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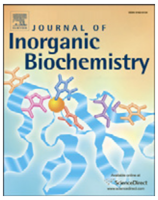

Bio-inspired citrate-functionalized apatite thin films crystallized on Ti–6Al–4V implants pre-coated with corrosion resistant layers

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ABSTRACT

In this paper the crystallization of a bioinspired citrate-functionalized apatite (cit-Ap) thin film (thickness about 2 μm) on Ti–6Al–4V supports pre-coated with bioactive and corrosion resistant buffer layer of silicon nitride (Si_3N_4), silicon carbide (SiC) or titanium nitride (TiN) is reported. The apatitic coatings were produced by a new coating technique based on the induction heating of the implants immersed in a flowing calcium–citrate–phosphate solution at pH 11. The influence of the buffer layers and the surface roughness of the substrate on the chemical–physical features and adhesion of the cit-Ap films were investigated. The best plasticity, compactness and adherence properties have been found in the Ap layer grown on Si_3N_4, followed by the Ap grown on SiC and TiN, respectively. The adhesion property was likely related to the roughness of the buffered substrates, whereas the compactness and plasticity were closely related to the operating conditions during the Ap crystallization (flow rate of the solution and increase of temperature) rather than to the nature of the buffer layer.

1. Introduction

Titanium and its alloys (Ti–6Al–4V, Ti–5Al–2.5Fe, etc.) are among the most commonly used materials to manufacture dental and orthopedic implants [1,2]. This success is related to their excellent mechanical properties (relative high strength and fatigue) and good corrosion resistance in physiological fluids due to the passivating native oxide layer acting as a protective barrier naturally formed on the surface. However, several problems associated with their implantation in the body were described, such as stress shielding, metal ion release, etc. [3–5]. This drawback is due to different degradation processes that range from simple chemical dissolution to mechanical wear of the articulating surfaces. The released metallic ions may also induce foreign body reactions, leading to inflammation and eventually to implant failure. For these reasons, research effort is focusing on modifying their surface to enhance osseointegration and to improve the corrosion and wear resistance. Among different surface treatments to enhance osseointegration one is the application of a coating consisting of components mimicking the bone extracellular matrix and in particular calcium phosphates (CaPs) in the form of apatite (Ap) phase. In this way, the mechanical strength of the titanium substrate can be combined with the bioactive feature of Ap in order to improve the biocompatibility of the implant as well as the adhesion to bone [6–10]. De Groot and Geesink [11] introduced CaP coatings for orthopedic and dental implants for the first time in 1987. Since then, numerous reports have been published about the evidences of the osteoconductive properties of CaP coated implants. They are described to increase the bone-to-implant contact, to improve the implant fixation and to facilitate the bridging of small gaps between implant and surrounding bone. In addition, since the Furlong and Osborn's first clinical trials [12] several studies revealed that the long-term performance of the CaP coated implants is comparable to the non-coated ones, however the coated implants demonstrated their benefit for a faster bone healing [13].

From a commercial point of view, the most successful method to deposit calcium phosphate coatings to implants is the plasma-spraying technique, due to the high deposition rate and the ability to coat large areas [14]. Although the osteoconductive and bone-bonding behavior of CaP plasma-sprayed coatings is confirmed by numerous studies, still some serious concerns are related to this technique. In particular, these concerns deal with the composition and thickness that is also related to the adhesion strength of the coatings. The thickness of plasma sprayed coatings, as applied by various manufacturers, varies between 50 and 200 μm. A coating thickness >100 μm can introduce fatigue under tensile loading and the higher thermal expansivity of Ap with respect to titanium alloy yields tensile stress that produces through-thickness cracking. Excessive thickness can favor coating delamination and fragmentation, which in time can result in implant mobility. The adhesion strength

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between CaP coating and substrate varies from 5 to 65 MPa depending on the plasma-spraying technique used. Filiaggi et al. [15] reported that the strength of Ap coating is dependent on the coating thickness and in particular coatings less than 50 μm provided higher adhesion strength than coatings of 240 μm. Coatings of > 100 μm thickness reduce the fatigue resistance of the titanium alloy substrates, whereas 25 to 50 μm thick coatings did not delaminate during fatigue or final fracture. To overcome these problems pre-treatments of the substrate by mechanical methods such as grit blasting to roughen the substrate and to increase the mechanical interlocking of the coating-substrate system were also developed. In fact, the surface roughness is a desired characteristic to produce large contact areas between metal substrate and Ap improving their adhesion, but on the other hand it enhances the rate of released cations [4]. Both well polished passivated surfaces and thick Ap layers (up to 200 μm) have been reported to reduce the metal ion release kinetics [4], simply by acting as physical or chemical barrier. Taking into account all these limits, other methods to prepare thinner and more compact CaP films were developed, such as: magnetron sputtering [16], electrophoretic deposition [17], pulsed laser deposition, ion beam dynamic mixing [18], laser excimer [19], electrostatic spray deposition [20,21] and induction heating method [22].

