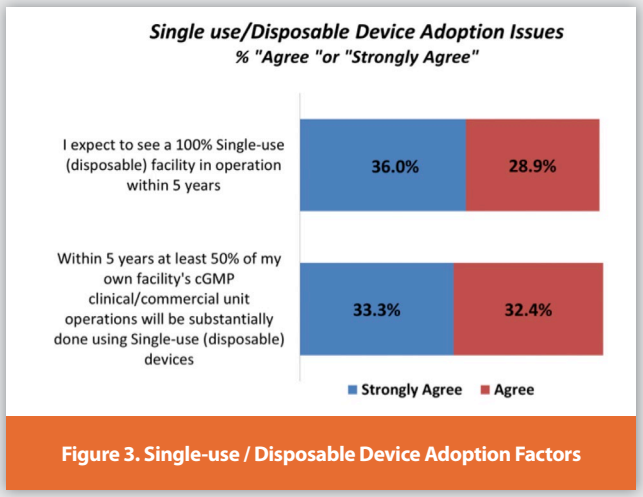

Figure 3. Single-use / Disposable Device Adoption Factors

will be a 100% fully disposable facility in operation in 5 years. This is up from 57.2% of respondents last year. It's likely these facilities would be new and using devices like single-use upstream bioreactors and downstream disposable chromatography and filtration systems. Nearly two-thirds of respondents (65.7%) said they anticipated their own facility's cGMP clinical/commercial operations would be substantially using single-use devices in five years. This response keeps going up, from 51.8% in 2016 and 49.1% in 2015.

Conclusions

Single-use systems, which are being used at clinical scale for well over 80% of bioprocessing operations, will continue to be adopted by biomanufacturers and CMOs at larger commercial scale as pipeline products being produced in SUS are approved, and move into commercial production. Because most single-use disposable systems are already being used in scale up/clinical production, much of the future growth will come from the growth of larger commercial scales, increasing market growth of SUS since these are much costlier systems to implement. As the industry matures, vendors are creating improved disposable technologies to differentiate themselves from competitors. This bodes well for manufacturers and CMOs as it will drive down prices and increase competition.

Disposable processing equipment is now being considered increasingly for more strategic reasons, such as reduction in overall costs, and improved productivity. The 'tactical' reasons such as reductions in cleaning and validation requirements and in cross-contamination events are still important decision factors, but are being seen as relatively less critical.

As better upstream productivity in recent years has required lower and/or less frequent dosing, and production requirements can be made at a tenth of the scale from a decade ago, more production lines can be specified at single-use scales. At this scale, e.g., 2,000L or less, disposable bioreactors are viable and cost-effective. Further, some facilities, such as CMC Biologics in Bothell, WA, and Copenhagen Denmark are chaining multiple 2000 liter bioreactors to create up to 12,000L batch sizes. Thus, further reducing the need for large stainless steel tankage.

Complete single-use upstream processes can compete with larger commercial-scale manufacturing in cost. And now, suppliers and innovators are turning to downstream processing single-use systems to find cost-effective and efficient solutions. Even facilities with conventional steel facilities are creating hybrids with SUS to optimally incorporate disposables for production.

As regulatory agencies become more comfortable with the performance of SUS, the industry will see a wider adoption at commercial scale. This will result in the market for SUS rapidly growing far past its current size. The availability of current SUS has benefited new biopharmaceutical start-ups in particular, allowing them to spend much less capital and quickly advance the development of new products. Single-use systems may therefore increase the competition within the biopharmaceutical manufacturing industry as a whole, allowing smaller and medium-sized companies to gain a quick foothold, whereas in the past, they would have been prevented from doing so by huge up-front facilities costs.

References

1. 14th Annual Report and Survey of Biopharmaceutical Manufacturing Capacity and Production, BioPlan Associates, Inc. April 2017, www.bioplanassociates.com

Author Biography

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Survey Methodology: The 2017 Fourteenth Annual Report and Survey of Biopharmaceutical Manufacturing Capacity and Production yields a composite view and trend analysis from 227 responsible individuals at biopharmaceutical manufacturers and contract manufacturing organizations (CMOs) in 25 countries. The methodology also included over 131 direct suppliers of materials, services and equipment to this industry. This year's study covers such issues as: new product needs, facility budget changes, current capacity, future capacity constraints, expansions, use of disposables, trends and budgets in disposables, trends in downstream purification, quality management and control, hiring issues, and employment. The quantitative trend analysis provides details and comparisons of production by biotherapeutic developers and CMOs. It also evaluates trends over time, and assesses differences in the world's major markets in the U.S. and Europe.

Keys to Successful Implementation of Single-Use Technology

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As biopharmaceutical projects occupy an increasingly larger share of the development pipeline, drug makers are striving to implement strategies that bring affordable therapies to market quickly and cost effectively. Over the past several years, the adoption of single-use technology has emerged as one important strategy for improving downstream and upstream processing while avoiding the downsides of traditional stainless-steel bioreactors.

Uptake of single-use technology shows no signs of slowing. A recent market report predicted the single-use market will become a $6-billion industry by 2024, marking a compound aggregate growth rate of 11.1% from 2015 to 2024.[1]

While drug companies are highly motivated to use single-use solutions to speed the development of new molecules, increase production efficiency, and decrease capital expenditures, they still face several complicated challenges such as on-time delivery of materials, regulatory issues, and quality questions.

How are innovators addressing these issues and bringing single-use solutions to the next level?

Single-Use Challenges and Opportunities

While many teams can implement single-use systems to some degree, not everyone has the knowledge and experience to do it well. Some fi rms believe they are restricted to a one-size-fi ts-all approach for single-use assemblies. In reality, "single use" cannot be implemented the same way for every molecule and every project. A knowledgeable third-party expert can effi ciently develop single-use assembly elements tailored to a given project while the sponsor company focuses on making its molecule as productive as possible.

At the M Lab™ Collaboration Centers, sponsor companies have access to the Mobius® MyWay portfolio, allowing them the fl exibility to choose from three single-use assembly routes. Mobius® Stock solutions can ship within 24 hours for clients with immediate needs. Mobius® Select assemblies give sponsor companies with accelerated timelines the option of using custom assemblies from an optimized component library (six-week lead time). Last, Mobius® Choice offers fully customized solutions for end-users with specialized requirements (standard lead time). This diverse portfolio helps us address the many different needs and challenges that users face.

But with the many choices available to them, how do companies choose the best assembly for their project?

We believe having input from a knowledgeable expert as well as state-of-the-art process development tools are key for designing the best prototypes possible. This pairing—expertise and innovative tools—enables projects to quickly move from the draft stage to one that is fully optimized.

Using our non-GMP facility for this work helps spark creativity and allows end-users to explore the full breadth of options available to them. Clients can troubleshoot unit operations freely with modern tools for both small- and large-scale projects without being bound by regulatory restrictions and standard operating procedures. Experts are committed to helping with demonstrations, evaluations, and education about single-use solutions to quickly optimize and implement applications across various processes.

1 Single-use Bioprocessing Systems Market: Customizability as per Consumer Requirements Key Feature Driving Adoption, reports TMR," Sept. 27, 2016, http://www.transparencymarketresearch. com/pressrelease/single-use-bio-processing-systems-market.htm

To support such intricate process development teamwork, we chose to establish our nine innovative M Lab™ Collaboration Centers across the globe which include a host of virtual tools for remote discussions and troubleshooting. Centers are located in North America, Latin America, Europe, and Asia, and we tailor our approaches to the various regional dynamics.

The individualized support and guidance offered at the global M Lab™ Collaboration Centers also helps with any regulatory and validation concerns that arise. For instance, sponsor companies often want to know how to generate the best data for testing processing materials for extractables and leachables. The team at the M Lab™ Collaboration Centers is committed to addressing questions like this and creating a transparent way to supply databacked critical information and solid best practices about our technologies in support of process validation and optimized manufacturing protocols. Regardless of where in the world this work takes place, we align our training and educational materials and tailor it to the situation at hand.

This collaborative effort is not only critical for new projects, but also for facilitating the streamlined transfer of projects from a traditional stainless- steel manufacturing process to one designed around single-use technology. Working with a knowledgeable partner helps avoid time and resources lost to errors and retesting.

Collaboration in Action

A collaborative approach brings together great people and great minds, overcomes barriers, and accelerates progress. We feel this strategy leads to robust best practices that customers can confi dently implement in their manufacturing processes. What follows are four examples that illustrate how partnering in a creative M Lab™ Collaboration Center environment played an important role in the success of customer projects.

CASE STUDY 1: Importance of global network.

A contract manufacturer located outside of the United States did not have a fully automated single-use TFF system to produce clinical material for a US-based client. The manufacturer needed to see what such a system would look like and immediately decide on a strategy to implement. Using a virtual demonstration, we responded very quickly and showcased appropriate systems. We then invited the client to an M Lab™ Collaboration Center in the United States, so they could not only see the most appropriate system, but also discuss the intricacies of the process and how it would translate into recipes they could run. This type of customer engagement would not have been possible without the interconnectivity of our various regional M Lab™ Collaboration Centers. The contract manufacturer and their client were both very happy with the end result and implemented the system successfully.

CASE STUDY 2: Higher protein concentrations.

The need for higher protein concentrations in bulk drug substances is increasing. One client asked us to collaborate on the use of a 500-L single-use mixer to uniformly mix a viscous drug product without risking protein aggregation. With a joint project team, we designed a set of experiments and showed in an M Lab™ Collaboration Center that our mixers worked well while maintaining drug product quality.

