ACADEMIA ROMÂNĂ
Revue Roumaine de Chimie
http://web.icf.ro/rrch/

Rev. Roum. Chim.,
2014, *59*(6-7), 523-526

Dedicated to the memory of
Professor Eugen Segal (1933-2013)

SURFACE CHARACTERIZATION OF PERITONEAL DIALYSIS CATHETER CONTAINING SILVER NANOPARTICLES

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Received April 28, 2014

For the end-stage renal disease patients, peritoneal dialysis is a kidney replacement therapy. Despite being a lifesaving treatment, the rate of mortality in patients is elevated, mainly due to the chronic peritoneal dysfunction which is characterized by inflammation, peritoneal fibrosis and neoangiogenesis. This paper reports on a study concerning helium plasma followed by silver nitrate treatments on peritoneal dialysis catheter in order to obtain antimicrobial proprieties and to reduce the infection rate on patients.

INTRODUCTION

Over 150,000 people in 2009 were on continuous ambulatory peritoneal dialysis treatment[1] for end-stage renal failure, representing some 15% of the global dialysis population, the alternative being expensive and life-style-limiting haemodialysis, or renal transplantation where possible. It is expected in the next years that the numbers of patients requiring dialysis will increase, especially in developing countries.[2,3] Peritoneal dialysis catheters are at risk from infection throughout their use, which is intended to extend for months or years, the most frequent routes of infection being from the skin exit site, the tissue tunnel associated with the catheter and the catheter lumen. The silver-ion treated catheters in reducing dialysis-related infections are included in the attempts to reduce infection in patients undergoing peritoneal catheters,[4] but the methods of obtaining silver ions at catheter surfaces differs and the antimicrobial activity time differs. To date, there are only few clinical studies[4-8] evaluating the efficacy of silver-ion implanted peritoneal access devices. The potential benefit of silver-ion treated peritoneal dialysis catheters was extrapolated from favorable experience with intravascular appliances. Sterling silver disk attached to the anterior abdominal wall protected against catheter infection-related peritonitis up to 4 weeks postimplant in a short-term animal study.[9] New surface-treatment procedures for peritoneal dialysis catheters that can reduce the rate of infectious complications without adversely affecting basic design and function would constitute a major advance in the field.

The aim of the present study was to obtain silver-treated catheters in order to reduce dialysis-related infections in chronic renal failure patients. Catheters were processed using a method in two

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steps: helium plasma treatments followed by silver nitrate chemical modification.[10-12]

EXPERIMENTAL

1. Materials and methods

1.1. Plasma treatments

The experiments were performed in an EMITECH RF plasma device at a power of 60 W for 5 min. As background gas was used helium at a pressure p = 10^{-2} mbar, the catheter being immersed into the plasma that fills the gas vessel by diffusion. For helium plasma, only etching and cross-linking of the catheter material without introduction of chemical groups take place during the treatment. This is produced by forming free radicals at or near the surface which interact forming cross-linking structures and unsaturated groups. Most catheters are manufactured of either silicone elastomer or polyurethane. Silastic tears easily and has limited tensile strength; urethanes are soluble in organic solvents and can be physically degraded by aldehydes, alcohols, and ethers.

1.2 NaOH/AgNO3 chemical modification

In order to have the highest yield of silver possible in a reproducible fashion for the NaOH/AgNO3 chemical modification, the plasma-treated catheter was immersed for 14 days at room temperature in a 0.2 m solution of $AgNO_3$, protected from light.[10] When 2-week period expired the samples were rinsed with deionized water, subsequently analyzed by the different characterization techniques.

2. Characterization methods

The catheters surface morphology was investigated by atomic force microscopy (AFM) methods which were performed at room temperature (22–24 ºC) on a Solver PRO-M (NT-MDT Co., Zelenograd, Moscow, Russia) setup. The last version of the NT-MDT NOVA software was used for image acquisition and analysis. Topographic images, obtained over an area of 5 x 5 µm for each sample, were recorded by using AFM tapping mode. A rectangular silicon cantilever NSG10 with the typical force constant of 11.8 N/m, resonance frequency of 213 kHz and tip curvature radius and height of 10 nm and 14–16 mm, respectively, was used. The surface textures were characterized in terms of roughness parameters, such as root mean square roughness (Sq) and average height. Following X-ray diffraction (XRD) method, the X-rays were generated by using a Cu Ka source from a D8 Bruker diffractometer with an emission current of 36 mA and a voltage of 30 kV. Scans were collected over the 2h = 2 – 60 º ranges using a step size of 0.01º and a count time of 0.5 s/step. The semi-quantitative analysis of the diffractograms was performed with an EVA soft from DiffracPlus package and a ICDD-PDF2 database based on the patterns' relative heights. Small angle X-ray scattering (SAXS) experiments were performed on a Bruker Nanostar instrument. The X-ray radiation employed was generated from a Cu sealed tube fine-focus X-ray source (Kα = 1.54184Å with a potential of 40 kV and a current of 35 mA). The X-rays were filtered through cross-coupled Gobel mirrors and collimated by pin-holes. The detector was set at 107 mm from the sample and the sample chamber and x-ray paths were evacuated. The detector distance and beam intersection was calibrated employing a silver behenate standard. The instrument was controlled with the SAXS software suite and the data was collected in the still (add) mode. The sample chamber and x-ray beam paths were evacuated and a 1800 sec scan was preformed. The glassy carbon standard was then inserted between the sample and the detector and a 300 sec standard scan was collected. Finally a 1800 sec background scan (air only) which was followed by insertion of the glassy carbon and a 300 sec standard scan were preformed. Transmission factors were then calculated for the treated sample and the blank. The blank was then subtracted from the treated sample employing the scale factor calculated by dividing the sample transmission factor by the blank transmission factor. An area integration was employed to reduced the data to a one-dimensional q (4πsinθ/λ) versus ln(Intensity) trace.

