Chapter 41
Efect Of Sterylization And Long-Term Exposure To Artificial Urine On Corrosion Behaviour Of Metallic Biomaterials With Poly(Glikolide-Co-Kaprolactone) Coatings

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Abstract. Novel implants based on metallic alloys (stainless steel 316 LVM, Ti6Al4V ELI and Ti6Al7Nb alloys) prepared by self-developed method of surface modification are presented in this study. Implants were coated with biodegradable and bioresorbable layer of poly(glycolide-co-caprolactone). Next, half of the perpetrated specimens was subjected to radiation sterilization. After that, samples were divided in to two groups: I - initial state (samples with sterilized "S" and non-sterilized polymer "NS"; II - samples with sterilized "S90" and non-sterilized polymer coating "NS90" after 90 days of exposure to artificial urine. As a result of the conducted research, it was found that after long-term exposure the polymer degradation was even and inconsiderable. However, reduction of adhesion and finally, retraction of the coatings on the titanium alloys has been observed. Moreover, the influence of sterilization process on the kinetics of metallic ion release to corrosion environment for all metallic biomaterials has been observed.

Keywords: artificial urine, corrosion resistance, metallic biomaterial, polymer coatings, poly(glycolide-co-caprolactone)

41.1 Introduction

Stenosis of the ureter or urethra may lead to urinary retention, the consequence of which is a direct threat of the patient health. It may cause permanent damage to some internal organs, especially kidneys [1]. The main reasons for stenosis of ureters and urethra are prostatic hyperplasia, cancer of an ureter and urethra, and narrowings as a consequence of the operation on the urinary tract [1]. In order to

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reduce the consequences of urine retention, various procedures are used to expand and maintain urethral or ureteral patency. One of the basic method of treatment is the use specially designed urological catheters - urological stents. Continuous development of techniques are increasingly demanding for materials used for this type of implants. This applies particularly to their physical and chemical properties, which should be tailored to the urine environment, in which urological implants are staying.

Still unsolved, a very important issue is the problem of encrusting the surface of these implants. It involves a partial or complete overlap of the surface of the crystals of calcium phosphate in the form of hydroxyapatite and magnesium ammonium phosphate. The gradual overgrowing of implant lumen leads to obstruction and may stop the flow of urine. This contributes the need of removing the stent in a surgical revision procedure and provide a new one. [2, 3, 4].In many studies on the problem of encrustation it was observed that urinary tract infection in the presence of the implant increases the deposition process of these crystals [5]. Degradation of the implanted biomaterial takes place in an environment of human body fluids predominantly from the operational and mechanical loads and corrosive action of the environment [7]. Corrosion resistance is dependent on the type of metallic biomaterial and condition, morphology and the crystal structure of its surface layer. Therefore, improvement of functional properties of metal implants used in urology, but also in other fields of medicine, such as interventional cardiology or orthopaedics and traumatology, can be obtained by mechanical and electrochemical processing and modify the surface layer of biomaterials [7, 8]. Besides modification of the surface itself of the metal biomaterial, coatings made of polymeric biomaterials or biodegradable polymers which release substances can be used. The purpose of these layers is to prevent the development of inflammatory conditions caused by bacteria or fungi, reducing the risk of incrustation. Additionally, for long-term implants, biodegradable polymer coating may assist in the healing process by enabling the epithelial tissue coverage of the stent [10, 11, 12]. At the same time, the metal structure of the implant will provide continuous treatment of the narrowed sections of the ureter or urethra [13].

Considering the problems, involved use of metallic biomaterial in urology and their surface modification perspectives, in this study were presented the results of corrosion resistance of metallic biomaterials with bioresorbable polymer coatings in the environment simulating human urinary tract. The aim of the work was to determinate the influence of long-term exposure to artificial urine and radiation sterilization on corrosion resistance of metallic substrate and degradation process of the polymer coating.

