The evolving role of automation in process development and manufacture of cell and gene-based therapies

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Abstract

The need for automation in cell and gene therapy manufacturing processes is primarily driven by the regulatory requirements for reproducibility, clinical need for consistent, efficacious therapies and commercial constraints of developing scalable, cost-effective processes. Automation has already played a pivotal role in healthcare manufacture, including that of successful vaccine production. However, the role of automation continues to evolve and technologies and platforms are now emerging which succeed in expediting efforts in process development, facilitating high-throughput approaches, thereby effectively reducing the cost of development and time to market. Moreover, early integration of automation facilitates process transitions at later stages of clinical development and commercialisation. However, meeting the capital costs of developing an automated solution can be challenging, particularly in early development; a strategic decision needs to be made regarding the point in the development pathway automation is introduced, and to what extent. Factored into this decision is the nature of any solution, for example a turn-key automated system or a bespoke platform. Ultimately the extent and nature of any approach will depend on our level of process and product understanding; as this improves, we are likely to see a significant shift toward the adoption of automated solutions.

Introduction and scope

Automation for cell and gene therapy production has often been associated with high capital costs and concerns that it may not be amenable to accommodate the significant level of process complexity associated with cell-based products. Moreover, many early-stage cell and gene therapy candidates are developed as manual-based processes and emanate from

academic/clinical centres and SMEs which often do not have the resources nor the translational capability to automate the bioprocess. It is becoming increasingly clear, however, that in the pursuit of a cost-effective, scalable, reproducible production process for cell and gene therapy products, automated technologies and/or processes are likely to form an intrinsic part of any manufacturing strategy. This is driven by the need to reduce variation in the manufacture of inherently complex, living products, improve quality, comply with regulatory standards, enable comparability between manufacturing sites and reduce production errors (Thomas *et al.* 2007, Williams *et al.* 2012).

Early automated systems for cell-based therapy applications were focused on systems to mimic human operator processes. For example, the SelecT and CompacT SelecT systems (TAP Biosystems – now a Sartorius Stedim company) are completely automated platforms that were designed to undertake routine cell culture activities such as passaging, fully equipped with an incubator to house the tissue culture flasks post culture. Such systems demonstrated increased levels of process consistency for a range of therapeutically relevant cell types and enabled the application of further process improvement techniques such as six-sigma (Thomas *et al.* 2008). Through automation, process variance associated with interbatch and interoperator variation (Veraitch *et al.* 2008) is significantly reduced resulting in a more reproducible process.

In contrast to the early reticence concerning automation, automated solutions for cell processing continue to be developed and evolve, with increasing levels of adoption across industry and academia. Approaches include complete integrated and closed process solutions through to automation of discrete operations within a process chain. The development and application of automation is driven by the market/regulatory requirements and enabling technological progress. The former are continually changing with advances in the nature of products that are based on cells and their underlying processes, whilst the latter progresses with improvements in multiple relevant areas such as hardware, analytical techniques, process

understanding and operational software. This article introduces some important areas of focus driving implementation of different automated solutions in the therapeutic cell based process and product market.

Automation objectives

Automation in cell culture is driven by common core objectives: accuracy and precision of process parameter control to deliver reproducibility and repeatability (table 1) in process outcomes. Frequently, scalability of these factors is also an important consideration to allow increased throughput experimental design or commercial scale production with reduced risks of serious deterioration in process control. In industrial settings, economic scalability is a further important characteristic; control and reduction of cost of goods can be achieved when increased capital spend on automation is more than offset by reduction in costs associated with manual labour and cost of low quality product or batch failures arising from variable manual processes. There is, however, a fine balance with regards to the implementation of automation in any clinical development process; introduced too early in development and it may result in high, unmanageable capital costs and a degree of inflexibility, unable to adapt to changes as process/product understanding improves with time. Too late in development and the costs and time associated with repeating clinical trials and/or demonstrating comparability after making a significant process change render automation non-viable, even if in the long term, scalable and reproducible manufacture via a manual-based process is limited. Therefore a clear and coherent strategy is required to balance when, and to what extent, a process is automated. This is dependent on a multitude of factors including level of process and product understanding to facilitate effective comparability studies, type of automation required (i.e. a turn-key solution

or a bespoke platform), the company's financial capacity and risk appetite and the predicted market demand for the therapy amongst others.

