POLITEHNICA UNIVERSITY OF BUCHAREST
FACULTY OF APPLIED CHEMISTRY AND MATERIALS SCIENCE

DOCTORAL THESIS SUMMARY

DELIVERY AND VECTORIZATION SYSTEMS OF BIOLOGICAL ACTIVE PRINCIPLES

SISTEME CU ELIBERARE ȘI VECTORIZARE DE PRINCIPII BIOLOGIC ACTIVE

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I dedicate this thesis to my grandparents.

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KEYWORDS: polyvinyl chloride, surface modification, catheters, spin coating, anticoagulant surfaces, antibacterial surfaces, cardiovascular stents, titanium oxynitride.

INTRODUCTION

Catheters are medical devices used on a large scale for medical treatments. Unfortunatelly these devices present side effects due to bacterial adhesion and proliferation on the surface. The most important disadvantages of medical devices are related to calcification, structural flaws, infections, thrombosis. A very important characteristic of medical devices is biocompatibility, meaning the ability to perform their functions without presenting side effects. Medical devices behaviour depends on the physical properties of the material (rigidity, surface roughness), but also the chemical nature [1]. Adherence and proliferation of bacteria and living cells on the surface that comes in contact with the living tissue are the main causes of nosocomial contamination, thus a special attention is needed on how to prevent or minimize the occurrence of infections due to medical devices [2, 3]. Polyvinyl chloride (PVC) is a biocompatible, easily available material with a low price, thus it was chosen for this study with the aim of obtaining a biomaterial that reduces the side effects described earlier.

The objective of this doctoral research study is to modify the surface of polyvinyl chloride so that the material obtained does not allow the adhesion and proliferation of bacteria and cells on its surface, does not allow blood clotting or the formation of calcifications. These aspects are very immportant when the material is intended for use in the medical field for obtaining catheters. The most common problems encountered in catheters are due to their blockage or the appearance of an infection at the place of their implantation. Due to these side effects, the catheters must be removed and replaced frequently, which requires another surgery, high costs and causes major discomfort to the patient.

In order to achieve the desired objective, in the Doctoral Thesis entitled ***"Delivery and vectorization systems of biological active principles"***, both physical and chemical modification methods of the PVC surface were studied. Physical modification was performed in order to eliminate the unevenness caused by the manufacturing process of PVC films and to obtain a smooth surface. The smoother the surface, the less likely it is for cellular organisms to adhere to the surface of the material. If the material presents a rough surface, at the site of these flaws bacteria or cells can anchor and can not be washed when the biological fluids pass du to the hydrophobic nature of PVC. In order to increase the hidrophilicity, the chemical modification of the surface was performed by introducing hydrophilic groups. To prevent blood clotting and thrombosis, a coumarinic compound (dicoumarol and warfarine) with anticoagulant activity was incorporated on the material surface. This class of compounds has been used because they are known anticoagulants and also, in addition to anticoagulant activity, they also have antibacterial, antioxidant, antiallergic, antitumor activities [4, 5]. The study of the synthesis of some active substances representative of this class of compounds was another objective of this study.

Another research direction approached in this doctoral research was to study the behavior of metal stents made of stainless steel, modified on the surface by depositing a TiOxNy film. This deposition prevents the migration of nickel, molybdenum, chromium or other metals from the surface of stainless steel, reducing inflammation and toxicity [15]. The physico-chemical and biological properties of TiOxNy films differ depending on the deposition method, the N / O ratio present on the surface. Stents are devices used in percutaneous coronary surgery or coronary angioplasty. These procedures are performed to determine revascularization in the implanted area [12, 13]. The first embodiment of stents was developed in 1980, but since then numerous changes in their structure and composition were performed [12]. At the beginning, most stents were made mostly of metals (stainless steel,

cobalt-chromium alloys, nickel-titanium alloy (nitinol), etc.). Over time, other materials have been also studied, such as biodegradable or non-biodegradable polymers, but also multilayer materials with different surface coatings, in order to be used in these applications. However, the complications associated with stent implantation still remain a problem. For a stent to be considered ideal, it must prevent inflammation, restenosis, prevent the formation of thrombosis, but at the same time to initiate endothelialization [14].

The thesis was structured in three main chapters, each of which is divided into several important subchapters. In the first chapter the scientific objectives of doctoral research activity are presented. Chapter II (**INTRODUCTION**) consists in a short introduction, followed by a detailed description of the main topics studied in this paper. This description was made following a detailed study of the literature in the field of this research study. The bibliographic research activity was structured on subchapters as follows: **II.1 CATHETERS** – this subchapter presents information about the formulation of catheters, the types of materials used in their manufacture, the types of catheters available on the market, their use, the properties that these devices must meet depending on the intended application, but also information about the disadvantages of available catheters on market. The next subchapter, **II.2. POLYVINYL CHLORIDE**, focuses on the description of the material chosen to be studied, namely polyvinyl chloride, its properties, but also methods of surface modification, to improve its characteristics, in order to be used in the formulation of catheters. To determine the ways in which the incorporation of active principles on the polymer surface can be achieved, in order to obtain a suitable release behavior for the intended application, the types of controlled-drug release systems were studied in detail, and also the delivery mechanisms of the active substance (**II.3. CONTROLLED DRUG DELIVERY SYSTEMS**). In experimental studies, the surface of PVC was physically modified by deposition of thin films using the spin coating method. Subchapter **II.4. SPIN COATING METHOD** presents the detailed description of this coating technique, and also the research direction on this subject, following the literature journals. **II.5. CARDIOVASCULAR STENTS** is a subchapter dedicated to a second type of medical devices studied in this doctoral research activity, respectively medical stents. This part of the paper contains numerous studies related to the synthesis, properties, but also on the behaviour of stents when they come in contact with the biological fluids, the current state of research in this field, but also the trends of further development.

Chapter **III. SCIENTIFIC RESULTS OBTAINED IN THE DOCTORAL RESEARCH ACTIVITY** presents the experimental part of the doctoral thesis, in which the original contributions and the results obtained by performing laboratory experiments, and also experimental data interpretations. In the beginning, as described in chapter **III.1. PHYSICAL MODIFICATION OF THE PVC SURFACE USING SOLVENTS FOR SURFACE SWELLING,** physical modification of medical grade PVC surface, that present flaws due to the production process, was studied with the aim to reduce these surface defects. For this, the immersion of flat PVC samples in various solvents was carried out, for different periods of time, so that the polymer swelled to the surface, without changing the properties in bulk. This treatment led to the correction of defects present on the PVC surface. Although after performing this modification, the surface roughness is semnificatively reduced, this material has another immportant disadvantage, PVC being a hydrophobic polymer with reduced biocompatibility. In order to improve the hidrophilicity, PVC surface was chemically modified by introducing ester groups on the surface. This modification and the results obtained are described in detail in Chapter **III.2. CHEMICAL MODIFICATION OF THE PVC SURFACE**. In chapters **III.3 DICOUMAROL SYNTHESIS** and respectively **III.4. WARFARIN SYNTHESIS** the experimental procedure and the optimal parameters required for the synthesis of a pure product with a high yeld, are presented. Further, the incorporation

of these compounds was studied using the spin coating method (**III.5. SURFACE MODIFICATION OF PVC USING SPIN COATING TECHNIQUE**), by depositing a film also of PVC that contains the active principle. Following the experiments, it was determined that warfarin is not a suitable compound for the intended application because it has a good solubility in water and is quickly released from the inside of the film. Thus, the study was concentrated on PVC/Dicoumarol film deposition. The experimental procedure, results and experimental data interpretation were described in chapter **III.5.1 Modification performed using PVC/Dicoumarol film deposition.** Here, a detailed description of the release behaviour of dicoumarol from the surface of the film deposited, protein adsorbtion behaviour and the ability to prevent bacterial adhesion on the material surface. In order to obtain a surface with antibacterial properties, silver deposition on the surface of PVC was also studied. Spin coating technique was used to incorporate silver nanoparticles by performing the deposition of a thin film of PVC/$AgNO_3$. The experimental procedure, the materials and methods of analysis, experimental data interpretation and a detailed discution of the surface composition and morphology, the silver ions release behaviour, albumin adhesion on the samples surfaces, and also the modified PVC resistance to *Gram negative* and respectively *Gram positive* bacterial adhesion, are described in chapter **III.5.2. Silver nanoparticles deposition on the surface of PVC**.