RESULTS AND DISCUSSIONS

Significant changes in the surface topography induced by plasma followed by silver nitrate treatments on catheters were also evidenced by using tapping-mode AFM experiments. The bi- and tridimensional phase images of the native catheter film (see Fig. 1a) reveal a surface, with Sq of 9.5 nm. In addition, the histogram taken along the solid line from 2D image and the average profile, shown in Fig. 1b), and Fig. 1c), allowed the calculation of the average height, the value of 25 nm being obtained.

Following silver reduction onto catheter plasma treated surfaces, the AFM images highlight the distribution of silver nanoparticles, evidencing particles agglomeration of irregular stick appearance (formed by small spherical-like silver particles of sizes about 40-60nm), (see Fig. 2 a,b,c). The surface was characterized by a Sq of 13.5 nm and an average height of 45 nm.

The crystallographic structure and composition of the treated samples was studied by X-ray diffraction.

The XRD peaks of peritoneal dialysis catheter (Fig. 3) indicate the presence of silver (68,8 %) – ICDD card 00-001-1164, silver oxide (30,3 %) – ICDD card 00-019-1155, and a small amount of silver nitrite (0,9 %) – ICDD card 00-001-0823.

It is important to notice from Fig. 4 that the SAXS pattern changes significantly after plasma followed by silver nitrate treatments. This shows significant morphological changes within the catheter sample, indicating the Ag nanostructures can rearrange themselves depending upon counterionic environment. The Guinier analysis:

$$\ln(\Delta R(\theta)) = 1 - \left(R_g^2/3\right)q^2 \qquad (1)$$

refers to the analysis of the SAXS scattering curve at very small scattering angles. This analysis allows for the direct estimation of two SAXS invariants, the radius-of-gyration, Rg, and the extrapolated intensity at zero scattering angle, I(0). We present the Primus version for determining

Rg[13] and the results are shown below. Fitting of the obtained SAXS profiles with spherical models gives the size distribution profiles for Ag nanostructures in the case of treated catheter and the results are in good concordance with AFM measurements (Rg = 50 nm).

The availability of ionic silver for bacterial contact affects antibacterial activity of silver containing materials.[14] Structural changes and deformation in bacterial cell walls and membranes that lead to disruption of metabolic processes followed by cell death are caused by the positively charged silver ions which can interact with negatively charged biomacromolecular components (phosphate and disulfide or sulfhydryl groups of enzymes) and nucleic acids.[15] It is believed that the antimicrobial action of silver nanoparticles also depends on the surface area of nanoparticles which are most likely to act as reservoirs for releasing the Ag ions. The antimicrobial mechanism of silver nanoparticles has also been suggested to be related to membrane damage due to free radicals derived from the surface of the nanoparticles.[16] Moreover, silver nanoparticles may accumulate in the bacterial cytoplasmic membrane, causing a significant increase in membrane permeability and leading to cell death.[17]

