

Corrosion behaviour and bioactivity of electrophoretically deposited hydroxyapatite on titanium in physiological media (Hanks' solution)

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Hydroxyapatite (HA) coatings were developed on titanium by electrophoretic deposition at various deposition potentials from 30 to 60 V and at a constant deposition time of 5 minutes using the synthetic HA ($\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$) powder in a suspension of dimethylformamide (DMF, $\text{HCON}(\text{CH}_3)_2$). The electrochemical corrosion behavior of the HA coatings in simulated body fluid (SBF Hanks' solution) at 37 °C and pH 7.4 was investigated by means of open-circuit potential (OCP) measurement and potentiodynamic polarization tests. The OCP test showed that the values OCP for the coated samples shifted to more noble potential than for uncoated titanium, especially after addition of dispersants. The polarization test revealed that all HA coated specimens had a corrosion resistance higher than that of the substrate, especially after addition of dispersants such as polyvinyl butyral (PVB), polyethylene glycol (PEG) and triethanolamine (TEA) to the suspension. The coating morphology after polarization, characterized by scanning electron microscopy (SEM), showed penetration of electrolyte into the HA coats. Bone bioactivity of the coatings was also studied by immersion of coated specimens in Hanks' solution for 3 and 7 days. Apatite granules growth on the surface of the HA layers was observed.

Keywords: *hydroxyapatite, electrophoretic deposition, titanium, corrosion, bioactivity*

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1. Introduction

Titanium metal and its alloys are used in dental and orthopedic implants on account of their excellent corrosion resistance, biocompatibility and osseointegration behavior. Commercially pure titanium (CP-Ti) is widely used as an implant material. The biocompatibility and corrosion resistance of the titanium metal is the result of passive TiO_2 films of thickness of 2–6 nm, formed on the titanium surface [1]. However, metallic materials are susceptible to corrosive attack by body fluids, with subsequent release of metallic ions, which might cause adverse effects to the surrounding tissues. Moreover, metallic surfaces are in general not adequately bioactive,

and surface treatment is usually needed to enhance their bioactivity so as to improve osseointegration with bone tissues. In view of these, Ti implants are commonly coated with hydroxyapatite ($\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$, HA) – bioceramics which resembles the mineral constituents of human bones and teeth. The HA coating would reduce the release of metallic ions by acting as a barrier, and at the same time, enhance the bone bioactivity by virtue of its chemical constituents [2].

Many coating techniques, such as plasma spraying [3], sol-gel [4], dip coating [5], chemical solution deposition [6], biomimetic coating [7] and electrophoresis [8, 9], have been employed for the preparation of HA coatings. Electrophoretic deposition (EPD) presents several advantages such as an ability to control thickness and morphology of the deposited layer by controlling

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the electrochemical parameters, an ability to deposit relatively uniform coatings on complex shapes, a higher deposition rate as compared with most coating processes, and the low cost to purchase the equipment [10]. EPD is essentially a two-step process. In the first step, charged particles suspended in a liquid migrate towards an electrode under the effect of an electric field (electrophoresis). In the second step, the particles deposit on the electrode, forming a relatively dense and homogeneous compact film [11].

A suspension for EPD is a complex system in which each component has a substantial effect on the deposition efficiency. It is very important to obtain well dispersed and stable suspensions. In general, suspensions can be dispersed by electrostatic, steric, or electrosteric stabilization mechanisms. Ceramic particles must be electrically charged to permit forming by EPD. A wide variety of additives can be used to control the particle charge, so as to produce well-dispersed suspensions. Organic macromolecules could be utilized to induce steric stabilization, where the macromolecules are attached to the particle surface, or depletion stabilization, in which the macromolecules are free in suspension. The steric stabilization is effective in both aqueous and non-aqueous media. The intercalation of polymer particles is achieved by their adsorption on the surface of colloidal particles, which are produced near the cathode and form a cathodic deposit [12]. The charge on a colloidal particle could also originate from dispersants. It was concluded according to [12] that the most effective are phosphate ester (PE), menhaden fish oil and ethoxylate. In this work, we have used organic dispersant macromolecules such as polyvinyl butyral (PVB) and polyethylene glycol (PEG). Also we used triethanolamine (TEA) as a source of three OH⁻ groups which can be adsorbed and ionized on the surface of the HA particles too, so as the conductivity of the suspension increases with the amount of TEA. Consequently, the addition of TEA is in favor of the stable suspension for EPD [13]. Hence, a dispersing agent may be added to enhance the coating stability.