Another direction in the doctoral research activity was related to the characterization of stainless steel stents surfaces coated with TiOxNy. Chapter **III.6. SURFACE EVALUATION OF OXYNITRIDE COATINGS (TiO_xN_y) USED FOR OBTAINING LAYERED CARDIOVASCULAR STENTS** presents the materials and methods used, as well as an extensive discussion of the stability of these stents in vitro, the way that these surfaces interact with the proteins found in biological fluids and the resistance to the adhesion of bacteria on the surface.

In the last part of the thesis (**CONCLUSIONS**) with the title *DELIVERY AND VECTORIZATION SYSTEMS OF BIOLOGICAL ACTIVE PRINCIPLES* are presented the general conclusions and the original contributions that resulted from the doctoral research activity: **C1. GENERAL CONCLUSIONS AND ORIGINAL CONTRIBUTIONS.** The **C2. PERSPECTIVES FOR FURTHER DEVELOPMENT** are also presented in the final part of the paper, these perspectives being the objective of a future research study.

The content of this paper also contains the **DISSEMINATION OF RESULTS** in scientific articles published in international journals listed ISI, having a cumulative impact factor .0.41 + 4.184 + 4.421 + 1.205 + 5.88 = 16.1. At the end is listed the studied bibliography, placed in the order of its appearance in the text.

*Below, the content of Chapter **III. SCIENTIFIC RESULTS OBTAINED IN THE DOCTORAL RESEARCH ACTIVITY** will be briefly presented, organized in subchapters, maintaining the numbering used in the thesis, but also a selective bibliography also keeping the numbering used in the text of the thesis.*

III. SCIENTIFIC RESULTS OBTAINED IN THE DOCTORAL RESEARCH ACTIVITY

III.1. PHYSICAL MODIFICATION OF THE PVC SURFACE USING SOLVENTS FOR SURFACE SWELLING

The aim of this study is to eliminate the flaws (hill-valey appearance) that appears during the manufacturing process, thus obtaining a smooth, uniform surface. The smoother the surface, the less likely it is for cellular organisms to adhere to the surface of the material. If the material presents flaws, bacteria and cells can anchor around them to the surface and cannot be washed by the biological fluids due to the hydrophobic nature of PVC. Therefore in these areas the proliferation of bacteria or cells takes place, which leads to infections or, in the case of blood, to the formation of clots due to coagulation.

III.1.1. Materials and methods

Standard flat sample of polyvinyl chloride Plăci standard de policlorură de vinil plasticized with dioctyl phthalate, acetone (Silal Trading), heptan (Merck), dimethilformamide (DMF), demineralized water, were used without prior purification. The samples were characterised using an infrared microscope, Thermo Scientific Nicolet iN10 Infrared Microscope, in order to analyze the sample surface. Scanning Electron Microscopy (SEM) was used in order to have a better view on the surface morphology before and after performing the modification by immersion in solvent.

III.1.2. Experimental

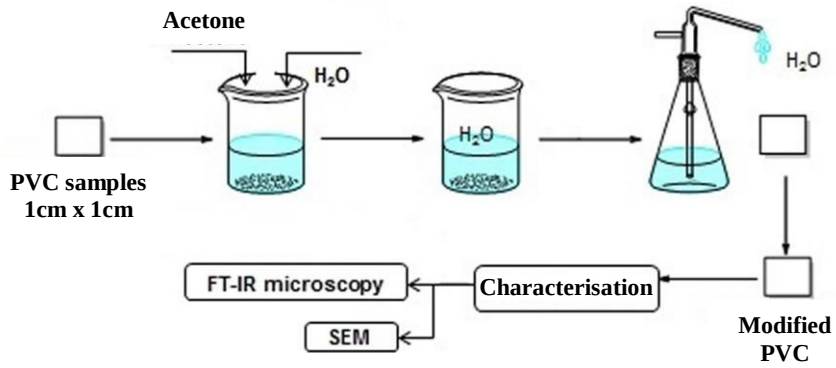


Fig. III-1. Schematical representation of the experimental procedure used for surface modification of PVC by immersion in solvent

In addition to the experiments performed in acetone, experiments were also performed in 40% aqueous dimethylformamide solution and in heptane, using the same working method.

III.1.3.Results şi discutions

From the FT-IR analysis it was observed that using heptane and dimethylformamide no satisfactory results were obtained. The surface morphology of the PVC samples did not change or the changes were insignificant.

In the case of samples modified by immersion in acetone solution 50% and respectively 100%, significant changes are observed after 10 minutes of immersion. In Fig. III-2 are presented the results obtained by FT-IR microscopy in the case of the samples modified using acetone, before and after surface treatment.

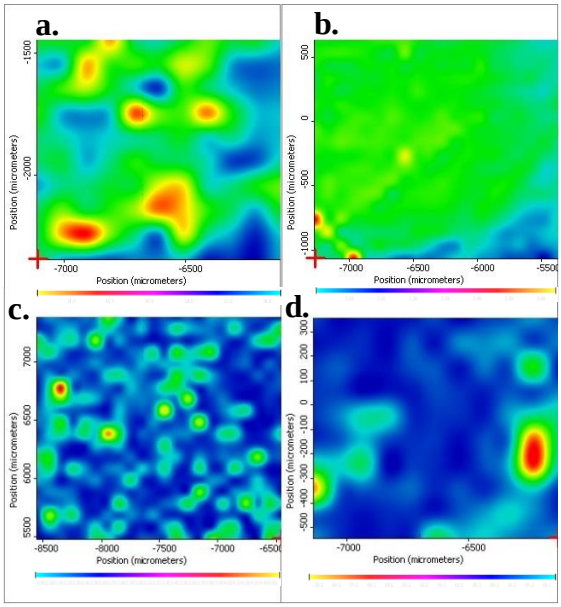


Fig. III-2. FT-IR microscopy images for: a – the sample before acetone immersion, b – the sample after immersion in acetone for 10 min., c – the sample before immersion in aqueous solution of acetone 50%, d - the sample after immersion in aqueous solution of acetone 50% for 10 min.

In order to confim the obtained results, the samples were also analyzed by scanning electron microscopy (SEM)(Fig.III-3).

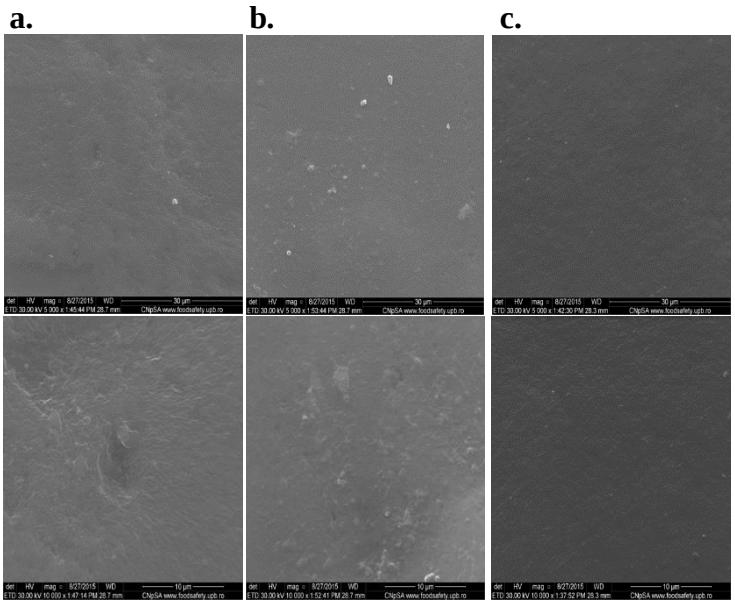


Fig. III-3. SEM images performed at 2000x, 5000x and respectively at 10000x, corresponding to samples: a – unmodified PVC, b – PVC modified by immersion in acetone 50% for 10 min., c – PVC modified by immersion in acetone for 10 min.

