

## **Apatite Coating on Titanium Samples Obtained by Powder Metallurgy**

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**Abstract.** Titanium and its alloys are widely used as orthopedic and dental implant materials. However, they cannot bond with bone directly and promote new bone formation. It is desirable to provide a bone-bonding ability to Ti metal and its alloys. This ability can be achieved by surface modification such as chemical treatments. The aim of this study was to evaluate in SBF the apatite-forming ability of Ti subjected to different pre-treatments. Titanium laminated and samples Ti obtained by powder metallurgy were compared. The pretreatments studied were the alkali-treated; alkali and heat-treated; acid and alkali-treated; alkali-CaCl<sub>2</sub>-heat and hot water treatment. The groups were soaking in SBF for 1, 3, 6 and 9 days in equipment with constant agitation at 36.5°C. The obtained coatings were analyzed by diffuse reflectance spectroscopy on the infrared (DRIFT) and scanning electron microscopy (SEM). The apatite formation was present in all groups; however, the apatite-coating was more effective in samples obtained by powder metallurgy.

### **Introduction**

Commercially pure titanium and its alloys are major implant materials for dental and medical applications. It is well known that titanium shows good properties such as a high strength-to-weight, good corrosion resistance in physiological environment, fatigue resistance and low elastic modulus. However, metallic materials are not able to bond with bone [1, 2]. For this reason, investigations with the purpose of modifying its surface in order to turn it bioactive have been frequently reported in the literature [1-3]. The surface modifications of bioinert materials can be divided into physical, chemical and electro-chemical methods. Plasma spraying, ion sputtering, ion implanting are examples of physical methods. Electro-chemical methods can be electrocrystallization, electrophoretic deposition and anode oxidation. Chemical methods include acid-etching, oxidation with hydrogen peroxide, alkali-heating or acid-alkali procedure, etc. [4]. After chemical and thermal treatments, titanium and their alloys can become bioactive, using the technique of biomimetic coating.

The biomimetic route uses supersaturated aqueous solutions with ionic composition similar to that of human plasma (SBF), it allows to coat and co-precipitate apatite crystals onto metal producing calcium phosphate layer [5,6]. The alkali treatment with NaOH solution is the most commonly used surface conditioning, however, several studies have shown better apatite-forming ability when performed heat treatment [7, 8]. During the alkali treatment, the TiO<sub>2</sub> layer presents on surface titanium partially dissolves in the alkaline solution because of the attack by hydroxyl groups. A further hydroxyl attack on the hydrated TiO<sub>2</sub> produces negatively charged hydrates on the surfaces of the substrates. These negatively charged species combine with the alkali ions in the aqueous solution to produce an alkaline titanate hydrogel layer. During heat treatment, the hydrogel layer is dehydrated and densifies to form a stable amorphous or crystalline alkali titanate layer [9]. The Ti-OH groups formed on the surface of sodium titanate after soaking in SBF are negatively charged and hence, combine selectively with the positively charged Ca<sup>2+</sup> ions in the fluid to form calcium titanate. As the calcium ions accumulate on the surface, the surface gradually gains an overall positive charge. As a result, the positively charged surface combines with negatively charged phosphate ions to form amorphous calcium phosphate. The calcium phosphate spontaneously transforms into apatite in the body environment because apatite is the stable phase [10].

The acid-alkali procedure can also be used as a treatment prior to immersion in SBF. The pre-treatment with highly concentrated solution of HCl and temperature (in range 40°-50°C) increases corrosion rates. As a result, the initial passive oxide film was removed and a lot of acid-etched pits with a new oxide layer were formed to increase the surface area [11]. The second step with alkali treatment might have two concurrent effects: that was the formation of a microporous surface layer on the acid-etched surface and an amorphous sodium titanate layer on this microporous layer.

A recently pretreatment assessed was reported by Kizuki et al.[12]. They demonstrated that a fairly large number of calcium are easily incorporated into surface of titanium when it is first a NaOH solution and then soaked in a CaCl<sub>2</sub> solution. This is because the sodium hydrogen titanate that formed on the Ti metal surface during the first NaOH treatment has a layered structure and the Na<sup>+</sup> ions in it are easily replaced with Ca<sup>2+</sup> ions by CaCl<sub>2</sub> treatment. Subsequent heat treatment, there was a low mobility of the Ca<sup>2+</sup> in the calcium titanate transformed from the calcium hydrogen titanate. The apatite-forming ability of heat-treated titanium increased remarkably when it was subsequently treated with hot water.

