Contents lists available at ScienceDirect



Materials Science and Engineering C

journal homepage: www.elsevier.com/locate/msec



Bacterial adhesion on amorphous and crystalline metal oxide coatings



Argelia Almaguer-Flores^a, Phaedra Silva-Bermudez^{b,c,*}, Rey Galicia^c, Sandra E. Rodil^c

^a Facultad de Odontología, División de Estudios de Posgrado e Investigación, Universidad Nacional Autónoma de México, Circuito exterior s/n, Ciudad Universitaria, 04510 México D.F., Mexico ^b Unidad de Ingeniería de Tejidos, Terapia Celular y Medicina Regenerativa, Instituto Nacional de Rehabilitación, Calzada México-Xochimilco No. 289, Col. Arenal de Guadalupe, 14389 México D.F., Mexico

^c Instituto de Investigaciones en Materiales, Universidad Nacional Autónoma de México, Circuito Exterior s/n, Ciudad Universitaria, 04510 México D.F., Mexico

ARTICLE INFO

Article history: Received 29 January 2015 Received in revised form 10 May 2015 Accepted 10 July 2015 Available online 20 July 2015

Keywords: Titanium oxide Zirconium oxide Atomic ordering Surface energy Bacterial adhesion Surface topography

ABSTRACT

Several studies have demonstrated the influence of surface properties (surface energy, composition and topography) of biocompatible materials on the adhesion of cells/bacteria on solid substrates; however, few have provided information about the effect of the atomic arrangement or crystallinity. Using magnetron sputtering deposition, we produced amorphous and crystalline TiO₂ and ZrO₂ coatings with controlled micro and nanoscale morphology. The effect of the structure on the physical–chemical surface properties was carefully analyzed. Then, we studied how these parameters affect the adhesion of *Escherichia coli* and *Staphylococcus aureus*. Our findings demonstrated that the nano-topography and the surface energy were significantly influenced by the coating structure. Bacterial adhesion at micro-rough (2.6 μ m) surfaces was independent of the surface composition and structure, contrary to the observation in sub-micron (0.5 μ m) rough surfaces, where the crystalline oxides (TiO₂ > ZrO₂) surfaces exhibited higher numbers of attached bacteria. Particularly, crystalline TiO₂, which presented a predominant acidic nature, was more attractive for the adhesion of the elevisition of amorphous or crystalline oxide coatings, offers a route for the rational design of implant surfaces to control or inhibit bacterial adhesion.

© 2015 Elsevier B.V. All rights reserved.

1. Introduction

The current research on biomaterials includes the modification of the surfaces as a strategy to tailor the surface properties aiming to improve the biocompatibility or to enhance the functionality of the implant material. Among the surface properties that are usually modified are the topography at the micro- and nano-scale, the surface chemistry and the surface energy [1–3].

In the particular area of dental implants, titanium and its alloys are the selected metallic materials due to their good biocompatibility and adequate mechanical and osseointegration properties [4]. Titanium is a reactive metal that in contact with the atmosphere rapidly forms a passive oxide film that inhibits subsequent dissolution processes making the material corrosion resistant. In the biological media, it has been shown that the surface oxide allows apposition of Ca and P ions, as well as hydroxyl and hydrate groups, leading to the nucleation of apatite (CaP) crystals, which enhances the osseointegration processs [5]. Despite these good properties, some surface modifications have also been introduced to improve the bone-to-implant contact area

* Corresponding author at: Unidad de Ingeniería de Tejidos, Terapia Celular y Medicina Regenerativa, Instituto Nacional de Rehabilitación, Calzada México-Xochimilco No. 289, Col. Arenal de Guadalupe, 14389 México D.F., Mexico. trying to reduce the loading-time of the implant [6,7]. It has been shown that Ti surfaces with micro-scale roughened surfaces, in the range of $1-10 \mu$ m, maximize the interlocking between mineralized bone and the implant surface as well as enhance the proliferation and differentiation of osteoblastic cells [8,9]. Some studies have shown that roughness at the nanometer level also plays a relevant role [10, 11]. The effect of the nano-scale on the cell adhesion phenomena might be related to changes in the wetting properties, leading to different protein adsorption schemes, or because the process used to induce the nanotopography introduces also modifications in the surface chemistry [12,13].