III.1.4. Conclusions

This modification was performed with succes by immersion in acetone and respectively in aqueous solution of acetone 50%, as it can be seen in the FT-IR microscopy images. These results were also confirmed by scanning electron microscopy images.

III.2. CHEMICAL MODIFICATION OF PVC SURFACE

Chemical modification of PVC surface using silver acetate (AcOAg) and respectively silver lactate (LAg) was studied with the aim of increasing surface hidrophilicity. The transformation takes place as ilustrated in Fig. III-4.

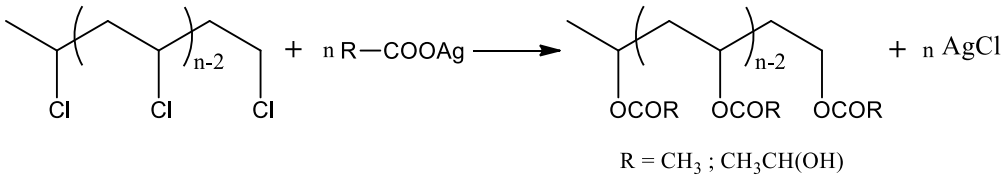


Fig. III-4. The chemical reaction that takes place between PVC, silver acetate and respectively silver lactate

III.2.1. Materials and methods

Standard flat samples of PVC, silver acetate (Alfa Aesar), silver lactate (Alfa Aesar), acetone (Silal Trading), demineralized water, were used without prior purification.

The sample were analyzed using infrared spectroscope Thermo Fischer Nicolet iN10 Attenuated Total Reflection Infrared. The contact angle on the sample surfaces was also measured.

III.2.2. Experimental

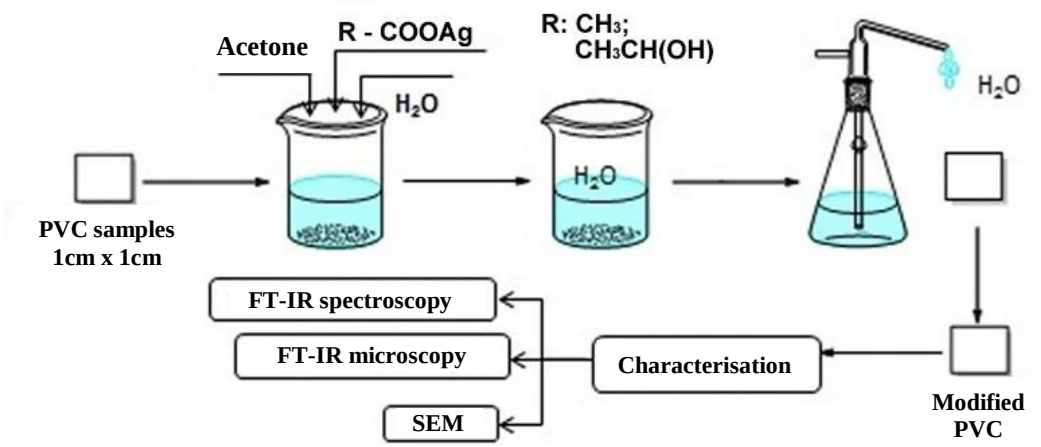


Fig. III-5. Schematical representation of the experimental procedure used for chemical modification of the PVC surface

PVC samples were chemically modified by the introduction of esteric groups on the surface, using silver acetate, and also silver lactate. The experimental procedure (Fig. III-5) consisted in the immersion of the samples in silver acetate and respectively in silver lactate solution, at room temperature and also at 50°C, for various periods of time.

III.2.3. Results and discutions

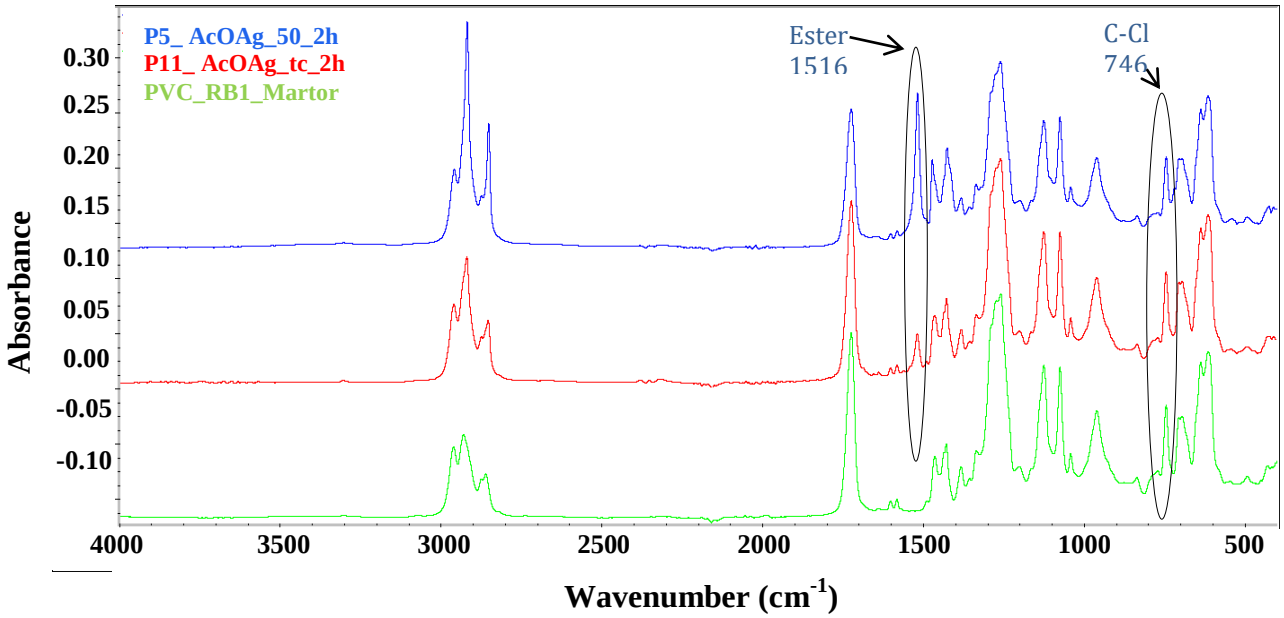


Fig. III-8. Comparison between the initial PVC spectrum and the spectra obtained in the case of the 2 experiments performed with AcOAg, after 2 hours of reaction