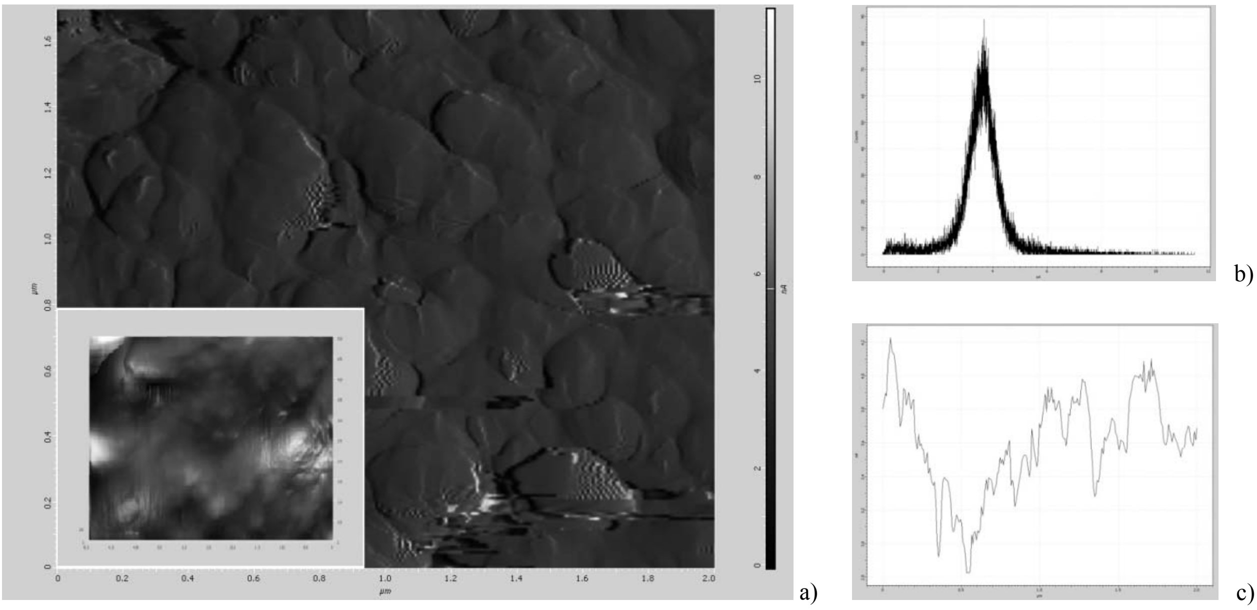

Fig. 1

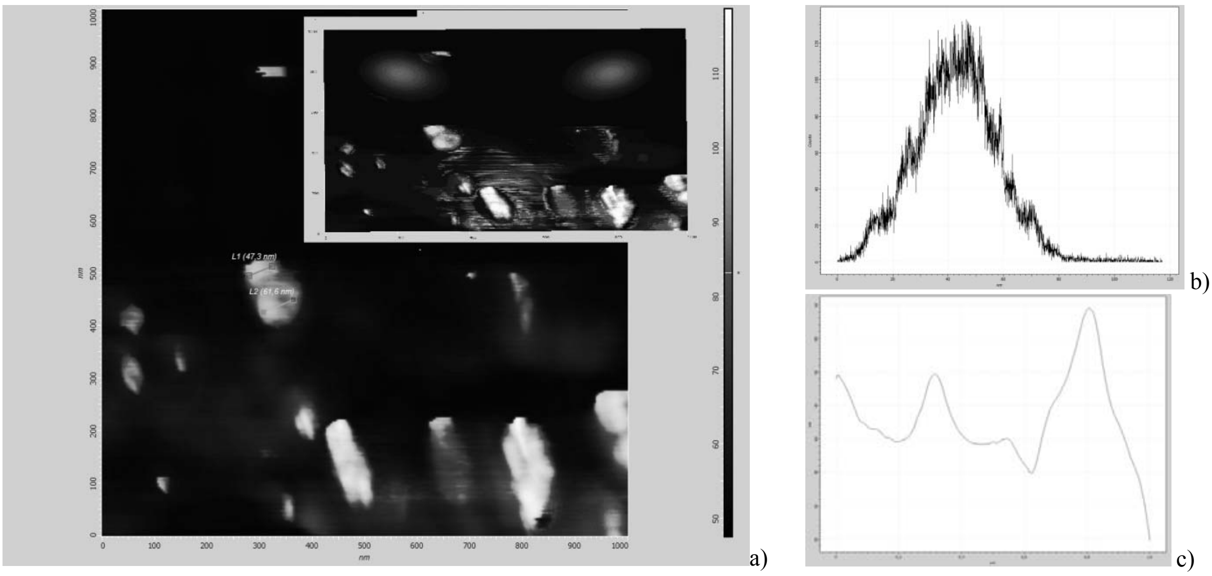

Fig. 2.

