Development of a Novel Serum-Free Medium for Mesenchymal Stem Cells, Complying with the Japanese Standards for Biological Ingredients

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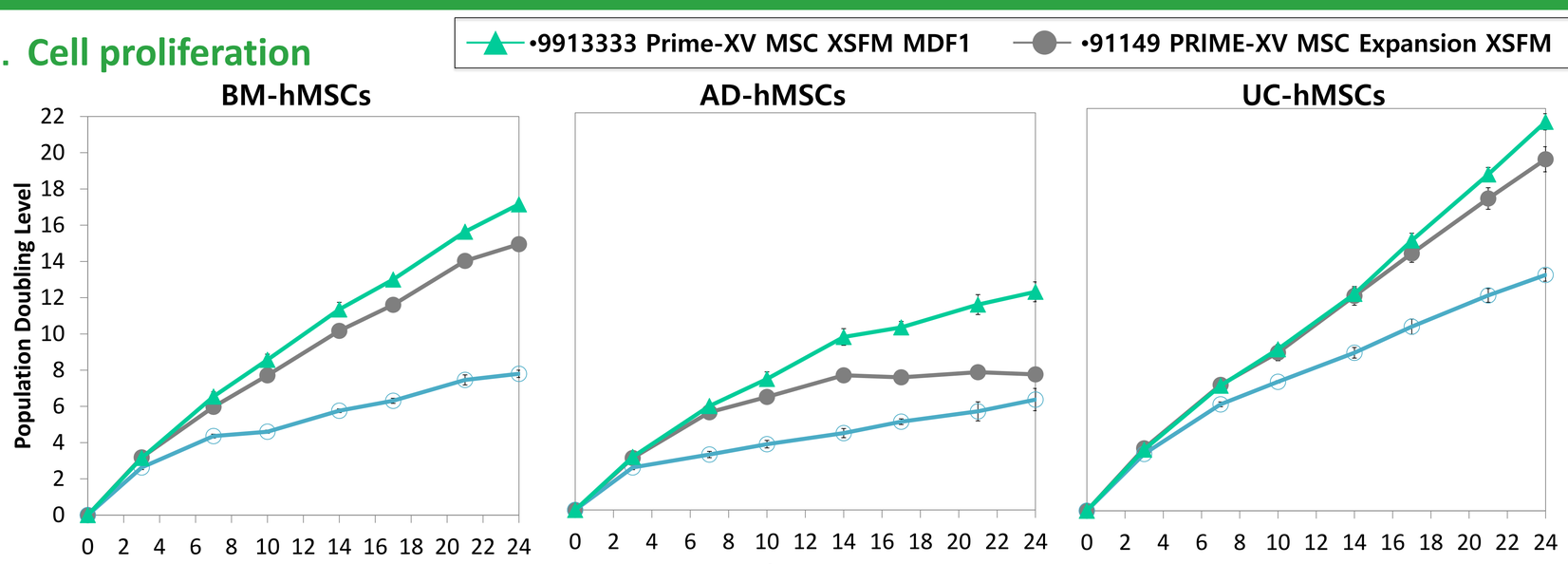
Introduction

Serum-containing culture media have been widely used for basic research and manufacturing of regenerative medicine products in the past. However, serum contains a potential risk for virus infection and lot-to-lot variability. To overcome these problems, serum-free media have been developed by various media suppliers and as a result, many alternatives are available on the market. On the other hand, each country poses different standard and requirements for the registration of final products. As an example, Pharmaceuticals and Medical Devices Agency (PMDA) of Japan requires cell culture media used in regenerative medicine products to comply with its "Standards for Biological Ingredients." The purpose of such requirement is to ensure the quality, efficacy and safety of the final products.

In this study, we modified PRIME-XV MSC Expansion XSFM (XSFM; FUJIFILM Irvine Scientific Inc.), a serum- and Xeno-free medium for expansion of human mesenchymal stem cells (hMSCs), to respond requirements posed by the PMDA. The modified medium, Prime-XV MSC XSFM MDF1 (MDF1), was evaluated on its cell growth capability, morphology and immunophenotype with hMSCs derived from three different sources.