The role of automation extends beyond the scope of manufacture; its importance is increasingly recognised for robust, high-throughput process development, effectively reducing the cost of development and time to market. Recent innovations in downscaled automated cell culture include commercially available micro-scale bioreactor systems offering control at the sub-10ml level usually associated with larger-scale reactors, with a focus on producing representative micro-scale systems to enable truly high throughput identification of scaled automated operating parameters (Nienow *et al.* 2013, Bareither and Pollard 2011, Jain *et al.* 2011). Such downscaled systems, or modular components of automation, reduces process risk and makes introduction of automation earlier in processes feasible, where previously capital costs would have been prohibitive for early stage R&D. Earlier integration of automation facilitates process transitions at later stages of clinical development and commercialisation where automation becomes imperative and simplifies process comparability work during transitions of location and scale. Such systems also facilitate the investigation of large numbers of process parameters in comparison with manual experimentation, and in conjunction with the additional level of process control associated with automation, provides statistical rigour to experimental findings.

Evolving requirements of new products

Human cell types likely to form the seed stocks or intermediates for clinical cell products have shown extreme sensitivity in lineage selection and proliferation to aspects of the bioprocess environment such as nutrient, gas supply or pH (Oburoglu *et al.* 2014, Studer *et al.* 2000). This

will demand a corresponding level of process control to maintain product quality. Automation allows increased frequency of monitoring and response through removal of restrictions on sampling frequency and feedback algorithms imposed by manual operators. This opens the avenue to more advanced process control strategies such as closed loop and PID control (Caldwell *et al.* 2015); process intensification or cell types with high sensitivity to process environment are likely to necessitate such approaches. As mentioned previously, however, there is a perception that automation limits flexibility and may be unable to adapt to the evolving requirements as process and product understanding improves. The strategy of automation implementation needs to account for this and due consideration given when automated technologies are selected.

Need for cell culture models to support automation

The capability of hardware with integrated sensors and responsive control can only be exploited to the degree that an appropriate model of control exists for the process and product. Cell based products have specific requirements for modelling for production; that models are complex enough to facilitate optimisation to target endpoints but cost efficient enough in development to justify investment. Whilst computational biology has made progress in development of complex mechanistic and empirical modelling approaches necessary to represent biological systems these are often challenging to develop with expensive and relatively sparse data available in development (Wilkinson 2009); the corollary to this is that simple empirical approaches such as Design of Experiments often fail to adequately represent complex cell culture dynamics in any meaningful way. An understanding of the model sophistication and structures most appropriate for the field will be critical to leveraging the potential control opportunities of automation and moving from simple fixed control to optimised systems with

active feedback control. Advanced process modelling, such as dynamic mechanistic models, population based models or combinations of such approaches need to evolve with automated platforms. These will provide confidence in required precision and allow cost-benefit (risk) calculation in determining design precision of automation and process operation.

Modular automation and closing systems

Operational risk is increased in open systems with multiple manual interventions; automation has the attraction that closing the process from the external environment becomes simpler due to reduced or eliminated human intervention. As cell therapy processes cannot be sterile filtered, the need to minimise, or ideally eliminate, open manipulations is paramount. Fully closed systems are naturally simpler when the unit operations they encompass are more limited, for example, Terumo BCT's COBE® 2991 Cell Processor which enables cell washing and concentration, or indeed cell expansion platforms such as automated stirred-tank bioreactor platforms. Some systems, such as Miltenyi Biotec's CliniMACS Prodigy® and Invetech's approach of leveraging modular and/or integrating off-the-shelf equipment facilitate the combination and integration of a wider range of unit operations including selection, activation (in the case of T-cell therapies), expansion and cell processing. In contrast, end to end closure is more challenging for lengthy processes encompassing multiple operations due to the operational specificity imposed, however some ideas at reconfigurable modular closed automation are currently being investigated such as those being developed by Tokyo Electron Smart Cell Processing Technologies and the IPT Fraunhofer (StemCellFactory 2015, Rafiq *et al.* 2016).

Integrated closed units either have to demonstrate a marked improvement in manufacturability and capture a large enough market to justify their single purpose or retain the tunability to cater to multiple markets. With the opportunity and challenges associated with the production of autologous (patient-specific) therapies, there is likely to be a market requirement for tuneable, automated systems. This is necessary to accommodate the constraints of autologous material and inherent donor variation associated with such cells arising from differences in patient characteristics, disease state, method/process of tissue isolation and cell selection and stochastic events. There is precedence, however, of being able to accommodate and adapt to specific process requirements from the biopharmaceutical industry, whereby automated systems can respond to the variation in product titres and glycosylation profiles for monoclonal antibody production (Carson 2005).

