FORMATION OF APATITE LAYER ON Ti-7.5Mo ALLOY WITH DIFFERENT ROUGHNESS

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Abstract. *Titanium and its alloys have been used in dentistry due their corrosion resistance and biocompatibility. These metals are covered with a thin oxide layer formed spontaneously in their surface when contact with the air. However, titanium coating is bioinert and it cannot bond chemically to bone tissue. The purpose of this work was evaluated bioactivity of Ti-7.5Mo alloy after processing and chemical treatment surface. Ingots were obtained from titanium and molybdenum by using an arc-melting furnace. They were submitted to heat treatment at 1100ºC for one hour, cooled in water, cold worked by swaging and turning on a CNC lathe in order to achieve microstructure and morphology next to dental implants. Samples were divided in two groups according to the roughness media (Ra): Group I (1.3µm) and Group II (2.6µm). Discs with 10 mm in diameter and 4 mm in thickness were cut for each one. They were ultrasonically cleaned with distilled water, acetone and air dried prior being treatment surface. Samples were immersed in NaOH aqueous solution with 5.0M at 80ºC for 3 days, washed with distilled water and dried at 40ºC for 24h. Then, Ti-7.5Mo discs were immersed in SBF (Simulated Body Fluid) for 7 days to form an apatite layer on the surface. The surfaces were characterized by scanning electron microscope. Contact angle measurements were carried out in order to evaluate the wettability of surface alloy. Analysis indicates that the surface was covered with a thin oxide layer after NaOH treatment. Results showed that a bioactive Ti-7.5Mo alloy can be obtained by chemical treatment.*

Keywords*: titanium alloys, surface roughness, apatite.*

1. INTRODUCTION

Titanium and titanium alloys have been used for dental implants due to their characteristics, such as mechanical resistance, biocompatibility, non-toxicity, early osseointegration, corrosion resistance ,among many others (Le Guehennec *et al*, 2007).

They are considered bioinerts and when these materials are inserted into the human body, they are generally encapsulated by fibrous tissue and therefore cannot form a chemical bond with bone. As a result of this limitation, several studies have been undertaken to improve the bioactivity of these materials (Kokubo, 1998).

The bone-implant fixation is influenced by two main mechanisms: mechanical and chemical. The chemical composition is responsible for the interaction of the implant with proteins, biological fluids, cells and tissues dependents on surface wettability (Huang *et al,* 2004). The roughness improves the mechanical properties and surface characteristics of the implant, because it allows bone growth through pores. The roughness can also modify the characteristics such as surface energy, changing the character of a surface hydrophobic or hydrophilic, therefore roughness and chemical composition surface influence the success of the implant (Bagno and Bello, 2004).

Vanzillotta *et al*. (2006) used various surface treatment techniques to modify titanium implants surfaces to become bioactive. Bioglass, glass-ceramics, bioceramics such as hydroxyapatite and other calcium phosphates can be considered bioactive materials, because these materials accelerates the osseointegration. However, bioglass and bioceramics coatings suffer degradation in body environments (Cooper, 2000).

Kokubo (1998) showed that a bioactive titanium surface can be obtained via an alkaline treatment in NaOH followed by immersion in SBF (Simulated Body Fluid). Barrère *et al.* (2002) studied several compositions of Simulated Body Fluid (SBF) to assess the influence of $NaCl^-$ and HCO_3^- on the process of calcium phosphate (Ca-P) mineralization on a Ti alloy (Ti-6Al-4V). Ca-P coatings formed at the same time that Ca-P precipitated for solutions of SBF x 5, SBF x 5 (HCO_3^- x 0), SBF x 5 (HCO_3^- x 3) and SBF x 5 ($NaCl^-$ x 3). The layer of Ca-P was uniform and dense, and it comprised elongated crystals. For solutions of SBF x 5 ($NaCl^-$ x 0), due to the low ionic strength, the Ca-P coating formed on the Ti-6Al-4V substrate after precipitation from the solution. Under these conditions, the Ca-P layer exhibited smaller crystals, and the substrate was not fully coated.

