Superfine drug-eluting polyvinyllpyrrolidone based coating for biliary stents

I V Zaporotskova*, R V Shinkarev

Volgograd State University, Volgograd, 400062, Universitetskii prospect, 100

Corresponding author's e-mail: irinazaporotskova@gmail.com

Received 15 October 2014, www.cmnt.lv

Abstract

The paper describes a chemical method for the fabrication of superfine polivinillpirrolidon based coating for doxorubicin-eluting polymer biliary stents. The optimal conditions for the fabrication were defined and the effectiveness of the coating when exposed to bile in static conditions was proved. The study found that the obtained drug-eluting coating reduces the process of bile crystallization on the sample polymer stent surface. The mechanism of "PVP- doxorubicin" stable adsorption complex formation was studied using quantum - chemical method DFT. A number of positions of doxorubicin molecule in respect to polyvinylpyrrolidone fragment were analyzed, and the basic characteristics of adsorption processes were defined. It was found that energetically more favorable interaction occurs via the doxorubicin molecule oxy group.

Keywords: biliary stents, N-vinylpyrrolidone, polyvidone, doxorubicin, crystallization process, adsorption, semi-empirical scheme DFT.

1 Introduction

Stenting as a method for treating narrow or weak arteries by placing inside metallic tubes was first implemented about fifty years ago [1, 2]. Currently, as many as four hundred types of stents made of various materials are applied in medicine. They differ from one another in length, design and surface coating that comes in contact with bodily fluids [3, 4]. Apart from being intensively used in the treatment of coronary heart disease, stents are universally required to recover natural passages in the body such as the biliary tract and ureter. For this particular purpose, stents made of plastic (polyethylene, PVC) are used [5]. These so-called biliary stents counteract disease-induced flow restrictions caused by stones or malignant tumors or help to ensure the passage of stones or drainage after surgery or injury [6-8]. However, the efficiency of stents can seriously decline in case infection sets in or salts build up on the stent surface [9]. To eliminate the possible negative effects an effective coating is required that when applied on the stent surface will ensure high concentration of bactericidal medication, prevent the buildup of salts and stones and provide systemic treating effects while the stent releases drug into the body [10-12]. At present, drug-eluting stents are considered to be a most important tool of treatment. When combined with modern medical therapy they enable to quickly, efficiently and safely eliminate disease-induced symptoms. That is why a demand for a broader range of possible drug carriers that prolong drug release and possess additional antibacterial properties increases and the development of new simple and inexpensive methods to apply coating on the surface of polymer stents seems to be vital.

In this paper we describe a new method to prepare thin film coating that is based on a well-known antioxidant polymeric substance N- vinylpyrrolidone made from the monomer polivinillpirrolidone (PVP or polyvidone) [13-16]. As a drug to saturate the polymeric coating applied on the biliary stent surface we chose doxorubicin [17, 18]. As drug carrier we chose PVP because it has a number of valuable characteristics, namely non-toxicity, solubility in most organic solvents and water, good adhesive properties and high tendency to form compounds [19].

We have designed a technique that can be used to apply drug-releasing thin film coating on biliary stents. The experimental results are reinforced by theoretical quantum-chemical calculations for the interaction processes between the coating components (PVP and doxorubicin). The theoretical data turned out to be in good agreement with the obtained experimental results.

2 Experimental studies of possibility for application of superfine drug coating on the biliary stent surface

A biliary stent represents a polymer sample in the shape of a flexible cylinder made from polyurethane. For application of drug coating on the biliary stent surface an aqueous solution was prepared, which is a complex of drug and polymer that acts as a carrier for the drug substance. The solution of «PVP + doxorubicin» was applied on the biliary stent surface by chemical means and formed the expected drug coating. Polyvidone (PVP) can be described as a mixture of linear amorphous polymers with varying degrees of viscosity. PVP is a yellow-white or white hygroscopic powder with a faint characteristic odor, it becomes soft at a temperature of 140° –160° C. The substance is soluble in water, alcohol, concentrated and diluted mineral acids, aromatic hydrocarbons. The polyvidone (C6H9NO) n macromolecule structure is shown in Figure 1.