CASE STUDY 3: Virtual solutions.

One client had key team members based in Asia, Europe, and the United States, and wanted employees from all these areas to discuss a specifi c unit operation. M Lab™ Collaboration Center specialists ran the experiment at our Massachusetts Center with the client's team members in the United States, while other individuals watched the experiment in real time from sites in Europe and Asia using our virtual tools.

CASE STUDY 4: The power of education.

The authorities in Singapore wanted to prepare the local workforce for the infl ux of new biopharmaceutical R&D and manufacturing projects coming into the country. Because our experts have been deeply entrenched in the industry and regional regulatory issues for years, we were able to train employees of biopharma companies based there and well as regulatory personnel. This collaboration was important to us because we believe an educated workforce is vital to the success of a project—especially in emerging markets where employees may not all have the same degree of regulatory and practical experience in the biopharmaceutical industry. Our involvement in industry consortia like the BioPhorum Operations Group plays a major role in our ability to help clients on this front.

Summary

As companies move away from traditional stainless- steel bioreactors and explore new technologies for accelerating timelines and slashing costs, single-use solutions have come to the forefront as an important option. To fully take advantage of this powerful technique, collaborating with a third-party provider that has an established framework and tools for testing and exploring possible single-use platforms alongside clients is essential for maximizing effi ciencies and cost savings.

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BPOG Five-Year Vision for Single-Use Technologies

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Executive summary

Single-use technologies (SUT) for biomanufacturing, otherwise known as disposable technologies, have the potential to transform the industry through more cost effective solutions and solve crucial manufacturing and compliance problems. Today, suppliers have made great advances in SUT, but the vision of better, faster and lower-cost operations has not been fully realized. Over the past two years, the BioPhorum Operations Group (BPOG, see box) has been painstakingly developing best practices for SUT and work streams for extractables and leachables, user requirements and change notifications are advancing and improving the implementation of SUT. Collectively, these efforts represent thousands of man-hours and pool the knowledge and real-life experiences of many of the leading biomanufacturers embracing this technology. But much more is on the horizon. BPOG and its member companies are developing a five-year vision (see Figure 1) for SUT, targeting a selection of SUT and auxiliary systems that are critical to ensure that SUT are a mature and established technology for biomanufacturing.

Introduction

Over the last five years, biopharmaceutical manufacturers have been implementing SUT from clinical to commercial production processes in their main North America and Europe manufacturing sites, and their secondary sites in other continents. The impressive uptake of SUT has been mainly driven by its promising and wide-ranging advantages:

1. Speed: The installation of SUT can be significantly faster than traditional stainless steel (SS) installations. With SUT there is no need for cleaning and sterilization between runs, and so Clean-In-Place (CIP) and Steam-In-Place (SIP) piping and controls are not required, greatly reducing design engineering and field installation times. Also, SS equipment is often custom designed, while SUT hardware is usually a standard vendor offering with much shorter delivery lead

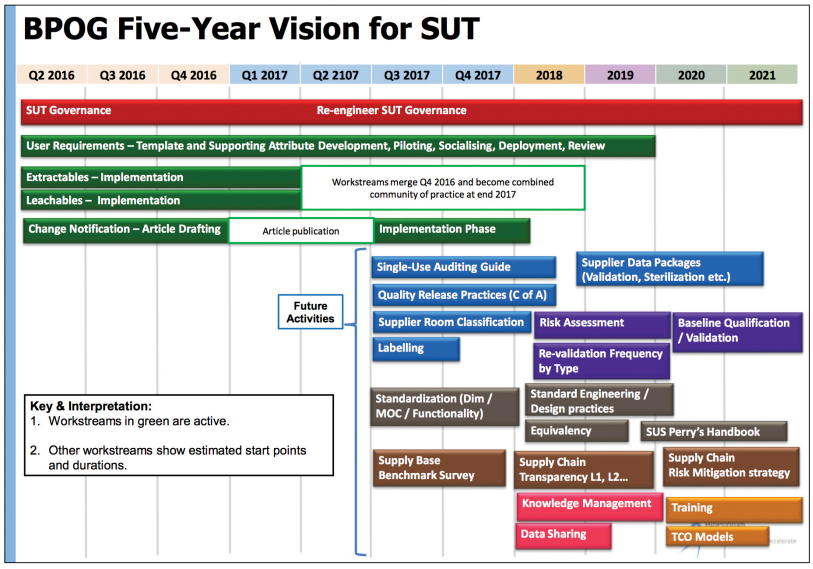

Figure 1. BPOG Five-Year Vision for SUT

Time	Data	Process	Risk
• Too short to qualify by end-user • No end-user input prior to implementation • Inventory management • Time and resource spent on minor or out of scope changes	• Often does not meet end-users' specifications • Inconsistent expectations from end-users • Do we buy this part ?	• Lack of single point of contact (SPOC) • Inadequate handover package • Over- or under-estimation of change requirements • No feedback to supplier • Unclear how changes will impact end-users • Lack of standardization for addressing customer-specific designs	• Imprecise understanding of intended application • Resistance to continuous improvement • High volume of changes simultaneously • Unclear or misaligned understanding of risk • Risk of lost of in-process and bulk materials due to failures/investigations

Figure 2.

times. Benchmark times for completing SS facilities is often considerably more than two years, while SUT facilities could take less than 18 months but the goal would be <12 months with the flexibility SUT provide.

2. Cost: Due to the factors mentioned in Speed above, SUT facilities require significantly lower capital costs than SS. CIP and SIP can require up to 70% of the piping and process controls in large biotech facilities, but as SUT does not require CIP or SIP then the capital costs are also much lower – in some cases capital cost reductions of over 50%.

3. Flexibility: SUT provides flexibility for facility design and the scalability/selection of equipment. By eliminating CIP and SIP, the scope of a single-use facility is significantly reduced in terms of the demands for electrical, water for injection (WFI), automation, air supply and HVAC. The equipment is mostly mobile and can be easily relocated within a facility or to a different location. Additionally, equipment can be qualified in an R&D space and moved to a GMP space at a later time, providing flexibility for training and personnel movement for qualification.

Also, the consumables part of single-use equipment including tubing diameter and pump sizes can be changed out to suit a range of protein production processes These changeable consumable parts allow the use of the same hardware without losing efficiency and accuracy over a wide range of operating parameters.

4. Closed system: The use of SUT and aseptic connectors/tube welding allows for a fully closed system, making it feasible to do bioprocessing (upstream and downstream) in one suite and reduce a facility's footprint. Closed processing also allows operation under reduced room classification conditions. Additionally, closed processing with the use of SUT reduces the risk of microbial contamination/bioburden as well as safety concerns relating to potent molecules such as ADCs [1-3].

5. Environmental impact: While it is obvious that SUT requires disposal of the single-use components, the environmental impact of cleaning and sterilization SS systems is also very significant. SUT processes can require 80% less WFI than SS, and none of the cleaning agents required for CIP. A number of life cycle analyses have been completed comparing SUT and SS, and the consensus is that SUT has a similar or lower environmental impact than SS [4].

Although SUT haven't been promoted as a 'disruptive technology' to biopharmaceutical manufacturing, these innovative of SUT have started to spark our imaginations about how we can exploit its advantages in meeting the new era of medicine manufacturing.

Disruption to current GMP operating model

SUT has been incorporated in our process designs from 100% SS production lines, when mega drug was the norm of the past, toward hybrid or fully disposable lines. The rapid implementation of SUT has also begun to strain our operating systems/models to a point where many GMP controls established over a decade ago are now the limiting factors that prevent us from attaining the full advantages of this disruptive technology.

BioPhorum Operations Group (BPOG)

BPOG is a company-to-company consortium representing around 90% of the commercial biologics manufacturing capacity. Being sponsored at senior levels by entire leadership teams means engagement is strategic and team representation is strong, ensuring learning is actively and routinely adopted. Best practices and user requirements are developed as a single voice of the companies and leverage network-wide data, knowledge and experience rather than collating the perspectives of a few industry experts or supplier organizations. BPOG's aim is to facilitate the implementation of operational best practices within manufacturers and supply partners, alike, to deliver measurable progress and benefit. This enables a wide consensus and drives the harmonization of industry approaches and standards. BPOG's model is inclusive and freely disseminates papers and models to interested parties as its aim is to encourage rapid and widespread buy-in and adoption of newly developed best practices.

So, knowing what you know now, how would you design a five-year SUT roadmap for your company's future? Would you handle all of the challenges yourself or in collaboration with other talents (peers, suppliers, academia, regulators, etc.) and supported by the expert guidance of a professional facilitator? For most of us, the consortium approach would be most effective. There are occasional visionaries who see unique opportunities and pursue the vision by themselves, but the vast majority of individual successes are achieved by targeting calculated risks and goals consistently over time, supported by knowledge-based best practices that are shared by diverse groups of experts.

Below are some of the constraints that many biopharmaceutical companies are experiencing and are represented as an upcoming working group (color block) in the five-year vision diagram (Figure 1):

1. Flux of supplier change notifications (SCNs): Unlike a clinical production, which can be of 1-5 years' duration and have lower levels of process validation and change management, SCNs are a complex and necessary process for GMP manufacturing sites. It is quite common for a commercial production site to receive over 100 SCNs per year, assuming the site manufactures 5-10 commercial products per year. Several key challenges encountered by end-users and suppliers [5] are shown below and can be grouped into four categories: Time, Data, Process and Risk.

