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Surface Modification of Ti6Al4V Alloys by Dielectric Barrier Discharge (DBD) Plasma for Enhance Bioactivity

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Abstract
Surface modifications of Ti6Al4V alloys are essential for improving their performance when implanted into the human body. Surface modification by dielectric barrier discharge (DBD) plasma is a simple and practical method, ensuring ease of use, economic viability, and reliable results. In this paper, the surface of the Ti6Al4V alloy was modified with DBD plasma and then compared with the unmodified surface after both were coated with strontium-doped hydroxyapatite (Sr-HA) by the hydrothermal method. The characterization of the surfaces was assessed with respect to crystalline structure, surface morphology, wettability, antibacterial activity against Klebsiella pneumoniae colonies, and corrosion behavior. The bioactivity was also evaluated in vitro by immersing both coated surfaces in simulated body fluid (SBF) solution for 28 days. The results show that the surface modified with DBD plasma enhanced the adhesion of nanostructured coating, became more hydrophilic with a contact angle of 51.5°, and exhibited the higher polarization resistance value (Rp=13.3 kΩ.cm^2) compared with the unmodified surface (Rp= 5 kΩ.cm^2). In addition, the bone-like apatite was improved on the modified surface. Thus, surface modification with DBD plasma is an effective way to enhance the nucleation sites and surface hydrophilicity, thereby improving the biological activity of the implant surface.

Keyword: Surface modification • Ti6Al4V alloy • DBD plasma treatment • hydrothermal method • hydroxyapatite coatings • hydrophilic surface.

Introduction

Metallic biomaterial implants are typically needed to restore the functionality of bones that have been damaged or degenerated due to a variety of factors, such as aging, sports injuries, and accident[1]. Thus, requirement for the fabrication of superior-quality implants with better performance[2].

Titanium (Ti) alloys like Ti6Al4V are frequently used as base metallic materials in biomedical applications, including dental and orthopedic implants, particularly for load-bearing implants due to their high strength to weight ratio, favorable mechanical properties, acceptable corrosion resistance, good biocompatibility, and low cytotoxicity[3, 4]. On the other hand, the key to successful implantation of Ti alloy is that bone tissue or bone cells can integrate on its surface.

However, Ti6Al4V alloys are biologically inert and lack sufficient tissue response, making it difficult for bone cells to be effectively stimulated to proliferate on their surface[5]. Therefore, surface modifications are crucial for improving the bio-functional surface properties of Ti alloys for long-term performance[6].

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The most effective approach to surface modification of Ti alloys involves controlling the implant surface roughness and preparing a bioactive material coating, such as Hydroxyapatite (HA), which it shares chemical similarities with the mineral found in genuine bone[7-9]. HA is a calcium phosphate compound that can be used for enhance the surface characteristics of the interface between the implants and human tissues[10]. Nevertheless, the mechanical characteristics of the artificially synthesized pure HA are weak.; therefore, strontium-doped HA (Sr-HA) coating can enhance mechanical properties and much better cell attachment than the pure HA coating[11-15].

Among various methods used to form Sr-HA coatings on Ti6Al4V surfaces, the hydrothermal method is relatively simple, compatible, low-cost, and effective in promoting chemical interactions between the sample surface and the coating[16-19]. on the other hand, despite the ability of these coatings to improve the bioactivity of Ti6Al4V, the poor adhesion of these coatings is a major concern[4]. Thus, modification of Ti6Al4V surface before coating by plasma is advantageous, owing to its ability to influence the surface selectivity, but the bulk composition of the material remains unchanged[20]. Recently, Dielectric Barrier Discharge (DBD) plasma has significantly improved titanium surface wettability, antibacterial behavior, and corrosion resistance[21, 22]. Therefore, DBD plasma treatment is used as a surface modification of Ti6Al4V alloy before coating to enhance the efficiency and adhesion of the hydrothermally grown (Sr-HA).