41.2 Materials and Methods

In the tests, samples of austenitic stainless steel 316 LVM as well as Ti6Al4V ELI and Ti6Al67Nb titanium alloys were used. Their chemical composition, microstruc-

ture and mechanical properties met the requirements according to standards ISO 5832-1, ISO 5832-3 and ISO 5832-11 respectively. The samples were obtained from rods of 6 mm diameter. Prior to polymer coating the surfaces were ground with grade 600 grinding paper. Titanium alloy samples were additionally subjected to anodic oxidation in phosphoric and sulphuric acid bath at potential 97V. Next, the polymer coating based no poly(glycolide-co-caprolactone) - G-Cap (10/90) was applied to the metal substrate. G-Cap was synthesized in bulk at temperature equal to 120°C by the ring opening polymerization of glycolide with the use of Zirconium (IV) acetylacetonate (Zr(acac)4) as a non-toxic initiator. Specimens were coated with the G-Cap by dip-coating method. Metal rods were immersed into polymer (10% w/w) solution in dichloromethane for 30 seconds and dried at air for one day and then in a vacuum dryer. The G-Cap polymer used in the research was both developed and deposited on metallic substrate at the Centre of Polymer and Carbon Materials, Polish Academy of Sciences in Zabrze, Poland. The samples with polymer coatings were divided in to two groups. One of them was subjected to a radiation sterilization (sterilized sample was marked as "S", no sterilized as "NS"). A radiation dose of 25 kGy with an energy of 10MeV was used. Next half of the specimens ("S" and "NS") was long-term (90 days) exposed to 0.1 dm^3 of artificial urine [14] with the following composition: NaCl - 6.17 g/dm^3, KCl - 4.75 g/dm^3, $NaHPO_4$ - 4.35 g/dm^3, Na_2SO_4 - 2.41 g/dm^3, $MgSO_4$ - 0.99 g/dm^3, $C_6H_5Na_3O_7$ - 0.94 g/dm^3 distilled water, (pH = 5.5). The samples kept in artificial urine were marked as "S90" and "NS90" respectively for sterilized and no sterilized ones.

The morphology of the samples before and after corrosion study was analyzed using multimode atomic force microscopy (AFM) (MultiMode, di-Veeco, USA, CA), and stereoscopic microscope Zeiss Stereo Discovery V8 with digital camera MC5s with magnification 4x-24x.

Pitting corrosion tests were carried out by potentiodyamic method as recommended by the PN-EN ISO 10993-15 in artificial urine solutine. The test set up consisted of the VoltaLab PGP201 potentiostat, saturated calomel electrode SCE as the reference electrode, the auxiliary electrode (platinium wire), the working electrode (test samples) and the PC with VoltaMaster 4 software. The corrosion test started with establishing the open circuit potential E_{OCP} at currentless condition during the time T= 120 min. The polarization curves were recorded starting with the potential value $E_{start} = E_{OCP}$ - 100 mV. The applied scanning rate was equal to 3mV/s. Once the anodic current density reached the value of 1mA/cm^2, or potential E reached the value of +4V, the polarization direction was changed. On the basis of the obtained curves the breakdown potential, E_b and the repasivation potential E_{cp} were determined. Moreover the value of the corrosion potential E_{corr} and the polarization resistance R_p were calculated with the use of Stern method. The potentiodynamic test of pitting corrosion resistance was carried out for all groups of samples.

Metal ion concentration in artificial urine, following 90-day exposition of samples was measured with JY 2000 spectrometer by Yobin-Yvon with the use of inductively coupled plasma-atomic emission spectrometry (ICP-AES).

Degradation process of polymer coating was conducted with the use of nuclear magnetic resonance spectroscopy - NMR spectroscopy (Bruker-Avance II Ultra-

shield Plus spectrometr, 600 MHz). Especially change of the comonomeric unit composition during degradation were monitored. The molar mass and molar mass distribution of the polymers were determined by gel permeation chromatography - GPC (Physics SP 8800 chromatograph and detectors: UV, Spectra-Physicsi differential refractometer, Shodex SE 61, Viscotek RI VE3580 detector and viscometer Viscotek 270 Dual Detector Array).

41.3 Results and discussion

The AFM (Fig. 41.1) micrographs of the samples before degradation show that G-Cap coating was corrugated and homogenously distributed on the metal substrate. The surface of the implants was smooth with roughness parameter of RMS = 137.34 nm and Ra = 64.009 nm.

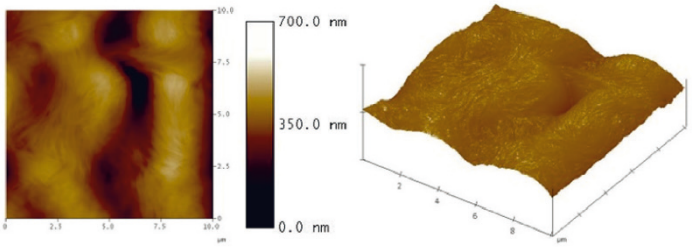

Fig. 41.1: Example AFM micrographs of the G-Cap coating on sterilized Ti6Al4V alloy (initial state - before degradation)

Macroscopic observations of specimens surface after long-term (90 days) exposure to artificial urine solution (Fig. 41.2b) showed a decohesion of polymer coating on 316 LVM stainless steel compared to its initial state before exposition. Particularly, the cracking of the coating and partial disintegration with the surface of the metallic biomaterial were observed. However, for Ti alloys, a complete delamination of the polymer coating was observed. The consequence of this was the sliding and exposing of the metal substrate - Fig. 41.2e and Fig. 41.2f. In addition, local thickening of the coating and air bubbles on the border of the polymer-metal substrate were observed - Fig. 41.2c and Fig. 41.2d.