The stability of biomaterials still originates from their electrochemical passivity, and their ability to avoid passivity breakdown in the highly aggressive physiological environment. Different surface modifications have been employed to provide a specific resistance to the implants corrosion; these surface modifications mainly consist in physical coating process on the metals involving ceramic materials, such as bioglass, cerabone and glass ceramic. Titanium implants are commonly coated with hydroxyapatite layer [14] that provides the biocompatibility and corrosion resistance of titanium substrate.

At this stage, there is little information on the electrochemical behavior of biomaterials coated with ceramics, especially with surfactant addition, in the scientific literature [15]. The corrosion behavior of EPD HA coatings on the Ti alloys has not been fully studied with respect to the surfactant addition and a size of the starting HA particles.

Enhanced biocompatibility of titanium and titanium-based materials is achieved by coating them with ceramic biomaterials, which makes them the most widely used material combination for dental and orthopaedic implants. The material response is governed by ion leaching and by corrosion with the release of particles [15].

This work aims to investigate the effect of electrophoretic HA-coatings on corrosion behavior of Ti substrates. The corrosion behavior of the metallic biomaterials was investigated in physiological media (Hanks' solution) at 37 °C and pH 7.4 by means of open-circuit potential measurement and potentiodynamic polarization tests. In vitro bioactivity of HA coatings and the effect of additives on the corrosion behavior were also studied.

2. Materials and methods

2.1. Preparation of HA powder

Preparation of HA powder was based on the previous work [16] in which a simple co-precipitation method was proposed to prepare nano-sized HA powders using calcium nitrate tetrahydrate [Ca(NO₃)₂ · 4H₂O 99 %, WINLAB

U.K] and phosphoric acid [H_3PO_4 85 %, SDS, France] as starting materials. $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$ was dissolved in distilled water to form a 0.5 M solution, into which H_3PO_4 was added at a Ca/P ratio of 1.67. After 30 minutes of mixing, ammonium hydroxide was poured into the mixed solution to precipitate HA until the pH value of the mixture reached 10 (about 10 mL NH_4OH was added). After vigorous stirring for 30 minutes, HA powders were obtained by washing the precipitate repeatedly two times to remove the unwanted ions (NH_4^+ and NO_3^-) and then drying at 70 °C in a drying chamber for 24 hours.

2.2. Treatment of titanium substrate

Commercially pure titanium (99.66 %) of 0.25 mm thickness was used in this study. Titanium (Ti) sheets of 12 x 12 x 0.25 mm were degreased with acetone for 20 min in an ultrasonic bath. The substrates were etched in a solution containing nitric acid and hydrofluoric acid (1:1), rinsed with distilled water, and then allowed to dry in air.

2.3. Preparation of suspensions and electrophoretic deposition

HA powder synthesized as in section 2.1 was used as coating material. 1 g of the HA powder was added into 100 ml DMF (99 %, LOBA Cheme). After magnetically stirring for 15 min, the suspension was dispersed ultrasonically for 30 min at 50 kHz in an ultrasonic bath (BRANSON U.S.A). A titanium sheet was used as the working electrode (cathode) and platinum sheet was used as the counter electrode (anode) The distance between the electrodes was 2 cm. Electrophoretic deposition experiments were performed at various voltages of 30, 40 and 60 V (because of deterioration of the coating at deposition voltages higher than 60 Volt) at deposition time of 5 minutes. The weight of HA deposited at various applied voltages was recorded. Dispersing agents such as polyvinyl butyral (PVB), polyethylene glycol (PEG) and triethanolamine (TEA) were added to the suspension to improve the corrosion resistance of the HA coating.

Table 1. The chemical composition of SBF (Hank's solution).

