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OSTEOBLAST MINERALIZATION IS INHIBITED BY SIMULATED MICROGRAVITY USING
RANDOM POSITIONING MACHINE**Abstract**

Purpose: Microgravity-induced bone loss is one serious risk for the astronauts' health and an obstacle for human exploration of deep space. Previous studies have demonstrated that the decrease of mature osteoblasts and bone formation is one main cause for microgravity-induced bone loss. In this study, a desktop random positioning machine (RPM) was applied to simulate microgravity condition and the effect of simulated microgravity on osteoblast mineralization was studied. **Methodology:** Confluent murine MC3T3-E1 preosteoblastic cells were induced to differentiate with osteogenic medium (α MEM supplemented with 10% FBS, 50 μ g/ml ascorbic acid and 10 mM β -glycerophosphate) in 1 g condition for 7 days, when cells entering mineralizing stage, and then were set as two groups: 1 g condition (Control) and simulated microgravity condition (SM). Cells were treated for 24 h. The mineralized nodules formation was detected by alizarin red s staining. The alkaline phosphatase (ALP) activity was measured using pNP colorimetric assay. The mRNA expression of osteogenic genes including osteocalcin (OC), type I collagen α 1(Col I α 1) and dentin matrix 1 (DMP1) was determined by real time RT-PCR. **Results:** The results showed that 24 h treatment of simulated microgravity (SM) significantly inhibited the mineralized nodules formation compared with 1 g condition (Control). In addition, the alkaline phosphatase (ALP) activity was significantly decreased. Consistent with above results, real time RT-PCR detection revealed that the expression of OC, Col I α 1 and DMP1 was all down-regulated by 24 h treatment of simulated microgravity. **Conclusions:** These findings suggested that 24 h treatment of simulated microgravity using random positioning machine showed inhibitory effect on the mineralization of mineralizing osteoblasts, and the inhibitory effect may be through regulating the ALP activity and the osteogenic genes' expression.