The results of pitting corrosion test of metallic biomaterials with poly(glycolide-co-caprolactone) - G-Cap coatings in initial state and after long-term exposure to artificial urine are shown in table 41.1.

The polarization curves for all types of 316 LVM stainless steel samples coated with the polymer G-Cup coating were characterized by a hysteresis loop. Breakdown E_b and repasivation E_{cp} potentials have been identified. Occurring of pit-

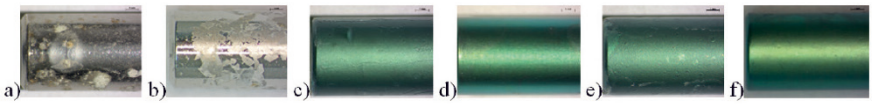

Fig. 41.2: Examples of samples surface, 316LVM: a) NS - no sterilized, b) NS90 - after 90 day of exposure, Ti6Al4V alloy: c) NS and d) NS90, Ti6Al7Nb alloy: e) NS and f) NS90

Table 41.1: Results of the pitting corrosion tests

		NS	S	NS90	S90
316LVM	E_{corr}, mV	18.0±4.5	91.5±4.9	-31.2±2.1	-112.7±22.8
	E_b, mV	+1550.5±22.1	+2286.5±269.5	+1396.0±152.6	+1410.0±43.2
	E_{cp}, mV	-10.5±4.7	-61.2±9.8	60.0±14.1	-71.0±18.7
	R_p,k$\Omega \cdot$ cm^2	26.8±3.6	208.2±48.4	0.4±0.02	0.5±0.2
Ti6Al4V ELI	E_{corr}, mV	55.2±4.8	-162.9±31.6	-75.0±11.5	-358.9±30
	E_b, mV	-	-	-	-
	E_{cp}, mV	-	-	-	-
	R_p, k$\Omega \cdot$ cm^2	55.1±9.6	60.9±5.3	89.9±24.7	2.2±0.4
Ti6Al7Nb	E_{corr}, mV	-15.5±1.2	-250±49.0	-167.5±18.4	-385.0±67.5
	E_b, mV	-	-	-	-
	E_{cp}, mV	-	-	-	-
	R_p, k$\Omega \cdot$ cm^2	191.1±3.4	14.0±3.3	47.3±17.6	12.0±1.4

ting corrosion for these samples were confirmed by macroscopic observations. Figs 41.2a and 41.2b show characteristic pitting corrosion pits.

It should be emphasized that for the samples in the initial state (not exposure to artificial urine) the corrosion products were stopped under polymer surface - Fig. 41.2a. After long-term exposure, the copolymer coating was cracked. As a result, partial decohesion of the coating and release of corrosion products into the electrolyte occurred.

The values of repasivation potential E_{cp} determined on the basis of the polarization curves were similar to the corrosion potentials E_{corr} for these samples. As a result of sterilization process it was observed increase of corrosion E_{corr} and breakdown E_b potentials in comparison to non-sterilized "NS" samples. Moreover, the value of polarization resistance R_p of the sterilized samples "S" were higher than non-sterilized ones "NS" - table 41.1. Long-term exposure of "NS90" samples in artificial urine caused reduction of all parameters describing the corrosion resistance in comparison to non-sterilized "NS" samples in the initial state. A similar trend was observed for sterilized in initial state "S" and after long term exposure "S90" 316 LVM stainless steel samples - table 41.1.

The results of corrosion tests of titanium alloys with polymer coatings shows resistance to pitting corrosion in whole measuring range (+4V). Neither the breakdown E_b nor transpassivation E_{tr} potentials were observed It needs to be highlighted that, for all titanium samples "S90" and "NS90" exposed 90 days in artificial urine,

decohesion of the polymer coating with metal substrate was observed. The process of separation of polymer coating from metal substrate was initiated around 75 days of exposure Figs. 41.2d and 41.2f.