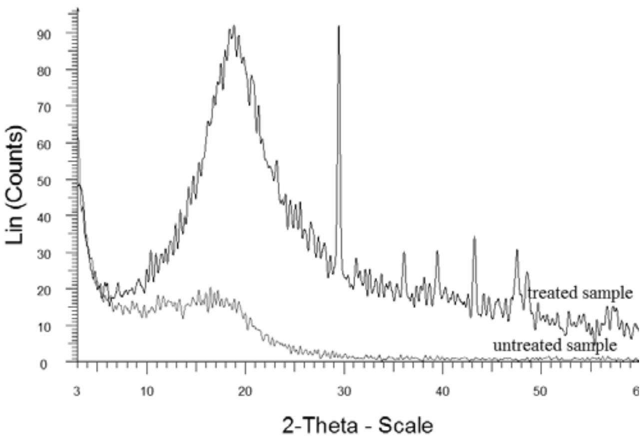

Fig. 3

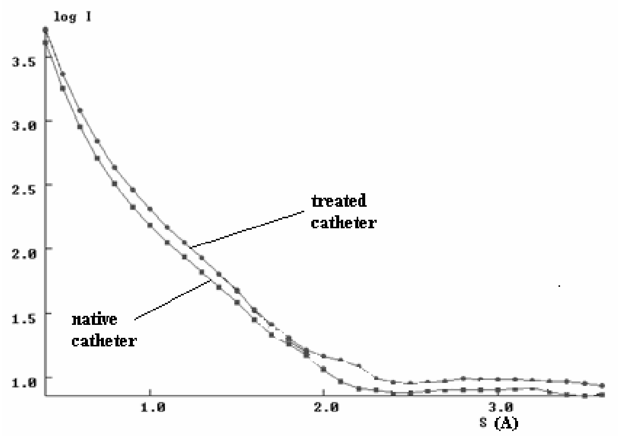

Fig. 4

CONCLUSIONS

The addition of metallic silver components to peritoneal dialysis access devices has been tried as a means of preventing dialysis-related infections. The AFM, XRD and SAXS methods confirm the presence of silver nanoparticles in the treated peritoneal dialysis catheters.

Thus, it is reported that the helium plasma followed by silver nitrate treatment is an appropriate method for obtaining peritoneal dialysis catheters with possible antimicrobial proprieties.

Acknowledgments: The author thanks to dr. Iuliana Stoica for AFM acquisition data. The research leading to these results has received funding from the CNCSIS- UEFISCD project PN-II-RU-TE-0123 no. 28/29.04.2013, "Phosphorus-containing polymers for high performance materials used in advanced technologies and/or biomedical applications". Paper dedicated to the 65th anniversary of "Petru Poni" Institute of Macromolecular Chemistry of Roumanian Academy, Iaşi, Roumania.

REFERENCES

1. E. Thodis, P. Passadakis, N. Lyrantzopooulos, S. Panagoutsos, V. Vargemezis and D. Oreopoulos, *D. Int. Urol. Nephrol.*, **2005**, *37*, 379.
2. V.C. Chitalia, A.F. Almeida, H. Rai, M. Bapat, K.V. Chitalia, V.N. Acharya, *Kidney Int*., **2002**, *61*, 747.
3. S. Vikrant, *Indian J. Nephrol*., **2007**, *17*, 165.
4. J.H. Crabtree, R.J. Burchette, R.A. Siddiqi, I.T. Huen, L.L. Hadnott, A. Fishman, *Peritoneal Dialysis International*, **2003**, *23*, 368.
5. R. Bayston, L.E. Fisher, K. Weber, *Biomaterials*, **2009**, *30*, 3167.
6. C.Y. Kim, A. Kumar, L.A. Sampath, K. Sokol, S.M. Modak, *Am. J. Kidney Dis.*, **2002**, *39*, 165.
7. M.K.Dasgupta, *Perit. Dial. Int*., **1997**, *17*(Suppl. 2), 142.
8. M. Aflori, "RF plasma. Diagnosis and applications", 2013, ed Technopress.
9. S.R. Ash, *Perit. Dial. Int*., **1988**, *8*, 64.
10. D.J. Balazsa, K. Triandafillub, P. Woodc, Y. Chevolota, C. van Deldenc, H. Harmsb, C. Hollensteind, H. Mathieua, *J. Biomaterials*, **2004**, *25*, 2139.
11. M. Aflori, M. Drobota, D. Gh. Dimitriu, I. Stoica, B. Simionescu, V. Harabagiu, *Mat. Sci. Eng. B*, **2013**, *178*, 1303.
12. M. Drobota, M. Aflori, V. Barboiu, *Dig. J. Nanomater. Bios.*, **2010**, *5*, 35.
13. P.V. Konarev, V.V. Volkov, M.H.J. Koch, D.I. Svergun, PRIMUS SAXS interpretation program. Version 2.2 12.03.03
14. M.A. Busolo, P. Fernandez, M.J. Ocio, J.M. Lagaron, *Food Addit. Contam. Part A Chem. Anal. Control Expo Risk Assess.*, **2010**, *27*, 1617.
15. S. Egger, R.P. Lehmann, M.J. Height, M.J. Loessner, M. Schuppler, *Appl. Environ. Microbiol.*, **2009**, *75(9)*, 2973.
16. T.H. Kim, S.H. Jung, K.H. Cho, *FEBS Lett.*, **2007**, *581*, 4899.
17. I. Sondi, B. Salopek-Sondi, *J Colloid Interface Sci.*, **2004**, *275*, 177.