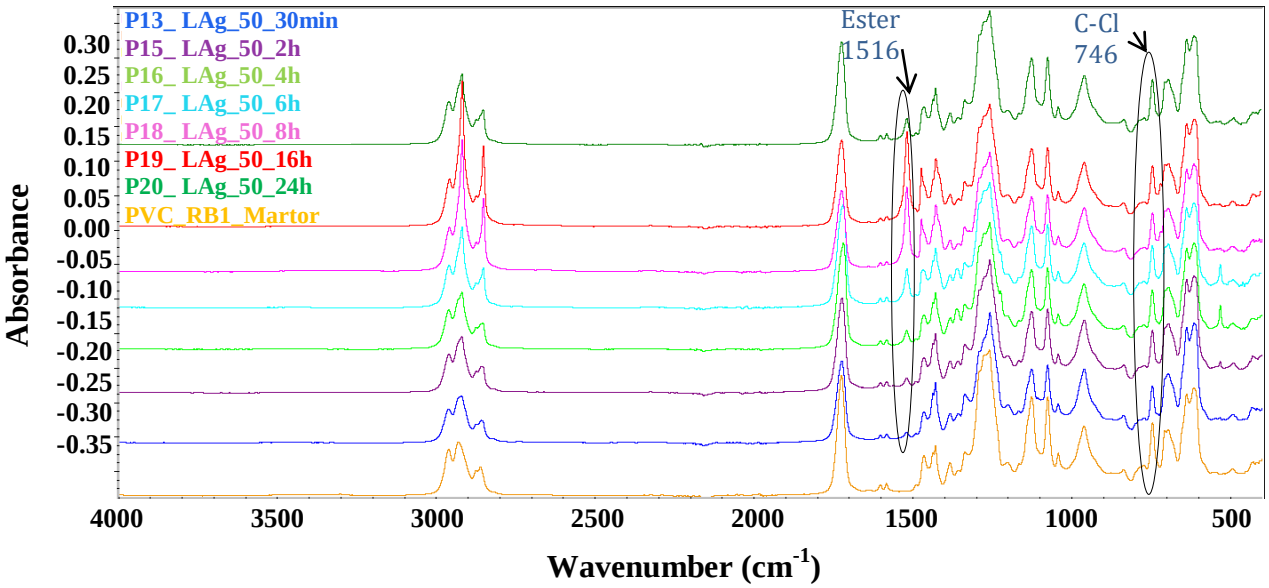


Fig. III-9. IR spectra for the samples resulted after the reaction with LAg at 50ºC

When LAg is used, the reaction speed is semnificatively lower even when the reaction is conducted at 50ºC (Fig. III-9). The appearance of the ester bond on the surface of PVC modified using LAg, at room temperature, takes place only after 6h (Fig. III-10). After 8h the intensity of the peak corresponding to the esteric bound is significantly increased. Increasing the temperature to 50ºC, the reaction speed is also increased, the corresponding ester bond appeared on the surface of PVC after 30 min, even though only in reduced intensity. In order to obtain a similar yeld with the one obtained after performing the reaction with AcOAg, the reaction with silver lactate should be performed at least 7-8h.

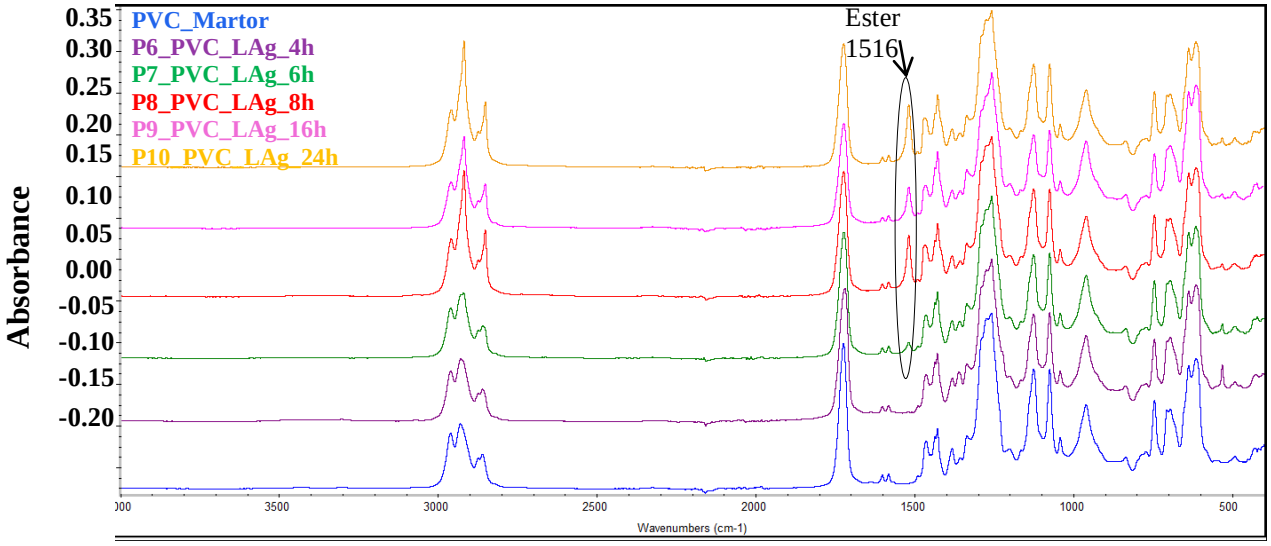


Fig. III-10. IR spectra for the samples resulted from the reaction with LAg at room temperature

In order to determine if the hidrophilicity of the surface is modified by introduction of esteric groups, the contact angle for the samples modified using AcOAg and respectively LAg, at various reaction times, was determined and compared with the contact angle of the initial, unmodified PVC (Fig. III-11).

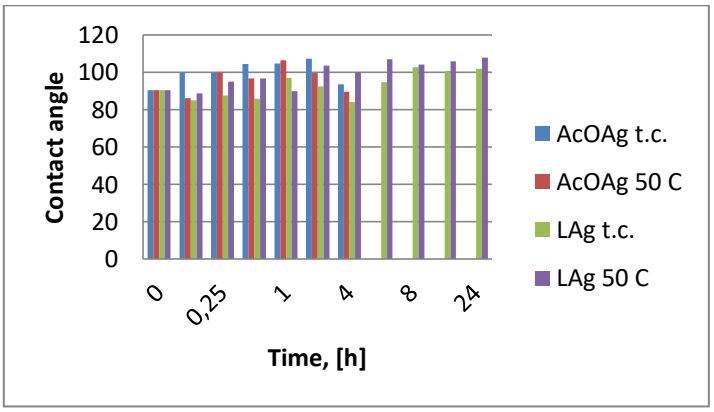


Fig. III-11. Contact angle modification after performing the reaction with AcOAg and respectively with LAg at various reaction time

III.2.4. Conclusions

Chemical modification of the surface was performed in order to increase the hydrophilicity of the polymer surface and the wetting properties so as not to allow the adhesion of bacteria and cells on the material. This modification was successfully performed by the reaction with silver acetate and also with silver lactate, after introduction of ester groups on the surface of PVC. The presence of the ester bounds on the samples surfaces was confirmed by infrared spectrometry. In the spectra corresponding to the modified surfaces, the appearance of the band corresponding to the ester (1516 cm-1) and the decrease of the band intensity corresponding to the C-Cl bond (746 cm-1) are observed. In case of chemical modification of the PVC surface with silver acetate in 50% acetone medium at room temperature, the time required to complete the reaction is 2 hours, and if the change is made at

50ºC the time required is 1 hour. If silver lactate is used to introduce ester groups on the PVC surface, a much longer reaction time of more than 4 hours is required, which can lead to polymer degradation.

The maximum contact angle achieved using this type of modification has a similar value in both AcOAg and LAg but the difference between the required reaction times is very large, the reaction with AcOAg being preferable. The longer the polymer is in contact with the solvent for a longer period of time, the greater the possibility that changes will occur in the PVC properties.

III.3. DICOUMAROL SYNTHESIS

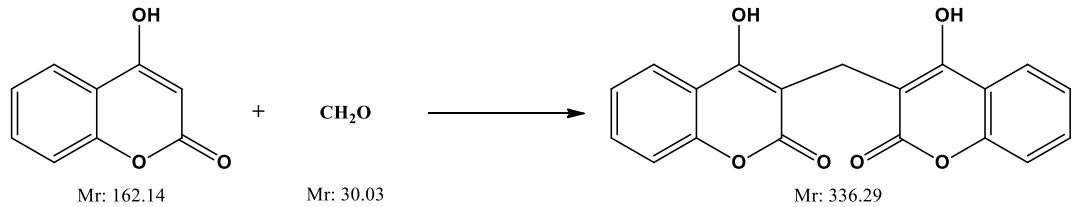


Fig. III-12. Dicoumarol synthesis reaction

The synthesis methods used in this research study were prior studied and proposed by K. M. Khan şi co. [349] and respectively by M. Prabhakar [352]. These synthesis techniques were improved, the experimetal procedure was optimized and the yeld was increased significantly. Also, the purity of the compounds obtained was improved.

