Challenges for Particle Control in Cell Therapy Products

Introduction

Cell therapy is rapidly gaining momentum as a therapeutic modality, with several licensed market products and a robust clinical development pipeline. Beyond the commercially available cell therapy products and those under development, stem cells have been a standard treatment in hematology and oncology for many years.

Despite the promising therapeutic efficacy, several challenges persist in the development and production of cell therapy products. The manufacturing process significantly deviates from traditional automated, high-volume biopharmaceutical manufacturing in closed isolator systems. In fact, only a few cell therapy manufacturers have successfully transitioned from early lab-based research to a commercial process designed to support a complex cell therapy pipeline.

The cell therapy manufacturing process is often operator-driven with handling in an open cleanroom. The target patient populations are typically small—sometimes as small as one, in the case of autologous therapies that rely on a patient's own cellular material as the therapeutic vehicle. Furthermore, the manufacturing material, including product-contacting equipment and consumables, are often sterile but not optimized for reduced particle shedding.

Challenges in the Control of Visible and Sub-visible particles in Cell Therapy Products

Cell therapy manufacturing possesses unique challenges, given that cells are the active pharmaceutical ingredient. Consequently, there are inherent limitations to the purification processes that can be employed, and the effective removal of particulates from the final formulations is constrained. This limitation arises from the fact that cells cannot pass through a sterile filter capturing particulates without also entrapping the cells. Consequently, particles introduced into the product during manufacturing have the potential to be present in the final product.

Additionally, the risk of introducing particles into the product is higher when compared to traditional biopharmaceutical products. Manufacturing operations often take place in a conventional

clean room or a simple laminar flow bench, involving numerous manual steps. In the case of a flexible isolator solution, gloves are used to execute these manual procedures. Furthermore, cell therapy single-use manufacturing equipment often represents tailored solutions without extensive market history, typically not optimized to minimize particle shedding into the product.

In addition to the increased risk of the introduction of particles in the product and the inability to filter the product, conventional analytical methods for visible and sub-visible particles may be constrained. For example, the visual inspection process is hindered by the elevated turbidity of cell therapy products, coupled with the temperature sensitivity and small batch sizes inherent to these products. These factors often prevent the implementation of a conventional visual inspection process, including a 100% inspection as an integral part of the manufacturing process, with subsequent AQL sampling. Likewise, pharmacopoeial sub-visible particle methodologies, such as light obscuration, face limitations in distinguishing cells from other extrinsic or intrinsic sub-visible particles.

Considerations to Control Visible and Sub-visble Particles in Cell Therapy Products

Visible and sub-visible particles are typically considered critical quality attributes given the general concern for patient safety and the potential impact on cell viability / cell aggregation.
It is the responsibility of the sponsor of the cell therapy product to characterize and understand the load, source, composition and potential impact of particulates in the final formulation of their product. In order to address this challenge, sponsors need to assess the risk for introduction of visible and subvisible particles in their manufacturing process, considering factors such as potential sources, number, size, composition, and criticality.

Typical particle sources are:

- Raw materials: formulation buffer components, process solutions
- Manufacturing equipment: pumps, isolator gloves, HVAC, capping equipment, tubings, bags, flasks, regents, lab ware, etc
- Primary packaging: bags, vials, stoppers
- Gowning material and manufacturing operators

Effective collaboration between suppliers and sponsors is crucial to proactively control and minimize the introduction of particulates. While complete removal of all particulates may be challenging, it is essential to assess each manufacturing step for particulate control by scrutinizing the sources outlined in the list above.

Apart from the manufacturing challenges, conventional analytical methods for particles in biopharmaceutical products have limitations when applied to cell therapy products.

Compared to conventional biopharmaceutical drug products, visual inspection poses significant challenges, as cell therapy products are typically turbid and opalescent suspensions. Moreover, factors like cell density, cell type, formulation characteristics, and the nature of primary packaging introduce additional complexities for operators attempting to identify visible particles. For instance, inspecting cell therapy bags proves more challenging compared to drug products filled in conventional glass vials.

Developing an effective visual inspection method requires careful consideration of various factors, including establishing appropriate inspection times, determining optimal light intensity, and refining sample handling procedures. It's important to note that the final inspection parameters may deviate from those recommended by pharmacopoeias. A product-specific visual inspection qualification test kit should be established representing actual product characteristics such as viscosity, turbidity and fill volume. Furthermore, the product specific visibility size limits can be established by performing a threshold Knapp test which accounts for the significant interference caused by the turbid cell suspension in the detectability of visible particles.

Likewise, testing for sub-visible particles (SvP) poses a significant concern, particularly in the context of cell therapy products. Both the US and European pharmacopeia provide two methods for determining SvP contamination in parenteral products. Method 1 relies on light obscuration, while method 2 involves a microscopic particle count. However, these methods present limitations when applied to SvP testing in cell therapy products, primarily due to the inherent presence of cells in the measurement.

Light obscuration, under Method 1, has the drawback of detecting and classifying cells as subvisible particles, lacking the capability to distinguish cells from other SvP. On the other hand, the microscopic method faces challenges as sub-visible particles may be obscured by cells. Addressing these limitations, image-based methodologies, such as flow imaging microscopy analysis powered by artificial intelligence image analyzing algorithms, emerge as a valuable approach for characterizing sub-visible particles in cell therapy products.

Flow imaging microscopy has the distinct advantage of discriminating between cells, cell debris, and other sub-visible particles originating from the formulation or manufacturing process. Despite not being a pharmacopeial method, flow imaging microscopy analysis stands out as the most promising methodology for assessing sub-visible particles in the context of cell therapy products.

An alternative strategy for assessing visible and subvisible particles involves monitoring particulates external to the actual cell manufacturing process. This can entail conducting media fills or placebo fills to demonstrate that the manufacturing process itself contributes minimal levels of particulate contamination. Adopting this approach also enables the utilization of the pharmacopoeial sub-visible particle Method 1. While this presents an alternative to testing the actual cell therapy product, it is crucial to acknowledge its inherent limitations and risks. Notably, the levels of particulate contamination may vary during the actual production runs. Additionally, relying on aseptic media or water runs for such testing overlooks potential particulates originating from the cell therapy formulation.

Summary and Outlook

Cell therapy, despite its promising therapeutic efficacy, faces challenges in manufacturing and for setting up a particle control strategy. The process deviates from traditional biopharmaceutical manufacturing, often being operator-driven and conducted in open cleanrooms. The inability to filter cells without entrapping them limits the removal of particulates from final formulations. The risk of particle introduction is higher, and both compendial visual inspection and sub-visible particle testing exhibit subpar performance. Sponsors must understand the load, source, composition, and potential impact of particulates in their products and should closely collaborate with manufacturing equipment supplies to minimize particle shedding into the product. Ultimately, there is a need for harmonization within the global regulatory landscape, taking into account the unique challenges associated with cell therapy products and addressing them through dedicated guidance documents.