There are several studies on the biomimetic coating applied to smooth titanium surfaces, however, few studies about this coating technique applied to titanium substrates obtained by powder metallurgy with roughness characteristic of the technique. The aim of this study was to evaluate and compared in SBF the apatite-forming ability of Ti laminate and obtained by P/M, subjected to different pre-treatments.

## Materials and Method

### Sample preparation

Commercially pure laminate titanium plates (NAS titânio) (0,5x0,5x1,5mm) and Ti cylindrical (5,5 mm diameter x 4 length) samples obtained by powder metallurgy were used in this study. The plates received sanding with silicon carbide sandpaper (n#600) to regularize the surface. For cylindrical samples, titanium hydride powder with particle medium size 92µm was encapsulated under vacuum in flexible rubber molds and cold isostatically pressed at 200 MPa during 60s. The bodies were sintered in high vacuum condition at 1400° C/1h with heating rates of 10°C/min. The densities and roughness of laminate and P/M samples were measured.

### Pre-treatments

All samples were washed in acetone and ultrapure water (MilliQ, Millipore) with an ultrasonic cleaner. The first pre-treatment (Alkali-treated) consisted in soaked the samples a NaOH (5M) solution at 60°C for 24h, and washed with ultrapure water. The second pre-treatment (Alkali-heat-treated) received alkali-treated and subsequent heated to 600°C at a rate of 5°C/min for 1h in an air atmosphere. The third pre-treatment (Acid-alkali-treated) consisted in acid etching (HCl-37%) for 40 min at 50°C and subsequent alkali-treated. The last pre-treatment (Alkali-CaCl<sub>2</sub>-heat-hot water-treated) used was alkali-treated and soaked of 100mM CaCl<sub>2</sub> solution at 40° C for 24h, and then gently washed with ultrapure water. After the samples were received heat treatments and subsequently subjected to hot water at 60°C for 24h. All samples were washed with ultrapure water, and then dried at 40°C.

### Biomimetic coating

The SBF solution used was prepared by dissolution in ultrapure water of the following analytical grade reagents: NaCl, KCl, K<sub>2</sub>HPO<sub>4</sub>, CaCl<sub>2</sub>.2H<sub>2</sub>O, MgCl<sub>2</sub>.6H<sub>2</sub>O, NaHCO<sub>3</sub>, Na<sub>2</sub>SO<sub>4</sub>. The pH was adjusted with HCl 0.1M and tris (hidroximetilaminomethane) at 7.25 (36.5°C) [13]. The solution was kept in closed polyethylene containers.

All the samples were placed in polyethylene recipient, immersed in SBF for 1, 3, 6 and 9 days in equipment with constant agitation at 36.5°C. The solution was renewed every three days. Finally, the samples were washed again in ultrapure water and dried at 40°C.

The coated specimens were analyzed by diffuse reflectance FTIR spectroscopy (Thermo Nicolet 870-NEXUS) and scanning electron microscopy (SEM- Philips XL-30). The cross sections of samples were made for evaluate the layer thickness.

## Results and Discussion

The morphological appearance of laminated plates and cylindrical samples obtained by P/M can be assessed by SEM (Fig. 1). The difference in surface is clearly identified. It's known that higher surface roughness induces a better osseointegration due to the higher specific surface in contact with bone [14]. The roughness (Ra pattern) and hydrostatic density of commercial plates and cylindrical samples of titanium were measured. The density of commercial plate Ti was around 98% of the estimated density and 0.92µm (Ra) of roughness. For the samples obtained by powder metallurgy (P/M), the density presented 88% of the estimated density and 5.76µm(Ra) of roughness.

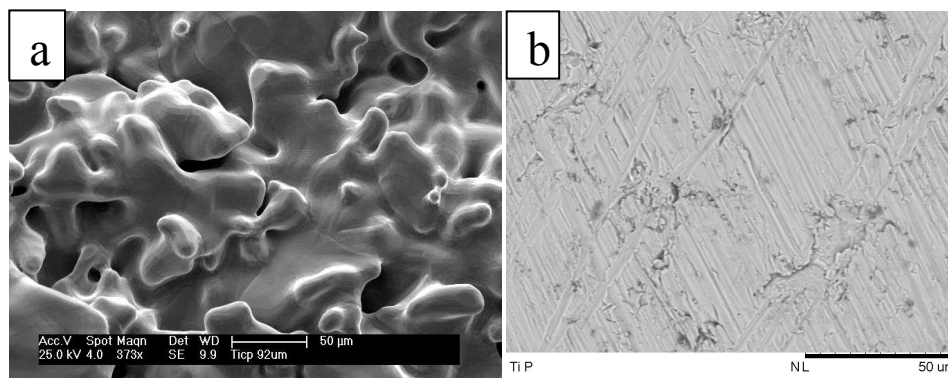


Fig. 1: SEM micrographs of surface samples titanium without treatments. a) sample obtained by powder metallurgy – general aspect, note the typical roughness and b) commercial plate titanium presenting surface after sanding with SiC sandpaper (n# 600) - general aspect.