On reviewing the challenges, it is easy to see that the fundamental issue can be summed up as a lack of mutual understanding from both end-users and suppliers. To address this constraint, a BPSA/BPOG cross-functional team (already formed in 2015) is devising a solution. Watch for new announcements in the coming months (http://www.biophorum.com).

2. The demands of qualifying new suppliers: Individual engineers from a specific project within a part of the production process, at each site across the entire company, may select and qualify the SUT of their choice where there is a lack of internal standardized processes and procedures. In searching for the best technologies, picking the best from the field means an increased demand to qualify new suppliers into each company's cGMP supplier management systems. The supplier quality organization was already strained without the new need for an early understanding/forecasting of the demands from a tsunami of new supplier qualification requests. Depending on the audit schedule, it can take up to 12 months to accommodate a new request into an already tight audit visit schedule. Such a delay can impact a project timeline from securing the necessary SUT from development studies to validation, as well as delaying the full cGMP implementation for an improvement project of an existing commercial process. The new single-use audit guide subteam will be formed to collaborate with suppliers in developing a new approach for efficient audit and qualification practices without a proportional increase in head-count.

3. Complicated supply chain and logistics: SUT supply chains are very complex and often are both horizontally and vertically integrated. Vertically integrated supply chains, have suppliers manufacturing components, or films which are use across the suppliers product line. This provides the supplier with additional control over the design specifications and the control of raw materials can be more strictly monitored. Horizontally integrated supply chains utilize insourcing or purchasing single use components from other suppliers, this added complexity which can be difficult during investigations, since the end-users typically will only be able to interact with the primary supplier. Most suppliers offer a mix of both supply chain models offering various designs with both their own components and other suppliers' components as options. The expansion of SUT implementation and adoption across numerous sites around the globe has led to thousands of custom designs. These are specific to individual organizations and sites that accomplish the same operations, but mat not benefit from the industry's vast design experiences, improved robustness from automation and cost reduction through the scale of manufacturing. These designs complicate the manufacturing supply chain and delivery of SUT, preventing end-users from realizing the full benefits of the technology, causing production delays and missed schedules. While there are specific situations that may require occasional unique design solutions, several BPOG member organizations have eliminated thousands of redundant designs through internal standardization and have therefore saved resources and improved on-time delivery. Suppliers often will be able to provide standard solutions most effectively in reducing supply chain complexities.

4. Maintain control of current GMP system: The new reality of SUT is that end-users will surrender a portion of the control for their production equipment to SUT suppliers, yet the end-users will still be held accountable for every deviation and failure. Such failures have collectively cost the industry millions of dollars in lost materials, decreased productivity and investigations. However, the suppliers' controls have the potential to greatly improve equipment robustness as they can incorporate quality when they design and manufacture

single-use components and share test data. To date, this benefit has not been fully realized due to restricted information access and barriers to sub-suppliers tractability and information. To maintain cGMP integrities without wasteful controls, it is crucial for the SUT industry to move away from the traditional supplier-customer relationship and toward a collaborative partnership relationship.

5. Increased uncertainties from regulatory expectations: Although the use of SUT was encouraged for clinical manufacturing, the transition into a cGMP commercial environment hasn't been smooth and 483 observations are common. Clinical manufacturing was the early adopter of SUT and provided end-users, suppliers and the regulatory agencies with the opportunity to identify knowledge gaps. The recent translation of SUT from clinical applications to large-scale cGMP commercial manufacturing resulted in an increased number of SUT-specific agency observations. Agency observations are driven by a lack of maturity of the SUT, disparities in single-use experience between companies or suppliers and lack of industry standards and best practices, which are all compounded by evolving agency expectations.

6. Shorter cGMP production facility readiness: There is a constant drive to reduce timelines from research to a first in-patient study and then launch. While SUT can be a great asset to reduce manufacturing readiness timelines, the complexities of SU assembly customizations, long lead times, the need to modify the custom automation of sub-systems, the quality of available data packages, and the constraints from locked hardware require tremendous effort from end-users to design, qualify and validate a new production facility based 100% on SUT. BPOG is forging partnerships between end-users and suppliers to find a mutually beneficial solution to this challenge.

7. Lack of standardization: Fifty years ago, there was a lack of standardization in SS design. There was no standard SS connector or filter housing, for example, and so spare pumps could often not be utilized due to the wrong fitting and one vendor's filter would not fit into another vendor's housing. Today, we take for granted the tri-clamp fitting and the code 7 filter as the SS standard. In many ways, SUT is in a similar position to SS 50 years ago – there are no standard connections, tubing sizes, material of contact (such as 316L in SS), tubing hanger/tubing management design, etc. Hopefully, it will not take 50 years to achieve SUT standardization. While many aspects of SUT are still too new to standardize without slowing the needed innovation, there are others that we can start working on, such as dimensional standards. BPOG will work together with suppliers, BPSA, ASTM, ASME and others towards standardization, tackling the easy wins first before moving on to more complex areas as the industry matures.

As illustrated from the constraints listed above, many of the issues are not the technology itself but rather the direct and indirect supporting processes and systems that are required in a cGMP and lean-manufacturing environment. If we truly want to fully realize the advantages of disposable technologies in meeting a new era of challenges – from speed to lower-cost innovative medicine and establishing SUT as a mainstream biomanufacturing technology – it is imperative to transform our operating paradigm in reducing any uncertainties/obstacles. Since BPOG can't tackle all uncertainties at once, it is crucial to form a multi-year plan that enables us to coordinate various work streams that will be initiated over a multi-year time span, with a common shared vision of creating a new operating paradigm.

The BPOG Five-Year Vision for SUT is "By April 2021, to attain equivalent or better working knowledge and application of single-use technology as the stainless steel system today (2016)"

Conclusion

SUT are disruptive technologies offering the biopharmaceutical industry the value proposition of increased speed to commercialization, reduced capital and manufacturing costs, flexible plant and equipment design, closed systems to reducing contamination risk and demands for air classification, while lowering the impact on the environment.

Drug manufacturers initially embraced SUT in clinical settings and more recently in large-scale commercial manufacturing. However, commercialization of SUT has presented new opportunities for enhanced knowledge in the both science and GMP compliance. Implementation of SUT requires a disruption to the current GMP operating models and systems. In the new model, manufacturers realize there will be an increased number of suppliers and accept the compliance responsibilities for these suppliers and their materials. Successful implementation then requires suppliers and manufacturers to work together to address the new challenges, such the increased number of SCNs, complicated supply chains and logistics, and the lack of adequate data packages and standardization. These challenges must be addressed while ensuring that practices meet evolving global regulatory requirements.

BPOG has taken a leading role in enabling the adoption of SUT by stimulating collective industry discussions and providing common solutions to SUT challenges. Key deliverables already provided to the industry are establishing and implementing BPOG's extractable protocol by many BPOG members(http://www.biophorum.com/category/resources/extractables/about-us/) [4], publishing leachables best practice guide (the official copy is under final editing for publication on BPOG website) [5,6], developing user requirements and SCN best practices, which are all aimed at advancing and improving SUT implementation.

The BPOG five-year vision for SUT implementation will continue to unite and galvanize the industry to advance its training, develop the supply chain and increase the impact of knowledge management, with a vision to attain equivalent or better working knowledge and application of single-use technology as the stainless steel system today (2016).

References

1. Mahajan E., Microline: A Fully Disposable Manufacturing Facility, ISPE Annual Conference, Nov 2011.
2. Boedeker B., Facility of the Future: Effect of Disposables and Continuous Processing on Plant Design, BioPharma Asia, July/Aug, 24-27, 2014.

Leachables and Extractables from Single Use Bioreactors and their Impact on Cell Culture Performance

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Amgen

Introduction

In the biopharmaceutical industry, drug substance production from cell culture lines is a historically proven, effective process.[1] Selected cell lines with desired compound expression are grown suspended in nutrient broth under optimal conditions. Until recently, the primary choice of vessel used for cell culture was fixed stainless steel reactors, due to their physical durability and resistance to the harsh chemicals. The downsides of these steel bioreactors were their high initial set-up costs and stringent requirements of cleaning and sterilization after use to prevent cross-contamination. All of these add up to a reliable system, but at a cost of flexibility, which is crucial in today's rapidly changing market.

An alternative for the stainless steel reactor is the single use bioreactor (SUB).[2,3] A typical SUB is a bag composed of a multilayer polymer film, with various connection ports with tubing, agitator, and systems for gas inlet and outlet. As the name implies, cells are cultured inside these bags, which are then simply disposed afterwards. This process cuts all post production processes related to cleaning and sterilization. Capital investment is minimized, making development stage work more flexible with better managed risks.

As in any contact materials used in the production of drug substances, possible leachable compounds from the SUB are a matter of concern.[4] The SUB is used for cell culture, so leachable concerns toward the final drug product are negligible. This is due to cell culture being the upstream stage of manufacturing and the presence of numerous purification processes in the subsequent downstream stages. On the other hand, SUB leachable concerns toward the nurture of the cell culture itself are highly relevant as any negative factors that inhibit cell growth could decrease the yield of the drug substance or even make it necessary to abort the production run. As other basic factors affecting cell growth like growth media/nutrient composition, gases, pH, temperature, humidity, density, etc. have been examined and tested thoroughly in the past, any new factors specific to SUBs should be reviewed as well, so they can be used as effectively and reliably as the previous stainless steel reactors.