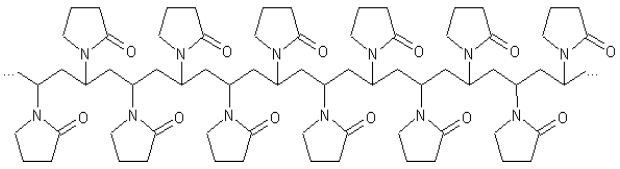


FIGURE 1 Polyvidone macromolecule structure

The choice of polyvidone as substance carrier was determined by the following reasons. The electronic nature of the PVP functional groups (fragments of the macromolecule) provides this polymer with a number of unique features that ensure its wide application in industry and medicine. It is characterized by high absorption capacity and tendency to form compounds. This property of polyvidone to bind to many substances (low molecular compounds) including drugs, dyes, etc., is used in medical practice to prolong the effect of drugs and remove disease-causing toxins from the body of humans and animals [13-16]. Polyvidone does not display any effect on the body, since it cannot be split by enzymes and it passes unchanged through the kidneys [15]. When binding to substances of protein origin including toxins, products of tissue decomposition, products of bacterial origin, PVP forms compounds that can easily pass through the kidneys. But for polyvidone these organic compounds would have accumulated in the body. Polyvidone based compounds help to normalize permeability of cell membranes and restore electrolyte composition. As a result, the liver and kidneys regain their normal functions, enzymatic processes as well as protein synthesis restore, etc. [16].

Doxorubicin (gross formula $C_{27}H_{29}NO_{11}$) (Figure 2) is a crystalline or amorphous orange-red or red powder. The substance solves well in water, aqueous acids, acetone, butanol, chloroform. Doxorubicin is used as a chemotherapy drug in the treatment of certain cancers [17].

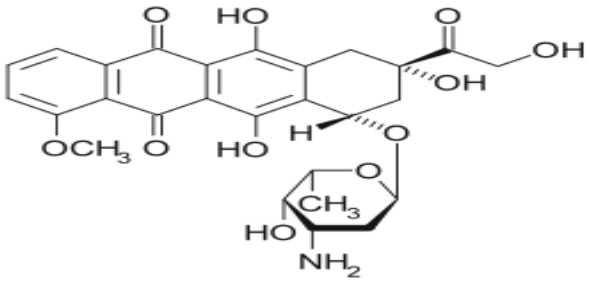
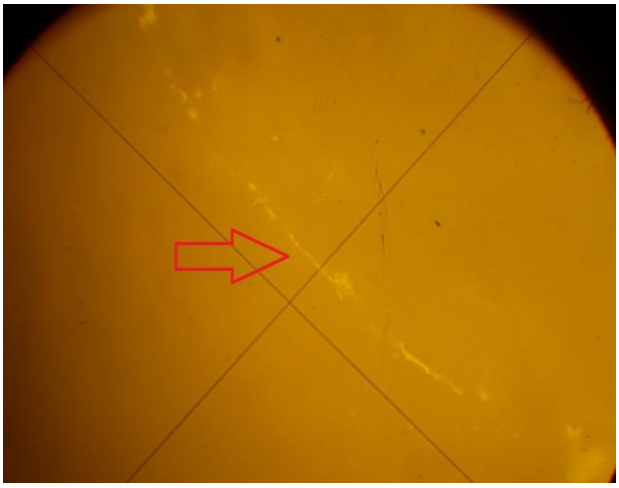
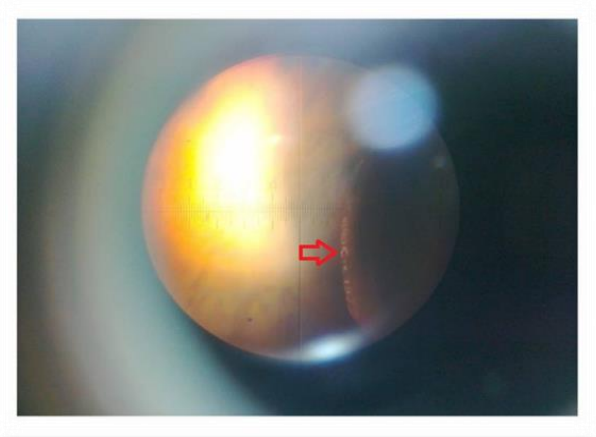


