ISOLATION AND CHARACTERIZATION OF RABBIT PERIOSTEUM-DERIVED MESENCHYMAL STEM CELLS

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Key words: periosteum, mesenchymal stem cells, regenerative medicine, rabbit

Introduction: Periosteum represents a good source for mesenchymal stem cells (MSCs). They have been regarded as a promising therapeutic tool for muscoskeletal regeneration. Periosteum-derived MSCs have higher osteogenic potential in vitro than MSCs from other tissues and organs. The aim of this study was to isolate and characterize rabbit periosteum-derived MSCs for further their application for bone tissue engineering.

Methods: Rabbit periosteum was isolated from the proximal epimetaphysis tibia under ketamine anesthesia. Periosteum-derived cells were isolated from rabbit periosteum after short-term collagenase digestion treatment. The cells were cultivated and expanded in complete growth medium α-MEM. Characterization of periosteum-derived cells was performed with proliferation assay, CFU-assay, multilineage differentiation assay and morphological analysis.

Results: Our data on isolation revealed that the average yield of periosteum-derived cells from 1 mg of the rabbit periosteum was about 1 – 1.2 × 104 cells. Most isolated periosteum-derived cells adhered to the surface of culture flask and have spindle-shaped and fibroblast-like morphology. Proliferative assay showed that optimal seeding density of periosteum-derived cells is 1 × 104 cells/cm2. At this seeding density periosteum-derived cells reached 80-90% confluence monolayer in 4 days after subculturing. CFU-assay and multilineage differentiation assay revealed that periosteum-derived cells have a high capacity to form colony forming units and can effective differentiate into adipocytes, chondrocyte and especially osteoblasts. Together our data indicate that rabbit periosteum-derived cells have all features of MSCs.

Conclusion: Thus, in this study we showed that the periosteum is a good source of MSCs which can be easily isolated in large numbers and quickly expanded in vitro for preclinical study and tissue engineering research.