Differents surface treatment methods were reported in the literature to modify surfaces of titanium implants, such as the machining, chemical and electrochemical treatments, and plasma coatings (Ogawa *et al,* 2000). Therefore, the selection of a method or a combination of different methods is one of the important factors to obtain the desired surface

Wong *et al*. (1995) investigated the influence of surface roughness in bone implants and concluded that surfaces with higher roughness resulted in a better tissue response to titanium implants. According to Citeau *et al.* (2005) surface roughness is one of the essential factors for osseointegration of dental and orthopedic implants. It improves the mechanical anchoring of calcium phosphate coating that acts as a support to colonization and adhesion of osteogenic cells. Ronold and Ellingsen. (2002) demonstrated in previous studies that primary bone anchorage of titanium implants was improved by surface roughness with Ra ranging from 0.5 to 1.5 μm. Ronold *et al.* (2003) concluded in their studies that roughness values greater than 3.6 μm decreased the adhesion of bone cells, contra-indicating high values of roughness.

In the present work, we evaluate the formation of bone-like apatite layer on Ti-7.5Mo alloy with different roughness, after alkaline treatment and soaking in SBF.

2. MATERIALS AND METHODS

2.1 Materials

The Ti-7.5Mo alloy was produced from sheets of commercially pure titanium (99.9%) and molybdenum (99.9%). Melting was realized in arc melting furnace in an argon atmosphere (Figure 1). The ingots were then homogenized under vacuum at 1100ºC to eliminate chemical segregation. The resulting samples were finally cold-worked by swaging, producing a 13 mm rod.

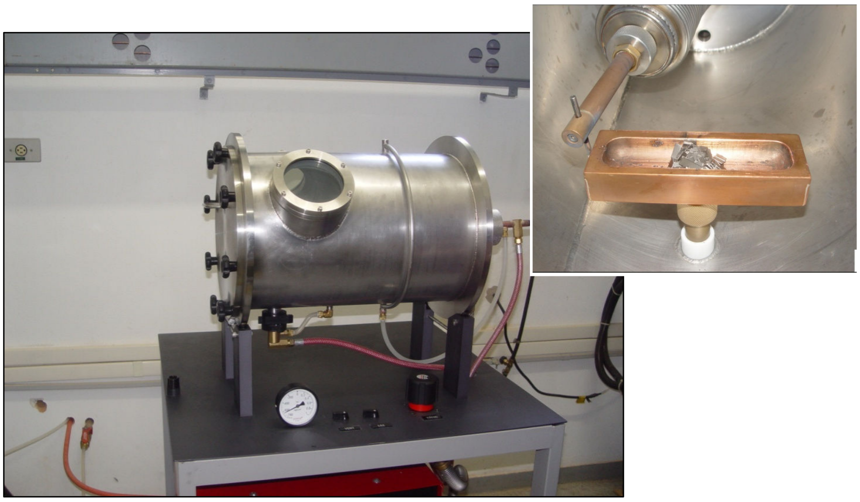


Figure 1 – Arc melting furnace

Bars of this alloy were machined using a CNC lathe ZIL(CENTUR 30S, ROMY, Brazil) with a rotation speed of 1000 rpm to obtain grooved surfaces. Samples were prepared by cutting out discs (10 mm in diameter and 4 mm in thickness). Media roughness (Ra) was measured with a roughness meter (1.3 and 2.6 µm). Samples were then divided in two groups according to roughness: Group I (1.3µm) and Group II (2.6µm). These samples were ultrasonically cleaned with distilled water and acetone and air-dried prior to surface treatment. Machined samples were used as control group and were not subjected to further surface treatment.

For alkaline surface treatment, samples were immersed in a 5.0M NaOH aqueous solution at 80ºC for 3 days, washed with distilled water, and dried at 40ºC for 24 h using a methodology proposed by Wei *et al*. (2002).

2.2 Biomimetic coating

Ti-7.5Mo discs were immersed in SBF (Simulated Body Fluid) for 7 days to form an apatite layer on the sample surface. The SBF solution composition proposed by Barrère *et al*. (2002) was used in this study. SBF with an ionic concentration approximately 5.0 times greater than blood plasma was prepared by dissolving reagent grade NaCl, $MgCl_2.6H_2O$, $CaCl_2.2H_2O$, Na_2HPO_4, and $NaHCO_3$ in distilled water and buffering to pH 7.4 at 36.5°C. The solution was refreshed every second day.

The sample surfaces (after NaOH treatments and soaking in SBF) were examined using a scanning electron microscope (SEM, LEO 1450 VP, Zeiss, Germany). The hydrophilicity of the surfaces was evaluated by contact angle analysis using the sessile drop method.