FIGURE 2 Doxorubicin formula

The application of drug coating on the polyurethane stent surface was performed as follows. Firstly, 50% aqueous polyvidone solution was prepared. Full dissolution of the required concentration of PVP in water required 24 hours. The obtained solution had a yellow color and sufficient viscosity that allowed it to mount well on the polyurethane sample surface. We also prepared another solution of the same polyvidone concentration, but this time we added doxorubicin into the solution in an amount of 8.5 % of the total volume. On thorough mixing and complete dissolving of the substances in water we obtained a bright orange color solution. Further, we degreased a biliary stent surface by placing it in a solvent (acetone) for 30 minutes and then chemically applied coating by placing the stent in each of the prepared solutions. The dwell time of the stent in the solution was 24 hours. After being dried, the coated samples were examined under an optical microscope with magnification 100 x. Analysis of the images (Figure 3) allowed us to determine the coating thickness, which turned out to be equal to 0.03 mm.



a)



b)

FIGURE 3 Fragments of the polymer stent with drug coating: a) 50%-PVP solution, b) 50%-PVP solution + 8,5% doxorubicin; (magnification x100)

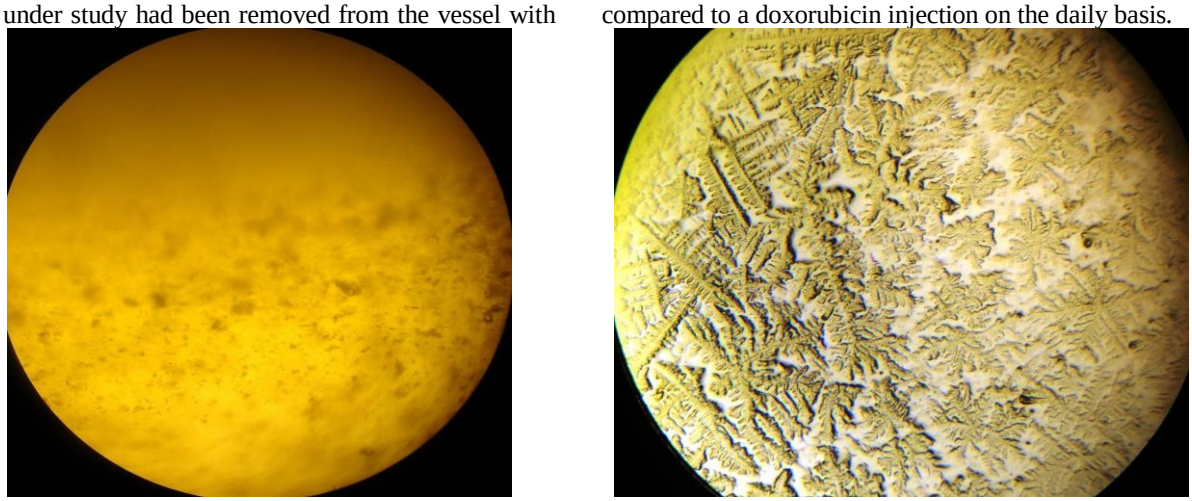
The next stage of the experiment was to test the effectiveness of the obtained biliary stent drug coating by exposure to bile. According to the fundamental research by K. Juniper that studied the bile elements it was found that the main ones that play a decisive role in the crystallization process, are cholesterol (CH), calcium carbonate (CC), calcium bilirubinate (BC) crystals, or a combination of them [20-22]. The study of bile by using polarized light microscopy showed that it can undergo a multi stage crystallization process. Homogeneous micellar solution is an evenly spread dark field. In polarized light the crystals form optical shapes – textures that have various shapes depending on the phase and type of crystal [17]. Taking into consideration these data, we conducted an experiment to test possibility to reduce the intensity of bile crystallization on the biliary stent surface by means of "PVP + doxorubicin" complex coating application.