Radiation sterilization has reduced the corrosion potential both for the Ti6Al4V ELI and Ti6Al7Nb alloys substrates. In case of the sterilized "S" and no sterilized "NS" Ti6Al4V ELI samples, the same values of polarization resistance R_p were observed - table 41.1. For the Ti6Al7Nb, it was found that the sterilization reduced the value of polarization resistance R_p in comparison to the non-sterilized samples. A similar relationship was observed for Ti6Al4V ELI and Ti6Al7Nb alloys with PLGA coating after long-time exposure to Ringer's solution [15].

Results of release of metal ions to artificial urine after long-time (90 days) exposure were converted into surface mass density and presented in table 41.2. The results, for the sterilized and non-sterilized samples after exposure, showed that the sterilization increased corrosion resistance of the metallic substrate. In particular, a decrease in mass density of Fe, Cr, Ni and Mo ions release was observed for 316 LVM stainless steel - table 41.2. The same was observe for the titanium alloys (Ti6Al4V ELI and Ti6Al7Nb) for Ti, Al, V and Nb ions - table 41.2. Similar effect of the radiation sterilization on metal ion release from metallic biomaterials coated with the PLGA coatings after long-term exposure to Ringer's solution were observed by authors [15].

Table 41.2: Results of surface mass density for metal ions

316LVM			Ti6Al4V			Ti6Al7Nb		
	NS90	S90		NS90	S90		NS90	S90
Surface mass density, $\mu g/cm^2$								
Fe	0.314±0.019	0.257±0.019	Ti	3.200± 0,038	2.467±0,024	Ti	2.905±0.019	2.062±0.019
Cr	3.933±0.099	3.252±0.038	Al	1.971± 0.038	1.762± 0.024	Al	1.714±0.019	1.157±0.048
Ni	1.610±0.038	1.219±0.071	V	1.124± 0.019	0.786± 0.043	V	-	-
Mo	0.429±0.038	0.210±0.019	Nb	-	-	Nb	1.200±0.038	1.019±0.038

NMR analysis revealed even way of degradation of the poly(glycolide-co-caprolactone) G-Cap coatings - the content of caproyl units in the polymeric chain remained on the same level during 90 days. There was significant decrease of molecular weight of PGCap (from 65 kDa to 15 kDa after 90 days), However, the dispersity (Mw/Mn) of the samples did not change - it was equal to 2. In the NMR spectra of poly(glycolide-co-caprolactone), there was no signals corresponding to the short chains of oligomers.The degradation process of the poly(glycolide-caprolactone) coatings proceeds quite slowly during 90 days. This behaviour is connected with the different microstructure of the chain, comparing to poly(lactide-co-glycolide): caproyl units are characterized by greater hydrophobicity and therefore have a slower hydrolysis rate.

41.4 Conclusion

On the basis of obtained results, it was shown that radiation sterilized poly (glycolide -co-caprolactone) - G-Cap polymer coating on 316 LVM stainless steel increased its corrosion resistance in comparison to non-sterilized ones. Especially increase of breakdown potential and polarization resistance were recorded - table 1. Moreover it was observed that long-term exposure to the artificial urine decreased corrosion resistance independently to the sterilization process. Despite NMR and GCP studies results which shows that the degradation process of the poly(glycolide-caprolactone) coatings proceeds quite slowly during 90 days, has also been found that, polymer coating was cracked contributing to unveling of the metal substrate. In addition especially for 316 LVM samples, it has been observed in the macroscopic study that prior to exposure, for polymer modified sample, the polymer coating contributes to the retention of corrosion product under its surface and effectively limits its penetration to the electrolyte. For Ti6Al4V ELI and Ti6Al7Nb alloys with G-Cap coating, there was no significant effect of radiation sterilization on corrosion resistance parameters determined by potentiodynamic studies. The same as 316 LVM stainless steel for titanium alloys, long-term exposure to artificial urine resulted in a decrease of corrosion resistance despite the process of sterilization. In addition, the polymer coating was removed from both titanium alloys substrate before the end of the 90-day test period. Separation of the coating from the substrate occurred about 75 days of exposure.

Results of surface mass density of metal ions released to the artificial urine for all kind of metallic biomaterials, both sterilized and non-sterilized after 90 days of exposure, showed that sterilization increased the corrosion resistance of the metallic substrate.

