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Comparative analysis of platelets adhesion to the surface of Ta-based ceramic coatings deposited by magnetron sputtering

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Abstract. The adhesion of platelets to the surface of implants and stents depends on the material's properties, such as surface roughness, topography and wettability. The present study analyzes the composition and surface properties of nanostructured Ta, Ta₂O₅ and TaON coatings and their correlation with the adhesion of platelets. The surface morphology and topography were observed by atomic force microscopy and electron scanning microscopy; while the elemental distribution and chemical composition were analyzed by energy dispersive X-ray spectroscopy. The coated surfaces' advancing contact angles were evaluated by tensiometric measurements. The surface free energy (SFE) was calculated by the Owens, Wendt, Rabel and Kaelble method. The results of *in vitro* tests demonstrated that the deposition of Ta-based coatings leads to a decrease in the platelets adhesion and agglomeration ability.

1. Introduction

The progress in the implementation of anti-proliferative coated stents in cardiology and cardiovascular surgery results in the minimization of the risks of postoperative complications, inflammations, thrombosis, and repeated revascularizations [1-4]. Modifying the surface of a medical implant or stent by forming ceramic coatings is a promising way of improving the functional properties and biocompatibility of modern medical devices [5, 6]. Hemo-compatible materials and coatings should prevent coagulation and adhesion and aggregation of platelets. The platelets have specific receptors in the cell membranes which interact with fibrinogen. After fibrinogen is fixed to the surface, the process of platelets adhesion, aggregation, and secretion of granules is activated [7]. The adhesion of platelets to the surface of an implant or stent depends on the material composition, surface roughness, topography and wettability [8].

The main requirements to the coating materials for biomedical applications are good coating-substrate adhesion strength, good mechanical properties, and biocompatibility. The Ta-based coatings possess high hardness, elastic recovery and resistance to cracking in comparison with metals and other

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modern implant materials [9]. In addition, these coatings have demonstrated a good biocompatibility in *in vitro* tests. On the other hand, the challenging problem exists of reducing the adhesion platelets to the surface of Ta-based stent coating materials, which increases the risks of postoperative complications and in-stent restenosis. The evaluation thus becomes necessary of the degree of platelets adhesion, aggregation, and structural changing in view of further biomaterial applications.

The aim of the present study was to study the effect of the structural and morphological properties of the surface of Ta, Ta₂O₅ and oxynitride TaON films on the functional characteristics of ceramic coatings and their correlation with the adhesion of platelets in *in vitro* tests.

2. Materials and methods

Stainless steel samples (AISI 316) were used as substrates. The process of deposition of Ta, Ta₂O₅ and TaON coatings by magnetron sputtering was performed in a high-vacuum system with a base pressure of about 1×10^{-3} Pa using a tantalum target with a diameter of 170 mm. The magnetron discharge power was 4 – 5 kW. An ICP source was used for oxygen activation with an RF power of up to 1 kW. The main parameters of the deposition process were as follows: Ar pressure $p_{Ar} = 2.3 \times 10^{-1}$ Pa, oxygen mass flow rate of 35 sccm, nitrogen mass flow rate of 27 sccm, air mass flow rate of 25 sccm, magnetron voltage $U_m = 500 - 520$ V, magnetron current $I_m = 7.0 - 7.6$ A, total pressure $2.8 - 3.0 \times 10^{-1}$ Pa, coating deposition rate 6 – 8 $\mu\text{m}/\text{hour}$. An ion source (Radical type) was used for cleaning the samples surface before deposition [9].