In addition to automated manufacturing systems, dedicated automated cell isolation and selection platforms, such as the 'Stem Cell Factory', are beginning to emerge in important areas such as colony picking that are seen as key to quality in cell reprogramming (StemCellFactory 2015). We are seeing confluence of 'smorgasbord' automation where multiple unit operation automation are plugged together with single solution automated systems.

In terms of automation of individual unit operations or processes, some parts of the market are better catered than others; QC/analytical techniques, in particular, cell counting and flow cytometry are good examples of process operations with multiple automated solutions. Cell counting, typically a variable and time consuming process when undertaken by a human operator using a haemocytometer, is now considered to be more reproducible and repeatable when using automated cell counting systems. However it is also a good example of the conflict

between turn-key solutions and tuneable equipment; cell counters working simply on cell size can introduce systematic bias across a process due to cell size changes with phenotype or proliferative state. More complex tuneable and multi-parameter systems, such as those using viable and non-viable dyes add complexity in a production setting and potentially increase opportunity for operator variability and sampling error. A solution to this is the introduction of pre-calibrated and pre-qualified consumables such as cartridges or cassettes. Indeed Chemometec's automated mammalian cell counter (the NC-3000 Nucleocounter®) operates on the basis of immobilised viable and non-viable dyes (acridine orange and DAPI respectively) contained within pre-calibrated cassettes providing a precise and reproducible result.

Processes are likely to require a combination of approaches for measurement of key parameters; more importantly, whilst a given automated monitoring approach, such as viability, may be sold as a turnkey solution, the appropriate interpretation and response to the measurement may take considerable work to appropriately develop for individual cell types and products. Indeed, the onus is on the cell therapy to undertake installation, operational and performance qualification of the equipment and must ensure that the equipment, and its parameters, are suitable for the specific process at hand.

What does automation need to drive forward?

Early adoption and integration of automated platforms and systems is an important factor in the implementation of automation at later stages of clinical development. Sophisticated automated control and models at the small scale are essential to enable process design for larger-scale systems. With the emergence of the ambr15 cell culture® and ambr250 modular

systems as relevant small-scale models for larger-scale stirred-tank bioreactors, high-throughput experimental designs can now be developed. Moreover, as such systems are more reasonably priced compared to more expensive larger-scale systems, no longer is effective early R&D work limited to the confines of industry. The need for relevant, automated scale-down models is necessary to reduce R&D costs and enables academic centres to conduct translational R&D, thus contributing and improving the pre-competitive landscape. However, not every process will employ a stirred-tank bioreactor and the dearth of relevant small-scale, high-throughput models for other systems such as the rocking motion (Wave®-type) bioreactors and other expansion/cell-processing equipment, means that early adoption of automated systems may be limited. Tuneable low scale automation allows data to be generated that defines/justifies spec of larger scale automation.

Adoption of automation is also inextricably linked to process and product understanding. Understanding the process and product in greater detail allows for a greater awareness and clarity of what aspects of the process are amenable for automation, enables effective automation and identifies what the act of implementing automation is expected to achieve. Better process understanding allows for the development of a more refined automated solution This helps to redress the balance mentioned earlier and reduces the risk associated with moving toward an automated process.

Automation in the cell and gene therapy sector has focused primarily on developing platforms and underpinning technologies associated with cell manufacture, however there is a need to consider the role of automation in the broader process including cell collection, administration, dispensing, logistics, traceability and tracking. Moreover, there is a need to focus on the

'information automation', i.e. operational automation, and integrate data management; this will, in particular, have significant value for patient-specific therapies. This may facilitate the establishment of a validated system that enables batch 'release by exception', resulting in a significant reduction in QA and verification costs.

Although the challenges of developing and integrating automated approaches for cell and gene therapies are significant, the clinical and commercial opportunity of administering advanced, patient-specific therapies will continue to drive innovation in automation and allow us to realize a new-age of therapeutics.

Table 1: Definition of measurement system terminology

Accuracy	Refers to the distance of the measurement of a specific measurand from the "true" value
Precision	Refers to the distance of a measurement point relative to another for a specific measurand (i.e. spread of the data)
Reproducibility	The degree of agreement between measurements of a specific measurand when undertaken by different operators
Repeatability	The degree of agreement between measurements of a specific measurand when undertaken by the same operator

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