The experiment was conducted in three stages. First, the clean polymer stent was placed in a vessel filled with bile for 24 hours and after being removed from the bile the sample was immediately examined in polarized light by microscope MIKMED -5. The images captured show that in polarized

light crystals are well observable whereas other (non-crystalline) elements are not visible, which eliminates errors in the identification of the observed objects. The obtained photographs of the sample are presented in Figure 4. Crystallization began about 4-5 minutes later after the sample had been removed from the bile. Further, we studied bile crystallization process on the stent with PVP based coating. It was found that crystallization process started six-seven minutes later after the stent had been removed from the vessel with bile. Finally, a study of the bile crystallization process on the stent surface coated with "PVP + Doxorubicin" complex was conducted. The sample stent coated with drug under study was placed in a vessel with bile for 24 hours, then removed and immediately studied under a microscope. The obtained pictures are shown in Figure 5. The process of crystallization began in 18-20 minutes after we had removed the sample from the bile. That is, the time interval until the onset of crystallization increased approximately 3-fold. The observation showed that the crystals on the surface of the drug-eluting stent were substantially smaller in size.

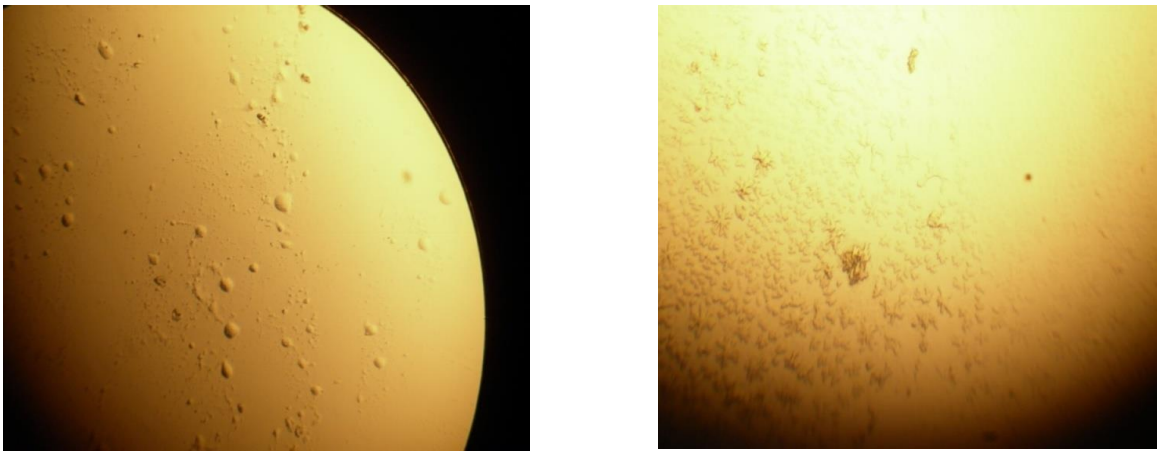
Then, we estimated the size of crystallization area at magnification 100 x that took place 5 minutes later after the stents under study had been removed from the vessel with bile and compared the one in all the three cases: 1) on the surface of the clean stent, 2) on the stent surface with PVP based coating, 3) on the stent surface with "PVP + doxorubicin" complex coating. The observations showed that the surface of the clean stent in the time interval of 5 minutes was found to be 100 % covered in crystals. In the case with PVP based coated stent as much as 80 % of the surface area was estimated to be crystallized whereas the stent coated with «PVP + doxorubicin» complex displayed no signs of crystallization process.

