

## **PR-I-1. In vitro biocompatibility of a super-fine grain pure titanium with osteoblast-like cells**

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### **Background**

Recent studies have been focused on the biocompatibility of biomaterials with ultra-fine surface features, their nanostructure improved cell-material interaction. Equal channel angular processing (ECAP) produces ultra-fine grain pure titanium with enhanced strength comparable to that of Ti6Al4V (Ti64) alloy. In this study, we investigated the in vitro biocompatibility of nanostructured titanium surface produced by ECAP, and compared with conventional coarse-grain pure titanium and Ti64 surfaces for future biomedical application.

### **Materials and Methods**

ECAP was used to produce ultra-fine grain titanium (ECAP) from coarse-grain titanium (Ti-1) rods. Ti-1, conventional coarse-grain c.p. titanium (ASTM grade 3; Ti-2), and Ti64 alloy (Ti64) surfaces were used as control in this study. Experimental and control surfaces were roughened by hydroxyapatite blasting. Osteoblast response to four different surfaces were investigated using MC3T3-E1 pre-osteoblasts. Cell attachment and proliferation were evaluated by mitochondrial dehydrogenase activity using a highly water soluble tetrazolium salt (WST-8). To determine whether nanostructured ECAP surface has influence on the differentiation of osteoblast cells, ALP activity and matrix mineralization assay were performed. The level of cytokine production in the culture supernatant was determined using an ELISA kit.

### **Results**

The number of attached cells on the surface of ECAP was significantly higher than those on the control surfaces ( $P < 0.01$ ). After 4 days of culture, ECAP surface showed significantly greater number of proliferated cells compared to control surfa-

ces ( $P < 0.01$ ). After 5 days of culture, cells grown on ECAP surface showed significantly increased ALP activity compared to the control surfaces ( $P < 0.05$ ). Consistent with the result of ALP assay, cell cultures on ECAP surface also showed increased number of nodules as indicated by the intense Alizarin Red S staining. The levels of  $\text{TNF-}\alpha$  and  $\text{IL-1}\beta$  produced by MC3T3-E1 cells cultured on titanium surfaces were slightly increased compared to those of tissue culture plates. But, the  $\text{TNF-}\alpha$  and  $\text{IL-1}\beta$  levels on ECAP surfaces were not significantly different from the control surfaces.

### Conclusion

These results suggest that ultra-fine grain pure titanium implants produced by ECAP may have advantages in the osseointegration over the conventional coarse-grain pure titanium and Ti64 alloy implants because of their improved osteoblast response and mechanical properties.