3. RESULTS AND DISCUSSION

Figure 2 shows scanning electron micrographs of the surfaces of samples from group I (Ra=1.3 μm) and group II (Ra=2.6 μm), respectively.

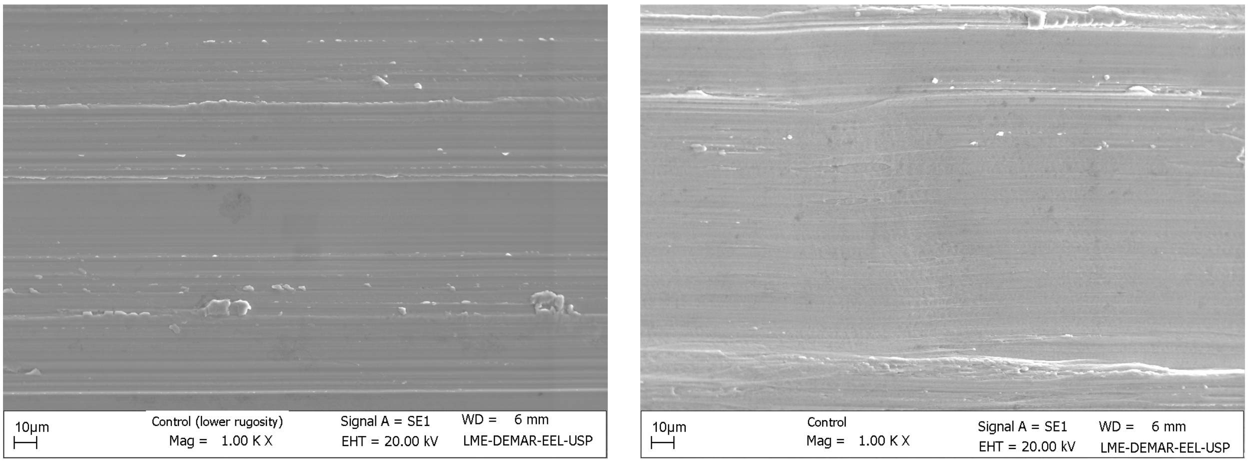


Figure 2 - Surfaces of samples with roughness media (Ra): (a) 1.3 μm; (b) 2.6 μm

Figure 3 shows scanning electron micrographs of the surfaces of samples after alkaline treatment for both roughness (1.3 e 2.6 μm).

After alkaline treatment, a porous layer formed on the surface was observed. This porous film formed on the surface was similar to verified by Jalota *et al.* (2006) in their studies. Some cracks originated on the surfaces of the samples with lower and higher roughness, were probably formed during drying after the alkaline treatment.

Miyazaki *et al*. (1999), evaluating the bioactivity of the tantalum, also noted the presence of cracks after NaOH treatment. They reported that the number and depth of the cracks increased with increasing concentrations of the NaOH for pure tantalum, 0.2, 0.5 and 5 M, respectively.

Although the results reported by these authors were bad for solutions to 5 M, Wei *et a*l. (2002) found that for the Ti - 6AL - 4V this concentration was that gave a more porous film with any cracks.

