Single-Use Solutions for Scale-Up and Technology Transfer

The development of systems specifically for use with single-use bioreactors will help facilitate scale-up and enable both inter- and intra-company process transfers.

Scale-up generally involves taking a lab-scale bioprocess and replicating it as closely as possible to produce larger amounts of product, as specified by either a client or the regulatory authorities. A typical scale-up sequence starts with a one-litre shake flask, moves to a five- to 10-litre glass bioreactor, and then scales through stainless steel vessels of varying sizes (20L, 100L and 500L) up to several thousand litres in production (1,000L to 20,000L) (see Figure 1). Most scale-up processes are concerned with more than simply increasing the volume of cells produced, but also with maintaining cellular productivity, increasing product titer and preserving product quality. The profitability of a drug is determined by the effectiveness of the scale-up and the resulting cost of the transition into manufacturing; nothing is more critical than the production titer, product integrity and time-to-market.

CHALLENGES IN cGMP SCALE-UP AND TECH TRANSFER

Today, mammalian cell products dominate the biotechnology industry. The top four product types in clinical trials are monoclonals, cytokines, recombinant proteins and – increasingly – vaccines. The most commonly used cell types – human embryonic kidney (HEK) 293 cells and Chinese hamster ovary (CHO) cells – are easily adapted to suspension culture. All of these cell lines have undergone scale-up to reactors having a volume of up to tens of thousands of liters. However, in many cases, scale-up to manufacturing level has taken years to perfect.

Scale-up, and especially technology transfer efforts, often involve moving processes between different types of bioreactors. It is not uncommon for different types of bioreactors to have significantly different designs and control algorithms. Often, a process developed in one group or company must be re-created by the next group, and completely re-optimised for the larger bioreactor. This situation is often aggravated by the fact that it is quite common for the different bioreactors to utilise entirely different control platforms. This means that the algorithms used to control and/or measure the process parameters can differ significantly between the two groups. The result is that the efficient and accurate transfer of process knowledge is often impeded. Even worse, the flow of information associated with the batch process is often disrupted and sometimes even lost during the transfer process. These issues must clearly be addressed by the bioprocessing professional – even before the more significant biological and physical challenges of scale-up can be addressed.

Both process monitoring and early analytical characterisation, as encouraged by the process analytical technology (PAT) initiative, have significant value in scale-up and technology transfer activities. This is especially true if these activities are clearly and uniformly defined; as previously noted, if the bioreactors or fermentors used during different scale-up stages have widely varying methods by which the process parameters are measured, any potential scale-up modelling and predictive calculation is compromised, and each scale-up step must be independently engineered or, more accurately, re-engineered. This is not an efficient method for bringing a new product into production.

In fact, the more complicated the process, the more tightly the product characteristics and production process must be controlled. Off-line data, often captured in parallel with process development, must be incorporated into the process history in order to provide a record of the continuous process improvement during scale-up. Although electronic data management tools are prevalent in the
manufacturing and operations divisions of many companies, these tools are rarely integrated with process development and pilot scale-up databases. In addition, because the bioreactor control systems are supplied by different vendors, the databases are not unified, and most of the communication occurs with off-the-shelf data programs (like Microsoft Excel) or paper records. The lack of uniformity and accessibility of the data make it difficult to manage and leverage for technology transfer and scale-up.

**SINGLE-USE BIOREACTORS**

Given the high capital and operating costs associated with conventional bioreactors, single-use bioreactors have become increasingly prevalent in upstream processing. Such systems also offer clear economic and time-to-market advantages, as well as evidence of enhanced performance. The same scale-up issues discussed above apply to single-use systems, although the predominant designs use the more conventional stirred-tank approach with plastic impellers that essentially mimic traditional bioreactors.

These bioreactors are available in sizes ranging from 50L to 2,000L (see Figure 1), and include gas/liquid inlet and outlet ports, filters, pressure control valves and additional ports for sensors. The primary advantage of single-use bioreactors is that bags can be easily reconfigured to add additional measurements, ports or features – thereby facilitating a more timely process transfer and optimisation. A significant disadvantage of single-use bioreactors is that bags can be easily reconfigured to add additional measurements, ports or features – thereby facilitating a more timely process transfer and optimisation. A significant disadvantage of single-use bioreactors is that the performance of traditional electrochemical sensors for pH and dissolved oxygen (DO) is often compromised, so that direct comparison with glass or stainless steel vessel processes is difficult or simply impossible. Moreover, until recently, the control systems used for larger single-use systems lacked both performance and sophistication when compared with their stainless steel counterparts.

**MEASUREMENT TOOLS FOR SINGLE-USE SCALE-UP AND TECH TRANSFER**

Single-use bioreactors and systems are now at a pivotal stage in their evolution; they need sensors and control systems matched to their physical properties, mode of operation and mode of sterilisation. The addition of single-use sensors will alleviate many of the issues that currently plague scale-up activities in traditional bioreactors/fermentors, and that can be engineered out of single-use systems. Put succinctly, without sensors there can be no control, and without measurement there can be no control. Equally true, if the sensors are not consistent, accurate, precise and reliable, then neither will be the measurement, control or efficacy of the process.