Single Use Bioreactors (SUBs)

Single use bioreactors (SUB) are formed primarily from a plastic film composed of multiple polymer layers. Each layer adds to the physical or chemical properties required for the SUB to properly function as a bioreactor. The specific polymer materials used in a given bag varies by manufacturer and model, and may include polyethylene, ultra-low-density polyethylene, linear low-density polyethylene, ethylene vinyl alcohol (EVOH), ethylene-vinyl acetate (EVA), polyesters, nylons, and others. Each layer may have its own set of additives like antioxidants, fillers, plasticizers, stabilizers, etc. to obtain the necessary characteristics, but the identities and levels of these additives are proprietary information and often not communicated to the biopharmaceutical manufacturer. SUBs are sterilized by gamma irradiation prior to shipping to the end user and are at that point ready to use.

Leachables/Extractables from SUBs

Leachables are any and all compounds that migrate from the contact material under normal usage and/or storage conditions. Identification and quantification of leachables allows the understanding of its effect on the used/stored material and assessing the suitability of that contact material for actual use/storage. For SUBs, direct leachable assessment is difficult, because the cell culture solution is a complex mixture of numerous compounds dissolved at high concentrations. This hinders the observation of leachables, which are initially unknown and of unknown (and typically low) concentration. The solution to this "needle in a haystack" problem is to study the extractables of the SUB first. By utilizing one or more simple extraction test solvents with elevated temperature conditions, enhanced migration of contact material compounds is forced into the non-complex solvent, which allows for easier detection. Once the extractable profile is known, key compounds of concern can be examined in the actual cell culture in the SUB by developing targeted analytical methods. By these complimentary routes, the SUB would be understood for its suitability in use.

In a study of extractables from commercially available SUBs,[5] sample bags made from representative films from four suppliers were filled with water or 40/60 organic/aqueous solvents to less than five percent total capacity and incubated for two days at 50 °C. The fill volume was limited, allowing liquid contact on all internal surfaces without excessively diluting the extracted compounds to facilitate detection. Pure organic solution was not used as it was deemed too harsh and the observed compounds would not be comparable to the actual leachable into aqueous cell culture solution. After extraction, the solutions were tested in a non-targeted approach by four analytical techniques: reverse phase liquid chromatography with UV detector (RP-HPLC/UV) for non-volatile organic compounds, gas chromatography mass spectrometry (GC/MS) for volatile compounds, reverse phase liquid chromatography mass spectrometry (RP-HPLC/MS) for oxygen and nitrogen containing organic compounds, and inductively coupled plasma mass spectrometry (ICP/MS) for inorganic elements. Observed compounds were identified and quantified by comparison to commercial database (UV and/or mass spectra) and/or against reference standard compounds, some customarily synthesized.

From the four bags, fifty three different organic compounds as well as five inorganic elements were observed (Table 1).[5] Of the observed organic compounds, the majority were degradation products of the polymer film and plastic additives, including antioxidants, plasticizers, and slip agents. For the most part there was significant variability in the observed extractable compounds across the four different bags; this result is not unexpected due to the different films composing the different bags. However, four compounds were observed in all four bags: 2-4-Di-tert-butylphenol (DtBP), 1, 3-Di-tert-butylbenzene, 2, 4-Di-tert-butylphenylphosphate (mDtBPP), Bis (2, 4-di-tert-butylphenyl) phosphate (bDtBPP). Further study on these compounds led to the understanding that they are all degradants of tris (2, 4-di-tert-butylphenyl) phosphite (TBPP) (CAS #31570-04-4) or trade name Irgafos 168®, which is a common antioxidant additive to many polymers (notably including most of the various types of polyethylene). TBPP itself was not present in the bag extracts at detectable levels. Further study showed that the above four degradation compounds

Table 1. List of Extractable Compounds from Four Different Single Use Bioreactor Bags. Listed examples other than the Antioxidant degradants and Unclassified were not observed in all four bags tested.

Total Observed	Extractable Type	Source	Example(s)	CAS #
14	Antioxidant degradants	Film	2-4-Di-tert-butylphenol	96-76-4
			1,3-Di-tert-butylbenzene	1014-60-4
			2,4-Di-tert-butylphenylphosphate	
			Bis (2,4-di-tert-butylphenyl) phosphate	
4	Plasticizers	Film	Diethyl phthalate	84-66-2
			Dibutyl phthalate	84-74-2
6	Slip Agents	Film	Nonanamide	1120-07-6
			Decanamide	2319-29-1
			Undecanamide	2244-06-6
2	Polymer degradant	Polycarbonate	Bisphenol A	80-05-7
2	Polymer degradant	Polyethylene film	Octane	111-65-9
1	Polymer degradant	EVA Film	Acetic Acid	64-19-7
1	Polymer monomer	Nylon Film	Caprolactam	105-60-2
23	Unclassified	Unknown	2-(2-butoxyethoxy)-ethanol	112-34-5
			Polyethylene glycol	25322-68-3
5	Inorganic elements	Whole Bag	Sodium	
			Silicon	

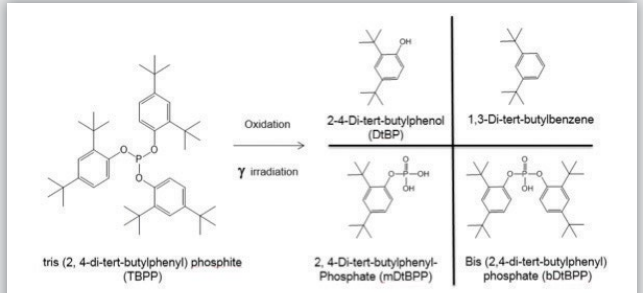

Figure 1. Degradation of Antioxidant TBPP by Oxidation and Gamma Irradiation.

were formed via oxidation and subsequent gamma radiation of TBPP (Figure 1).[6,7] The observation of these degradant compounds shows that extractables may be observed in forms altered from the original as they are added or present in the SUB's manufacturing.

Impact of SUB Leachables/ Extractables on Cell Growth

To understand the impact of SUB extractable on cell growth, an association study was performed on extractable profile versus cell growth.[8] The internal liquid contact side of the bag film of six different bags from five different vendors were directly extracted with water for 48 hours at 50 °C and those water extracts analyzed by RP-HPLC/UV. In a complementary set of experiments, cell culture media was incubated in each bag for three days at 37 °C and those incubated media were then used to grow Chinese Hamster Ovary (CHO) cells. The effect of the bag-incubated media was evaluating by measured the viable cell density (VCD) attained after three cell growth passages of three days each.

In comparing the bag film extractable profiles and VCD of CHO cells grown with bag-incubated media, only the observed high levels of bDtBPP correlated with low VCD (Figure 2). The bag with the highest level of bDtBPP lead to near zero VCD or complete cell death. None of the other TBPP degradant compounds (DtBP and mDtBPP) seemed to affect VCD.

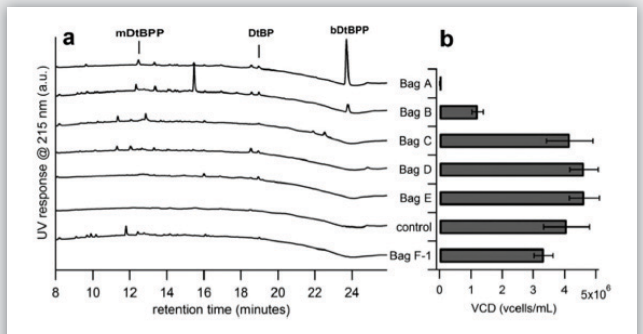

Figure 2. Comparison of SUB extractable profiles (a) and VCD of CHO cell culture grown with SUB-incubated media (b).[8]

The toxicity of the bDtBPP on cell culture growth was confirmed by a direct spiking study.[9] CHO cells were grown in medium spiked with three TBPP degradants (bDtBPP, DtBP, mDtBPP) and three other degradants of hindered phenol antioxidants at 0.8 to 1.0 mg/L. The VCD was measured for each cell sample and normalized against cells grown with non-spiked medium. Only bDtBPP had near fatal toxicity toward the cells at this concentration (Figure 3). The DtBP compound, which is known to be toxic to mammalian cells at concentrations near 30 mg/L,[10] did not affect the CHO cells in this experiment.

The toxicity and specificity of bDtBPP on cell growth was determined by spiking nine different CHO cell lines with varying levels of bDtBPP up to 1 mg/mL.[9] The viability and VCD were determined after three passages of three days, per dosage per cell line, and normalized to unspiked controls, ultimately giving dose-response curves. From the curves, the toxicity was determined as half-maximal effective concentration (EC_{50}) fitting to the Hill equation or the concentration that leads to 50% VCD/viability relative to control. The nine CHO cell lines showed varying levels of EC_{50} (Figure 4), but all showed impacts to cell growth at concentrations less than 1 mg/L. Note that while all the cell growth studies described so far were performed using CHO cells proprietary to Amgen, sensitivity to bDtBPP and/or SUB-incubated media (presumably due to bDtBPP) has been shown for other CHO

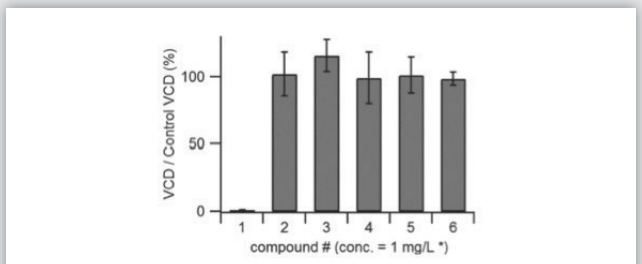

Figure 3. The normalized relative VCD of cells grown with medium spiked with antioxidant degradants. Compound 1: bDtBPP (*0.8 mg/L), 2: DtBP, 3: mDtBPP, 4-6: other hindered phenol compounds.[9]