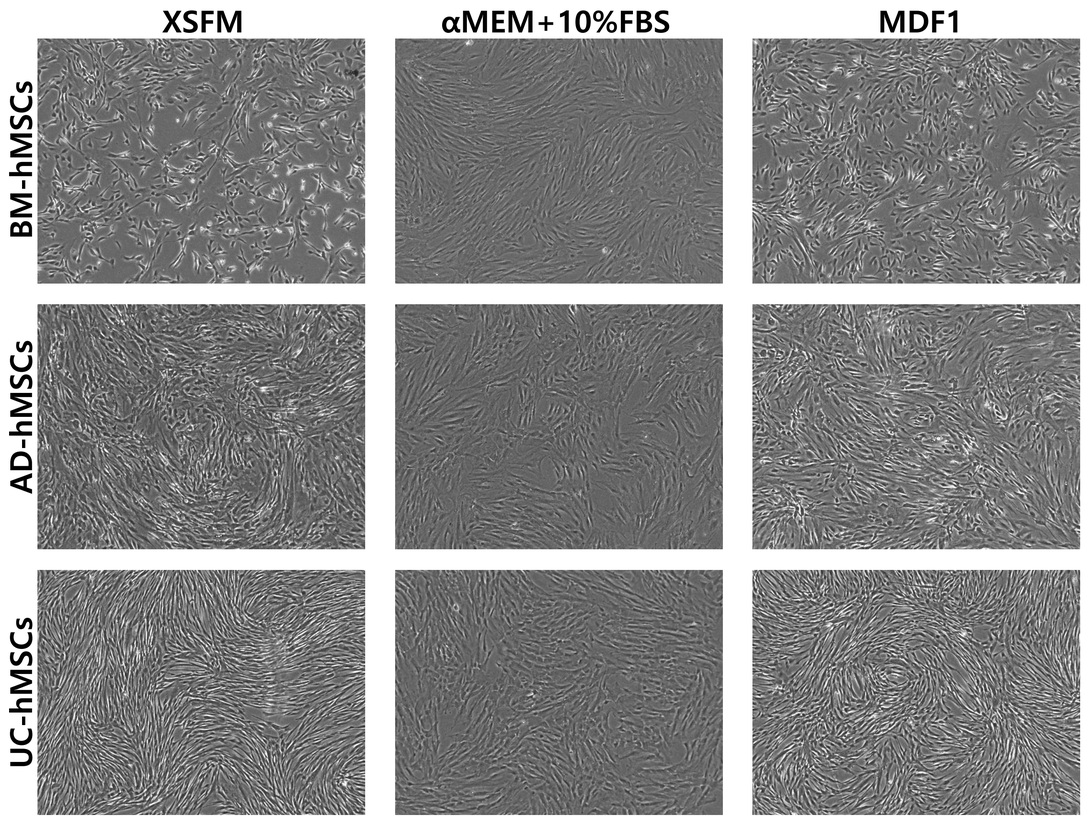
Results

1. Cell proliferation



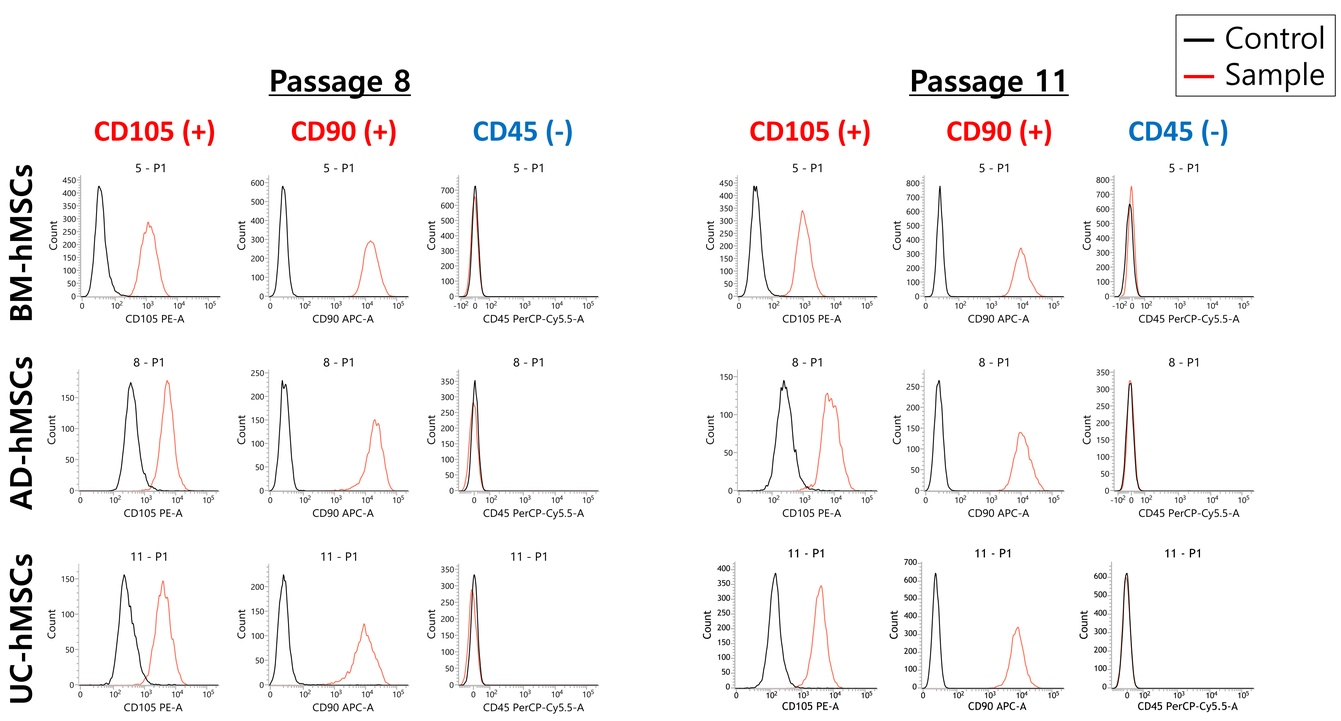
Bone marrow (BM), Adipose (AD) and Umbilical cord (UC)-derived hMSCs from ATCC were cultured for 24 days (Passage 5 to 11, n=3). CellBIND® 6-well plates (Corning) were used.
As a result, we confirmed Prime-XV MSC XSFM MDF1 supports faster cell growth of BM-, AD- and UC-hMSCs than serum-containing αMEM or PRIME-XV MSC XSFM does.