Traditional electrochemical sensors are not well matched to the single-use paradigm for a variety of reasons:

- Traditional sensors are typically sterilised via autoclave, whereas single-use containers are sterilised using gamma radiation.
- Traditional sensor calibration drifts due to the autoclave process, necessitating recalibration or simply introducing inaccuracy.
- Traditional sensors have significant unit-to-unit variation because of the non-uniformity in their manufacturing process (for example, membrane response, electrolyte levels and so on).
- Traditional sensors require contact with an electrically conductive housing (such as stainless steel) in order to have shielding from noise ingress and electromagnetic interference.

Additionally, in order to make a sterile introduction into the single-use container, the traditional electrochemical sensor must utilise an interface device, such as the Pall Kleenpak™ HT connector. The sensor is autoclaved in one component of the device, and then mated with a second component which is pre-inserted and gamma sterilised in the single-use container. The two components make an aseptic connection between the sensor and the bioreactor when the traditional sensor is pushed through the device and into the bioreactor. However, these devices incur additional cost and have finite failure rates (for example, leakage, sensor breakage, and so on) that compromise the process.

**SINGLE-USE SENSORS**

In order to enable single-use systems to scale up and down seamlessly, facilitate PAT concepts and meet cGMP requirements, single-use sensors with the appropriate features are required, namely:

- All wetted materials compliant with USP Class VI and/or ISO10993.
- Gamma radiation resistance or tolerance.
- Pre-calibrated to known standards.
- Economically viable.
- Able to provide electronic data traceability.

At Finesse, we have developed a range of sensors that utilise a completely disposable element that comes pre-inserted and pre-sterilised with the single-use container; this sterile barrier is never broken. The disposable elements are all gamma radiation resistant, and all wetted materials are either tested to USP Class VI standards and/or are a priori designated as ISO 10993 compliant.

A family of optical sensors (TruFluor™) provides DO and temperature, or pH and temperature, as process variables. Additionally, the disposable element comes with a pre-programmed radio frequency identification (RFID) tag that contains all the calibration information.
as well as the lot traceability information. This tag is read automatically by the associated transmitter. The calibration is performed against National Institute of Standards and Technology (NIST) traceable gas or pH standards so that consistency can be maintained.

A pressure system (TruTor®) is a disposable and fully calibrated plug-and-play pressure sensor, which utilises a built-in gamma radiation resistant memory chip. This device is calibrated against an NIST traceable pressure standard, and the calibration details are pre-programmed into the memory chip, allowing the same consistency of measurement and ease-of-use as the optical product.

The fact that these sensors come pre-inserted and pre-calibrated makes them convenient to use, while minimising operator error. Equally importantly, these sensors increase process repeatability and enable full traceability. The same sensor can be used in single-use vessels ranging from 1L to 2,000L, thereby allowing the user to bring accuracy, precision and – above all – consistency across the desired bioreactor set for scale-up. By using disposable sensors, the bioprocess professional can, for the first time, compare data across the full range of vessels being utilised, in order to clearly understand the scale-up trends affecting process development plans and experiments.

TURNKEY SCALE-UP CONTROL SOLUTIONS

For single-use reactor scale-up, the measurement problem has now been addressed – but the bioreactor and control system issues remain open. For volumes between 1L and 50L, the only single-use bioreactors currently available on the market are the popular pillow-bag reactors that use a rocking motion for mixing and aeration. The process dynamics that occur in these pillow-bag reactors are very different from their stirred-tank counterparts, and also vary widely between different manufacturers. There is, therefore, a need for either affordable stirred-tank single-use bioreactors in the 1L to 50L range, or for a control system that can unify the different bioreactor platforms and scale between them.

The Finesse TruBio™ control system was developed specifically with scale-up and technology transfer applications in mind. For example, the input and utilisation of data from off-line sampling or third-party devices (such as weigh scales, additional sensors or actuators) and all process data is stored in a single historical database for collection, retrieval and sharing of process results. Similarly, the thousands of parameters required to set up and operate a vessel can also be saved and re-loaded at will. In fact, these ‘configurations’ as well as recipes can then be shared between vessels and locations, thereby allowing two groups in physically distinct locations to work simultaneously on the same project.

Additionally, the system has the capability to be overlaid on different bioreactors using their serial communication protocol (with a Finesse TruConnect converter gateway) or ethernet connection, or to create an OPC data connection with other industrial control platforms. Once the software becomes the ‘master’ controller (for simple controllers) or main interface (for industrial controllers), it can apply its unique features to scale between different bioreactor engineering models or overlay its basic control loops and even advanced control algorithms for execution on the local controller, in order to scale a process and perform a batch consistently – irrespective of the reactor type.

CONCLUSION

Single-use bioreactors are reaching maturity very rapidly. Two new technology developments have the potential to resolve those issues preventing cost-effective and timely process scale-up and/or technology transfer of biological processes:

- First, the addition of disposable sensors that are matched to the single-use paradigm, thereby enabling these bioreactors to become the mainstream platform, from research through cGMP manufacturing.
- Second, the availability of flexible, intelligent control software that can both overlay on existing R&D infrastructure yet perform sophisticated control of large-scale single-use vessels.

Once these two solutions are fully accepted in the marketplace, it is anticipated that significant improvements in the utilisation of multi-product facilities and both inter- and intra-company process transfers will be realised. In turn, these improvements will provide quantifiable benefits in new product time-to-market and overall scale-up costs.

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