Chemical Name	Chemical Formula	Concentration g/L
Sodium Chloride	NaCl	8.0
Calcium Chloride	CaCl_2	0.14
Potassium Chloride	KCl	0.40
Sodium Bicarbonate	NaHCO_3	0.35
Glucose	H_{12}O_6	1.00
Sodium Dihydrogen phosphate	NaH_2PO_4	0.1
Magnesium Chloride	MgCl_2	0.1
Disodium Hydrogen phosphate	Na_2HPO_4	0.06
Magnesium Sulphate	MgSO_4	0.06

2.4. Electrochemical study

Electrochemical corrosion test of uncoated and coated samples was carried out in a simulated body fluid—Hank's solution (its composition is listed in Table 1) at pH 7.4 and temperature 37 ± 1 °C in open air. A saturated calomel electrode (SCE) and a platinized black platinum electrode were used as the reference and auxiliary electrodes respectively. The area of the coated surface exposed to the corrosion study was 1 cm². The other sides of the electrode and its edges were masked with lacquer and were dried in air. The electrodes were further dipped into the electrolytic solution to study the corrosion process. The electrochemical measurements carried out on the uncoated and coated samples included open circuit potential (OCP) time measurements and potentiodynamic polarization. The critical parameters like corrosion potential (E_{corr}), corrosion current (I_{corr}), corrosion resistance (Rp), corrosion rate were evaluated from the polarization curves by Tafel extrapolation with the aid of a commercial software (VoltaMaster 4 version 7.08).

The electrochemical experiments were performed using a VOLTA LAB 40 potentiostat (Model PGZ301, Radiometer Analytical, France) with the aid of commercial equipment. The samples were immersed in Hank's solution and the OCP was monitored for an hour. The potentiodynamic polarization scans were carried

out with the scan rate of 1 mV/s in the range of -500 mV (vs. SCE) to 2.5 V (vs. SCE).

2.5. Analysis after corrosion

2.5.1. Characterization of the coating surface

The corroded surfaces of different coating samples were characterized using scanning electron microscope SEM, (JEOL, JSM 5410, Japan).

2.6. In vitro bioactivity tests

The HA coating was soaked in 25 ml of simulated body fluid (SBF Hank's solution) at a pH of 7.4 at 37 °C for 3 and 7 days. After soaking, the samples were washed by distilled water then the coating surface was characterized with SEM, by energy dispersive spectrometry (EDS) analysis and using an X-Ray Diffractometer (XRD Bruker AXS-D8 X-ray diffractometer, ADVANCE, Germany).

3. Results and discussion

3.1. Open circuit potential–time measurements

The OCP–time plots for the uncoated and HA coated Ti without and with additives are shown in Figs. 1 and 2 respectively. The OCP was found to be shifted considerably in the noble direction compared to the uncoated sample (Fig. 1). This shift of OCP and its saturation at a certain potential indicate the formation of a stable HA/Ti interface. The OCP of uncoated sample in Hanks' solution reached a potential of -0.370 V (vs. SCE) after 60 minutes. This indicates the protective nature of the coatings on Ti. The noble behavior of the HA coatings could be due to the insulating nature of the surface. However, the obtained HA coatings are microporous in nature and hence the electrolyte seeps into the coating through the pores. Thus the OCP–time relationship for HA coatings developed at different potentials showed variations in the noble behavior. The coating obtained at a potential of 40 V performed better. It showed an initial OCP of -0.275 V (vs. SCE), reached a

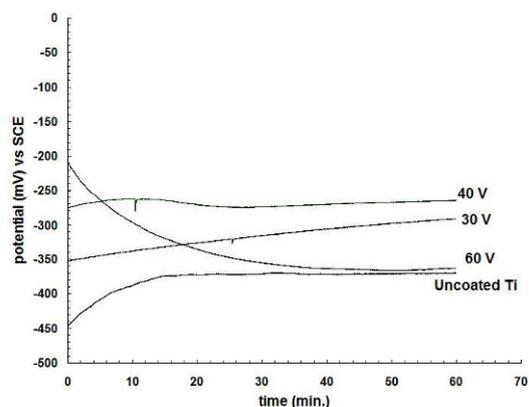


Fig. 1. OCP–time measurements in Hank's solution of uncoated and HA coated Ti obtained at different coating voltages at a constant time of 5 min.