The approach used in this work is to crystallize a thin citrate-functionalized Ap (cit-Ap) film on metallic implants pre-coated with a bioinert and corrosion resistant buffer layer using the induction heating method [22]. Ti–6Al–4V substrates pre-coated with silicon carbide (SiC), titanium nitride (TiN) or silicon nitride (Si_3N_4) layers were used. The remarkable physical and chemical properties of these buffer layers have been widely reported [23,24] and in particular their good diffusion barrier feature is worth to be highlighted. The control over the crystallized solid phase, the low processing time and the ability to coat implants with complex geometries are the most important advantages of the induction heating method [22]. This latter property is a very important feature of this method with respect to others deposition techniques that are able to produce thinner coatings but they cannot be applied to completely coat 3D targets or objects with irregular shapes such as dental screws. The main advantage of using citrate ions in the Ap reaction preparation deals with their chemical and biological features. Indeed, citrate is a medium strength Ca^{2+} complexing agent allowing the preparation of metastable solutions containing $Ca(cit)^-$ complexes in the presence of HPO_4^{2-} and OH^- ions, at low temperatures and elevated pHs. The increase in temperature favor the destabilization of the $Ca(cit)^-$ complex and, at the same time, the precipitation of CaP apatite because of the negative dependence of its solubility with the temperature (i.e. retrograde solubility). This fact in combination with the ability of free citrate ions to adsorb on apatite surface is conveniently exploited by this method to obtain thin apatite layers functionalized with citrate. Hu et al. [25] demonstrated by advanced multi-NMR spectroscopy that citrate molecules, strongly immobilized on bone apatite surfaces, accounts for 5.5 wt.% of the organic bone matter. Xie and Nancollas [26] hypothesized that the bio-inspired strategy of using citrate in the preparation of Ap can be a powerful tool to control the synthesis of this material. Recently, we reported on the synthesis of citrate-functionalized and biocompatible poorly crystalline apatite nanoparticles with sizes ranging from 20 to 200 nm by the thermal decomplexing batch method of Ca/cit/phosphate/carbonate solutions [27]. In addition, other papers reported that synthetic CaP materials modified with citrate improved the proliferation and differentiation of osteoblasts [28], and enhanced the bone remodeling at the early stages of bone healing [29]. Based on these previous results the bio-inspired strategy to coat the implants with a cit-Ap layer could potentially improve the materials in term of osseointegration. This work represents the first step in the set-up of these new coatings and, in particular, the influence of the nature of the buffer layers and the surface roughness of the substrates on the crystallization and adhesion properties of the cit-Ap films has been investigated.

2. Experimental

2.1. Materials

Analytical grade $CaCl_2$, K_3cit (where $cit^{3-} = C_6H_5O_7^{3-}$), K_2HPO_4 and KOH were purchased from Merck (Darmstadt, Germany). Ultra pure water (0.22 μS, 25 °C, MilliQ, Millipore) was used to prepare all solutions. Two types of Ti–6Al–4V substrates pre-coated by magnetron sputtering technique with thin TiN, SiC or Si_3N_4 buffer layers (supplied by VASKO Ltd) were used: square plaques of 6 mm in length and 3 mm in thickness and cylinders with dimensions of 3.5 mm-diameter and 14.2 mm long. Cylinders were chosen as the geometric prototype of dental implants, while plaques were used for characterization purposes. Unpolished and uncoated square plaques were used as blank substrates.

2.2. Induction heating coating technique

The induction heating system used in this work is comprised of three basic components: an alternating current power supply, an induction coil and a metallic support to be heated. The power supply sends alternating current through the coil, generating an electromagnetic field. When the support is placed in the coil, the electromagnetic field induces eddy currents (Foucault currents) on its conductor metallic surface. Then, the eddy current dissipates heat due to the Joule effect. The coating technique used in this work, developed by Gómez-Morales et al. [22], consists of applying this system to locally heat the surface of a metallic support immersed in a flowing metastable calcium/citrate/phosphate aqueous solution at high pH (pHs 10–11). In this way, the Ap precipitation takes place at the contact between the heated metallic implant with the solution and can be schematized (considering stoichiometric hydroxyapatite, $Ca_5(PO_4)_3OH$, as model solid phase) according to the following reaction:

$$3\ HPO_4^{2-} + 5\ Ca(cit)^- + OH^- \Rightarrow Ca_5(PO_4)_3OH + 5\ (cit)^{3-} + 3\ H^+ \qquad (1)$$

In detail, the experimental set-up is composed of an induction furnace (CELES, France) of 3 kW and 152 kHz provided of a copper coil with appropriate water cooling, a cylindrical quartz tube of 3 cm in diameter with open ends containing the specimen adhered to a Teflon holder, a thermostatic feeding tank, a thermostat-cryostat (Haake, Germany), a peristaltic pump, and tubes connecting the feeding tank to the bottom of the cylindrical quartz tube and the top of the tube to the tank to recover the residual flowing solution (Fig. 1). The feeding tank was kept at 10 °C to avoid uncontrolled CaP nucleation in the mother solution. The voltage (V) of the induction furnace can be regulated up to 6 kV, which yields anodic intensity (I) up to 0.5 A.

2.3. Ap crystallization

Freshly prepared solutions of compositions 0.001 M $CaCl_2$, 0.002 M K_3-cit and 0.006 M K_2HPO_4 at pH 11 adjusted with 0.1 M KOH were pumped through the quartz tube and the remaining solutions were recirculated to the cold (10 °C) feeding tank. Three Ti–6Al–4V square planar plaques pre-coated with TiN, SiC or Si_3N_4 were coated with Ap at 500 W for 1 h at solution flow rates (Φ) of 115, 100 and 80 mL/min, respectively. The heat dissipated (W) by each plaque toward the flowing solution by Joule effect was estimated according to the following formula:

$$W(\text{watts}) = 0.0167\Phi \cdot C_e \cdot \Delta T \qquad (2)$$

where Φ is the solution flow rate (mL/min), 0.0167 is a conversion factor from mL/min to g/s, C_e is the specific heat of water (4.18 J/g °C) and ΔT (±0.1 °C) is the temperature increase of the flowing solution during the coating process. ΔT was calculated by measuring the initial temperature