The coatings thickness was measured by a Calotest device. The surface morphology and topography were observed by atomic force microscopy (AFM, Bruker, USA) and electron scanning microscopy (SEM, JSM-7100F). The elemental distribution and chemical composition of the coatings were analyzed by energy dispersive X-ray (EDX) spectroscopy (Oxford Link ISIS 300). The advancing contact angles of the coated surfaces were evaluated by tensiometric measurements using a DSA 100 E apparatus (Kruss, Germany). Three test liquids were used: water, glycerol and diiodomethane. The surface free energy (SFE) was calculated following the Owens, Wendt, Rabel and Kaeble method [10-12]. Standard optical microscopy (Micro 200, Belorussia) and AFM methods for platelets observation were used. Deoxygenated blood was extracted and anticoagulated by sodium citrate Na₃C₆H₅O₇. Blood plasma with platelets was placed on the samples surface and incubated at 37 °C. After incubation of the samples with platelets plasma, the surfaces were washed by phosphate buffer Na₂HPO₄ (pH ~7) in order to remove the non-adhered platelets, and then dried. The adhesion, shape changes, aggregation degree, and granules secretion of the platelets were analyzed.

3. Results and discussion

X-ray diffraction profiles and XPS spectra of nanostructured Ta-based films have been analyzed previously [9] and the formation has been demonstrated of coatings with stoichiometric composition. Figure 1 shows SEM images of Ta, Ta₂O₅ and TaON coatings deposited by us by magnetron sputtering. The coatings surface was smooth and uniform, without cracks and delamination. The EDS spectra revealed the presence of the main characteristic elements, namely, tantalum (Ta), oxygen (O), and nitrogen (N).

The thickness and surface parameters of Ta, Ta₂O₅, TaON coatings – surface roughness and surface free energy (SFE), are presented in table 1. The deposition of Ta₂O₅ and Ta coatings smooths the surface and increases its hydrophobicity, in comparison with the stainless steel samples (AISI 316).

As a parameter closely related to the coatings' thickness, the roughness parameters exhibit minimal values in the case of Ta₂O₅ and Ta films. The results of the advancing contact angle measurements of the deposited films are in good agreement with values reported previously [12, 13]. The SFE values for the Ta and Ta₂O₅ coatings reveal a higher hydrophobicity of the respective surfaces.

Standard optical and advanced AFM technique (figure 2) were used to evaluate the platelets adhesion to the coated substrates. For a qualitative assessment of the activation degree of platelets on the surface, three morphological classes were selected: weakly activated cells (spherical with pseudopodia), strongly activated (completely flattened on the surface), and aggregates.

