Efficient GMP-compliant expansion of mesenchymal stromal cells (MSCs) from umbilical cord, bone marrow, and adipose tissue using a closed cultivation system

Introduction

Human mesenchymal stem cells (MSCs) hold great promise for cellular and cell therapy applications, and can be isolated from multiple tissues, e.g., bone marrow (BM), umbilical cord (UC), and adipose tissue (AT). To ensure high-quality and safety of the resulting clinical products, MSC lines have to be maintained using standardized cultivation conditions and procedures. To meet end-use requirements of GMP (Good Manufacturing Practice) and CACFP (Comprehensive Assessment of Cell Therapy Products), MSCs derived from different starting materials (BM aspirate, dissociated UC or adipose tissue-MSCs (AT-MSCs)) were pre-expanded prior to transfer to the CliniMACS Prodigy Adherent Cell Culture System, combining the process modules in a flexible way.

The process includes the following modules, which can be configured as required:
- Density gradient centrifugation (DGC)
- Surface coating
- Inoculation
- Media change

Here we show that MSCs from different tissue sources can be expanded and passed from primary tissue or single-cell suspensions using the CliniMACS Prodigy Adherent Cell Culture System, combining the process modules in a flexible way.

Methods

1 Manufacturing of MSCs using the CliniMACS Prodigy® Adherent Cell Culture System

The CliniMACS® System provides a range of options for setting up closed bags containing tissue, cells, reagents, and media, making different starting cells viable for a variety of applications, e.g., for the cultivation process, our two-step cultivation set (CliniMACS Prodigy® T20) which provides up to eight connections for bags. The system also offers options to perform solutions transfer from an external 4 °C storage compartment to the cultivate and centrifugation unit. DGC and all subsequent steps are controlled autarkically by the system, while the preparation of tissues and AT-MSCs (AT-MSCs) and BM aspirates was performed outside of the system. In this study, tissue aspirates were collected in the CliniMACS Prodigy Adherent Cell Culture System, including the initial isolation steps in the CACFP as well as all labelling steps, in inoculation, washing of cells, medium exchange, and in harvesting. Fig. 2 describes the system after the possibility of taking samples to determine the number and analyze marker expression.

Different human starting materials were used and processed to isolate and expand MSCs from umbilical cord, bone marrow, and adipose tissue using the CliniMACS Prodigy® Adherent Cell Culture System and manual processing. The MSC-Brew GMP Medium used in this process is xeno-free and meets the recommendations of USP <1043> on ancillary materials.

Results

Fig. 2. Expansion of MSCs using the CliniMACS Prodigy® Adherent Cell Culture System

Expansion of MSCs using the CliniMACS Prodigy® Adherent Cell Culture System

- The novel CliniMACS® Prodigy Adherent Cell Culture System enables continuous cultivation of adherent cells in a closed system.
- MSCs from different tissue types (bone marrow, umbilical cord, and adipose tissue) were expanded using the CliniMACS Prodigy Adherent Cell Culture System and analyzed for expansion using standard tissue culture methods to determine the expansion potential at all conditions (fig. 3).
- MSCs were expanded for at least two passages, which was sufficient to harvest cells in a clinically relevant number in giga-scale. This novel technology enabled the automated processing of bone marrow aspirates using a density gradient centrifugation within the system.
- The MSC-Brew GMP Medium used in this process is xeno-free and meets the recommendations of USP <1043> on ancillary materials.

Conclusion

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