Non-thermal DBD, alternatively referred to as silent discharge, is widely used in materials science to modify the surface properties, which enhance wettability, topography, and morphological changes[23-25]. DBD plasma garnered a lot of interest because of its stable discharge channel, straightforward device structure, short processing time, low operator cost, and environmental friendliness[25-29]. Operation at atmospheric pressure technique, at or near room temperature, and involves the generation of a high-voltage electric field between two electrodes separated by a dielectric barrier and a gas gap[30]. DBD offering key tool for improving metal–coating interactions[31].

This study aimed to compare the hydrothermally growth (Sr-HA) coating on the unmodified surface of Ti6Al4V alloy and the modified surface with DBD plasma. The characterization of the surfaces was assessed with respect to crystalline structure, surface morphology, wettability, antibacterial activity against Klebsiella pneumoniae colonies, and corrosion behavior. The biological activity of these coatings was also evaluated in vitro by immersing the coated surfaces in simulated body fluid (SBF) solution.

Experimental

Experimental Design

Two groups of Ti6Al4V alloy samples, unmodified and modified surface with DBD plasma, were prepared in this study. Sr-HA coating was deposited on both groups by the hydrothermal method to evaluate the effect of surface modification on coating growth and performance. Each group was prepared with six samples (n=6). The prepared samples were then used for different characterizations, including XRD, FESEM, measurement of contact angle, antibacterial test, corrosion resistance, and bioactivity in SBF solution under identical experimental conditions.

Preparation of Ti6Al4V Alloys

The samples of Ti6Al4V alloy discs (diameter in 2 cm, thickness in 3 mm) were ground using silicon carbide paper with grit sizes (400 to 2000 microns) to remove macro defects and contaminations. Then, polished using the Struers-RotoPol-21 system, employing polishing cloth and diamond paste. The polished samples were ultrasonically cleaned for 20 min in ethanol and deionized (DI) water to remove impurity particles and then dried at room temperature.

DBD Plasma Technique Treatment

The DBD plasma technique consists of two copper electrodes (8 cm length, 3 cm width, and 6 mm thick) and a glass plate (0.5 cm thick) serving as the dielectric barrier. The polished Ti6Al4V sample was placed on the ground copper electrode, and a 2 mm gap was maintained between the sample and the glass plate. An alternating high-voltage power source (Fanavaran Nano-Meghyas HV35P OV) operating at 8 kHz and 8 kV was applied to the external copper electrodes to generate a dielectric discharge between them. The feed gas was 99.99% pure argon at a flow rate of 30 L/min. The DBD plasma treatment was conducted at room temperature (25°C) and atmospheric pressure for 5 min to provide sufficient plasma interaction for effective activation and surface modification.

Preparation of Coating via Hydrothermal Method

The HA solution was prepared by mixing 1 M of calcium hydroxide and 0.06 M of dicalcium phosphate dehydrate with DI water for 25 min using a magnetic stirrer. Subsequently, a 0.5 M of Strontium nitrate dissolved in DI water with stirring for 15 min. After that, Strontium nitrate solution was added dropwise into the prepared HA solution under stirring for 20 min. The final concentration of the (Sr-HA) solution is 19.5 M. The unmodified and modified surfaces of Ti6Al4V alloys subjected to DBD plasma treatment were placed separately in a Teflon-lined stainless steel autoclave filled with a mixed (Sr-HA) solution for 4 hours at 180°C. Afterward, the autoclave cooled naturally to room temperature, and the sample was removed, rinsed with DI water, and dried in an oven at 100°C for 1 hour.

Surfaces Characterization

The phase analysis and crystal structure of the unmodified and modified surfaces of Ti6Al4V alloys before and after hydrothermal coating were characterized using X-ray diffraction (XRD) with Cu Kα radiation (λ = 0.1542 nm) at 40 kV and 30 mA, with a 0.05° step from 20° to 60°. Surface morphologies were analyzed using a field-emission scanning electron microscope (Tescan Mira3 FESEM, Czech).

