Effect of Surface Characteristics of Metallic Biomaterials on Interaction with Osteoblast Cells

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Objectives: The objectives of this study were to characterize the roughness, surface potential, surface free energy, electron-donor and electron-acceptor surface parameters, and composition of the oxide film formed on stainless steel and titanium-aluminium-vanadium metal alloys subjected to different surface treatments, and to investigate how these properties influence the attachment and growth of osteoblast cells. A more comprehensive understanding of such relationships can lead to better biomaterial and surface treatment selection, development of new material/surface preparation combinations and better understanding regarding of which surface properties elicit the desired osteoblast cell responses to increase healing rate of bone implants and reduce pain for patients.

Methods: The interaction of MC3T3-E1 osteoblastic cells with Ti-6Al-4V and 316L stainless steel (SS) subjected to different surface treatments including anodization, electropolishing, and passivation was studied. The resulting cell proliferation, morphology, and differentiation were examined with optical microscopy, scanning electron microscopy, and kinetic ELISAs for alkaline phosphatase (ALP) activity. Surface roughness characteristics were determined using white light profilometry and atomic force microscopy. The chemical composition of the oxide layers were analysed by Auger electron spectroscopy. The surface free energy and electron-donor and electron-acceptor parameters of materials were determined through contact angle measurements and using the Lifshitz-van der Waals Lewis acid-base interaction model.[1]

Results: Surface free energies of biomaterials ranged from 35 to 56 mJ/m^2 and influenced ALP activity. Materials with a higher surface energy produced increased differentiation of osteoblast cells when compared to substrates with a lower surface energy. Also, the electron-acceptor component of surface free energy was found to have a strong correlation to ALP activity. Surfaces with more electron-acceptor sites encouraged osteoblastic differentiation (Fig.1).

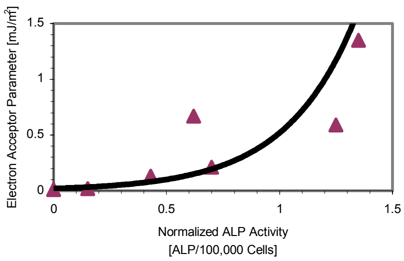

Figure 1. The effect of electron-acceptor parameter on ALP activity.

Both effects of the surface energy and electron acceptor parameter of the surface free energy on osteoblast differentiation suggest that protonation of the oxide surface is mainly responsible for an increase in the electron-acceptor properties of the oxide surfaces and increase osteoblastic differentiation. This hypothesis together with the hypothesis that the electron-donor sites on the osteoblast cell surfaces activate specific interactions with the electron-acceptor sites on the substrate surfaces may suggest that the density and distribution of hydroxide groups on the oxide surfaces is important in designing implant materials with enhanced biointeraction to bone cells. If cells are affected by the presence of hydroxides on the surface, then developing an understanding of the mechanisms that control this interaction could lead to the optimization of this parameter in current and future metallic biomaterials.

Investigating how chemistry affects the presence of electron-acceptor sites on the surface would aid in designing optimized surface treatments. For example the increased chromium content at the surface of the SS oxide layer from 2.1 to 5.5 at% correlated with increased osteoblast differentiation. Although a precise explanation for this correlation is unknown, it is most likely due to differences in surface hydroxylation and/or surface potential resulting from the differences in surface chemistry.

It was also found that surfaces with nano-scale roughness rather than micro-scale roughness had a more profound impact on the growth of osteoblasts. Surfaces with nanoroughness at a level of R_a = 7-10 nm were found to encourage cell differentiation (Fig.2).

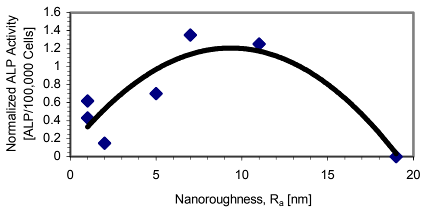

Figure 2. The effect of surface nanoroughness on ALP activity

The distribution and orientation of micro-scale surface features, the peaks and valleys that constitute surface topography were found to have an impact on cell morphology and differentiation. Random surface topographies were found to produce more randomly spaced teardrop-shaped cells with fibrils (which aid in adhesion) covering the entire cell. Oriented surface topographies produced cell morphologies that were long, linear, and oriented in parallel, with fibrils located only at the ends of the cells.

Conclusions: Osteoblast cell attachment, differentiation and morphology on oxide surfaces of Ti-6Al-4V and 316L stainless steel can be manipulated by a biomaterial's nanoroughness and its characteristics and Lewis acid-base properties. The results suggest that the density and distribution of hydroxide groups on the oxide surfaces is important in design implant materials with enhanced biointeraction to bone cells.

Reference: 1) C.J. van Oss, *Interfacial Forces in Aqueous Media*, Marcel Dekker, New York 1994.