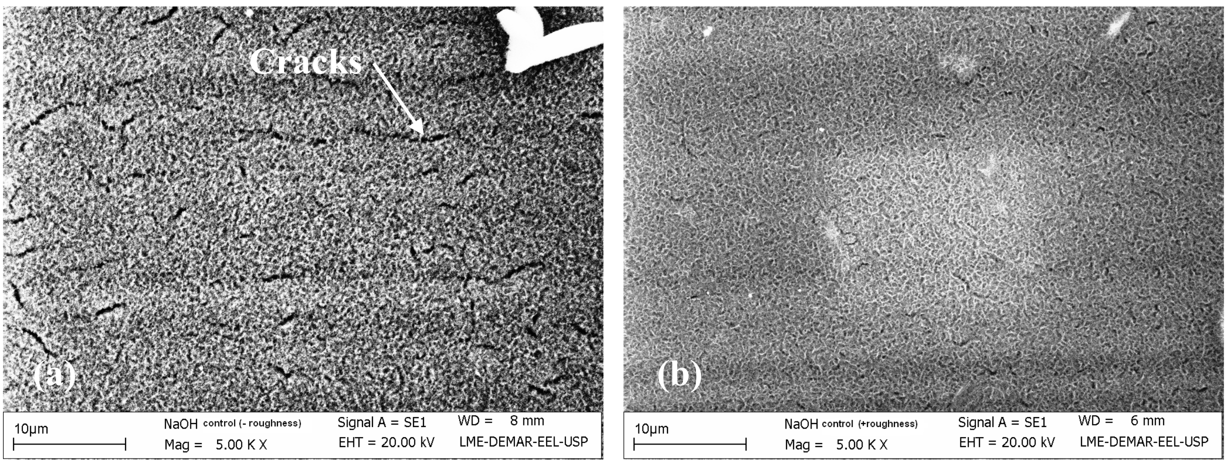
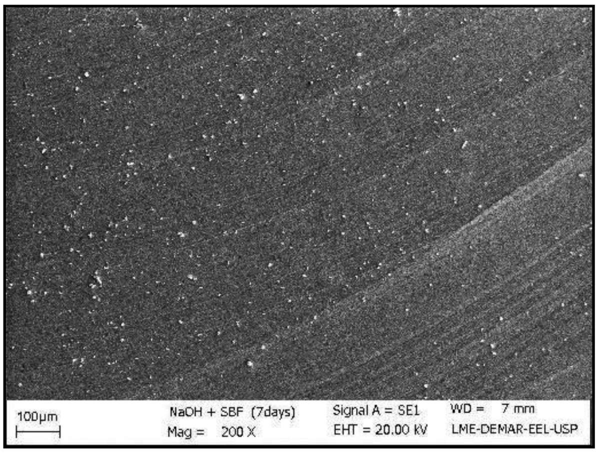


Figure 3 - Surfaces of samples after alkaline treatment with roughness media (Ra): (a) 1.3 μm; (b) 2.6 μm

After the alkaline treatment and SBF soaking for 7 days we observed the growth of calcium phosphate on the film. For this period, it was observed an increase in clusters, with higher nucleation of calcium phosphate in the samples with roughness media Ra = 2.6 μm. Figure 4 shows the scanning electron micrographs of samples surfaces for both values of roughness evaluated.



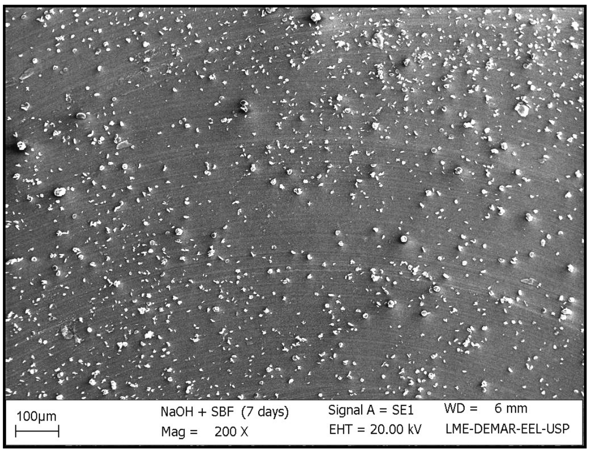


Figure 4 – Growth of calcium phosphate on surfaces of samples after immersion in SBF for 7 days: (a) 1.3 µm ; (b) 2.6 µm

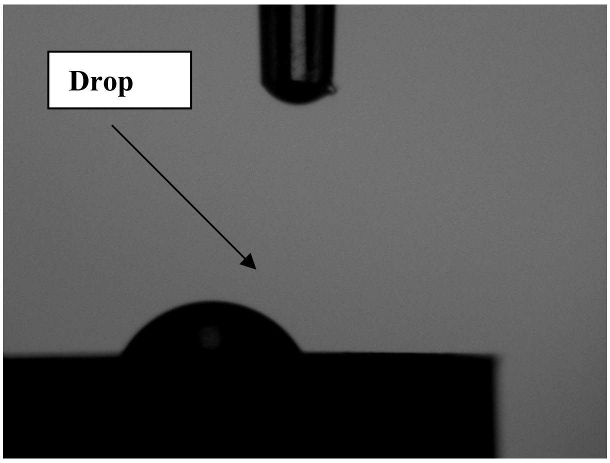
According to Feng *et al*. (1999), the desired composition and thickness of the coatings can be controlled by adjusting the conditions of NaOH-treatment and immersion. An increase of the NaOH concentration, NaOH-treatment time and NaOH solution temperature is favorable for Ca-P crystals to grow on Ti. Chen *et al.* (2003) reported that a microporous surface favors the formation of nuclei of apatite and improves the adhesion between the porous coating and the substrate, and the authors observed the presence of pores with diameter between 1-2 µm. According Citeau *et al.* (2005) the surface roughness is one of the factors essential to osseointegration of dental implants and orthopedic. It increases the mechanical anchor of calcium phosphate that acts as a support for the colonization and adhesion of osteogenic cells. Studies have reported that the primary anchor is increased by a surface roughness from 0.5 to 1.5 µm. Ronold et al. (2003) concluded in their studies that roughness values greater than 3.6 µm led to a decrease in adhesion of bone cells, contra-indicating high values of roughness.