Thus, we empirically tested that application of coating on the biliary stent is possible by chemical method when certain conditions are observed. In the course of this experiment, we found optimal proportions of polyvidone, water and doxorubicin volumes to produce the most stable coating. The effectiveness of coating was tested under exposure to bile, and it was found that the empirically obtained coating reduces bile crystallization processes on the sample surface. Resistance of the coating that provides uniform dissolution of doxorubicin and enables its prolonged effect due to which long-term presence of the drug in the body is possible was tested. This prolonged action is much more effective as compared to a doxorubicin injection on the daily basis.



a) b)

FIGURE 4 A fragment of the clean stent before the onset of bile crystallization process on its surface (a) and a fragment of the stent five minutes later it had been removed from the bile (b)



a) b)

FIGURE 5 A fragment of the stent coated with "PVP + Doxorubicin" complex before the onset of bile crystallization process on its surface (a) and a fragment of the stent five minutes later (b)

3 Quantum chemical study of the interaction between PVP macro- molecules and doxorubicin molecules

We studied the interaction mechanism between polyvidone and doxorubicin and performed calculations of the interaction process in the framework of quantum-chemical method DFT (density functional theory) using B3LYP functional [23]. To describe the polymer PVP a molecular cluster consisting of three monomers N-vinylpyrrolidone was modeled.

It is known that one of the most important properties of PVP is the ability to form stable complexes with phenolic compounds due to the formation of a hydrogen bond between the functional group of polyvidone > N- C = 0 and the oxy group of polyphenol molecule. In the structure of doxorubicin molecule five oxy groups can be identified. Taking this fact into consideration, we selected hydrogen atoms belonging to these groups as possible active adsorption centers. In Figure 6 they are numbered 1 – 5.

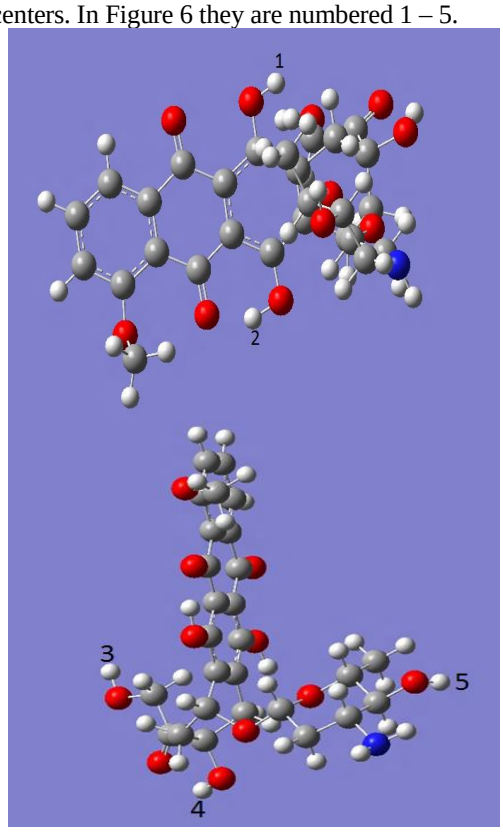


FIGURE 6 Possible adsorption centres of a doxorubicin molecule

We considered several positions of a doxorubicin molecule in respect to the fragment of PVP shown in Figure 7. As the figure shows interaction takes place at the doxorubicin active centre – the hydrogen atom of the oxy groups: a) adsorption center 1; b) adsorption center 2; c) adsorption center 3; d) adsorption center 4; e) adsorption center 5.

The adsorption process was modeled in increments of 0.1 Å of doxorubicin molecule to a fragment of Polyvidone macromolecule. The calculations allowed us to construct energy curves of these processes (Figure 8). The chart analysis showed that each of the three curves (variants 1, 4, 5) has an energy minimum that indicates the case of adsorption. The rest of the positions studied (namely, variants 2 and 3) correspond to the unstable state of the system since the minimum is located in the range of positive energies.