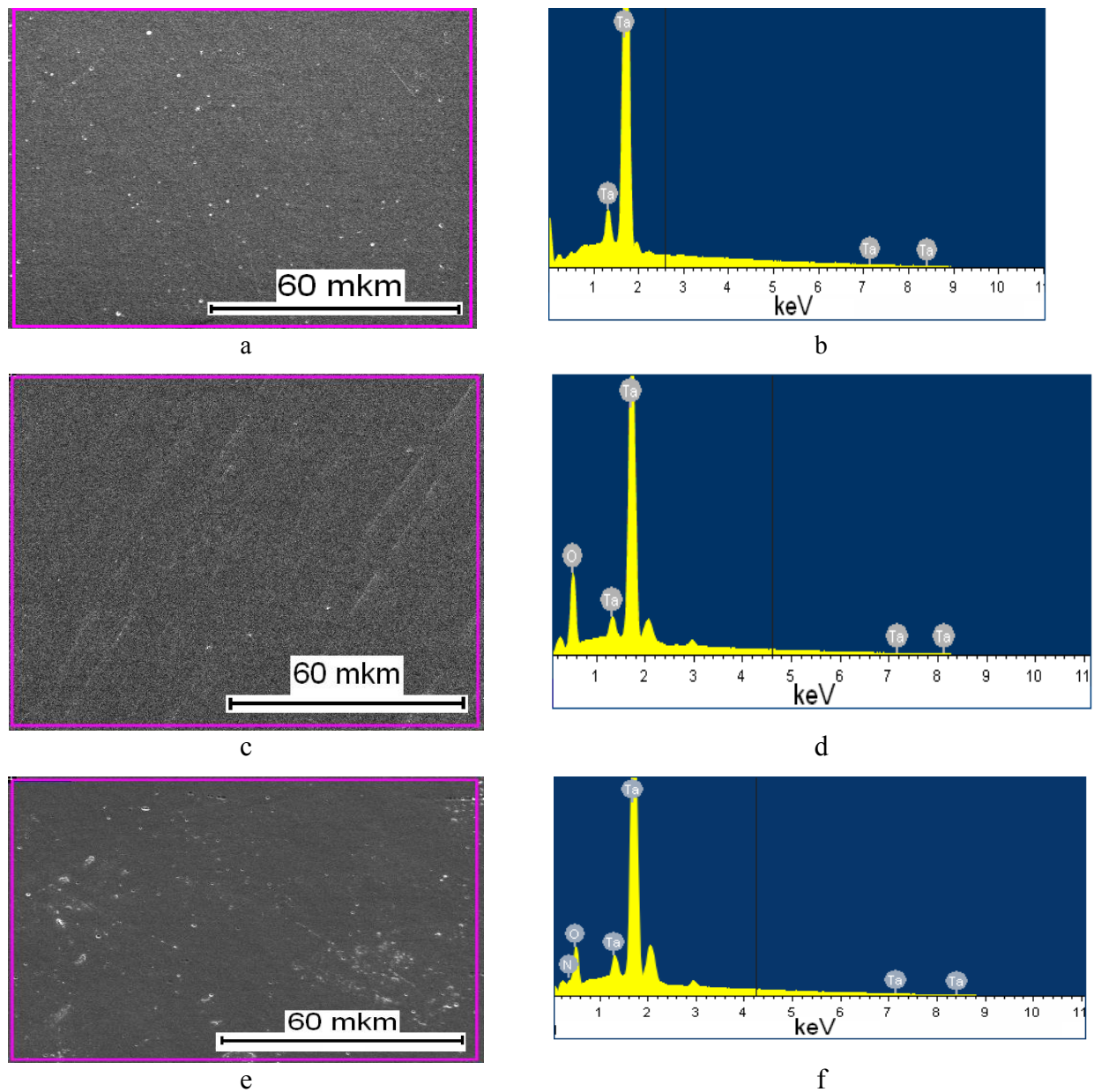


Figure 1. SEM images (a) and EDS spectra (b) of Ta, Ta₂O₅ (c, d), TaON (e, f) coatings deposited by magnetron sputtering on stainless steel (AISI 316) substrates.

Table 1. Thickness and surface properties of nanostructured Ta, Ta₂O₅, TaON coatings deposited by magnetron sputtering

	Thickness, μm	Roughness R_a , nm	Roughness R_q , nm	Advancing contact angle, °	SFE, mN/m
SS (AISI 316) substrate		7.4	9.0	75.0	31.7
Ta	0.89	6.2	8.3	84.2	29.1
Ta₂O₅,	1.12	4.4	7.6	89.0	31.1
TaON	0.91	7.5	9.9	94.1	37.8

Previous works have detected cytotoxic effects and major changes in the cells' metabolic activity on the surface of Ta-based coatings depending on the surface chemistry [14]. In the present study, the platelets adhesion was analyzed with regard to the chemical composition and surface properties of the Ta-based films.

The Ta-coated surfaces exhibited a uniform surface aggregation (figure 2 a). On the contrary, non-uniform surface aggregation was seen in the case of TaON films (figure 2 c).

The minimal aggregation was observed in the case of Ta₂O₅ coatings (figure 2 b). On their surface, the individual platelets were weakly activated and had a spherical shape with minimal pseudopodia. In addition, destroyed platelets were observed on both Ta₂O₅ and TaON coatings.