Summarizing the obtained results, it can be stand that biodegradable pol (glycolide-caprolactone) - G-Cap coating allows for controlled release of the active substance. Furthermore, radiation sterilization with an electron beam of 25 KGy and an energy of 10 MeV can be used as a surface treatment of metallic biomaterials intended for urological implants. Both polymer coating and sterilization process decrease corrosion resistance of metallic biomaterials substrates. However, it is still necessary to further improve the chemical composition of the G-Cap polymer coating. Especially proportion of components to ensure better adhesion of the layer to the metallic substrate during long-term exposure to artificial urine. Also, it is very important to improve process of polymer coating deposition of metallic substrate to avoid air bubbles and other disadvantages e.g. uneven layer thickness. It should provide greater accuracy, repeatability and thickness control.

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References

1. Borkowski A.: Urologia: Podręcznik dla studentów medycyny. PZWL, Warszawa (2008)
2. Jacobse S.M., Strickler D.J. Mobley H.L., Shirtliff M.E.: Complicated catheter–assiociated urinary tract infections due to Escherichia coli and Proteus mirabilis. Clin Microbil Rev 21(1), 26–59 (2008)
3. Shaw G.L., Choong S.K., Fry C.: Ecrustation of biomaterials in the urinary tract. Urol Res 33(1), 17–22 (2005)
4. Ostrowska K., Strzelczyk A., Różalski A., Strączek P.: Biofilm bakteryjny jako przyczyna zakażeń układu moczowego — mikroorganizmy patogenne, metody prewencji i eradykacji. Postepy HigMed Dosw 67, 1027–1033 (2013)
5. Klimek L., Różański W., Jabłonowski Z., Sosnowski M., Kliś R.: Obserwacje zmian mikroskopowych cewników typu Double J w zależności od czasu utrzymywania ich w drogach moczowych. Inzynieria Biomaterialow 43-44, 36–39 (2005)
6. Kajzer A., Kajzer W., Gołombek K., Knol M., Dzielicki J., Walke W.: Corrosion resistance, eis and wettability of the implants made of 316 LVM steel used in chest deformation treatment. Archives of Metallurgy and Materials, 61 2a, 767–770 (2016)
7. Szewczenko, J.; Marciniak, J.; Kajzer, W.; Kajzer A.: Evaluation of corrosion resistance of titanium alloys used for medical implants. Archives of Metallurgy and Materials. 61 2 695–699 (2016)
8. M. Bartmański, B. Świeczko-Żurek: Project of hip joint endoprosthesis for an individual patient with materials selection, Advances In Materials Science, 15, 1 (43), 30–36 (2015)
   Solid State Phenomena Corrosion and Surface Engineering 227 491–494 (2015)
9. Basiaga M., Paszenda Z., Karasiński P., Kajzer A., Walke W.: Mechanical Properties of Anodically Oxidized cpTi and Ti-6Al-7Nb Alloy Applications of Computational Tools in Biosciences and Medical Engineering. Adv Struct Mater 71. Chapter 8 Springer 123–132 (2015)
10. Roe D., Karandikar B., Bonn–Savage N., Gibbins B., Roullet J.B.,: Antimicrobial surface functionalization of plastic catheters by silver nanoparticles. J Antimicrob Chemother 61, 869–876 (2008)
11. Trupti A., Lauserpina C., Shanta M.: Efficacy of a silicone urinary catheter impregnated with chlorhexidine and triclosan against colonization with proteus mirabilis and other uropathogens. Infect Control Hosp Epidemiol 28, 596–598 (2007)
12. Hachem R., Reitzel R., Borne A., Jiang Y., Tinkey P., Uthamanthil R., Chandra J., Ghannoum M. Raad I.: Novel antiseptic urinary catheters for prevention of urinary tract infections: correlation of in vivo and in vitro test results. Antimicrob Agents Chemother 53, 5145–5149 (2009)
13. Lee G., Marathe S., Sabbagh S., Crisp J.: Thermo-expandable intra-prostatic stent in the treatment of acute urinary retention in elderly patients with significant comorbidities. Int Urol Nephrol 37, 501–504 (2005)
14. Opalko F.J., Adria J.H., Khanc S.R.: Heterogeneous nucleation of calcium oxalate trihydrate in artificial urine by constant composition. J Cryst Growth 181, 410–417 (1997)
15. Szewczenko J., Kajzer W., Grygiel-Pradelok M., Jaworska J., Jelonek K., Nowińska K., Gawliczek M., Libera M., Marcinkowski A., Kasperczyk J.: Corrosion resistance of PLGA-coated biomaterials. Acta of Bioengineering and Biomechanics 19 1 173–179 (2017)