Characterization of a silicon-based biomimetic treatment to improve the osteointegration of titanium alloy (Ti6Al4V)

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Introduction

One of the primary goals of biomaterials research for many dental and orthopedic applications is the direct osteointegration of titanium and its alloy through the modification of titanium oxide layer by mimicking hydroxyapatite mineralization and osteoconductive properties.

The aim of the present work is to investigate the physical-chemical and mechanical properties of new biomimetic treatments on titanium alloy (Ti6Al4V) and to assess their *in vitro* biological properties. The biomimetic treatments are obtained by Anodic Spark Deposition technique (ASD), which allows to develop a thin and porous surface layer enriched with selected ions from the electrochemical solution. The developed biomimetic treatment was compared to another ASD surface (SiB-Na) [1] which was recently developed and used in clinical practice with excellent results.

Materials and Methods

Five surfaces were prepared using commercially grade V titanium alloy and pure grade II titanium samples (10 mm diam., 0.5 mm thick.):

i) **SiB-Na** : Ca-P-Si-Na coated titanium grade II by ASD treatment [1] used ad control;

i) **Ti grade V**: machined titanium used as control;

ii)**VB-NaOH**: Ca-P-Si-Na coated titanium alloy by ASD treatment [1] (current density (I) = 40mA/cm2; Voltage (V) = 250);

iv)**VC-NaOH**: Ca-P-Si-Na coated titanium alloy by ASD treatments [1] (I = 40mA/cm2; V= 295);

v)**SVB-NaOH**: Ca-P-Si-Na coated titanium alloy by ASD treatment [1] (I= 40mA/cm2; V= 250), previously sandblasted using Al_2O_3 (mesh 36) and subjected to an acid sandblasting decontamination.

All the ASD samples obtained were finally treated in 5M NaOH solution at 60±1°C for 2 hours.

Surface properties were evaluated with scanning electron microscopy (SEM), electron dispersion spectroscopy (EDS), thin film x-ray diffraction (TF-XRD), laser profilometry (LP) and inductively coupled plasma optical emission spectrometry (ICP) . Optical contact angle measurements (OCA), in static conditions, were used to investigate the surfaces wettability. The potentiodynamic tests were performed in Ringer solution at 37°. Potentiodynamic current density-potential curves were determined in a potential range between -1000mV and +1000mV with a potential scan rate of 0.5 mVs^{-1}. MG63 (ECACC 86051601, human, osteosarcoma) cell line was cultured in EMEM. A suspension of 7×10^3 cells were seeded on the specimens previously sterilized in ethanol absolute followed by UV irradiation (254nm).

One, 3 and 7 days after cell seeding the direct cytotoxicity was assessed with Alamar Blue Assay. Viability was also confirmed by images obtained after staining of nuclei (using Hoechs) and F-actin filaments (using FITC fluorescent Falloidina).

Results and Discussion

The results show that biomimetic treatments on titanium alloy are characterized by a microporous surface (Fig. 1) enriched with silicon, calcium, phosphorus and sodium elements; in all treatments all the alkali etching treatment was found to enhance calcium/phosphorus ratio as in the SiB-Na control.

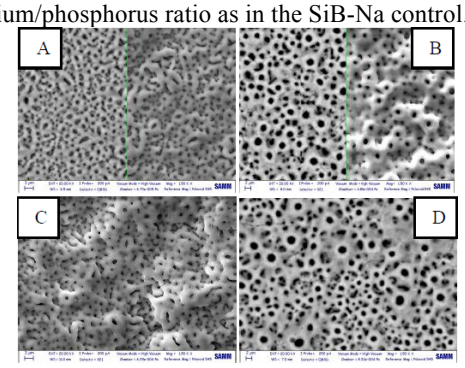

Fig. 1. SEM pictures of VBNaOH (A), VCNaOH (B), SVBNaOH (C), SiB-NaOH (D)

XRD analysis showed the presence of TiO_2 with anatase structure (2θ=25°) for all ASD treated materials. The biomimetic titanium alloy surfaces, in particular VB-NaOH, was found to possess an excellent hydrophilicity compared to machined titanium Ti grade V, showing a contact angle as low as 20° (Fig.2).

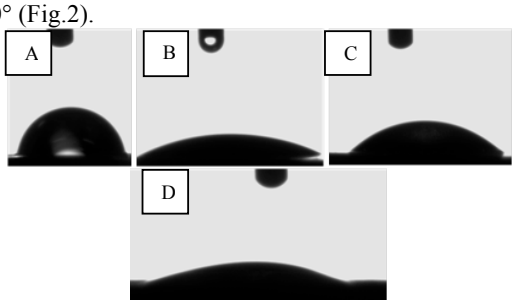

Fig. 2. Optical contact Angle measurements (OCA)Shape taken by the drop deposited on the surface of TiV (A), VBNaOH (B), VCNaOH (C) and SVBNaOH (D)

By potentiodynamic analysis it was possible to detect a decrease in passivation current of samples VBNaOH, VCNaOH, SVBNaOH, compared with control V. Moreover, this current is constant in the range of biological potential, confirming a good corrosion behavior. It was not found any cytotoxic effects on the the ASD coatings witch showed similar levels of cellular activity for at each time point Viability was also confirmed by images obtained after staining of nuclei and F-actin filaments.

Conclusions

Considering the results obtained in the present study the ASD titanium alloy treatments can be considered promising treatments as osteointegrative implantology materials

References [1] WO 2010/013120 A1 Silicon-based biomimetic treatmentfor the osteointegration of metal substrates.