III.3.1. Materials and methods

4-hidroxycumarine (Merck), fomaldehide (Merck), EDTA (Reanal Budapest), ethanol (Chemical Company), toluen (Merck), piperydine (Merck), NaOH (Merck), norit charcoal, were used without prior purification. The product obtained was analyzed using thin layer chromatography and FT-IR spectroscopy.

III.3.2. Experimental

1. Initially the reaction was led in the way described by M. Prabhakar in the work "EDTA – catalyzed fast and efficient eco-friendly synthesis of dicoumarol derivatives in water" [352]. The progress of the reaction was verified by thin layer chromatography (plates: Merck silica gel 60 F254, eluent: toluene: ethyl acetate: acetic acid 20: 5: 1) as shown in Fig. III-13, Fig. III-14, Fig. III-15. Using the reaction conditions described by the authors, the yield is low, the ratio between the product formed and the raw material remaining unreacted being about 1:1. By increasing the reaction time and adding an excess of formaldehyde (1.2 equivalents), the reaction yield was significantly improved. The product obtained cannot be purified by the method described in the article, so another purification method has been developed. The crude product was purified by dissolving in dilute sodium hydroxide (NaOH) (stoichiometric amount), decolorized with charcoal, and the clear solution was extracted with toluene. The aqueous phase was then acidified with sulfuric acid (H_2SO_4) to pH = 2. The resulting suspension was filtered in vacuum and the precipitate was washed with demineralized water to remove traces of acid. The white solid obtained was dried in an oven at 105 ° C. Yield obtained: η = 59%.

		
Fig. III-13. TLC of the reaction after 40 min.	**Fig. III-14.** TLC after performing the reaction using the improved conditions	**Fig. III-15.** Purified Dicoumarol

During the reaction, the suspension becomes very compact and very difficult to stir, and the yield obtained by the method presented above is low, so it was decided to perform the synthesis by another method.

2. K. M. Khan et al. [349] proposed the synthesis of dicoumarol using piperidine to catalyze the reaction in ethanol medium. The progress of the reaction was analyzed by TLC as shown in Figs. III-16, Fig. III-17, Fig. III-18 and Fig. III-19. This method can be significantly improved by conducting the reaction at the reflux temperature and by using an exces of formaldehide. Performing these modifications an increased yeld (74%) was obtained after purification. The crude product was purified as performed in the previous method.

			
Fig. III-16. TLC of the reaction mass when performing the reaction as described by K. M. Khan et al. [349]	**Fig. III-17**. TLC when performing the reaction using the improved conditions.	**Fig. III-18.** TLC of the crude product and the reaction solvent.	**Fig. III-19.** Purified Dicoumarol

III.3.3.Results and discutions

In Fig. III-20 is represented the IR spectrum of control dicoumarol, compared with the IR pectrum of the obtained product.

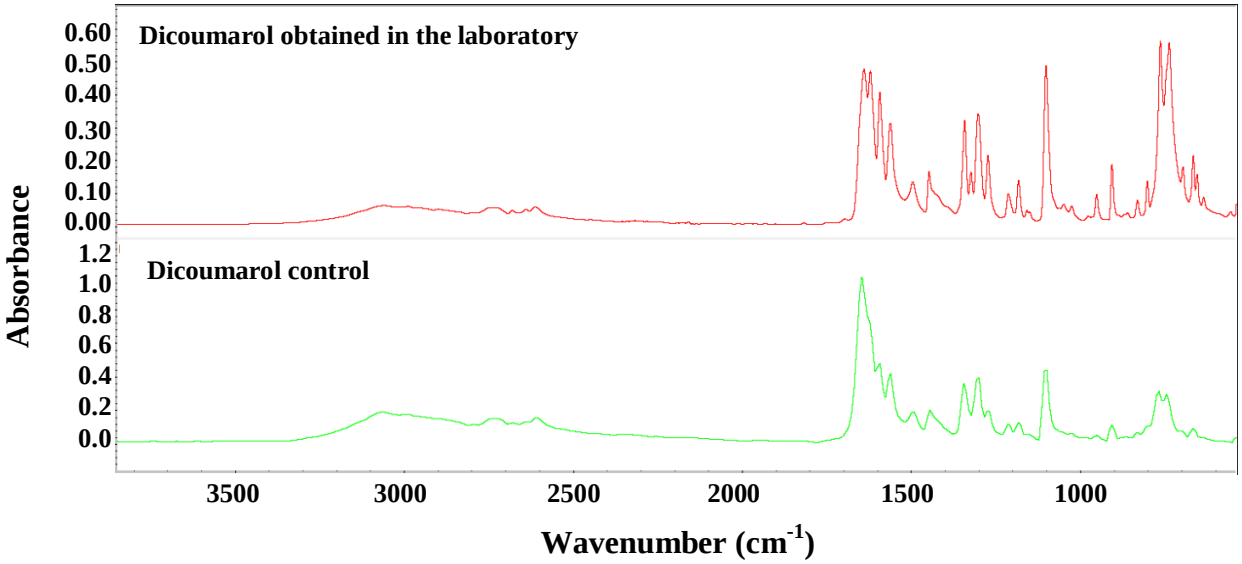


Fig. III-20. IR spectrum of the control dicoumarol, compared with the product obtained by synthesis.

By comparing the control spectra of dicoumarol with the spectra of the obtained product, it was concluded that the active principle obtained is dicoumarol.

III.4. WARFARIN SYNTHESIS

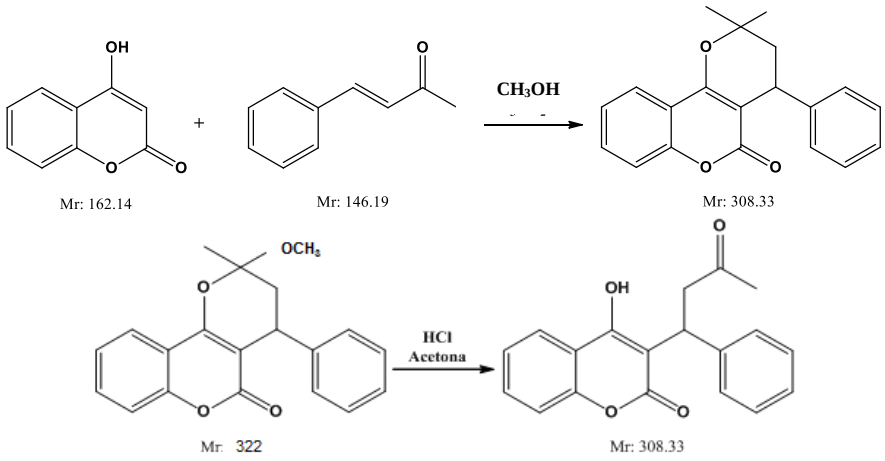


Fig. III-22. Warfarin synthesis scheme

III.4.1. Materials and methods

4-hidroxycumarine (Merck), benzilidenacetone (Merck), piperidine (Merck), methanol (Silal Trading), acetone (Silal Trading), hydrochloric acid (Silal Trading), sodium hydroxide (Silal Trading), demineralized water were used without prior purification. The product obtained was analyzed using thin layer chromatography and FT-IR spectroscopy.

III.4.2. Experimental

Initially, the synthesis was performed according to the method described by E. Bush and W. F. Trager [353]. This method has been studied and modified so that the working procedure is easier and the product obtained is easier to purify.

III.4.3. Results and discutions

As in the case of dicoumarol, in the case of warfarin, the reaction evolution was also controlled using thin layer chromatography (TLC plates: Silica gel 60 F_{254} Merck, eluent: toluen:dioxan:acetic acid 7:0,5:0,15, developing: UV 254 nm, iodine vapour). The product purity was determined also using TLC, in order to determin the elimination of impurities (Fig. III-23, Fig. III-24, Fig. III-25, Fig. III-26, Fig. III-27).

Fig. III-23. Reaction in the first step, compared with starting materials.