The four pre-treatments were performed on both types of surface (laminated and P/M samples). All the pretreatments chosen in this work was based on apatite-forming ability reported in literature. For this reason, a comparison between them becomes interesting. The coating of samples were evaluated by morphological appearance in surface and cross sections for all times (1, 3, 6 and 9 days in SBF). This allowed observing that the layer was increasing gradually in all pretreatments evaluated over the nine days.

The Fig 2 shows the micrographs of samples with pretreatments when immersed in SBF for 1 day. The differences in rate of apatite formation might be explained by differences in the surface composition depending on the treatment conditions. The alkali-treated group for P/M samples, showed the formation of apatite nuclei of spherical shape, however, in cross-section was observed a thin discontinuous layer around 1 µm of thickness. The alkali-heat-treated group had a better performance as reported in other studies [7, 10]. There was formation of nano-apatite involving the surface associated with larger nuclei apatite in the range of 10-20µm. Furthermore, the layer presents more continuous with thickness ranging from 1,5-4,5µm. The acid-alkali treatment also had a good performance presenting layer formation of apatite with thickness of about 3µm. The alkali-CaCl<sub>2</sub>-heat-hot water treatment was more effective than the others pre-treatments for apatite formation [12]. The layer presents regular way with thickness ranging from 2,5-5µm. It is important to emphasize for P/M samples the difference of thickness values represents the morphological appearance aspect. Then, the minimal value of thickness is found in the recessed areas on the surface. However, the predominance of thickness corresponding at the higher value.

The laminate surface samples after 1 day in SBF, in all pretreatments evaluated, had apatite formation with isolated nuclei. However, the layer presented thin and intermittent. The acid-alkali treated and the alkali- $\text{CaCl}_2$ -heat-hot water treatment presented a similar pattern of thickness (around  $1\mu\text{m}$ ).

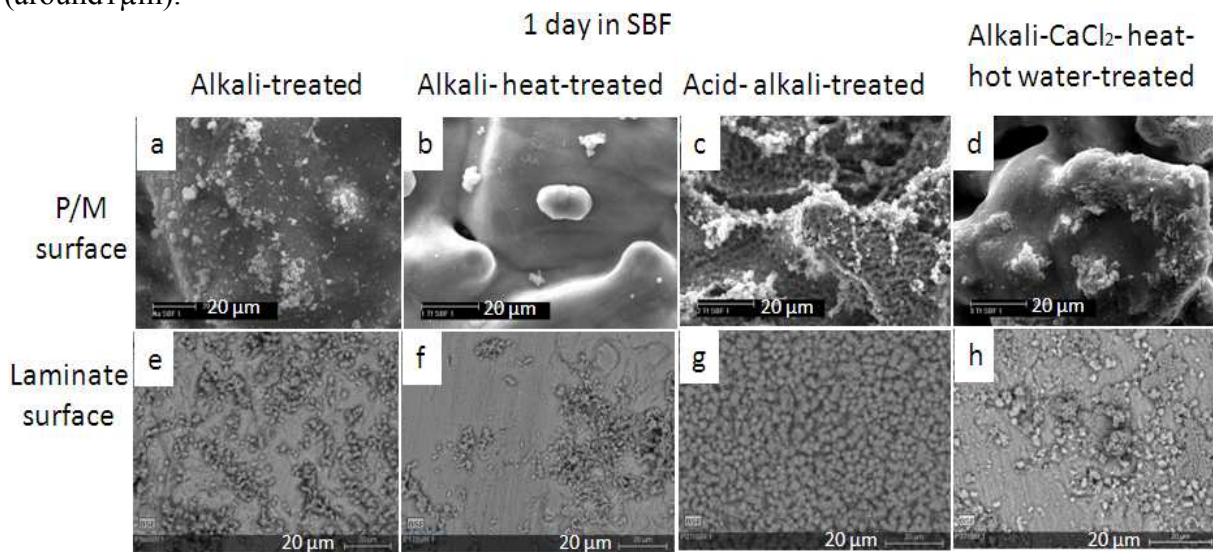


Fig. 2: SEM micrograph of surfaces by P/M (a,b,c,d) and laminate (e,f,g,h) Ti metals subjected to the pretreatments and soaked in SBF for 1 day.

