Progress report 2024

Project Leader: Dehua Chang, M.D., Ph.D Project Lecturer: Shuoji Zhu, Noriko Kajiwara

[Department Outline]

Mesenchymal stem cells (MSCs) have garnered attention in regenerative medicine because of their multipotency, anti-inflammatory properties, and pro-angiogenic paracrine effects. In particular, human umbilical cord-derived mesenchymal stem cells (UCMSCs) are regarded as a promising source of cells because they come from young tissue, demonstrate high cellular activity, can be collected non-invasively, and offer advantages from a quality control perspective.

MSCs are defined by their trilineage differentiation potential (adipogenic, osteogenic, and chondrogenic) and the expression of surface markers such as CD105, CD73, and CD90.

However, even if these markers are expressed, it is not always possible to determine whether the cells are high quality or capable of proliferation. Therefore, methods for evaluating the quality of MSCs are extremely important. This study aims to quantitatively assess the morphological and biological characteristics of UCMSCs and to clarify the impact of cell passage numbers on their properties.

[Research Content]

To comprehensively evaluate the quality of UCMSCs, it is necessary to measure

morphological indicators such as cell area, perimeter, and the aspect ratio of length to width. In this study, we examined the differentiation potential and the expression of marker proteins associated with mesenchymal stem cells in UCMSCs at Passage 5 (P5) and Passage 10 (P10). Additionally, using cell image analysis techniques, we observed morphological changes in P5 and P10 UCMSCs and compared their biological characteristics for quality assessment. In cell therapy for regenerative medicine, the safety and efficacy of cells as therapeutic products are critically important as they directly influence therapeutic outcomes. High-quality cells are characterized by high proliferative capacity, small nuclei, and uniform morphology. This study demonstrated that UCMSCs at a lower passage number (P5) exhibited higher morphological stability, greater proliferative ability, and enhanced angiogenic potential compared to P10 cells.

The results of trilineage differentiation experiments (adipogenic, osteogenic, and chondrogenic) showed successful differentiation in both P5 and P10 UCMSCs. Marker proteins associated with MSCs CD105, CD73, CD90, CD11b, CD19, CD34, CD45, and HLA-DR were confirmed to be expressed in both P5 and P10 groups. In morphological analysis, P5 UCMSCs displayed better cell morphology and higher viability than P10 UCMSCs. When comparing the angiogenesis ability of P5 and P10 UCMSCs, P5 cells generated more vascular branch points and had a longer total vessel length. P5 cells also showed higher trends in cell number, cell density, and OD values compared to P10. Comparison of the cell morphology and characteristics between P5 and P10 revealed that over time, P10 cells exhibited greater fluctuations in aspect ratio and a tendency for decreased cell density compared to P5.

[Future Outlook]

These results suggest that the number of cell passages influences the morphological characteristics and properties of the cells. Furthermore, image analysis technology, by utilizing indicators correlated with cell characteristics, can be useful for evaluating the quality of mesenchymal stem cells during the manufacturing process and in the final product. This approach is expected to contribute to developing safe and effective cell therapy products.

[References]

- 1. Nascimento, D.S., Mosqueira, D., Sousa, L.M., Teixeira, M., Filipe, M., Resende, T.P. Human umbilical cord tissue-derived mesenchymal stromal cells attenuate remodeling after myocardial infarction by proangiogenic, antiapoptotic, and endogenous cell-activation mechanisms. Stem Cell Res. Ther. 2014, 5.
- Guo, R.; Wan, F., Morimatsu, M., Xu, Q., Feng, T., Yang, H. Cell sheet formation enhances the therapeutic effects of human umbilical cord mesenchymal stem cells on myocardial infarction as a bioactive material. Bioact. Mater. 2021, 6, 2999-3012.
- Kuchroo, P., Dave, V., Vijayan, A., Viswanathan, C.; Ghosh, D. Paracrine factors secreted by umbilical cord-derived mesenchymal stem cells induce angiogenesis in vitro by a VEGF-independent pathway. Stem Cells Dev. 2015, 24, 437-450.
- 4. Colicchia, M., Jones, D.A., Beirne, A-M, Hussain, M., Weeraman, D., Rathod, K. Umbilical cord-derived mesenchymal stromal cells in cardiovascular disease: review of preclinical and clinical data. Cytotherapy 2019, 21, 1007-1018.
- S.J. Zhu, J.F., Xuan, Y.C., Shentu, K.Kida, M. Kobayashi, M. Ono., D.h. Chang. Effect of chitin-architected spatiotemporal three-dimensional culture microenvironments on human umbilical cord-derived mesenchymal stem cells, Bioactive Materials May (2024)35 https://doi.org/10.1016/j.bioactmat.2024.01.014