Wettability Measurement

Wettability measurement was performed to determine the hydrophilicity and contact angle (CA) of unmodified and modified surfaces of Ti6Al4V alloys after hydrothermal coating. DI water was dropped on each sample's surface, and a photograph of the droplet was taken after 10 s. ImageJ software was used to determine the droplet's CA, which is the angle of the interface between the solid surface and the liquid. The measurements of the CA were performed at three different positions on each sample, and the average values were recorded.

Antibacterial Activity

The antibacterial activity of the unmodified and modified surfaces of Ti6Al4V alloys after hydrothermal coating was evaluated against Klebsiella pneumoniae in the College of Science, Mustansiriyah University, similar to the procedure reported in the literature[32, 33]. First, 10 ml of sterile Tryptone Soya Broth (TSB) was inoculated with bacteria, and the mixture was then incubated at 37°C for 24 hours. During the inoculation process, a 0.1 ml from the diluted bacterial suspension was administered onto both surfaces. Afterwards, the samples were placed in petri dishes. A cover slip was used to ensure the bacteria were in contact with the sample's surface. The Petri dishes were then incubated at 37°C for 24 hours. Once incubation was complete, the samples were transferred to falcon tubes containing 10 mL of sterile phosphate-buffered saline (PBS). The bacteria were then released into the PBS solution by vortexing the falcon tubes for 20 second. This process was done three times for every sample to estimate the bacterial concentration. Upon completion of the previous steps, the bacteria were then transferred to petri dishes containing culture media, which facilitates the growth of bacterial colonies. These petri dishes were subsequently 24 hours of incubation at 37°C. The image of colonies was captured with a digital camera.

Electrochemical Corrosion Behaviors

The electrochemical analyses were performed to assess the corrosion behavior of unmodified and modified surfaces of Ti6Al4V alloys after hydrothermal coating using potentiodynamic polarization under in vitro conditions in SBF solution. The electrochemical assessments were conducted using a Potentiostat/ Galvanostat (Model CS310M, S/N 2109730, manufactured by CorrTest Instruments, China) with a three-electrode configuration in which AgCl was the reference electrode, a graphite rod as the counter electrode, and the two prepared samples of Ti6Al4V alloy as the working electrode, separately for each test. At a scan rate of 0.5 mV/s, the potentiodynamic scan was carried in a potential range ±0.25 V vs. open circuit potential. Origin software was used to determine the corrosion potential (Ecorr) and corrosion current density (icorr) from the Tafel slope extrapolation method.

Bioactivity in Vitro

The ability of bone-like apatite to form on the unmodified and modified surfaces of Ti6Al4V alloys after hydrothermal coating was investigated with SBF in vitro. SBF solution (pH = 7.4), which has an ion concentration similar to human blood, was prepared in the laboratory according to the procedure of Kokubo et al.[34]. The prepared surfaces were immersed in 20 mL of SBF solution each and then incubated at 37°C for 28 days. The SBF solution was refreshed every 3 days, and the samples were removed from SBF after 28 days, rinsed with deionized water, and dried at room temperature. FTIR analysis was performed to verify the successful preparation of the SBF solution, and FESEM was used to evaluate apatite deposition on the prepared surfaces.

Results and Discussion

XRD analysis of Sr-HA coated by hydrothermal treatment on un-modified and modified surfaces of Ti6Al4V alloys is shown in **Figure 1**. Both samples showed distinctive peaks corresponding to two distinct crystalline phases, HA (hexagonal, ICDD No. 96-901-4314) and the Ti6Al4V alloy (ICDD No. 44-1294). The main HA characteristic peaks in both prepared samples at 2θ values of 35.46° and 40.51°, corresponding to the (031) and (221) Miller indices, respectively. The absence of secondary phases in XRD confirms successful incorporation of Sr^{2+} ions into the HA lattice without formation of separate Sr-containing phases. FESEM analysis provides the surface morphology of the unmodified sample, and the sample modified with DBD plasma before and after hydrothermal treatment, as shown in **Figure 2**. The images reveal a clear difference in surface behavior between the unmodified sample and the sample modified with DBD plasma before hydrothermal growth coating. The unmodified sample appears smooth with a few polishing scratches; this suggests a lower effective surface area and small aspect ratios, as present in **Figure 2a1 and 2a2**.