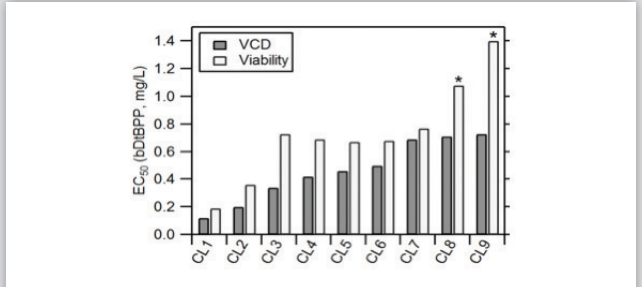

Figure 4. The half-maximal effective concentration (EC_{50}) of bDtBPP on nine CHO cell lines. *The viability of cell line 8 and 9 were extrapolated as values were higher than maximum concentration (1 mg/mL) tested. [9]

cell lines as well.[11-12] Given the generality of this effect, and in order to detect leachables problematic to cell growth that may arise in the future due to introduction of new materials, it will be highly beneficial to the SUS manufacturer and end-user communities to develop a standard cell-growth assay based on a non-proprietary CHO cell line.[13]

Conclusion

Given their location upstream in the biopharmaceutical manufacturing process, leachables from SUBs present very low risk of introducing undesirable compounds into final drug product. However, experiments in our laboratories and in others have shown that leachables from SUB assemblies can potentially affect cell culture performance adversely. Specifically, excessive quantities of leached bDtBPP were shown to be toxic to a wide array of CHO cell lines. This discovery was made possible by a thorough set of extractables experiments, careful identification of extracted compounds, and comparison of extractables with cell culture experiments. The bDtBPP example clearly demonstrates the need for robust extractables information for single-use bioprocess equipment, and complementary cell growth studies for SUBs and other single-use equipment meant for cell culture operations.

References

1. Butler M. Animal Cell Culture and Technology. 2nd ed. New York, NY: BIOS Publishers; 2004
2. Brecht R. Disposable bioreactors: maturation into pharmaceutical glycoprotein manufacturing. Adv Biochem Eng Biotechnol. 2009;115:1-31.
3. Eibl R, Kaiser S, Lombriser R, Eibl D. Disposable bioreactors: The current state-of-the-art and recommended applications in biotechnology. Appl Microbiol Biotechnol. 2010; 86: 41–49.
4. U. S. Food and Drug Administration CFR Part 211.65.
5. Marghitoiu L, Liu J, Lee H, Perez L, Fujimori K, Ronk M, Hammond M, Nunn H, Rogers G, Nashed-Samuel Y Extractables Analysis of Single-Use Flexible Plastic Biocontainers. PDA J Pharm Sci and Tech. 2015; 69 (1): 49-58.
6. Jenke DR. Linking extractables and leachables in container/closure applications. PDA J. Pharm.Sci. Technol. 2005; 59 (4):265–281.
7. Brandolini AJ, Garcia JM, Truitt RE. Spectroscopic characterization of the degradation products of phosphorus-containing polymer additives. Spectroscopy. 1992; 7 (3): 34 –39.
8. Hammond M, Marghitoiu L, Lee H, Perez L, Rogers G, Nashed-Samuel Y, Nunn H, Kline S A Cytotoxic Leachable Compound from Single-Use Bio-Process Equipment that Causes Poor Cell Growth Performance Biotechnol. Prog. 2014; 30(2): 332-337.
9. Hammond M, Nunn H, Rogers G, Lee H, Marghitoiu A-L, Perez L, Nashed-Samuel Y, Anderson C, Vandiver M, Kline S Identification of a Leachable Compound Detrimental to Cell Growth in Single Use Bioprocess Containers. PDA J Pharm Sci and Tech. 2013; 67(2): 123-134.
10. Moridani MY, Siraki A, O'Brien PJ. Quantitative structure toxicity relationships for phenolsin isolated rat hepatocytes. Chem.-Biol. Interact. 2003; 145 (2):213–223.
11. Jurkiewicz E, Husemann U, Greller G, Barbaroux M, Fenge C. Verification of a New Biocompatible Single-Use Film Formulation with Optimized Additive Content for Multiple Bioprocess Applications. Biotechonl. Prog. 2014; 30(5); 1171-1176
12. Eibl R, Steiger N, Fritz C, Eisenkraetzer D, Baer J, Mueller D, Eibl D. Recommendation for Leachable Studies: Standardized Cell Culture Test for the Early Identification of Critical Films for CHO Cell Lines in Chemically Defined Culture Media. DECHEMA Biotechnologie 2014; www.dechema.de/en/posis.html. Accessed 27 Dec 2016.
13. Tappe A, Cutting J, Hammond M, Nunn H, Kline S. The Case for a Standardized Assay to Test Suitability of Single-Use Systems in Cell Culture Applications. Bioprocess Int. 2016; 14(1): 10-13.

Author Biographies

Kioshi Fujimori *is a Senior Associate Scientist at Amgen Inc. in the Process Development department. For the last ten years, he primarily worked to study and investigate extractables and leachables from product contact material in production processes to final drug containers. B.S, Biochemistry, UCLA, 1996*

Matthew Hammond, PhD*, is a polymer science subject matter expert and focuses on assessments of material properties with an eye towards potential effects on manufacturing process performance and/or product quality, He is currently a senior scientist in the Attribute Sciences Group within Amgen's Process Development organization.*

Jian Liu, PhD*, is currently a scientist at Amgen Inc. leading analytical activities supporting cross-functional drug product and process development (upstream, downstream, fill-finish). Jian has wide experience from structure characterization of protens, peptides and small molecules to biomarker discovery with proteomics. Jian obtained his PhD in Analytical Chemistry from Purdue University.*

Hans Lee, PhD*, is a principal scientist at Amgen's Process Development organization. He joined Amgen in 2003 and is responsible for extractable/ leachable and non-conformance investigations related to single-use systems, primary containers and manufacturing processes.*

Mike Ronk *is a Principal Scientist at Amgen with nearly twenty years of experience in the biotechnology/pharmaceutical industry. Mike has an analytical chemistry back ground working in both Research and Process Development, and his current responsibilities encompass a variety of activities within Amgen's extractable and leachables group.*

Heather Nunn *is a Scientist in late stage process development at Amgen in Cambridge, Massachusetts. She received a BS in Biology from Willamette University and a Master of Pharmaceutical Bioengineering from the University of Washington. She has worked at Amgen for the last 15 years in roles ranging from cell line development to upstream commercial support.*

Yasser Nashed-Samuel, PhD*, is a principal scientist with 14 years of biopharmaceutical experience, and founder of the Extractables/ Leachables function at Amgen. Currently, leading a unique central analytical group supporting cross functional activities related to analytical methods, E/L, drug product development, device/primary containers development and qualification, single-use systems, raw materials and forensic investigations throughout the product lifecycle.*

Global Biomanufacturing Trends, Capacity, and Technology Drivers: Industry Biomanufacturing Capacity Overview

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Abstract

Biologic-based drugs are an increasingly important part of the product growth strategies for pharmaceutical and biopharmaceutical companies. As the number of commercial products and pipeline candidates grows, a crucial issue facing the industry is the current and future state of biomanufacturing capacity, the availability of that capacity, and the technologies impacting upstream and downstream bioprocessing. Pharmaceutical and biopharmaceutical companies and contract manufacturing organizations (CMOs) are aligning their strategies to not only address capacity but to address greater complexity in supplier risk and the adoption of advanced biomanufacturing technologies.

Biopharmaceutical products have rapidly become a larger percentage of overall pharmaceutical company revenue with sales of the top six selling antibody products, Humira, Remicade, Enbrel, Rituxan, Avastin, Herceptin, at just over $51B in 2015. The compound annual revenue growth rate for antibody products, which includes antibody conjugates, naked antibodies, and antibody fusion proteins, from 2003 to 2014 was 21%; however, this growth is expected to slow to the high teens in the coming years due to the maturation of many products, and emerging alternative technologies. Also, it is more difficult to sustain such growth rates the larger the market becomes.

To provide context about this growing segment of the market, BPTC estimates that there are over 900 biopharmaceutical products in some stage of clinical development in the United States or Europe, and the large majority of these products, 77%, are produced in mammalian cell culture systems. To further refine the biopharmaceutical manufacturing market, we evaluated the distribution of mammalian products by product type and phase of development. Figure 1 shows the distribution of the following product types, antibody products (which include naked monoclonal antibodies, Fc-fusion proteins, antibody fragments, bispecifics, antibody conjugates, and other antibody-related products), blood proteins, cytokines, enzymes, fusion proteins, hormones and other recombinant proteins, by phase of development. Antibody products are the dominant product type for all phases of development, but this product type is even more dominant for early phase products. Antibody products comprise nearly half, 48%, of currently marketed products. Recall that many of the early commercial biopharmaceutical products, such as growth hormones, insulins and interferons, are produced in microbial systems, but the use of microbial production systems is much less common now. The percentage of antibody products currently in the BLA, or equivalent, stage of regulatory submission is 67% (BLA/MAA stage in Figure 1). Antibody products make up 82% of products in Phase 3 development and 90% of products in Phase 1 and 2 development.