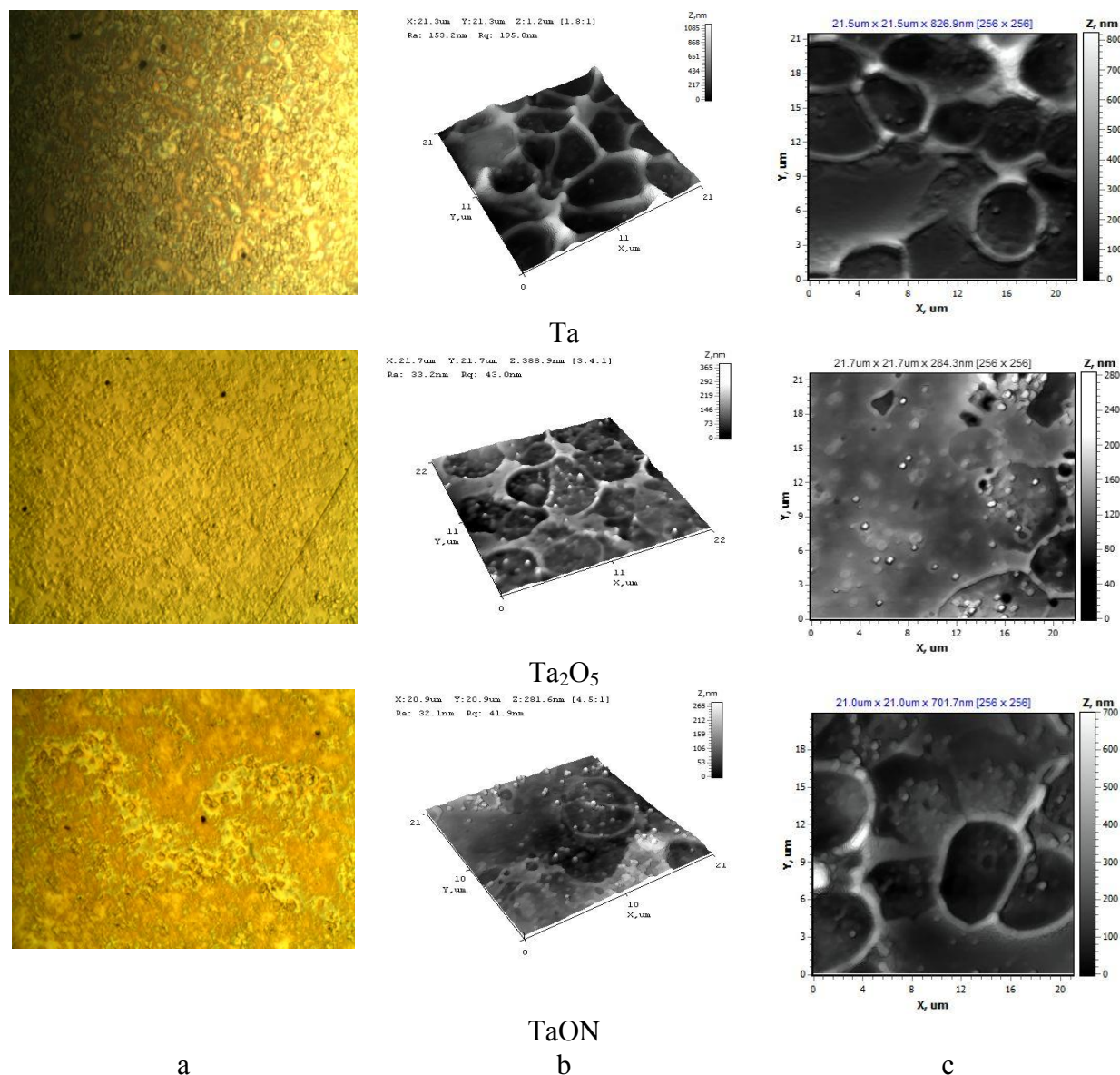


Figure 2. Optical (a, magnification $\times 400$) and AFM images (b, c) of Ta, Ta₂O₅, TaON coatings deposited by magnetron sputtering.

4. Conclusions

Our results demonstrated that the deposition of Ta-based coatings reduces the surface roughness parameters, while the surface properties acquire a more hydrophobic character in comparison with the stainless steel samples (AISI 316). Such surface modifications decrease the platelets adhesion and

agglomeration ability. Thus, the deposition of nanostructured coatings is an effective way of increasing the hemo-compatibility of modern implants and stents in view of biomedical applications.

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