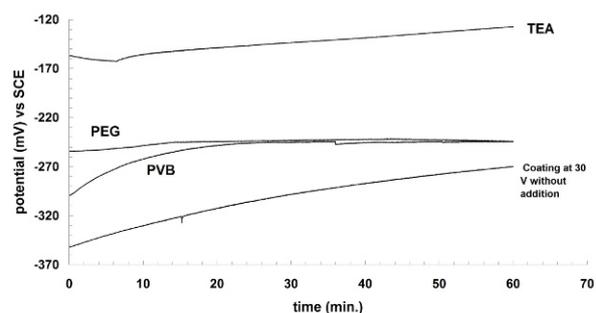


Fig. 2. OCP–time measurements in Hank's solution of HA coated Ti obtained with and without addition of dispersing agents at 30 V and constant time of 5 min.

steady state within 10 min and attained a potential of -0.265 V (vs. SCE) after 60 minutes. The curves for the coatings obtained at the deposition potential of 60 V showed minor variations in OCP (-0.362 V vs. SCE). This may be due to the existence of a large diffusion path within such thick and crack containing coatings, which are capable of causing large potential drops. From the OCP–time measurements it can be inferred that at the coating voltages of 30 V (-0.291 V vs. SCE) and 40 V (-0.265 V vs. SCE) the OCP is shifted to a maximum in the more noble region and the time taken to attain constant OCP is faster when compared with the uncoated samples.

The OCP–time curves of the specimens coated with HA after addition of dispersing agent to the

Table 2. Corrosion test parameters of uncoated Ti and HA coated Ti at different coating conditions.

Conditions	E_{corr} (mV)	I_{corr} (nA/cm ²)
Uncoated Ti	-342.4	299.3
HA coated at 30 V	-321.0	103.316
HA coated at 40 V	-220.7	128.68
HA coated at 60 V	-324.2	259.1
HA coated at 30 V with PVB	-275.4	77.16
HA coated at 30 V with PEG	-279.8	89.698
HA coated at 30 V with TEA	-279.4	44.71

suspension (Fig 2) are shifted towards more noble direction than for HA coating without any addition. The coating obtained by addition of 10 mL/L triethanolamine (TEA) at the deposition voltage of 30 V and deposition time of 5 minutes performed better. It showed an initial OCP of -0.156 V (vs. SCE), reached a steady state within 7 min and attained a potential of -0.127 V (vs. SCE) after 60 minutes. The coatings obtained by addition of 1 g/L of polyethylene glycol (PEG) and 4 g/L of polyvinyl butyral (PVB) at 30 V and time of 5 minutes exhibited similar behavior, reaching OCP of -0.244.5 V (vs. SCE). It can be noted that all coatings obtained with addition of dispersing agents (PVB, PEG and TEA) have a higher OCP potential than the coating obtained at 30 V without the additions.

3.2. Potentiodynamic polarization measurements

The corrosion current density (I_{corr}) was determined by Tafel extrapolation with the aid of a commercial software (VoltaMaster 4 version 7.08).

The polarization plots for the uncoated and HA coated Ti without and with additives are shown in Figs. 3 and 4. The polarization curves presented in Figs. 3 and 4 show very low corrosion current densities (extrapolated from the linear region of the cathodic and anodic curve to the corrosion potential, E_{corr}). The current density of the uncoated Ti specimens is of the order of 299.3 nA/cm², at the corrosion potential (E_{corr})

which is commonly observed for passive materials with protective surface films (Table 2). The current density difference between the coated and uncoated surfaces, along with the nobler E_{corr} with respect to SCE for the coated surface, suggests a beneficial effect of the coating on corrosion resistance (103.31 nA/cm², 128.68 nA/cm², 259.1 nA/cm², 89.67 nA/cm², 77.16 nA/cm² and 44.71 nA/cm² for Ti with HA coating deposited at 30 V, 40 V, 60 V, 30 V with PEG, 30 V with PVB and 30 V with TEA, respectively). Despite the increase in the current density as the potential was increased to approximately 0.2 V (SCE), the current density was essentially independent of the potential value up to approximately 1.3 V (SCE) for both uncoated and coated surfaces at 30 V, 40 V and 60 V. This suggests the establishment of a passive region, typically observed in the surfaces with a passive character (Fig. 3). The current density in this passive region was approximately 7.2 μ A/cm² for uncoated specimens, 10.8 μ A/cm² for Ti coated with HA at 30 V, 15.8 μ A/cm² for Ti coated with HA at 40 V and 19.2 μ A/cm² for Ti coated with HA at 60 V, 11.64 μ A/cm² for Ti coated with HA with PEG at 30 V, 14.8 μ A/cm² for Ti coated with HA with PVB at 30 V and 7.9 μ A/cm² for Ti coated with HA with TEA at 30 V. In this potential range, the coating had no substantial effect on the corrosion resistance of the substrate as discussed by Coelho et al [17]. At a potential value of approximately 1.2-1.3 V (SCE), the current density increased for both surfaces achieving the highest increase in magnitude for the Ti surface coated with HA at 30 V and the lowest increase for Ti coated with HA at 40 V. This behavior in this potential range was not observed for HA coated Ti at 60 V and HA coated Ti at 30 V with PEG, PVB and TEA addition.