The effects of other surface properties such as surface energy, surface charge, crystalline structure or phase have been less studied than those of topography or their results are not so conclusive [14,15]. The difficulty resides, partially, in that with the purpose of modifying the wettability or the surface energy, the surface chemistry and/or the topography are also altered. Our opinion is that, in most cases, the complex interplay among surface properties is neglected leading to inconsistent results. Surfaces are characterized by atoms presenting a different chemical ambient: dangling or strained bonds [16]. Surface atoms are in a higher energy state (larger surface free energy), compared to the bulk atoms of the same material, and in order to reduce the energy, many surface phenomena occur. Any pristine surface will interact with the environment trying to reduce the surface free energy (SFE). Therefore, the surface

E-mail addresses: suriel21@yahoo.com, pssilva@inr.gob.mx (P. Silva-Bermudez).

properties, unless they are measured in ultra-high vacuum conditions, are already modified by the surface–environment interaction [17]. Upon interaction with the biological environment, it will be this modified surface the one interacting with the ions, proteins, sugars and lipids present in the blood and tissue fluids [18,19]. Obviously, sterilization processes do also modify the surfaces and their properties [20]. In a similar manner, when surface topography is intentionally modified, both the surface chemistry and the wettability can be altered, and without a proper identification of those changes, no reasonable conclusions can be reached about the role played by a single surface-parameter.

We considered that due to this interplay among the different surface properties, the understanding of the effect of surface energy, surface charge and crystallinity has been more elusive, particularly for the implant applications discussed in here. The role of the atomic arrangement or atomic ordering; crystalline versus amorphous, has not been of interest partially because metallic implants are generally crystalline and their native oxide films are so thin that their structural organization is not well defined. However, nowadays the development of amorphous metals is an important research area due to their extraordinary mechanical and corrosion resistance properties [21,22]. Similarly, the use of biocompatible coatings to improve the biological response of the implants is an emerging area, in which the synthesis of crystalline or amorphous coatings is possible [23–25]. Therefore, the objective of this paper is to evaluate the effect of crystallinity on both the surface properties and the bacterial adhesion on titanium oxide (TiO₂) and zirconium oxide (ZrO₂) coatings. Moreover, the use of coatings provides us an important tool to evaluate the effect of different surface topographies, at both nano- and micro-scale, preserving the same surface chemistry. TiO₂ and ZrO₂ were chosen since it has already been demonstrated that both materials are biocompatible; TiO₂ as the passive coating on Ti-based implants [26] and ZrO₂ as a ceramic-based dental implant [27]. In the particular case of the interaction TiO₂ surfaces-biological media, experimental and theoretical works have shown that the surface atomic arrangement, which depends on both the crystalline structure and the exposed crystalline plane or facet, plays an important role for protein adsorption [28-30]. Sul et al. [31] studied the effect of the TiO₂ surface properties on the in vivo bone tissue response concluding that surface crystallinity and surface porosity were the surface properties with larger impact on the bone response; however, no further details have been given. Other works have studied or compared the biological response of different TiO₂ phases, but not comparison with amorphous TiO₂ is presented [32]. Similarly, no much information about the biological response of ZrO₂ surfaces presenting different grades of crystallinity was found in the literature. Sollazzo et al. studied the gene expression of MG-63 osteoblastic cells grown on crystalline ZrO₂ surfaces presenting the same structure, but different crystal sizes [33]. Di Virgilio et al. studied the cytotoxicity of colloidal silica spheres (\approx 830 nm diameter) coated with crystalline and amorphous ZrO₂; revealing that crystalline ZrO₂ showed an increased cytotoxicity and genotoxicity when compared to amorphous ZrO₂ [34].

Our hypothesis is that the atomic ordering through its influence on the surface energy, nano-topography and wettability might induce different bacterial adhesion patterns. To assess this hypothesis, amorphous and crystalline TiO_2 and ZrO_2 thin films were produced by magnetron sputtering. The film surfaces were extensively characterized; the wettability, surface energy, surface composition and topography were measured in order to take into account the complex relationship between the different surface properties. Meanwhile, the macro topography was controlled by using micro-structured cpTi surfaces [19]. In the present paper, the surface characterization and the surface-bacteria interactions are presented. It is worth mentioning that the atomic ordering also influences the protein adsorption schemes and cellular response; nevertheless, these are undergoing studies and such results will be reported separately.

