Human periodontal ligament stromal cells: the isolation and characterisation

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Restoring periodontal defect is still one of the most challenging clinical needs; this is due to the complex structure and diversity of cell types in this joint. The advancement in tissue engineering and cell therapy made it possible to utilise these approaches to overcome such a challenge. This study aimed to investigate the characteristics of isolated human periodontal ligament cells (HPDLCs), and the multi-linage differentiation capacity of these cells to enhance the regeneration process.

Human periodontal ligament cells were isolated from healthy extracted third molar teeth. Colony forming unit-fibroblasts (CFU-f) was used to determine the presence of progenitor cells within the isolated cell population. Further characterisation was conducted by examining the expression levels of mesenchymal stem cells markers (CD29, CD73, STRO-1) as well as the levels of hematopoietic markers (CD34 and CD45), using flow cytometry. Multilineage cells differentiation was induced using adipogenic, chondrogenic, osteogenic and fibrogenic media. Then, the HPDLCs were seeded on 3D Bombyx mori silk fibroin scaffolds in vitro for five weeks and analysed using histology, immunohistochemistry and biochemical assays to confirm proliferation and differentiation processes. Afterwards, the samples were implanted into the peritoneal space of nude mice using diffusion chambers for further seven weeks. All samples were then retrieved from animal models and processed for histology.

The data of CFU-f assay demonstrated attachment and proliferation of cultured cells subpopulation onto the vessel's surface indicating the presence of progenitor stromal cells. Also, HPDLCs expressed higher levels of MSCs markers in comparison to the lower levels of hematopoietic markers. These cells showed the ability to proliferate and differentiate into adipogenic, chondrogenic, osteogenic and fibrogenic cues. Both, in vitro and in vivo experiments demonstrated the ability of those cells to attach and spread on silk scaffolds; as well as performing other cellular activities like formation of collagen fibres, along with the deposition of extracellular minerals.

In conclusion, this study showed that HPDLCs contain the essential progenitor stromal cells, which have the capacity to differentiate into the main periodontal cells; this could indicate the role of these cells in enhancing the periodontal regeneration process.