In general, the increase in current density for titanium alloys at potentials around 1.3 V (SCE) has been associated with the first passive film breakdown and subsequent repassivation by secondary passive film formation, or by formation of Ti₂O₅ [16].

But the higher current density associated with the coated surfaces may result from the

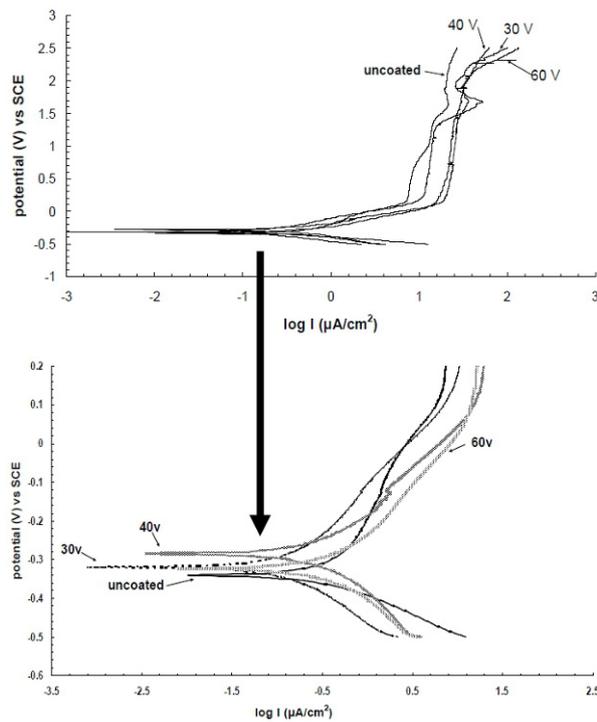


Fig. 3. Potentiodynamic polarization curves for Ti specimens, uncoated and coated at different coating voltages at a constant time of 5 min in Hanks' solution, at pH 7.4 and 37 °C.

porosity within the coating structure induced by polarization [17, 18]. This effect appears in case of HA coating deposited at 30 V and is smaller at higher deposition voltage due to the increased coating thickness. So, the solution takes more time to reach the surface of the base metal. But the microcracks induced by polarization also appear (Fig. 5). At the potentials around 1.9 V (SCE), the current stabilizes again suggesting the setting up of another passive region for both uncoated and HA coated surfaces at 30 V. However, the specimens coated with HA with addition of dispersing agents (PVB, PEG and TEA) do not exhibit any increase in current density around 1.3 V (vs. SCE). This may be due to the dense packing of HA particles (no microcracks, Fig. 5) on Ti surface, which gives no chance for solution to reach the base metal at this high polarization potential. This is also a very strong evidence showing the effectiveness of dispersing agent for dense particles packing and adherence to the substrate. Fig. 5 shows the