2. Materials and methods

2.1. Substrates

The metal oxide films were deposited on Si N/PH (100) wafers (UniSil Corporation, Sta. Clara, CA, USA) cut into 1 cm \times 1 cm square substrates (Si) to perform all the physical-chemical characterization of the thin films. Grade-2 commercially pure Ti disks, 1 mm-thick and 15 mm in diameter, with two different topographies, smooth pretreatment (PT) and rough sandblasted acid-etched (SLA), were used as substrates to obtain metal oxide coated surfaces with different topographies [19]. With the aim of identifying the different topographies induced by the amorphous/crystalline structure, a set of PT substrates were mirror-finished (PTmf) using 1000 grade SiC grit and polished using 50 μ m-diameter Al₂O₃ particles. Then, the PTmf substrates were ultrasonically cleaned before deposition of the metal oxide films.

2.2. Metal oxide coatings

Sets of PT, SLA, PTmf and Si substrates were coated with either TiO₂ or ZrO₂ thin films by reactive magnetron sputtering using, correspondingly, Ti (99.995% purity) or Zr (99.2% purity) 10 cm-diameter sputtering targets (SCI Engineered Materials Ohio, USA). Ar and O₂ of ultra-high purity grade were used as the atmosphere gases. The working pressure was 2.2×10^{-4} Pa (8:2 sccm, Ar:O₂ ratio) and the radio frequency (RF) power was set to 200 W.

In order to obtain crystalline or amorphous metal oxide structures, the substrate temperature used during deposition was changed. For the amorphous metal oxide films no substrate heating was applied during deposition, while the substrates were heated to 573 K and 523 K during deposition to produce crystalline TiO₂ and ZrO₂ films, respectively. Therefore, the final set of experimental samples was PT-*a*TiO₂, PT-*c*TiO₂, PT-*c*ZrO₂, SLA-*a*TiO₂, SLA-*a*TiO₂, SLA-*a*TiO₂, PT-*c*TiO₂, SI-*c*TiO₂, SI-*c*TiO₂, SI-*c*TiO₂, SI-*c*TiO₂, PTmf-*c*TiO₂, PTmf-*c*TiO₂, PTmf-*c*TiO₂, PTmf-*c*TiO₂, meter *c* stands for crystalline and *a* for amorphous oxides. Finally, the deposition time was adjusted to obtain coatings of similar thickness for the same metal oxide, independently of their atomic ordering; 1800 s and 2400 s were used for amorphous and crystalline films; respectively.

2.3. Surface characterization techniques

The as-received PT and SLA samples and the coated Si substrates were characterized by X-ray Photoelectron Spectroscopy (XPS) using a VG Microtech Multilab ESCA 2000 to obtain their surface chemical composition. The XPS was operated at 8×10^{-7} Pa using Al K α radiation $h\nu = 1486.6$ eV with a spatial resolution of 500 μ m and 50 and 20 eV pass energy to acquire the survey and high resolution spectra, respectively. High resolution spectra were acquired in the C 1s and O 1s photoelectron peak energy regions for all the samples and also in the Zr 3d energy region for the substrates coated by $aZrO_2$ or $cZrO_2$ films and in the Ti 2p region for the as-received PT and SLA samples and the substrates coated by aTiO₂ or cTiO₂ films. No Ar cleaning procedure was performed on the surfaces before the XPS measurements. High resolution spectra were fitted using the SDPv4.1 software® to obtain the surface elemental chemical compositions; all binding energies were referenced to the maximum of the C 1s peak (C-C + C-H) set at 285 eV. Results are reported as the mean of measurements in three samples per each kind of surface; the overall associated error was considered as the standard 5% measurement error of the XPS technique.

X-ray diffraction (XRD) patterns were obtained from the as-received PT and SLA substrates using a Bruker D8 XRD system in the coupled Bragg–Bretano mode and monochromatized Cu K α radiation. The atomic ordering of the metal oxide coatings was characterized from the films deposited on Si, using a Rigaku Ultima IV XRD system with a

Download English Version:

https://daneshyari.com/en/article/7868564

Download Persian Version:

https://daneshyari.com/article/7868564

Daneshyari.com