Quality Assurance of Clinical-grade Pulp Stem Cells Manufactured in GMP-compliant Facility

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Abstract

Purpose: Several clinical reports on MSC-based disease treatment have been published, evoking great excitement and therapeutic potential for a variety of diseases. There have been, however, no biosafety reports on clinical trials harnessing pulp stem cells. We must follow the principle of clinical trials using human stem cells to manufacture clinical-grade pulp stem cells. Thus, stringent controls in all the phases of cell manipulation using an isolator in a GMP-compliant facility were performed by the standard operating procedure (SOP) to determine the quality and safety of the pulp stem cells.

Materials and Methods: Clinical-grade human pulp stem cells were isolated and expanded according to GMP conditions. Pulp stem cells were isolated based on their chemotactic response to a granulocyte-colony stimulating factor (G-CSF) gradient from pulp cells separated by an enzymatic digestion method. The cells were expanded until the 7th, 15th and 20th passage of culture and were frozen by a programming freezer. The safety tests for bacteria, fungus, virus, endotoxin, and mycoplasma were performed in transportation solution after use, the primary cell culture medium and finally frozen cells at the 7th passage. Expression rate of stem cell markers was analyzed by flow cytometry. Human pulp stem cells were injected into testes or subcutaneously in NOD/SCID mice and KSN nude mice for tumorigenicity assay. The metaphases of pulp stem cells were Q-banded and karyotyped at the 20th passage of culture.

Results: The pulp stem cells were isolated, expanded until the 7th passage of culture and frozen in the isolator. The viability of the cells was over 80%, and the expression rate of cell surface markers CD29, CD44, CD73, CD90 and CD105 was over 95% and CD31 expression was negative, indicating their stemness. The pulp stem cells demonstrated negative results in all safety examinations including endotoxin (less than 1.0 pg/ml), no tumor formation and no abnormalities/aberrations in karyotype.

Conclusion: The quality and safety of human pulp stem cells manufactured in the GMP-compliant facility and a stable manner using the SOP were demonstrated. In the near future, novel treatments of pulpitis will become routine once the first clinical trial is successfully performed to confirm safety and efficacy, paving the way for more advanced stem cell therapies in dentistry and endodontics.

Key words: Quality control, Safety examination, Pulp stem cells, Pulp regeneration, Good Manufacturing Practice (GMP)