2. Cell morphology (Passage 8)



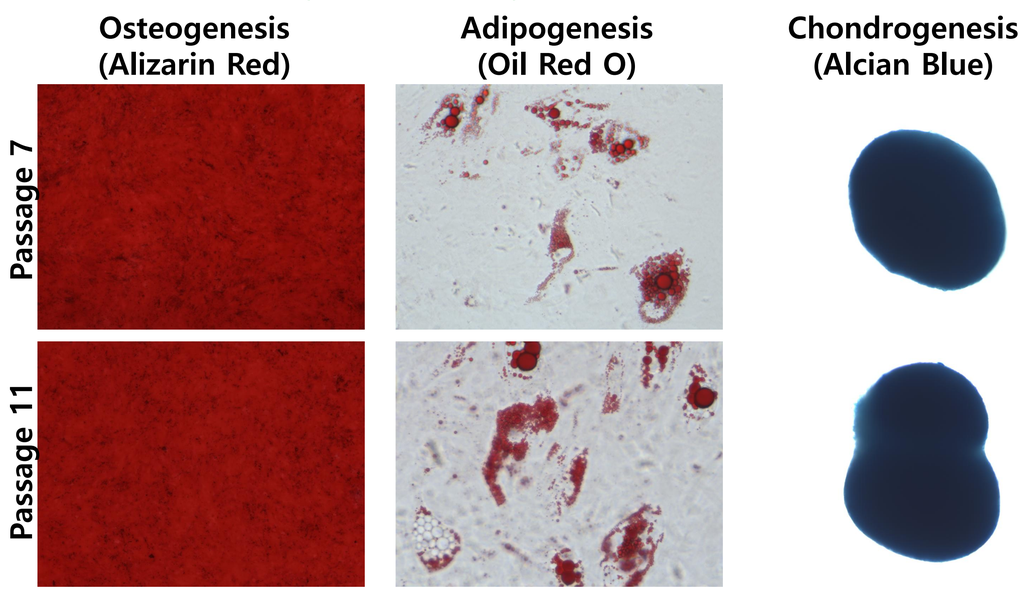
hMSCs cultured in MDF1 reveals a spindle-shape morphology, similar to the cells in XSFM or serum-containing αMEM.

3. Flow Cytometry analysis



Immunophenotypic profiles of the cells cultured in MDF1 meets the criteria of MSCs. These profiles are stable over multiple passages.

4. Differentiation (BM-hMSCs)



BM-hMSCs cultured in MDF1 were differentiated into adipocytes, osteoblasts and chondrocytes.

Conclusions

- Prime-XV MSC XSFM MDF1 was developed from FUJIFILM Irvine Scientific's proprietary formulation PRIME-XV MSC Expansion XSFM.
- Prime-XV MSC XSFM MDF1 is officially confirmed to comply with the Japanese regulation by Pharmaceuticals and Medical Devices Agency.
- Prime-XV MSC XSFM MDF1 was demonstrated to have a superior performance compared to PRIME-XV MSC Expansion XSFM and serum-containing medium.
- No negative impact on cell characteristics was observed by the media modification.
- This study indicates that Prime-XV MSC XSFM MDF1 complying with the Japanese regulation can be widely applied for manufacturing of regenerative medicine products.