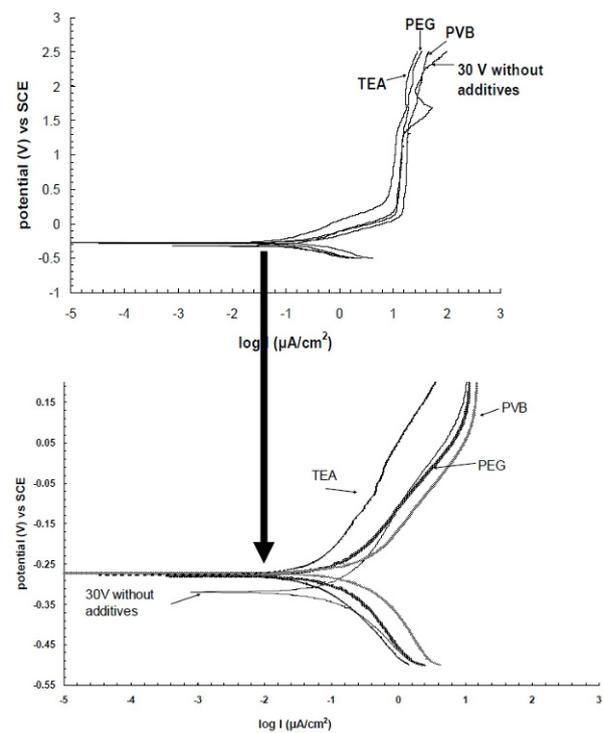


Fig. 4. Potentiodynamic polarization curves for Ti specimens covered with the coatings with and without addition of dispersing agents, obtained at the deposition voltage of 30 V at a constant time of 5 min in Hanks' solution, at pH 7.4 and 37 °C.

SEM images of HA coated Ti specimens after polarization test, which reveal the presence of microcracks in the case of the coating obtained at 30 V due to the penetration of solution during high polarization. The images of the coatings obtained in other conditions do not show clear microcracks because thick HA deposited on these samples prevents the solution penetration during polarization.

3.3. Bioactivity of the coatings

Fig. 6 shows the XRD pattern of a HA coated specimen after the immersion test of 3 and 7 days. It could be observed that the intensity of apatite peaks has increased significantly, indicating that apatite was grown on the specimen surface after immersion in Hanks' solution. These results reveal that the HA coatings formed by EPD are bone bioactive and hence may favor osseointegration.

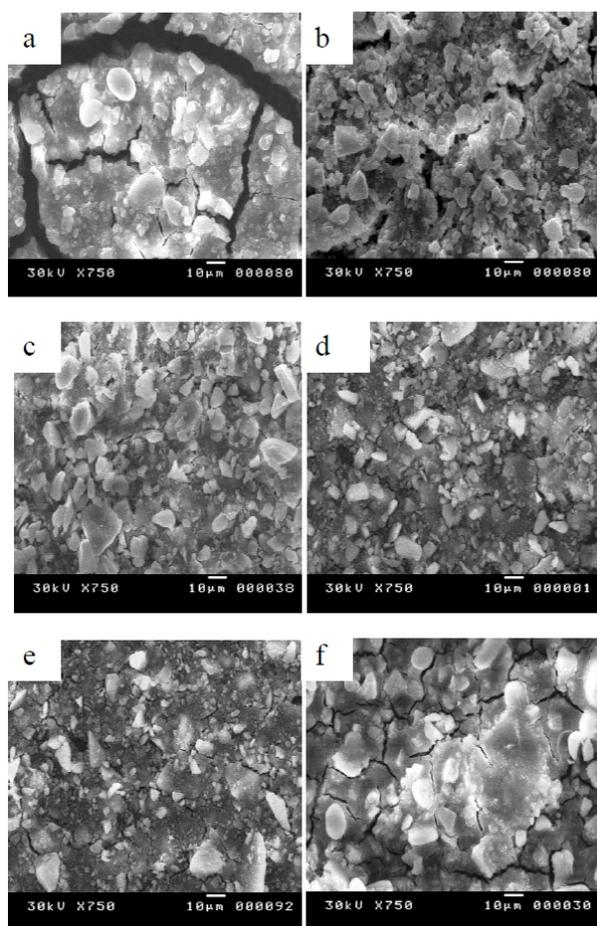


Fig. 5. SEM micrographs of the specimens coated with HA with and without additions, after polarization in Hanks' solution at different deposition voltages (a) 30 V, (b) 40 V, (c) 60 V, (d) 30 V with PVB, (e) 30 V with PEG and (f) 30 V with TEA.

Fig. 7 shows typical appearances (SEM images) of the specimen surfaces before and after immersion test. Apatite particles are clearly visible on these specimens after immersion in the test solution both for 3 and 7 days. At the beginning of immersion, the solution penetrates through the coating (Fig. 7), then the apatite particles start precipitating on the coating surface and close the cracks formed through solution penetration. The apatite particles are denser in case of immersion for 7 days, indicating that the apatite growth is more intense when the time of immersion increases. So it can be concluded the coatings become bioactive.