Whether approved or in development, all of these products need access to mammalian production capacity. To better understand the production requirements needed to meet the demand for all of these products, we created a demand forecast. The future demand for current commercially approved biopharmaceutical products is estimated from each product's reported annual sales data, along with

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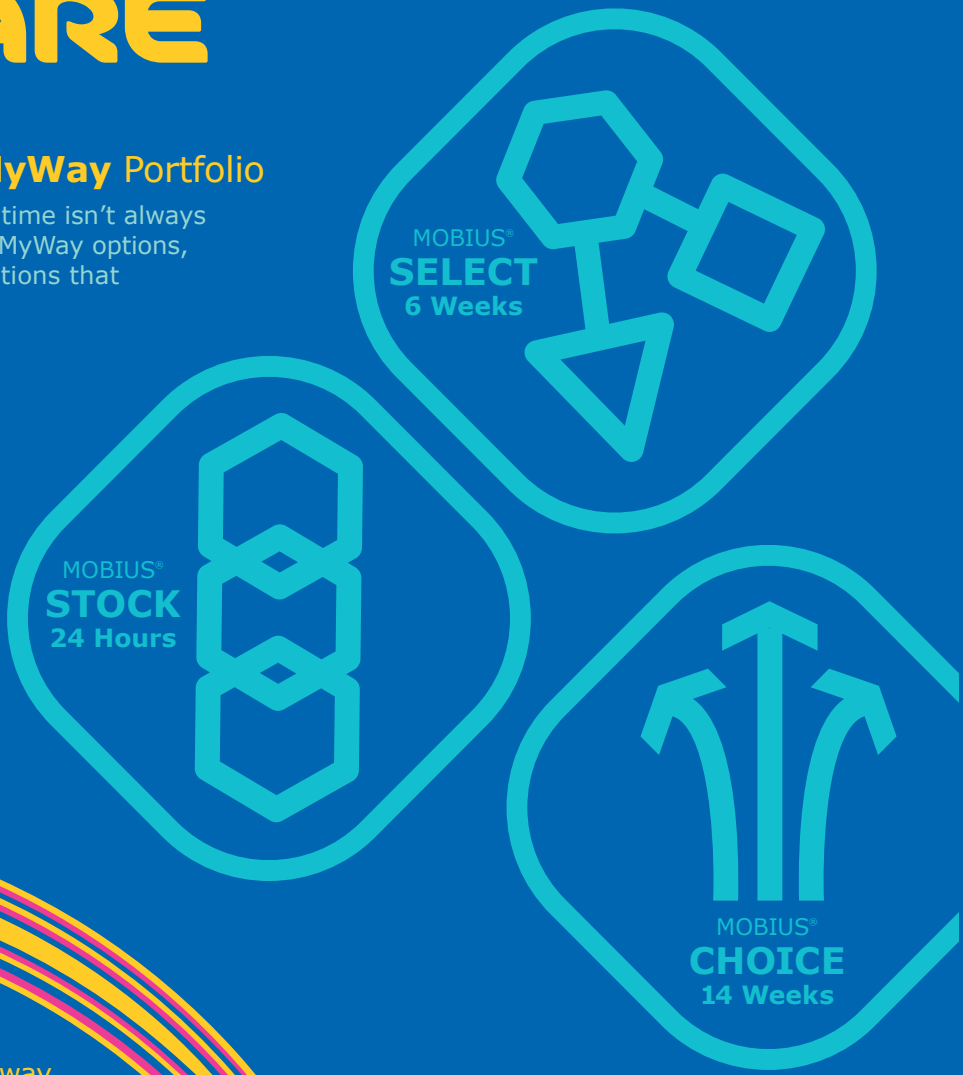

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estimates of each product's future growth rates. A product's growth in sales is calculated from actual sales data for the current and previous year. Where available, analysts' forecasts may also be used to estimate year-to-year sales growth for commercial products. Our calculated future product growth estimations also take into consideration a product's age; sales growth typically slows as a product matures, while newly approved products often do not reach full market penetration for several years.

Using the sales growth data along with the number of patients treated in the current year (based on price per mg and sales), an estimated treatment population for future years can be calculated for each year during the forecast period. This forecasted treatment population, combined with the yearly per patient dosing calculates the kilogram quantities of each product that will be required in future years. These forecasted product quantities along with an estimated expression level and overall yield estimates for each product can then be used to calculate the estimated amount of cell culture capacity (L/yr) that will be required for each product in future years.

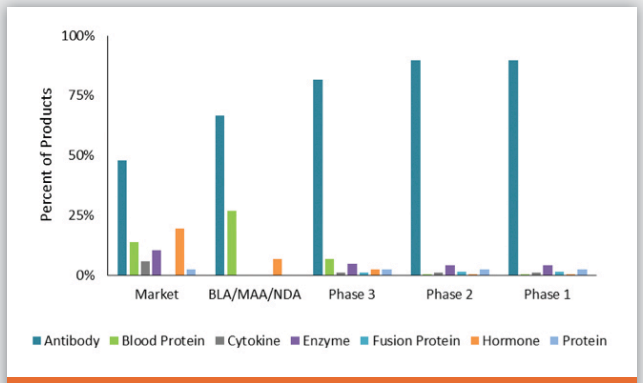

Figure 1. Distribution of Mammalian Products by Product Type and Phase of Development

Figure 2 shows the forecasted kilogram quantities of product needed to meet annual commercial demand for all products types produced using mammalian production systems. In 2014, approximately 13 metric tons of product are needed, and the large majority of the demand is for currently commercial products (teal band). The orange band labeled Clinical on each bar represents an estimate of clinical trial material manufactured to support the clinical development of all pipeline product candidates in a given year. The green band, present but not visible in the 2016 bar, represents products that have submitted a BLA, or equivalent, and are estimated to receive regulatory approval and enter the commercial market in 2016. The cycle time of 1 to 1.5 years from submission of BLA to approval is based on industry standard product development success rates. Similarly, the grey band first appearing in the 2017 bar represents products in Phase 3 clinical development in 2014 that are projected to receive regulatory approval and begin entering the commercial market in 2017. This grey band increases each year as the commercial demand for the products grows. The purple band and the light blue band represent the products in Phase 2 and Phase 1 development, respectively, that are forecasted to begin entering the commercial market in 2018 and 2019, respectively. As more products receive commercial approval each year, the overall kilogram requirements needed to meet commercial product demand increase from just over 13 metric tons in 2014 to nearly 40 metric tons in 2020.

Another way to view the total production capacity needed to meet product demand between 2014 and 2020 is to think of the demand in terms of total installed volume of mammalian cell culture capacity. Figure 3 shows the volumetric capacity required to support the clinical development and eventual commercial sales of all current pipeline product candidates in the year shown. An estimation of yield is

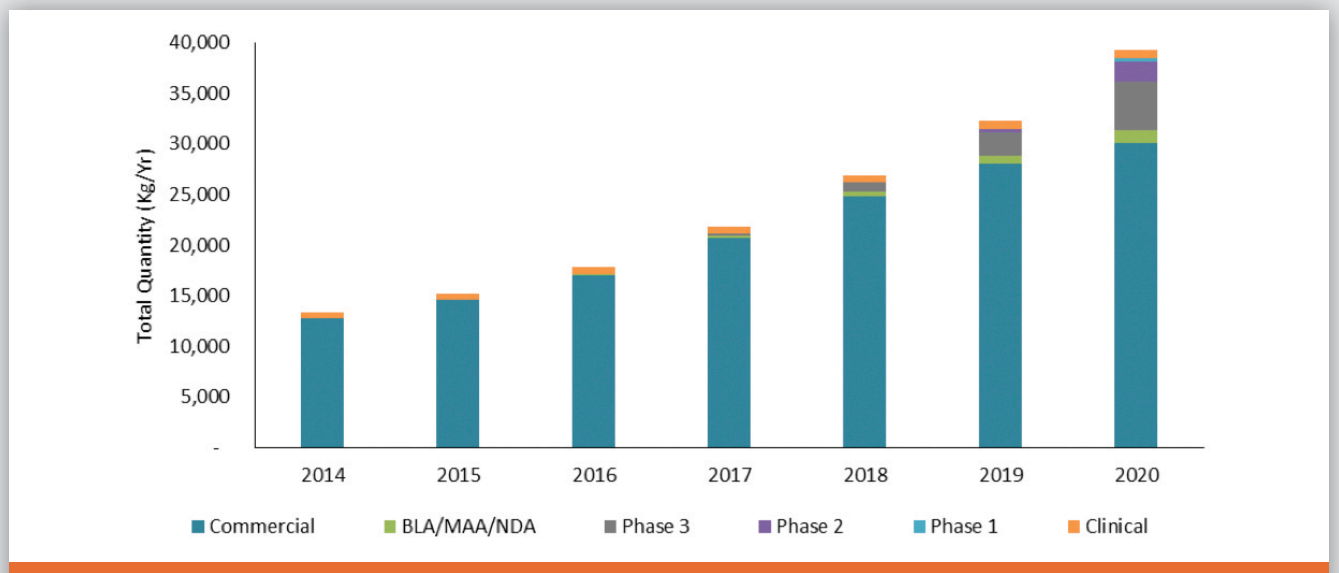

Figure 2. Forecast of Bulk Kilograms Needed to Meet Product Demand

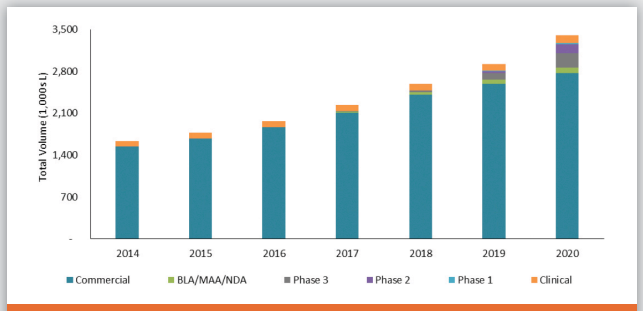

Figure 3. Forecast of Volumetric Capacity Needed to Meet Product Demand

required to go from kilogram demand to liter demand and introduces some uncertainty in the forecast due to the fact that most companies do not publish their production yields. For this reason, our analysis uses industry average estimates. As described for annual kilogram demand in Figure 2, each bar represents the volume required for those products in the indicated phase of development in 2014 that successfully reach the commercial market. In 2014, the annual volumetric requirements were just over 1,600 KL, while in 2020, the volumetric requirements will be just over 3,400 KL.