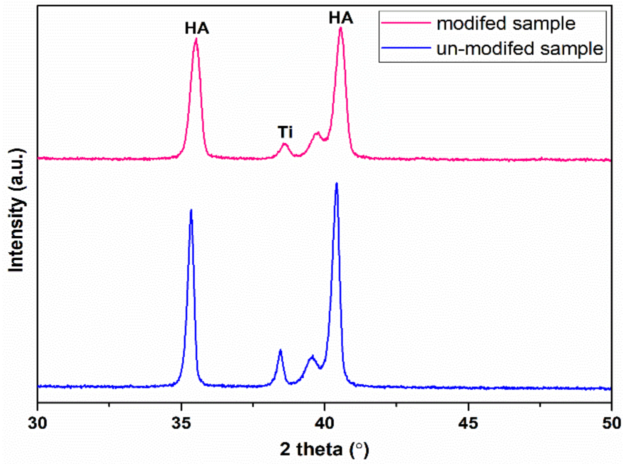

Figure 1. XRD analysis of the unmodified and modified samples by DBD plasma after hydrothermal coating.

In contrast, the sample modified with DBD plasma shows a change in surface roughness with finer surface etching and a high effective surface area with relatively large aspect ratios (**Figure 2b1** and **2b2**). After hydrothermal coating, the surface modified by DBD plasma exhibited a denser coating, greater coverage, and more uniform nanostructures (**Figure 2b3** and **2b4**) than the unmodified sample (**Figure 2a3** and **2a4**). The mean particle size, obtained from measurements of 10 randomly selected nanoparticle from FESEM images after hydrothermal coating using ImageJ software, was approximately 10 nm for the unmodified sample and 99.4 nm for the modified surface. The improvement in nanostructures coating growth on the surface modified with DBD plasma can be attributed to the plasma surface interaction mechanism. Thus, DBD plasma treatment modifies the Ti6Al4V alloy surface through ion bombardment and surface activation, which increases surface energy and generates reactive functional groups such as hydroxyl. These changes enhance surface roughness and provide additional nucleation sites for hydrothermal coating adhesion.

Wettability is defined as the capability and assessment of the interaction state between a liquid and its dispersion across a solid substrate[35]. Surface wettability is effect with surface chemistry and topography[36]. It is well-established that a decrease in CA corresponds to improved wettability of the surface and increased hydrophilicity. The average multiple measurements of the CA for both sample are presented in **Figure3.** Both samples are hydrophilic (<90°), and the CA decreased after treatment with DBD plasma compared with The modified sample. Thus, the CA changed from 80.2° for unmodified surface to 51.5° for modified surface, a decrease related to increased surface roughness. According to the Wenzel model, if a surface is already hydrophilic, increasing roughness makes it more hydrophilic, so that water fully penetrates the nanostructure, leading to a larger solid-liquid contact area and a decreased contact angle[37]. Implants exhibiting elevated hydrophilicity have the potential for enhance apatite deposition, thereby augmenting bioactivity on their surfaces[38].