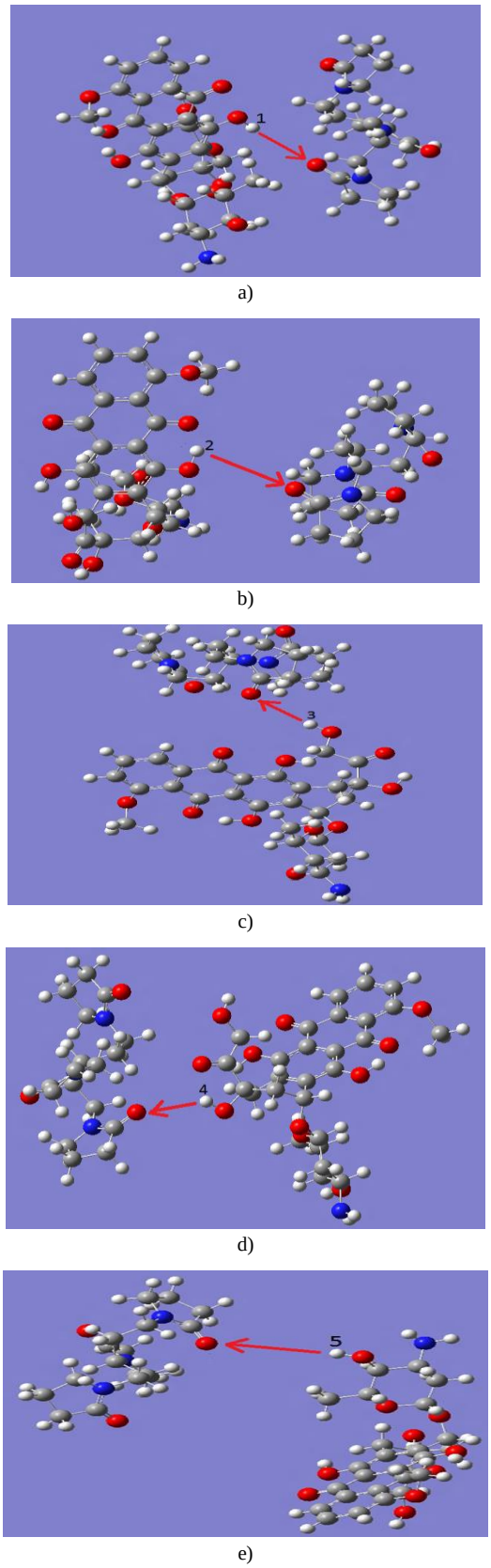


FIGURE 7 The process of doxorubicin molecule approaching an O atom of the PVP molecule; interaction takes place via the active centre of doxorubicin – H atom of the oxy group: a) adsorption centre 1; b) adsorption centre 2; c) adsorption centre 3; d) adsorption centre 4; e) adsorption centre 5

The curves show that energetically more favorable interaction is observed in variant 5 (Table 2, Figure 10) where adsorption energy EADS = - 0.57 eV, which corresponds to theoretically obtained values of hydrogen bond energy and indicates the formation of the most stable structure. The main characteristics of doxorubicin and PVP interaction process are shown in Table 1.

The obtained values of the main characteristics of the interaction determine the possibility of fairly easy desorption of doxorubicin from PVP and its gradual release into the body thus providing a long-term medical effect.

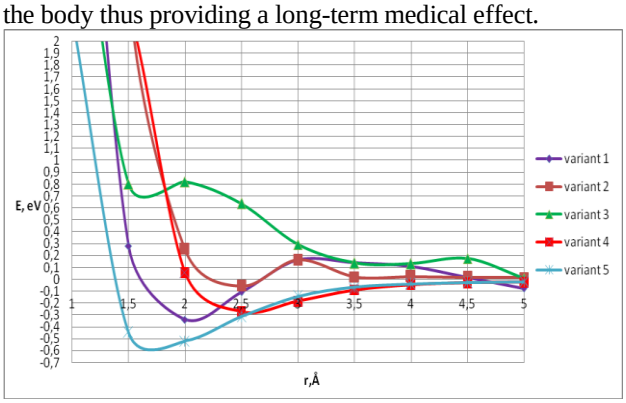


FIGURE 8 The dependence of doxorubicin and polyvidone macromolecules interaction energy values on a distance

TABLE 1 The main characteristics of PVP and doxorubicin interaction process for different positions: r_{ads}, Å - adsorption distance, E_{ads}, eV – adsorption energy

Variants	r_{ads}, Å	E_{ads}, eV
1	2.1	-0.34
2	-	-
3	-	-
4	2.6	-0.28
5	1.7	-0.57

4 Conclusions

We proposed a method for preparing drug-eluting coating for polymeric stent surface, namely, we experimentally found optimal proportions of polyvidone, water and doxorubicin volumes to produce the most sustainable coating, and we tested the coating efficiency under exposure to bile in static conditions. We found that the obtained coating reduces the process of bile crystallization on the sample surface.