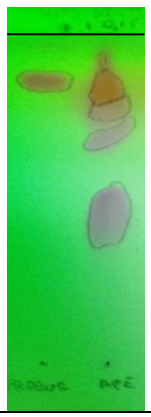


Fig. III -24. The ester resulted in the first stage of synthesis

Fig. III-25. The second stage of the reaction compared to the ester.

Fig. III-26. Toluen used for the extraction in the purification step.

Fig. III-27. Purified warfarin

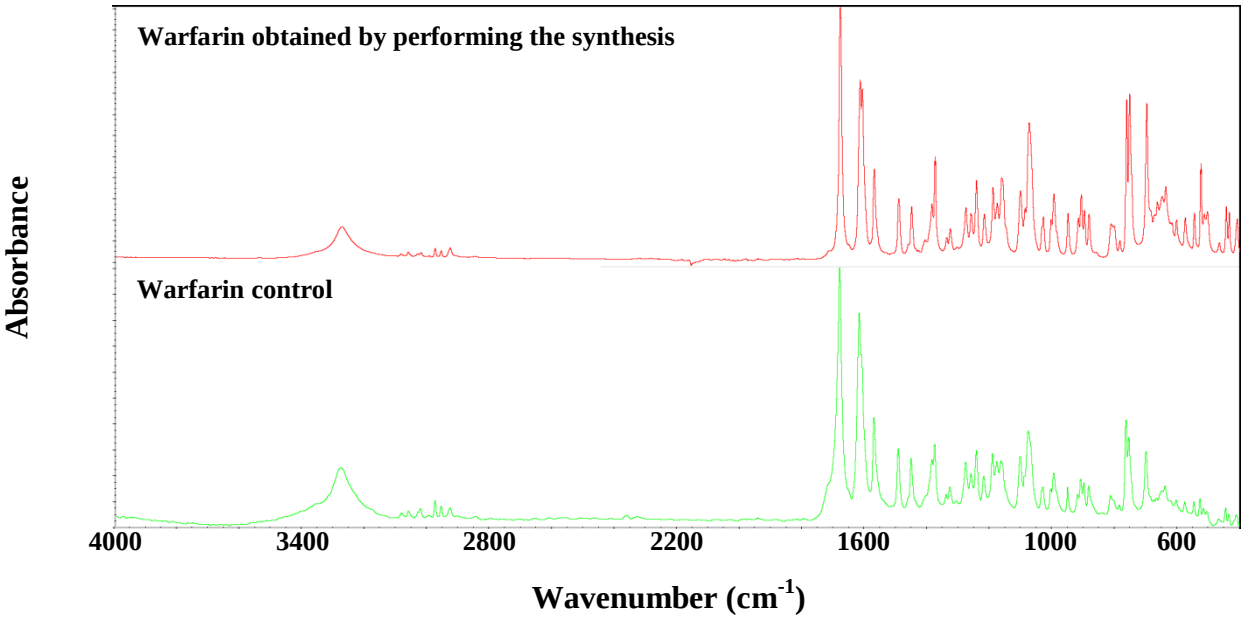


Fig. III-28. IR spectrum coresponding to warfarin control, compared with the spectrum of the product obtained by performing the synthesis

By comparing the spectrum of control warfarin with the spectrum of the product obtained by synthesis, it could be confirmed that the desired active principle was obtained. After purification, the warfarin obtained was in the form of pure product accordingly to the thin layer chromatography (Fig. III-27).

III.5. SURFACE MODIFICATION OF PVC USING SPIN COATING TECHNIQUE

In this study surface modification of PVC was performed in order to obtain a material that inhibits cellular adhesion and bacterial proliferation, and also to prevent blood clotting or the formation of calcifications. Therefor, the PVC surface was physically modified using spin coating technique to correct the irregularities arising from the synthesis, and to obtain a smooth, uniform surface.

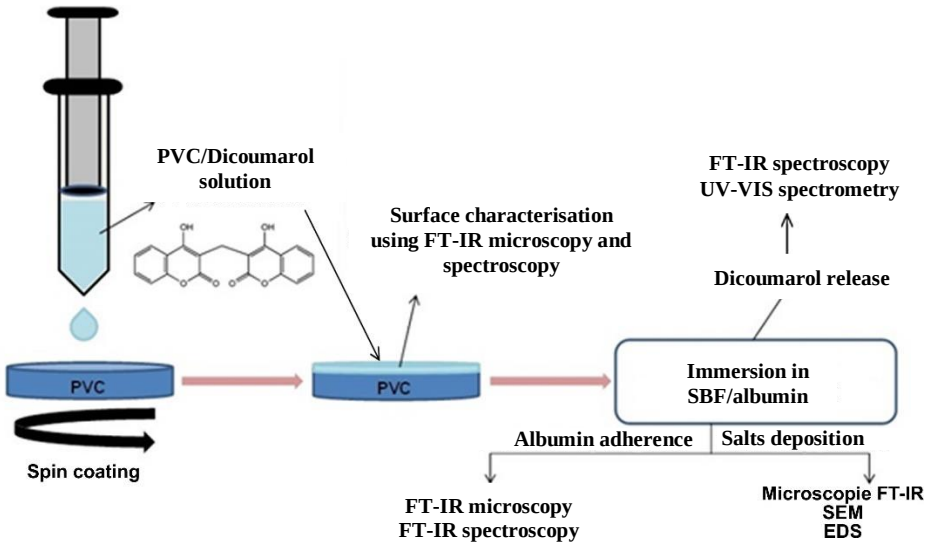
III.5.1. Modification performed using PVC/Dicoumarol film deposition

III.5.1.1. *Materials and methods*

In these experiments dicoumarol (previously synthesized), bovine serum albumin (Fluka), standard flat samples of PVC and tetrahydrofuran.

The modified samples were analyzed using a infrared microscop Thermo Scientific Nicolet iN10 in order to analyze the surface of the samples, and also using a infrared sprectroscope Thermo Fischer Nicolet iN10 Attenuated Total Reflection Infrared to determine the surface composition. Ultraviolet-visible spectrometry was performed using a Thermo Scientific Evolution 300 Spectrophotometer with the aim to determine dicoumrol release from the material surface in SBF (simulated body fluid). Scanning Electron Microscopy (SEM) and Energy Dispersive Spectroscopy (EDS) were performed in order to have a more clear view of the salts deposited on the surface of the modified PVC. Protein adsorbtion was studied using FT-IR microscopy and spectroscopy after several periods of immersion in SBF that contained albumin, mimicking the human body conditions. The antiaderent properties of the modified samples were analyzed by determining the values of colony forming units (CFU/mL). The samples were tested against 2 types of standard strains: Gram-positive (*Staphylococcus aureus* ATCC 25923) and Gram-negative (*Pseudomonas aeruginosa* ATCC 27853). For the performed tests, fresh cultures of 18-24 h were obtained after inoculation of bacterial strains on nutrient Agar medium.

III.5.1.2. *PVC/Dicoumarol film deposition*



III.5.1.3.*Results*

To confirm the presence of dicoumarol on the surface of the modified samples, the FT-IR spectra of the obtained samples were recorded and compared with the spectrum of

plain PVC and pure dicoumarol, respectively. Fig. III-29 presents a comparison between the FT-IR spectrum obtained at 4000 rpm, 7000 rpm and 10,000 rpm, respectively, the PVC and dicoumarol spectrum.