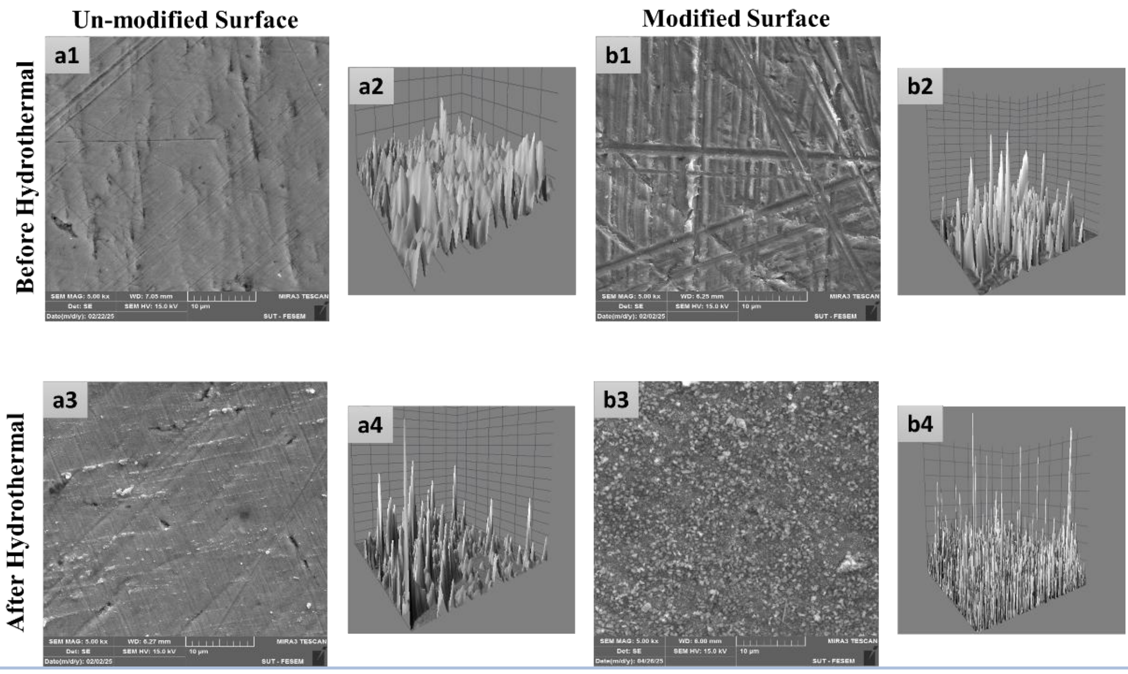

Figure 2. FESEM image of unmodified surface (a1-a4) and surface modified with DBD plasma (b1-b4) before and after hydrothermal coating.

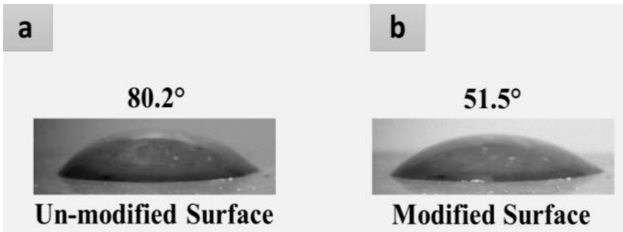

Figure 3. Average CA measurement of (a) unmodified and (b) modified surfaces with DBD plasma after hydrothermal coating.

Figure 4 illustrates the antibacterial performance of both samples: (a) unmodified and (b) modified surfaces with DBD plasma after hydrothermal number of bacterial colonies appears on the unmodified surface (**Figure 4a**). In contrast, the modified surface exhibited fewer bacterial colony-forming units when inoculated with the clinical isolate Klebsiella pneumoniae **Figuer 4b.** Consequently, these results suggest that enhancing antibacterial properties of the modified surface can be related to an increase in surface properties with DBD plasma, such as roughness, hydrophilicity, and coating density, which leads to increased mechanical damage to the bacterial cell wall and membrane, thereby enhancing antibacterial efficiency[39].

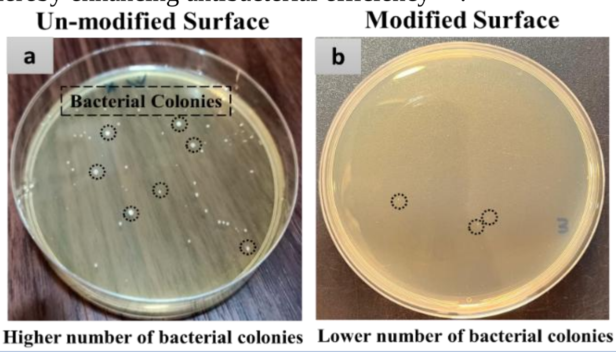

Figure 4. The antibacterial test of (a) unmodified and (b) modified surfaces with DBD plasma after hydrothermal coating.