We proposed and studied theoretically a plausible stable adsorption complex “PVP doxorubicin” formation mechanism that can be used as effective coating enabling prolonged release of drug into the body. We analyzed a number of positions of doxorubicin molecule with respect to polyvidone in the interaction process and calculated the main characteristics of adsorption processes. We found that energetically more favorable variant is the one where interaction takes place via the oxy group in a doxorubicin molecule. The obtained values of distance and adsorption energy indicate the possibility of a gradual release of doxorubicin into the body when it desorbs from PVP.

References

[1] Kalesan B, Pilgrim T, Heinimann K et al 2012 Comparison of drug-eluting stents with bare metal stents in patients with ST-segment elevation myocardial infarction *European Heart Journal* **33**(8) 977–87

[2] Cook S, Eshtehardi P, Kalesan B 2012 Impact of incomplete stent apposition on long-term clinical outcome after drug-eluting stent implantation *European Heart Journal* **33**(11) 1334–43

[3] Stone G W, Moses J W, Ellis S G et al 2007 Safety and efficacy of sirolimus and paclitaxel-eluting coronary stents *N. Engl. J. Med.* **356** 998–1008

[4] Patel M R, Holmes D R Jr 2008 Next-Generation Drug-Eluting Stents: A Spirited Step Forward or More of the Same *JAMA* **299**(16) 1952-3

[5] *Varianty biliarnyh stentov* [Types of biliary stents] URL: http://amberlife.com.ua

[6] Il'chenko A A 2004 *Zhelchnokamennaja bolezn'* [Cholelithiasis] Moscow: Anaharsis [Anacharsis] 199 p

[7] Kulikovskij V F, Soloshenko A V, Jarosh A L et al 2013 *Biliarnyj stent s almazopodobnym uglerodnym pokrytiem* [Diamond-like carbon coated biliary stent] *Jendoskopicheskaja hirurgija [Endoscopic surgery]* **19**(3) 16–20

[8] Zabolotnyj V T, Kolmakov A G, Sevost'janov M A, et al 2013 *Sovershenstvovanie medicinskih izdelij dlja jendovaskuljarnyh operacij [Improvement of medical devices for endovascular operations]* Moscow: Integral (3) 42–5

[9] *Vmeshatel'stva na zhelchevyvodjashhih putjah* [Biliary tract invasions] URL: http://www.rusmedserv.com/radiology/biliary-interventions/

[10] Cai X B, Zhang W X, Wan X J et al 2014 The effect of a novel drug-eluting plastic stent on biliary stone dissolution in an ex vivo bile perfusion model *Gastrointestinal Endoscopy* **49**(1) 156–62

[11] Lee Dong Ki 2009 Drug-eluting stent in malignant biliary obstruction *Journal of Hepato-Biliary-Pancreatic Surgery* **16**(5) 628-30

[12] Windecker S, Juni P 2009 The Drug-Eluting Stent Saga *Circulation* **119**(5) 53–56

[13] Kirsh Yu E 1998 *Poli-N-vinilpirrolidon i drugiye poli-N-vinilamidy [Poly-N-vinylpyrrolidone and other poly-N-vinylamides]* – Moscow: Nauka 252 p *(in Russian)*

[14] Sidel'kovskaja F P 1970 *Himija N-vinilpirrolidona i ego polimerov [Chemical properties of vinilpirrolidon and its polymers]* – Moscow: Nauka 160 p

[15] *N-vinilpirrolidon* 2014 *Bol'shaja jenciklopedija nefti i gaza [Encyclopedia of Oil and Gas]* URL: http://www.ngpedia.ru/id658762p1.html