Given this increase in volumetric demand over the next 5 years, the industry is concerned about the availability of production capacity. There is always some degree of uncertainty in balancing the demand and supply equation due to production problems, market demand over time and competitive factors. As shown in Figure 4, in 2016, the available mammalian cell culture supply is currently approximately 3,600 KL and is projected to grow to approximately 5,600 KL by 2021. However, not all capacity is equally available throughout the industry. In 2016, Product companies (companies focused on product development) hold approximately 73% of the installed mammalian cell culture capacity, while Excess companies (companies that are developing products, but also sell or make available any excess manufacturing capacity) and CMOs control significantly less capacity, 13% and 14%, respectively. The forecasted distribution of capacity changes only slightly in 2021, with Product companies holding 68% of the installed capacity, while CMO companies increase to 15% and Excess companies increase to 17% of the capacity.

When comparing clinical versus commercial capacity, it is evident that the total volume of capacity devoted to commercial manufacturing far exceeds that devoted to clinical production. In 2016, nearly 390 KL (11%) of capacity are designated for clinical manufacturing compared to nearly 3,250 KL (89%) for commercial products. In 2021, percentages do not change, with approximately 600 KL of clinical capacity and just over 5,000 KL for commercial capacity. This is not surprising because of the relatively small demand for clinical supply in comparison to the larger demand for commercial sale.

While Product companies control the majority of cell culture capacity, capacity is highly concentrated among ten companies. Table 1 shows the distribution of capacity among the top ten capacity holders in a given year. Capacity for companies not ranked in the top 10 are included in the "All Others" category. In 2016, the "All Others" category includes 120 companies, and in 2021 "All Others" include 128 companies. In 2016, 67% of the mammalian cell culture capacity is controlled by ten companies; in 2021, this changes to 61%. Based on substantial capacity investments, Samsung, Bristol-Myers Squibb and Novartis will displace Pfizer, Celltrion and Lilly from the top ten capacity holders by 2021.

Geographic distribution of capacity may also skew the accessibility to capacity. Figure 5 shows in 2016, North America holds the greatest percentage of capacity (52%), followed by Europe (32%) and Asia (16%). In 2021, the order remains the same but the percentages change slightly – North America (42%), Europe (34%) and Asia (24%). There has been significant growth of capacity in Asia, particularly in Korea and Singapore, due to government incentives and tax advantages. Asian locations for manufacturing also tend to be more attractive to companies with mature pipelines and the ability to manage complex global supply chains.

Irrespective of ownership or geographic location, there is a surplus of capacity as shown in the balance between a demand for mammalian cell culture and total available capacity in Figure 6. The light grey band in each bar represents the remaining available capacity. This analysis assumes an average capacity utilization of 18 batches per bioreactor per year. The demand for manufacturing capacity has been adjusted forward one year to account for the fact that bulk product is typically made well ahead of actual sales, on which demand calculations are based. For the majority of products sold in 2014, for example, bulk drug substance was manufactured in 2013.

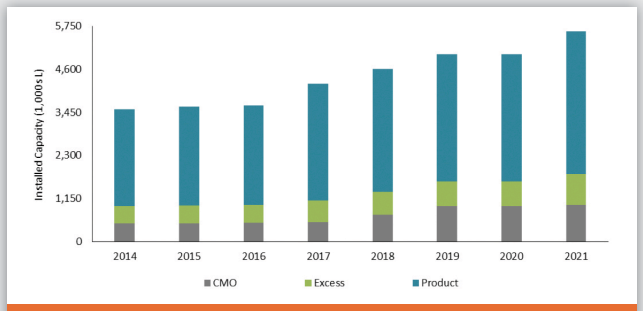

Figure 4. Current Mammalian Manufacturing Capacity

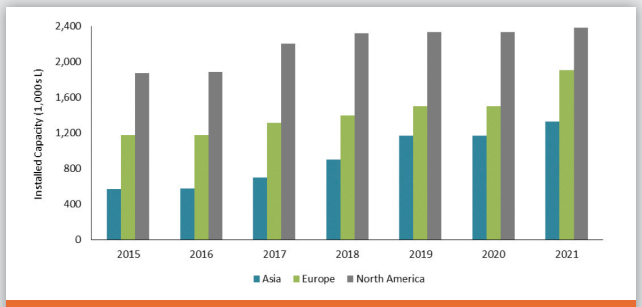

Figure 5. Geographic Distribution of Capacity

Table 1. Capacity Control				
2016 Rank	2021 Rank	Company	2016 Volume (1,000s L)	2021 Volume (1,000s L)
1	1	Roche	673	909
2	5	Lonza	261	281
3	8	Johnson & Johnson	230	230
4	6	Sanofi	223	243
5	3	Boehringer Ingelheim	205	338
6	9	Amgen	204	225
7	4	Biogen	196	316
8	-	Pfizer	149	-
9	-	Celltrion	140	-
10	-	Lilly	137	-
-	2	Samsung	-	362
-	7	Bristol-Myers Squibb	-	237
-	10	Novartis	-	205
		All Others (120/128)	1,214 (33%)	2,106 (39%)

Our analysis shows there is currently sufficient mammalian cell culture capacity world-wide to meet the total industry demand and that in 2014, only 50% of industry-wide cell culture capacity was utilized. This analysis of capacity utilization also indicates that while manufacturing capacity in general is projected to grow in the coming years, the demand for capacity will grow at a slightly greater rate so that by 2020 industry-wide capacity utilization will increase to 73%. At this anticipated level of utilization in 2020, some companies are likely to be challenged meeting the demand for specific products or gaining access to capacity at CMOs.

A utilization rate of 50% may give the appearance that the industry is not currently operating at "full utilization". However, manufacturers often consider "full utilization" in the range of 70-80% (or in some cases even lower) rather than 100% to account for change-overs, preventative maintenance, and facility upgrades. Product company manufacturers often take a proactive approach in protecting unused capacity to be able to respond to product demand surges and additional product indication approvals.

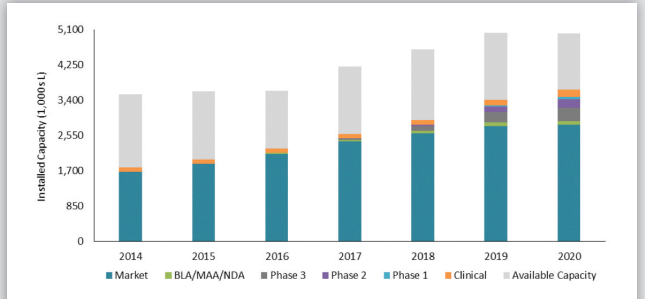

Figure 6. Overall Industry Supply and Demand Balance

As with any forecasting model, our assumptions are based on the mostly probable scenarios. However, if biopharmaceuticals being developed for certain large patient population indications such as Alzheimer's disease or those targeting the PDL/PDL-1 checkpoint in cancer are approved and covered by Pharmacy Benefit Managers, a significant increase in demand for manufacturing capacity will occur potentially leading to a serious capacity shortage.

Conversely, there are other manufacturing trends that will result in a lesser demand for some biopharmaceuticals, such as the increased focus on orphan drugs and a shift from full length naked antibodies to alternative antibody formats and more potent products, i.e., Antibody Drug Conjugates (ADCs), which would require lower doses, that in turn, would reduce the demand for manufacturing capacity.

As the biopharmaceutical industry has grown, the industry has built a certain type of capacity to meet the demands for the top six selling antibody products. The 2014 kilogram demand for each of the top six selling antibody products was >0.75 metric tons for a total 8.5 metric tons. The demand for all other antibody products combined was approximately 4 metric tons. The forecasted demand for approximately 70% of new products approved between 2016 and 2020 is expected to be less than 100 kg per year per product with the exception of Alzheimer's, PD-1/PDL-1, asthma, and possibly some PCSK-9 products. Future commercial manufacturing demands for 50% of products in Phase 2 and Phase 3 clinical development today can likely be met with a 5,000L bioreactor or smaller per product (See Table 2). This does not mean that large scale capacity is no longer needed. Our forecasts predict that the remaining 50% of products will need bioreactor capacity of 10,000L and greater to meet the predicted demand.