The corrosion behaviors were revealed by potentiodynamic polarization curves of the both sample in the SBF solution, as shown in **Figure5**. The values of corrosion parameters were obtained through the Tafel extrapolation method to evaluate and compare the corrosion behavior of both the samples are presents in **Table 1**. The results indicate that the icorr dramatically decreased from 4.44 (μA/cm^2) of the unmodified sample to 2.63 (μA/cm^2) of the modified sample. The unmodified sample exhibits the most negative corrosion potential (Ecorr = -0.054V), while the Ecorr of the modified sample significantly increased to -0.045 V. A higher polarization resistance value was obtained for modified surface (Rp=13.3 kΩ.cm^2) compared with the unmodified surface (Rp= 5 kΩ.cm^2). The enhancement in corrosion resistance on modified surface with DBD plasma is associated with the formation of a more uniform, chemically stable surface layer that acts as an effective barrier decrease charge transfer and ionic transport processes at the sample surface-SBF solution interface.

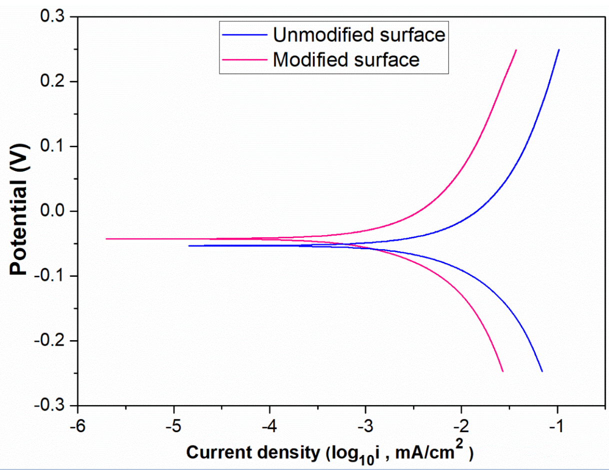

Figure 5. Potentiodynamic polarization curves for corrosion behavior of samples in SBF solution.

Table 1. Electrochemical corrosion parameters of the unmodified and modified samples.

Sample	E_{corr} (V)	I_{corr} ($\mu A/cm^2$)	β_a (V)	β_c (V)	R ($k\Omega.cm^2$)
unmodified	-0.539	44.4	0.103	-0.101	5
modified	-0.046	2.63	0.174	-0.151	13.3

Figure 6 shows the FTIR spectrum of the SBF solution. The spectra were acquired in the range of 4000-500 cm^{-1}.The obtained spectrum exhibited characteristic transmittance bands attributed to phosphate groups (PO_4^{3-}) observed at 1036, 662, 596 cm^{-1}. In addition, the bands assigned to O-H stretching and bending were detected at 3190 and 1630 cm^{-1}, respectively. Finally, carbonate group bands (CO_3^{2-}) were observed at 1460, 1404 cm^{-1}. The presence of these characteristic peaks confirms the existence of the major ionic constituents of SBF, indicating that the solution was successfully prepared.

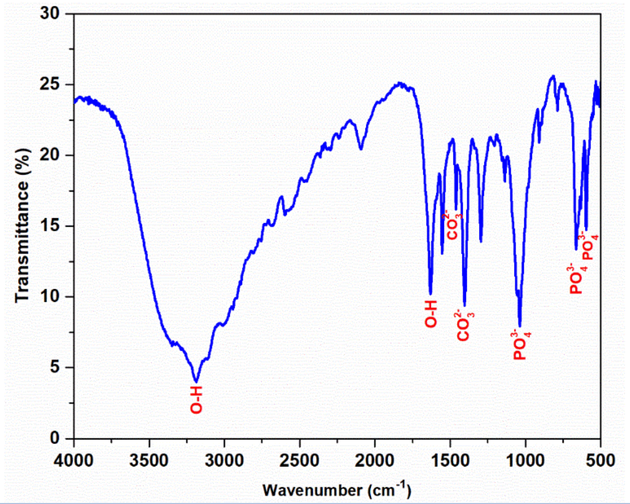

Figure 6. FTIR spectra for SBF solution.

