Response of osteoblast-like cells to poling titania coating

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Introduction:

Over several decades titanium (Ti) and its alloy have been used as medical implants due to their strength and good biocompatibility. Various surface modifications of the metal to form a bioactive titanium dioxide (TiO_2) layer have been applied to enhance bone bonding. We have reported the electrical polarization of hydroxyapatite (HAp) coating, and its stored electric charges and electric fields promote bone bonding. Although synthetic HAp coating is widely used coating process of calcium phosphates in orthopedic and dental devices, its weak adhesion strength to the substrate limits its use. Micro-arc-oxidation (MAO) is one of the surface modifications and MAO coating are uniformly porous and exhibit good adhesion to substrate but have no apatite-forming ability. To improve the bioactivity of the MAO formed TiO2 coating, electrical polarization treatment as subsequent activation method was tried.

In this study, we will investigate the possibility of electrical polarization for MAO-TiO_2 coating and how surface characteristic including the surface chemistry after the treatment modulates osteoblast-like cell function.

Materials and Methods:

Titanium discs (Φ10mm×2mm) were fabricated with commercially pure titanium (grade-2) and were ground using $320^{\#}$ and $600^{\#}$ waterproof abrasive paper. The titanium discs were subjected to MAO treatment in an aqueous electrolyte at 400V for 5 min by applying a DC field to the samples. The samples with MAO-TiO_2 coating were sandwiched between a pair of platinum (Pt) plate electrodes, electrically polarized in a DC field of $5.0kV\cdot cm^{-1}$ in air at 400°C for 1 hours, then cooled to room temperature under the applied DC voltage. The TiO_2 coating placed in contact with the anode was named negative surface (N-surface). The TiO_2 coating placed in contact with the cathode was named positive surface (P-surface). The un-polarized coating was used as control and named 0-surface. Thermally stimulated depolarization currents (TSDC) were measured for the polarized samples and calculated stored charges. The phases of the MAO-treated and polarized samples were analyzed by X-ray diffraction (XRD) and the surface morphology of the specimens was observed using scanning electron microscopy (SEM). The surface chemistry was analyzed by X-ray photoelectron spectroscopy (XPS). Cell-materials interaction was studied using osteoblast-like MG63 cells to evaluate the influence of the surfaces on attachment, proliferation and differentiation of the cells.

Results:

The MAO coating exhibits a porous surface with pores about 2~4 μm in diameter distributed at regular intervals. MAO-TiO_2 coatings are composed of both rutile and anatase type. It was confirmed that MAO-TiO_2 coating can be polarized at the experimental condition using TSDC measurements. Electrical polarization treatment significantly reduced the contact angle of water droplets although the surface roughness did not alter (Fig. 1).

Cell attachment was determined by counting the number of adherent cells. The number of adherent cells was higher on both N- and P-surfaces than 0-surfaces. Cells on N-surface were well-spreaded compared to other surfaces. Cell proliferation analysis was performed using MTT assay. The result of MTT assay indicated that cell proliferation on the N-surface can be significantly enhanced. The alkaline phosphatase (ALP) activity is an early marker of osteoblast differentiation. The ALP positive area on N-surface was larger than other surfaces.

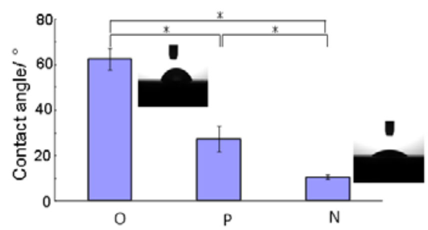

Figure 1. Change in hydrophilic status MAO-TiO2 coatings with and without polarization. Data are mean ± SD (n=4).*$p<0.05$, indicating a statistically significant difference between them.

Conclusions:

MAO-TiO_2 coating can be polarized and stored charges. The electrical polarization treatment resulted in change of surface chemical composition and surface wettability, though polarized MAO-TiO_2 coatings did not exhibit any obvious change in morphology and phase component with SEM and XRD observations. Activities of osteoblast-like cells were accelerated on the polarized MAO-TiO_2 coatings, especially N-surface.