The results of contact angle for the groups evaluated are shown in Table 1.

Table 1 - Measures of contact angle

Samples	Contact Angle	
	Roughness Ra = 1,3	Roughness Ra=2,6
Control (machined)	58,90 ± 0,45	43,20 ± 0,98
NaOH	43,62 ± 0,97	28,86 ± 0,41
NaOH + SBF (7 days)	33,47 ± 2,12	18,96 ± 0,21

Figure 5 shows image of the water drop on the control sample of Ti-7.5Mo and image of the water drop after growth of calcium phosphate on surfaces, showing coherence with the contact angle measures that shown in table 1.



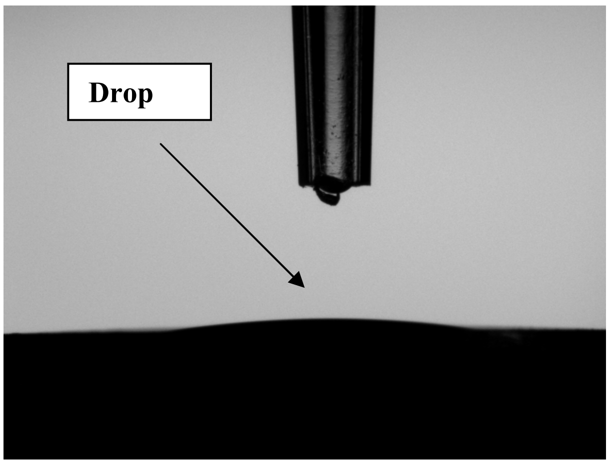


Figure 5 - Image of the water drop on the sample of Ti-7.5Mo : (a) control; (b) after growth of calcium phosphate on surfaces

The relationship between contact angle and wetting occurs proportional inversely in the same surface. Therefore, the diminution of this angle increases the capacity of the surface wettability (Lim and Donahue, 2004). Contact angle measurements give values ranging from 0◦ (hydrophilic) to 140◦ (hydrophobic) for titanium implant surfaces (Bagno and Bello, 2004). The surface chemical composition of titanium implants also affects the hydrophilicity of the surface. Highly hydrophilic surfaces seem more desirable than hydrophobic ones in view of their interactions with biological fluids, cells and tissues (Zhao *et al*, 2005)

In a recent animal study, Buser *et al.* (2004) found that a hydrophilic surface gave higher bone-to-implant contact than regular surface. Eriksson et al. (2004) demonstrated an early marker for bone formation and differentiation, on hydrophilic Ti discs than on hydrophobic discs after 1 week of implantation in rat tibiae. Furthermore, hydrophilic surfaces stimulate the biomineralization process. Rapid calcium phosphate nucleation was achieved on highly wettable Ti surfaces produced by glow discharge plasma treatment (Shibata and Miyasaki. 2002).

For both roughnesses, we observed that bioinert surfaces (machined) have contact angle values greater than bioactive surfaces (NaOH + SBF), so the bioactive surfaces have higher hydrophilicity. This result is in agreement with the studies of Chen *et al.* (2008) that attribute the increased hydrophilicity of the apatite biomimetics.

4. CONCLUSIONS

The results obtained using the biomimetic method in samples with different roughness provided a coating with a calcium phosphate uniform layer and homogeneous (1.3 and 2.6 μm). However, for samples with roughness media Ra = 2.6 μm, it was an increase in clusters, with higher nucleation of calcium phosphate. This greater macroroughness accelerated the formation of calcium phosphate and turned possible a smaller contact angle. Surfaces with small contact angle has greater hydrophilicity, which is important for the process of osseointegration. However, it was concluded that for samples with roughness media Ra = 2.6 μm, this roughness can be considered ideal for implants.

5. ACKNOWLEDGEMENTS

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