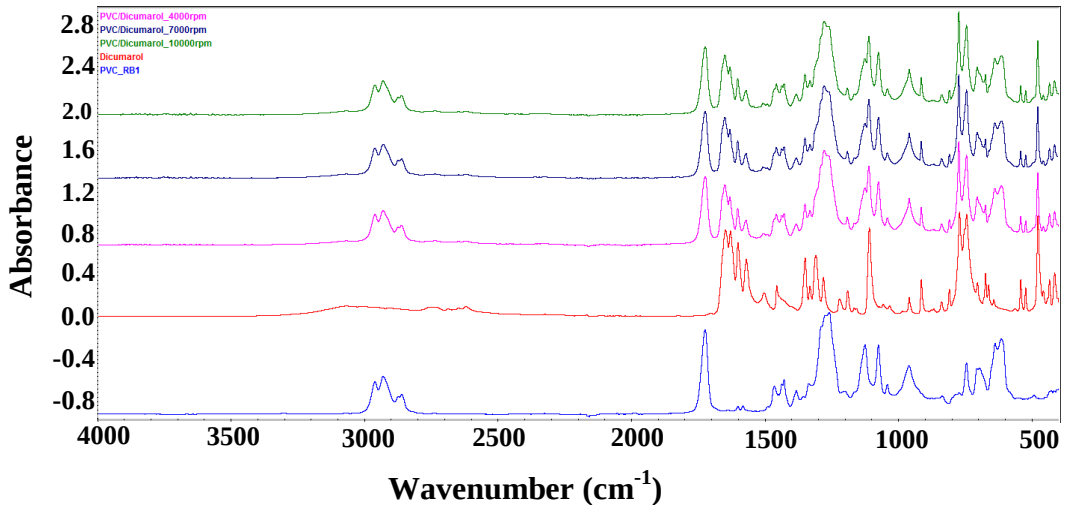


Fig. III-29. Comparison between the FT-IR spectrum obtained at 4000 rpm, 7000 rpm and respectively 10,000 rpm, the spectrum of PVC and of dicoumarol

In the obtained spectra the characteristic peaks of dicoumarol can be observed at 1650 cm^{-1} and 1350 cm^{-1}, corresponding to the ν (C = O) and ν (C – O) bonds in the rings. Also, the peaks present at 1110–1130 cm^{-1} indicate the presence of the ν bond (C – OH) and at 1070 cm^{-1} the presence of the ν (C – O – C) bond. In Fig. III-30.a-c. are presented the FT-IR microscopy images of the samples obtained at 4000 rpm, 7000 rpm and respectively 10,000 rpm.

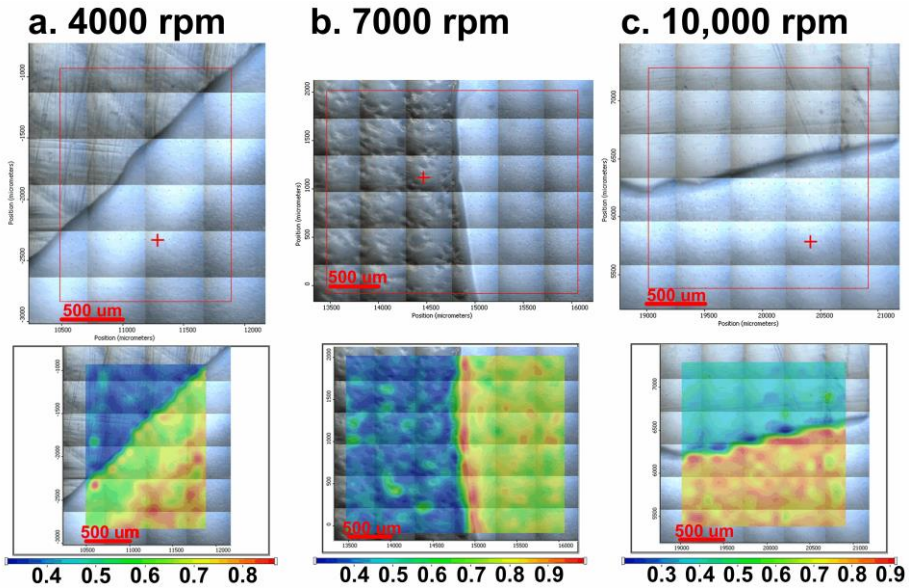


Fig. III-30. FT-IR microscopy images obtained at 1655 cm^{-1} for the deposition performed at 4000 rpm (a), at 1650 cm^{-1} for the deposition at 7000 rpm (b) and respectively at 1654 cm^{-1} for the deposition performed at 10,000 rpm (c).

a. Dicoumarol release from the film deposited on the surface of PVC

To determine whether the drug is being released from the surface, each sample was immersed in 100 mL of SBF solution at 36.5°C and thus maintained, with occasional stirring, for 30 days. The SBF solution was analyzed by UV-VIS at 303 nm (maximum adsorbtion wavelenght of dicoumarol) after various periods of time (1 h, 2 h, 4 h, 24 h, 2 days, 3 days, 7 days, 10 days, 14 days, 21 days and respectively 30 days) in order to observe the release behaviour of dicoumarol from the PVC film. Samples were also analyzed by FT-IR microscopy to determine if the surface morphology had changed in time after the contact with the SBF solution. FT-IR microscopy was also used to confirm the absence or presence of dicoumarol on the surface after a certain period of immersion. The results obtained using UV-VIS analysis are represented in the release graph corresponding to the samples obtained at 4000 rpm, 7000 rpm and respectively 10,000 rpm (Fig. III-31).

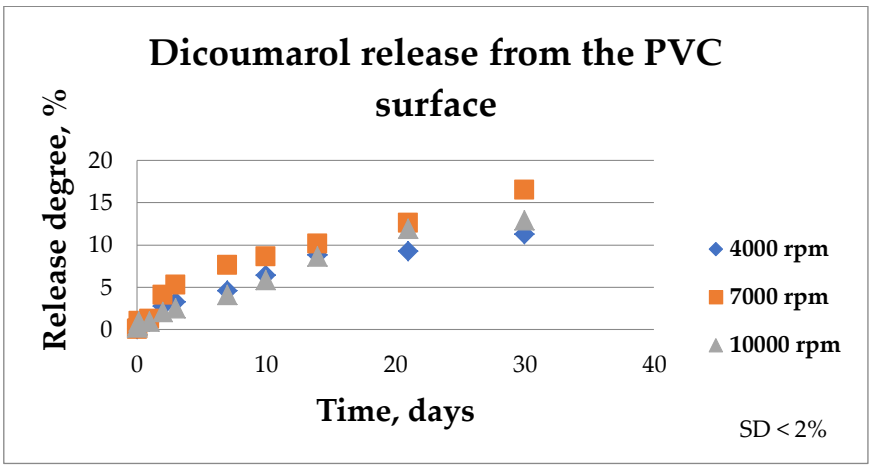


Fig. III-31. Dicoumarol release from the films obtained at 4000 rpm, 7000 rpm and respectively 10,000 rpm, analyzed using UV-VIS spectrometry at a wavelenght of 303 nm; standard deviation: SD < 2% in the case of all the represented points

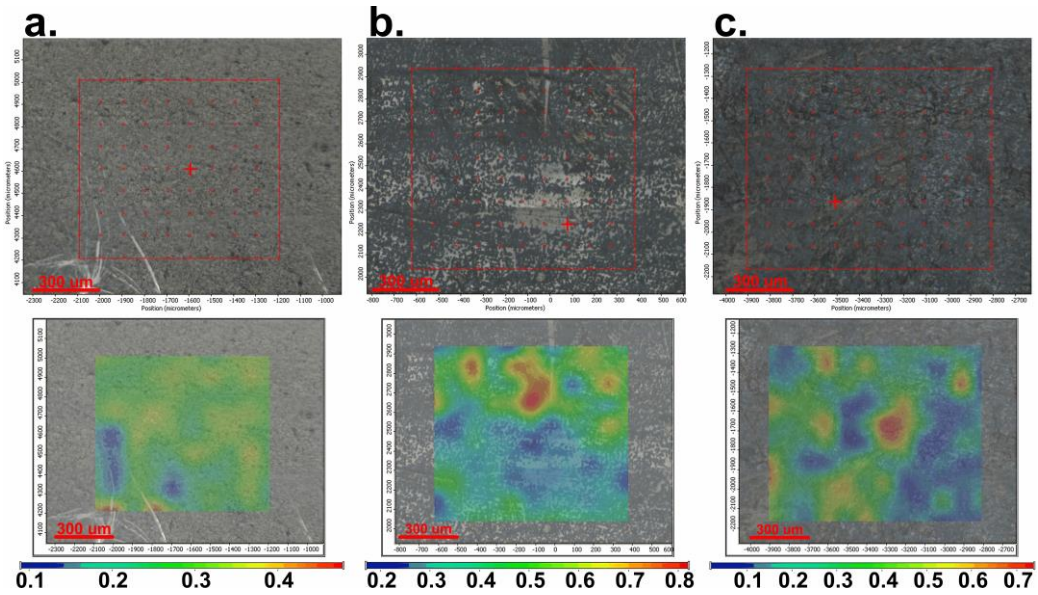


Fig. III-32. FT-IR images obtained at 1680 cm^{-1} for the film deposited at 4000 rpm: (a) after 7 days of immersion in SBF; (b) after 21 days of immersion in SBF; (c) after 30 days of immersion in SBF

