Biphasic ability of periosteum-derived cells between osteoblastogenesis and fibroblast genesis in vitro

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Background: Periosteum comprises a bilayer structure—an outside fibrous layer and an inside cambium layer. Incomplete preservation of alveolar bone is often observed because of compromised periosteum generating at the bony defect such as in a clinical setting. Thus, periosteum is considered indispensable for bone healing. We hypothesized the double-layered periosteum has reciprocative ability of fibroblastic differentiation and osteoblastic differentiation of the periosteal cells due to their environment.

Aim/Hypothesis: The aim of this study was to investigate the biphasic potential of periosteum-derived cells (PDCs) induced by osteogenic or fibrogenic cell culture condition.

Material and Methods: PDCs were isolated from calvarias of nine 4-week-female ICR mice. Bone marrow cells (BMCs) flushed from the femur of the same mice were used as control. All cells were identically passaged for 3rd or 4th generation. At 1 week of culture in Dulbecco's Modified Eagle Medium (DMEM), cells were divided into three culture medium treatments—continuous DMEM (Treatment D), 10 ng ml TGF-β1 supplemented DMEM (Treatment Dt), osteogenic induction DMEM (Treatment O). The cells were cultured for two weeks in each culture medium. Then the D and Dt groups were separated into two subtratements, respectively D-to-D and Dt-to-Dt, and D-to-O and Dt-to-O. On the other hand, Group O was separated into three subgroups—O-to-O, O-to-D and O-to-Dt. Then the cells in all groups were further cultured for 2 weeks. The cells were collected at 3 and 5 weeks to isolate total RNA. The expression levels of transcription factors related to osteoblast and fibroblast differentiation were analyzed by real-time PCR.

Results: Runx2 gene expression was higher in PDCs compare to BMCs in D-O and O-D. On the other hand, Osterix mRNA showed lower expression in PDCs than in BMCs over time except for Dt-Dt and O-Dt. Notably, Fgfr1 gene expression was observed elevated in PDCs compared to that in BMCs as well as the osteogenic transcription gene, Runx2 in both O-Dt and Dt-O. α-SMA gene expression profile was similar to that of Fgfr1. Briefly, Gene expression of the examined transcription factors for osteoblastic differentiation was relatively higher in PSCs cultured in the osteogenic condition at the first stage.

Conclusions and Clinical Implications: In the present study, PDCs prominently showed biphasic gene expression patterns for both osteoblastic and fibroblastic differentiation by switching the culture conditions, compared to BMCs.