[16] *Polivinilpirrolidon* 2014 *Farmmedhim* URL: http://fmchem.ru/catalog/115

[17] *Doksorubicin* 2014 *Reestr lekarstvennyh sredstv [The register of drugs]* http://www.reles.ru/cat/drugs/Doxorubicin/

[18] D'yachkov E P, D'yachkov P N, Zhdanov R I 2011 Vzaimodejstvie odnostennyh nanotrubok s doksorubicinom i poliehtilenglikolem po dannym metoda molekulyarnogo dokinga [Interaction of single-walled nanotubes with doxorubicin and polyethylene glycol from data of molecular docking method] *Doklady Akademii Nauk [Academy of Sciences reports]* **437**(4) 1–3

[19] Gao Y, Jiang P, Liu D F et al 2004 Evidence for the monolayer assembly of poly(vinylpyrrolidone) on the surfaces of silver nanowires *J. Phys. Chem.* B **108**(34) 12877–881

[20] Juniper K, Burson N 1954 Peculiar crystals occuring in bile and associated with biliary tract disease *Clin. Res. Proc.* **2** 153 – 41

[21] Juniper K, Burson E N 1957Biliary tract studies. The significance of biliary crystals *Gastroenterology* **32** 175 – 211

[22] Juniper K, Woolf W E 1956 Biliary tract studies. X-ray diffraction analysis of gallstones, correlation with occurance of tnicrospheroliths in bile *Am. J. Med.* **20** 385-591

[23] Koch W A 2002 *Chemist's Guide to Density Functional Theory* eds W Koch and M Holthausen Weinheim: Wiley-VCH, 19-28

Authors

Irina Zaporotskova, 12.11.1963, Melitopol, Ukraine

Current position, grades: professor, Doctor of Physical and Mathematical Sciences
University studies: Volgograd State University, Molecular physics, Solid State Physics
Scientific interest: nanotechnology, boron-carbon nonmaterial, nanocomposites.
Publications: 326 publications
Recent main: [1] Zaporotskova I V, Boroznin S V, Boroznina E V, Polikarpov D I 2013 Vacancy Transport Properties in Boron–Carbon BC3 nanotubes *Nanoscience and Nanotechnology Letters* **5** (11) 1164-8
[2] Zaporotskova I V, Polikarpova N P, Ermakova T A, Polikarpov D I 2013 Carbon Nanotubes, New Material for Purification of Water–Ethanol Mixtures from Isomers of Propanol *Russian Journal of General Chemistry* **83**(8) 1601–6
[3] Zaporotskova I V, Anikeev N A, Kojitov L V, Davletova O A, Popkova A V 2014 Theoretical Studies of the Structure of the Metal-carbon Composites on the Base of Acryle-nitrile Nanopolimer *Journal of nano- and electronic physics* **6**(3) 03035-1 - 03035-3
Experience: since 2008 - the managing editor of the journal "Bulletin of the Volga. Series 10: Innovation activities; since 2007 - the head of Research and Education Center VolSU "Nanomaterials and Nanotechnologies", repeatedly participated in conferences, symposiums, forums and academic seminars of national and international level.

Roman Shinkarev, 05.06.90, Kamyshin, Russia

Current position, grades: Assistant, postgraduate student
University studies: Volgograd State University 2013, Nanotechnology
Scientific interest: X-ray analysis, nanotechnologies in medicine, computer modeling of nanosystems.
Publications: [1] Zaporotskova I V , Shinkarev R V 2014 The superfine covering on base of polyvinyllpyrrolidone for MEDICAL stents *5 International Conference on NANO-structures Self-Assembly (NanoSEA 2014)* France Marseille 7 – 11 Jule 2014 *Book of abstract* 202
[2] Zaporotskova I V, Shinkarev R V 2013 The new medicinal superfine covering on base of polyvinillpirrolidone for biliary stents *Nanoscience & nanotechnology 2013* 14th International Workshop on Nanotechnology, 30 September – 4 October 2013 Frascati National Laboratories INFN *Book of abstract* 83-5
Experience: Currently - postgraduate student on "Condensed Matter Physics", several publications in Russian and foreign journals, won regional competition of innovative projects "UMNIK-2014"(2014)