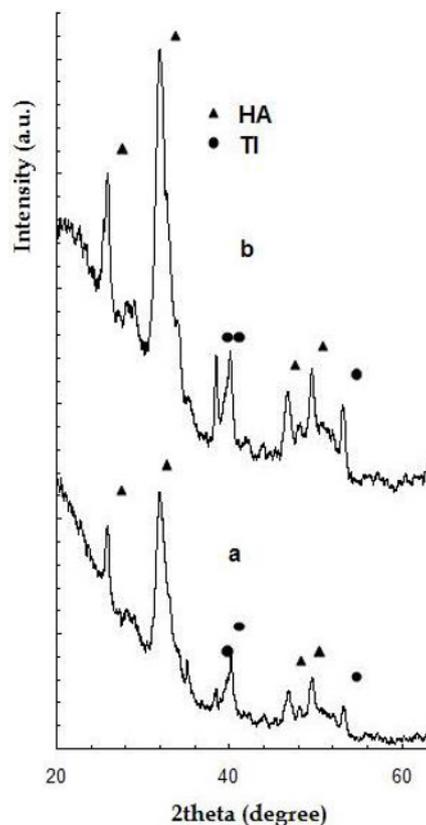


Fig. 6. XRD patterns of a HA coating at applied voltage of 30 V and deposition time 5 min. after immersion in Hanks' solution for (a) 3 days and (b) 7 days.

The chemical composition of the small granular particles was detected by EDS (Fig. 8). The molar ratio of $n(\text{Ca})/n(\text{P})$ is 1.7. The presence of sodium and chloride ions beside calcium and phosphorus ions indicates that HA coatings have the ability to induce nucleation and growth of ions and the bone-like apatite (Ca and P) on their surfaces from simulated body fluid (SBF) solutions (Hanks' solution). Other authors have also observed similar phenomena in pure HA coatings. Kwok [2] found the formation of apatite after immersion in Hanks' solution for 4 weeks. Xiao [13] found the formation of apatite on electrophoretically deposited Si-HA on Ti surface after immersion in simulated body fluid (SBF) solution for 7 days. The bone-like apatite layer has been found to be important to establish the bone bonding interface between bioactive materials and living tissues.

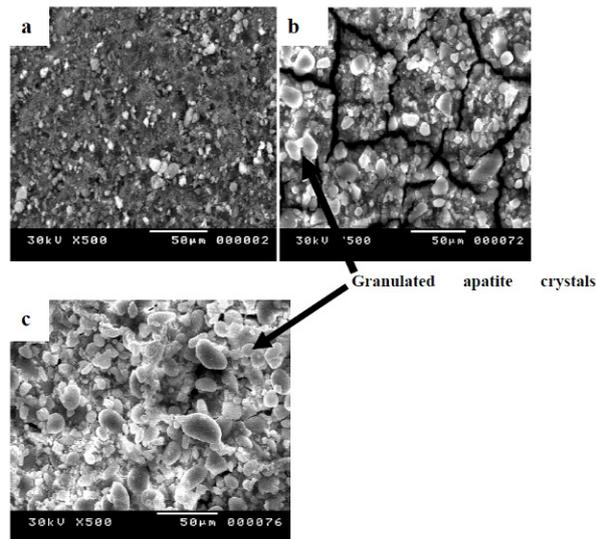


Fig. 7. SEM micrographs of HA coated specimens at deposition time of 5 min. and deposition voltage of 30 V before (a) and after immersion test in Hanks' solution for (b) 3 days and (c) 7 days.

It should be pointed out that the simulated body fluid used in this study (Hanks' solution) in the immersion test for bone bioactivity did not contain any biological matter such as bone-forming cells. Nevertheless, Kokubo and Takadama [19] in their recent review claimed that the results of immersion tests in inorganic simulated body fluids such as Hanks' solution are useful in predicting in vivo bone bioactivity.

4. Conclusion

1. The HA coated Ti had higher corrosion resistance compared to the uncoated Ti substrate. This improvement was supported by a noble shift of open-circuit potential and a lower corrosion current density.
2. The Ti coated with HA with addition of dispersing agents exhibited higher corrosion resistance compared to the HA coated Ti substrate without these additions. This improvement was supported by a noble shift of open-circuit potential and a lower corrosion current density (coating after TEA addition had nobler OCP and lower density of corrosion current).