Overall, the biopharmaceutical industry is expected to continue to have strong growth for the foreseeable future, and antibody products will be the dominant driver of this growth. Installed capacity is currently able to meet the manufacturing demand for these products, but control and location of capacity can affect how accessible certain capacity is. The majority of capacity is product based, as opposed to CMO based, making it difficult for companies without capacity to access it at the right time and under the right conditions. North America has the greatest percentage of installed capacity, but Asia has seen a surge in new capacity installation. To meet increased product demand, installed capacity is forecasted to increase from approximately 3.6 KL in 2016 to approximately 5.6 KL in 2021. While capacity will increase, demand for capacity will increase at an even

Table 2. Number of Product Demand Met by Bioreactor Scale					
# Products in Phase 2 and 3 Trials	# of Bioreactors	< 2,000L Bioreactor	5,000L Bioreactor	10,000L Bioreactor	> 10,000L Bioreactor
285	1	118 (41%)	25 (9%)	32 (11%)	110 (39%)
	2	139 (49%)	36 (13%)	23 (8%)	87 (31%)

faster rate potentially resulting in capacity shortages by 2021. We have noted that the industry is already experiencing some capacity constraints at the clinical scales due to very high clinical demand. The type and scale of capacity being installed will also be important as the demand for 50% of products in mid-to-late stage development can be met with 5,000L of capacity or less; while the other 50% of products will need larger, and potentially much larger, capacity to meet future demand. How the industry responds to these demands for capacity will certainly be critically important to ensure these products are available to the patients.

References

1. Ecker DM, Ransohoff TC, Jones SD, Levine HL. The state of mammalian cell culture biomanufacturing. Woburn (MA): BioProcess Technology Consultants, Inc.; 2011 Dec 12. 150 p. Available from: http://www.bptc.com/reports.php.
2. Pavlou AK, Reichert JM. Recombinant protein therapeutics-success rates, market trends and values to 2010. Nat Biotechnol. 2004 Dec;22(12):1513-9.
3. DiMasi JA, et al. Trends in risks associated with new drug development: success rates for investigational drugs. Clin Pharmacol Ther. 2010 Mar;87(3):272-7.

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How to Ensure a Trouble Free Countdown to One

Regulators see single use systems as a way to boost safety in biomanufacturing. However, novel technology can introduce new risks. Where are regulators focusing their attention – and how is the industry responding?

As you've already read in this supplement, single use technology has been one catalyst for a shift in the supplier–customer relationship in pharma. With single use, the responsibility for ensuring safety and regulatory compliance falls squarely on the shoulders of suppliers. "It means that even though we're not inspected by regulatory bodies, we have to be aligned for validation," says Janmeet Anant, Global Product Manager at Merck. Of course, end users must still conduct some tests, but they also need to trust that their supplier has performed due diligence and supplied a quality product.

Relinquishing control can be difficult, but there is a clear advantage: pharmaceutical companies can focus on the core mission of bringing drugs to patients. "If we do our job right, our customers can concentrate on getting that final drug product to clinical trials and onto the market," says Anant.

Supply of equipment is only half the story. Customers also need training and technical support; poorly-trained operators opening a box of single use bags with a knife or over-tightening an O-ring could spell disaster. Once again, a solid customer–supplier relationship is key.

In short, to meet current regulatory guidelines and pre-empt future requirements, manufacturers need the full support – and guidance – of their suppliers.

Single Use Rules

"Regulators are enthusiastic about single use, particularly as there are obvious benefits for safety," says Heike Michaelis, Director of the Emprove® program at Merck (see page 22). For example, with new developments in connectors, it's possible to create and maintain a closed system, even after multiple connections and disconnections. However, there are few detailed guidelines from regulators and, until recently, few industry standards that specifically cover single use.

But broad guidelines don't necessarily have to spell bad news. "Regulations are vague and rightfully so," says Anant. "I don't think a regulatory body should be prescriptive as it would limit innovation in the industry. From a technical point of view, we as an industry can propose best practices. And those can be ever-changing as we move forward."

With that in mind, the industry has taken matters into its own hands and started to develop recommendations, according to Michaelis. Efforts so far have concentrated on three key issues: extractables and leachables, particulate contamination and integrity assurance.

ExtractingProblems

A big focus for customers, regulators and suppliers alike in recent years has been extractables and leachables. Regulators are focusing on the risk of patient toxicity, but manufacturers must also consider how extracted or leached chemicals could affect cell growth or purification processes. "We're understanding more and more in terms of the quality of the plastics, but how does that affect a drug formerly made in a different system? There are no standards at this point," says Anant. "But the industry is working on it."

Organizations like the BioPhorum Operations Group (BPOG), which comprises more than 25 of the top multinational biologics manufacturers, are proposing standard approaches. And BPOG's offshoot Supply Partner Phorum is also getting involved by bringing together drug companies and suppliers to tackle key issues in the biomanufacturing supply chain. Michaelis draws attention to a 2014 white paper by BPOG, which set out recommendations to suppliers on how to perform extractable tests.[1] These recommendations are now being widely implemented.

Though standards are emerging for biopharmaceuticals, the increasing number of cell therapies in development (most of which are manufactured in single use systems) add a new dimension to extractables and leachables testing. "How will extractables and leachables affect very sensitive cells?" asks Anant. "For example, if the cell therapy is designed to produce beta cells in the pancreas to produce insulin in diabetics, will the plastics affect the differentiation or insulin-producing ability of the cells?" The cells may stay in the body for years, or even decades, so even subtle changes could have a cumulative effect.

A Particular Issue

Extractable and leachables aren't the only major concern for biomanufacturing. "Regulators tell us that the presence of particles causes over 20 percent of all pharmaceutical recalls," says Anant. It's of little surprise then that regulators are making the elimination of such particles a top priority.

Particles can be introduced into single use systems when plastic tubing is cut, welded or melted, from cardboard packaging, lint or fibers from operator's clothing, and so on. With single use assemblies, the supplier takes on responsibility for validation and quality, including inspecting and testing for particulate contamination. The debate currently centers on exactly what monitoring is necessary. As with extractables, there are no fixed standards for manufacturing systems. However, there are standards for final drug product containers, and industry groups are translating these into guidance on particulate monitoring in single use systems. "The BioProcess Systems Alliance has written a white paper on the topic, which lays out some good practices," says Anant.[2]

If particles are discovered in a single use system, it's important that the supplier has a robust process to investigate the root cause, correct any problems identified and prevent them happening again.

Building a Fortress

It's crucial for aseptic systems to remain closed, so that bacteria and other contaminants cannot enter and jeopardize quality. Some biopharmaceuticals pose a real risk to operators, so as well as making sure contaminants don't get in, it's important that the drug product can't get out. "Making sure the system remains closed – integrity assurance – is another crucial issue for manufacturers and users of single use technology," says Anant.

There are a number of different approaches to verifying the integrity of manufacturing systems. The American Society of Testing and Materials has a method based on pressure, with and without restraining plates. The single use system under scrutiny is sealed and pressurized, while very sensitive detectors measure any pressure drop over time. An alternative method uses helium as a tracing gas. The system is filled with helium and any helium detected outside the system indicates a problem. Gas and pressure systems have one flaw – the smallest "holes" they can detect in the system are still larger than some microbes. However, the results can be validated. Bacteria can be introduced via aerosol to the air around the system, followed by a test for contamination, which makes intuitive sense, as microbes are likely to come from the surrounding environment. An even more stringent approach is to immerse the system in a liquid spiked with bacteria, but it has met with controversy. "Some people say that immersion is too harsh – that it's never going to happen in reality. Others argue that we should apply the toughest test available, to provide another layer of safety," says Anant, adding that the optimal testing interval is also up for debate. "If every time we do the test it shows that there's no issue, do we do the test every three months, or is once a year enough?"

Regulators encourage the use of closed systems, and have started to relax cleanroom requirements for facilities making use of the technology. If nothing can get in or out, the environment around the system is theoretically irrelevant. In practice, regulators aren't ready to give up all environmental controls, but a drop in cleanroom classification can save companies millions of dollars per year.

Keeping Risk in Perspective

Anant believes that as companies carry out more testing, they are likely to find that the risks of single use technology are limited. "Right now, I think we are overdoing it a bit. That's understandable – it's better to be safe than to run into an unpleasant surprise later on. But as we do more tests, and more drug products reach the market, manufacturers and regulators will gain confidence in single use systems – and apply a more balanced risk assessment."

Michaelis agrees: "Although single use technology has been around for approximately 30 years, I still consider it a young industry. Very few customers have trialed full single use suites. The famous Amgen facility in Singapore is the flagship, and there are more to follow. As it's adopted more and more, we will learn more, and be able to make improvements."

One improvement that is needed, says Anant, is shoring up the supply chain. Pharmaceuticals make up a tiny proportion of the market for plastics, so it's crucial for single use suppliers to have solid relationships with plastics manufacturers. "Even a small change in the chemical composition of a plastic could have serious knock-on effects, so we choose to work with suppliers with a dedicated medical or food division, who understand the issues."

Further down the supply chain, the relationship between single use suppliers and pharmaceutical companies is strengthening, with increasing collaboration between the groups in setting standards and assessing quality. "Over the past two or three years, collaborations have started across the board, with many different industry associations around the world. Before that, everyone was checking their own agenda and focusing on their own needs. I think coming together to agree standards across the industry will help to advance the field," says Anant.

Michaelis believes the future is bright for single use systems: "Single use has yet to reach its potential, especially in the direction of personalized medicine. The pharma industry used to focus on large-volume drugs to treat millions of patients. Now, they are going after more complex personalized medicines, treating far fewer patients. Lower volumes mean that manufacturers need to be much more flexible with their production capabilities – a big plus for single use.

"Stainless steel won't die out, so I don't see a totally plastic future. But I do think single use systems will have a huge impact," concludes Michaelis.

References

1. BPOG, "Extractables Protocol", (2016). Available at: http://bit.ly/2gZq2TN. Accessed December 19, 2016.
2. BPSA, "Recommendations for testing, evaluation and control of particulates from single use process equipment", (2014). Available at: http://bit.ly/2gPdVoS. Accessed December 19, 2016.