Figure7 shown FESEM images in different scale bars (1 µm and 500 nm) of bone- like apatite formation on both unmodified and modified surface after immersion in SBF solution for 28 days. **Figure 7a1** and **7a2** shows the apatite formation in the unmodified surface is in its early stages, with particle diameters between 25 to 42 nm. This suggests a slow nucleation rate; as individual crystals haven't yet coalesced into a continuous layer. In comparison, the apatite formed in modified surface with higher density and the individual nanoparticle aggregated into large clusters, the measurement diameter ranging from 41 nm to 50 nm **Figure 7 b1** and **7b2**). This surface is covered in a three- dimensional porous network of apatite, which is more suitable for integration with biological tissue compare with unmodified surface. The enhanced formation of bone-like apatite on the modified surface can be attributed to its higher surface roughness and greater hydrophilicity. The increased hydrophilicity of the coating grown on the modified surface with DBD plasma promotes better interaction with physiological media, resulting in enhanced bioactivity. In addition, the nanostructure surface topography also offers more nucleation sites, facilitating faster and denser apatite growth.

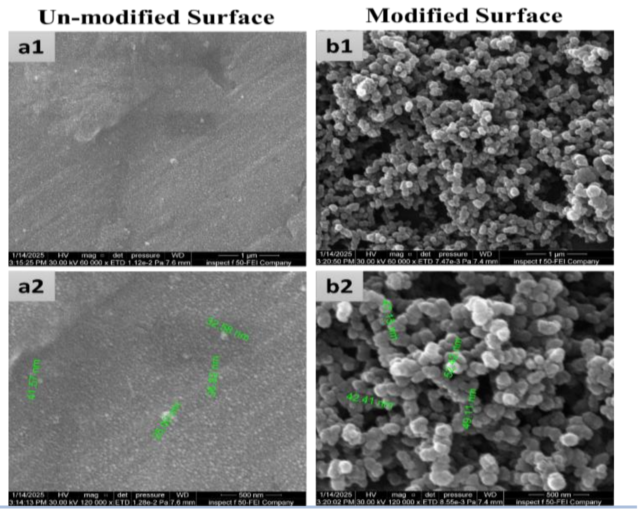

Figure 7. FESEM morphology at different magnification after immersion in SBF for 28 days. (a1 and a2) for unmodified surface, (b1 and b2) modified surface with DBD plasma..

Conclusion

This study compares the hydrothermal growth of Sr-HA coating on unmodified and modified surfaces of Ti6Al4V alloys with DBD plasma to evaluate the surface modification on their coating performance, as well as the biological response in vitro. The results indicate that DBD plasma etched the surface, increasing roughness, which led to a larger surface area, thus improving the adhesion of the nanostructure coating efficiency. In addition to increase in surface hydrophilic in which average CA changed from 80.2° to 51.5° for coating growth on unmodified and modified surfaces, respectively. Electrochemical corrosion measurements demonstrated a significant enhancement in corrosion resistance after coating growth on surface modification with DBD plasma which varying Rp from 5 kΩ.cm2 unmodified surface to 13.3 $k\Omega.cm^2$ for modified surface. The bioactivity in SBF solution revealed the DBD plasma enhance the formation of apatite growth. Overall, the use of surface modifies with DBD plasma enhance the hydrothermal coating density which subsequently improve the biological and corrosion properties of Ti6Al4V, suggesting its strong potential for biomedical implant applications. Future work could investigate the effect of DBD plasma exposure time on coating growth, and biological and corrosion properties of the Ti6Al4V.

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Authorship Contribution

The authors contributed equally to the article.

Declaration of Competing Interests

The authors confirm that the publication of this paper is not impacted by any conflicts of interest.

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