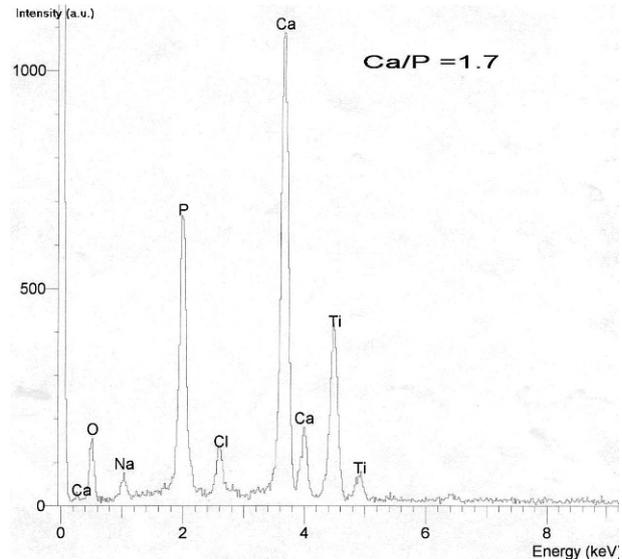


Fig. 8. EDS spectra of HA coating at applied voltage of 30 V and deposition time of 5 min. after immersion for 7 days in Hanks' solution. (O 5.0 %, Na 1.7 %, P 26.1 %, Cl 4.6 %, Ca 45.2 %, Ti 17.4 %).

3. SEM micrographs of HA coatings showed the presence of microcracks which indicate the penetration of solution into HA coatings during polarization.

4. Apatite particles were formed on the surface of HA coatings after immersion in Hanks' solution for 3 and 7 days indicating high bone bioactivity.

References

- [1] BALAKRISHNANA A., LEEA B. C., KIMA T. N., PANIGRAHIB B. B., *Trends Biomater. Artif. Organs.*, 22(1) (2008), 66.
- [2] KWOK C.T., WONG P.K., CHENG F.T., MAN H.C., *Appl. Surf. Sci.*, 255 (2009), 6736.
- [3] GARCIA-SANZ F.J., MAYOR M.B, ARIAS J.L., POU J., LEON B., PEREZ-AMO M., *J. Mater. Sci.: Mater. Med.*, 8 (1997), 861.
- [4] WANG D., CHEN C., HE T., LEI T., *J. Mater. Sci.: Mater. Med.*, 19 (2008), 2281.
- [5] AKSAKAL B., HANYALOGLU C., *J. Mater. Sci.: Mater. Med.*, 19 (2008), 2097.
- [6] KUO M.C., YEN S.K., *Mater. Sci. Eng. C*, 20 (2002), 153.
- [7] COSTANTINI A., LUCIANI G., BRANDA F., AMBROSIO L., MATTOGNO G., PANDOLFI L., *J. Mater. Sci.: Mater. Med.*, 13 (2002), 891.
- [8] WANG C., MA J., CHENG W., ZHANG R., *Mater. Lett.*, 57 (2002), 99.

- [9] ZHITOMIRSKY I., GAL-OR L., *J. Mater. Sci.: Mater. Med.*, 8: (1997) 213.
- [10] MENG X., KWON T., YANG Y., ONG J., KIM K., *J. Biomed. Mater. Res.*, 78B (2006) 373.
- [11] BOCCACCINI A.R., ZHITOMIRSKY I., *Current Opinion in Solid State and Mater. Sci.*, 6 (2002), 251.
- [12] ZHITOMIRSKY I., *Adv. Colloid Interface Sci.*, 97 (2002), 279.
- [13] XIAO X.F., LIU R.F., TANG X. L., *J. Mater. Sci.: Mater. Med.*, 19 (2008), 175.
- [14] NAVA-DINO C.G. et al., *Int. J. Electrochem. Sci.*, 7 (2012), 2389.
- [15] RICARDO M.S., MARÍA M.L., RUI L.R., *Biomater.*, 24 (2003), 4213.
- [16] ABDELSATTAR A.A., SHOEIB M.A., MOHAMED S.G., *Surf. Coat. Technol.*, 206 (2011), 43.
- [17] COELHO P. G., DE ASSIS S.L., COSTA I., THOMPSON V.P., *J. Mater. Sci.: Mater. Med.*, 20 (2009), 215.
- [18] ADVINCULA M.C., PETERSEN D., RAHEMTULLA F., ADVINCULA R., LEMONS J.E., *J. Biomed. Mater. Res. B Appl. Biomater.*, 80 (2007), 107.
- [19] KOKUBO T., TAKADAMA H., *Biomater.*, 27 (2006), 2907.

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