After 9 days in SBF, for P/M samples (Fig. 3) can be observed the full apatite coating in all pretreatments with difference at form of nucleation. The alkali-treated group presented regular amount of apatite. The cross section allowed observe a thickness in the range of  $1,5\text{-}6\mu\text{m}$ . Following the same pattern of 1 day in SBF, the alkali-heat-treated group (in 9 days in SBF) had a better response for ability apatite forming compared at alkali-treated group as reported in other studies [7,10]. It is possible verify higher amount of apatite in the surface and the thickness presented  $2\text{-}6,5\mu\text{m}$ . The acid-alkali-treated group presented similar amount of apatite spheres than alkali-heat group, however, the thickness was higher in the range of  $4\text{-}8\mu\text{m}$ . The alkali- $\text{CaCl}_2$ -heat-hot water group presented the higher amount apatite with smaller spheres compared at the others pretreatments. However, the thickness ( $3\text{-}6,5\mu\text{m}$ ) increased at a rate not higher as the others. The thickness values for P / M samples are closer to the higher as previously commented.

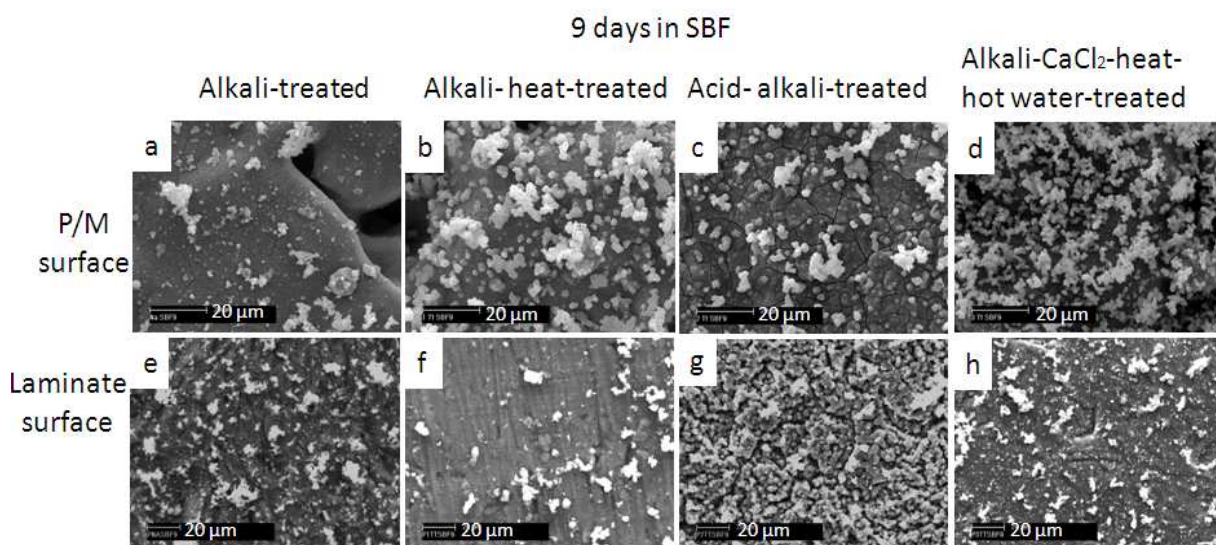


Fig. 3: SEM micrograph of surfaces by P/M (a,b,c,d) and laminate (e,f,g,h) Ti metals subjected to the pretreatments and soaked in SBF for 9 days.

The laminate samples for the alkali-treated group shows expressive amount of apatite, but the thickness of layer remained around 2,5  $\mu\text{m}$ . For alkali-heat-treated, can be observed less apatite. Nevertheless, the layer had thickness in range of 3 at 6,5 $\mu\text{m}$ . The acid-alkali-treated exhibited large amount of apatite with layer between 3 at 7  $\mu\text{m}$  of thickness. The alkali- $\text{CaCl}_2$ -heat-hot water group presents a satisfactory amount of apatite; but the layer was around 3  $\mu\text{m}$ .

The Fig. 4 shows an example of cross-section with the formed layer comparing between laminate and P/M surface at acid-alkali-treated in 9 days of SBF. It is possible observe the difference of thickness in both surface types. The P/M samples had a better performance in apatite-forming ability. This is due to roughness and higher surface area typical of technique.

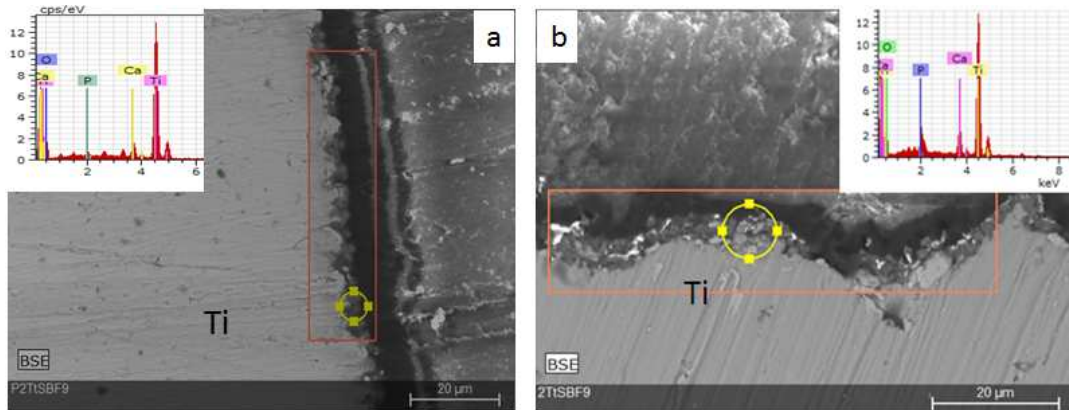


Fig.4: Cross-section of laminate Ti (a) and P/M Ti (b) showing the thickness of apatite layer with acid-alkali treatment when immersed for 9 days in SBF.

The DRIFT analysis presented a similar spectrum for all pretreatments and times evaluated. The Fig. 5 shows an example of spectrum for acid-alkali-treated (after 1 day in SBF) in Ti samples (laminate and P/M surface). In spectrum can be observed the presence of well defined wide band in the 3000-3600  $\text{cm}^{-1}$  region, characteristic of the O-H. Also observed bands of the  $\text{PO}_4^{3-}$ : 562 and 1020  $\text{cm}^{-1}$  and characteristic of the  $\text{CO}_3^{2-}$  ions vibration: 870 and 1450  $\text{cm}^{-1}$ . In 1650  $\text{cm}^{-1}$  is observed the band of  $\text{H}_2\text{O}$  (Fig. 5)[13]. The presence of these bands suggests the formation of apatite and  $\text{CO}_3^{2-}$  functional groups involves that carbonate ions are incorporated in the biomimetic apatite and form carbonate-hydroxyapatite.

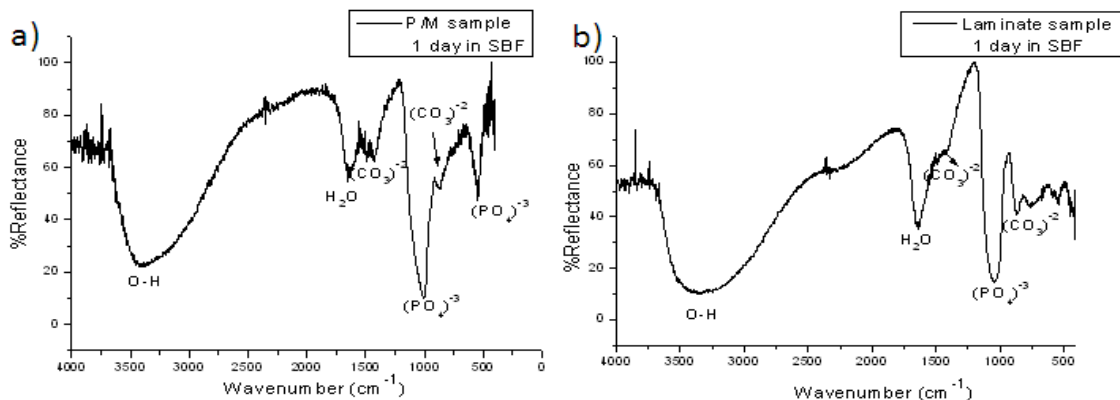


Fig. 5: Diffuse reflectance spectroscopy on the infrared (DRIFT) of P/M (a) and laminate (b) samples soaked at 1 day in SBF for acid-alkali treatment.

## Conclusions

The apatite formation was present in all groups studied since 1 day soaked in SBF. All pretreatments were effective for apatite-forming ability on titanium surface; however for samples obtained by powder metallurgy the apatite coating had better performance in comparison with

laminate surface. It is possible observed a layer apatite since 1 day for P/M samples. The alkali- $\text{CaCl}_2$ -heat-hot water treatment was more effective than others pretreatments for apatite formation in 1 day of SBF for P/M samples. However, after 9 days in SBF, the acid-alkali treatment presented a higher thickness layer of apatite.

The layer for laminate samples presents more continuous just after 9 days in SBF. For this surface, the acid-alkali treatment was better compared to others pre-treatments.

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