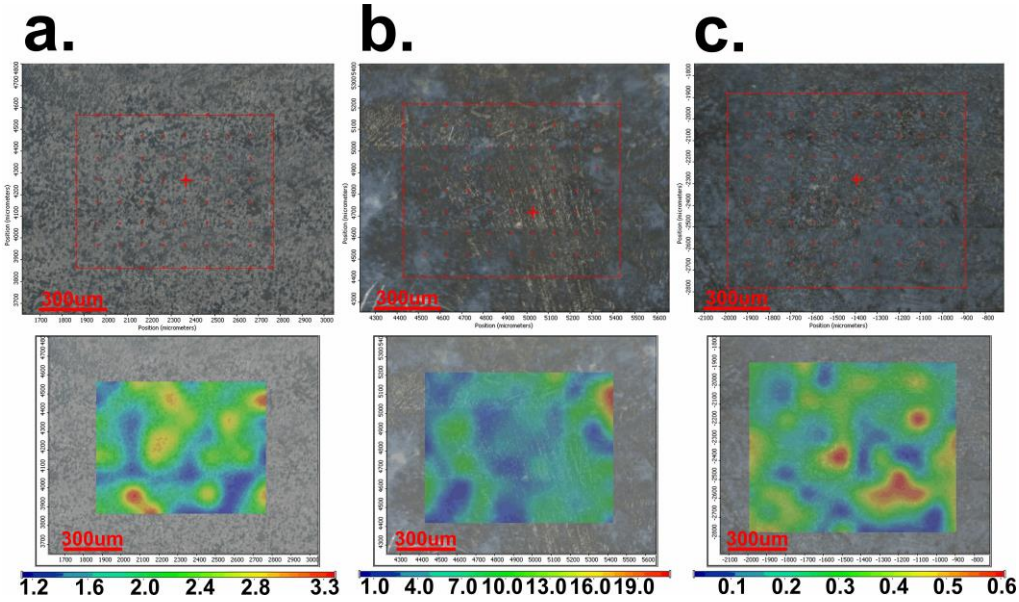


Fig. III-33. FT-IR images obtained at 1659 cm^{-1} for the film deposited at 7000 rpm: (a) after 7 days of immersion in SBF; (b) after 21 days of immersion in SBF; (c) after 30 days of immersion in SBF

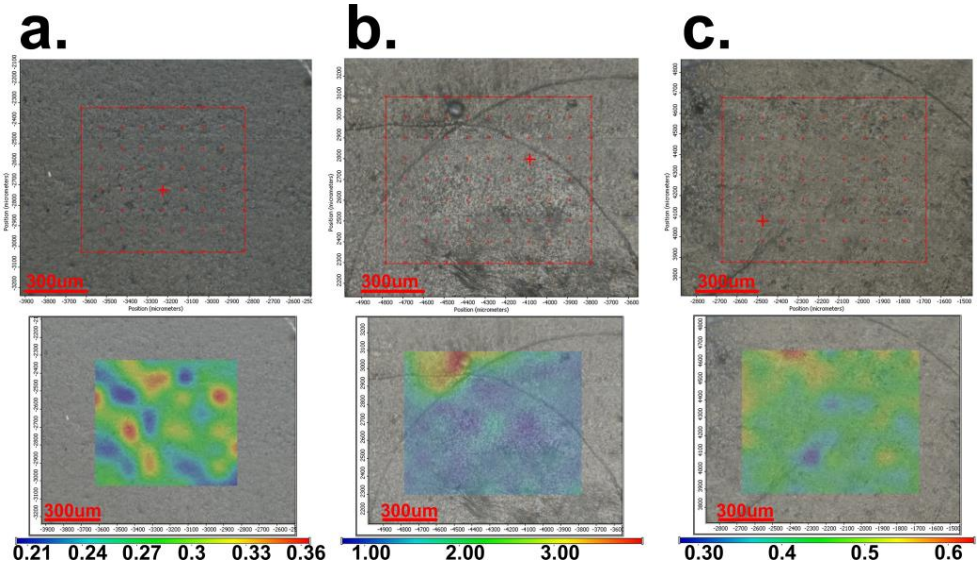


Fig. III-34. FT-IR images obtained at 1684 cm^{-1} for the film deposited at 10,000 rpm: (a) after 7 days of immersion in SBF; (b) after 21 days of immersion in SBF; (c) after 30 days of immersion in SBF

To determine whether the coating influenced the deposition of salts on the surface, a sample of unmodified PVC was also immersed in SBF for 30 days and then analyzed by FT-IR microscopy at 1723 cm^{-1} (characteristic wavelenght of simple PVC) for the same periods of time and under the same conditions as in the case of modified samples.

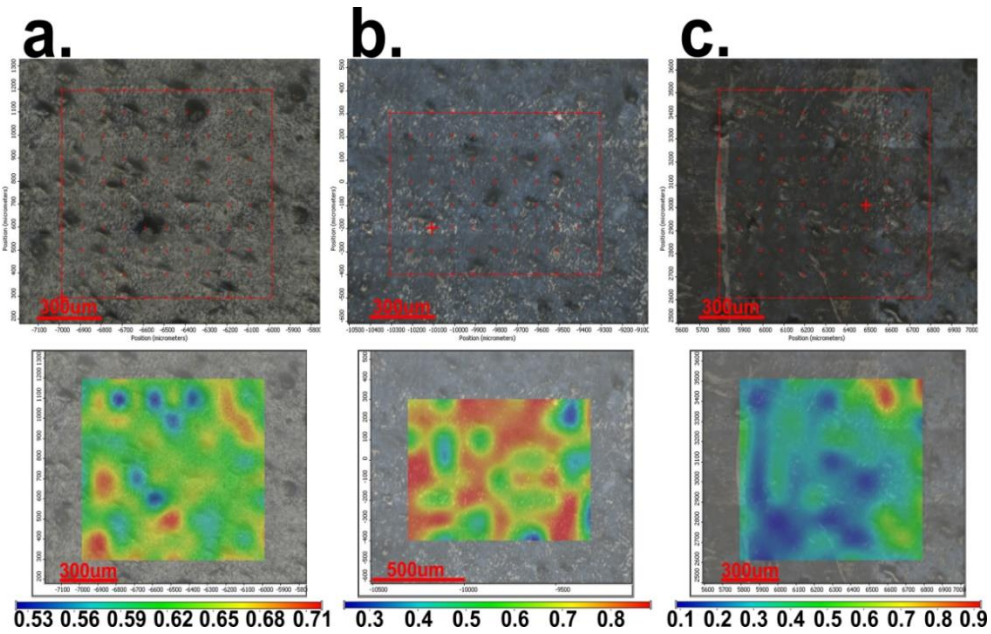


Fig. III-35. FT-IR images obtained at 1723 cm^{-1} for the initial, unmodified PVC: (a) after 7 days of immersion in SBF; (b) after 21 days of immersion in SBF; (c) after 30 days of immersion in SBF

Scanning Electron Microscopy (SEM) (Fig. III-36) and Energy Dispersive Spectroscopy (EDS) (Fig. III-37) were performed in order to have a clearer view of the salts that crystallized on the samples surfaces, and also to determine their composition.

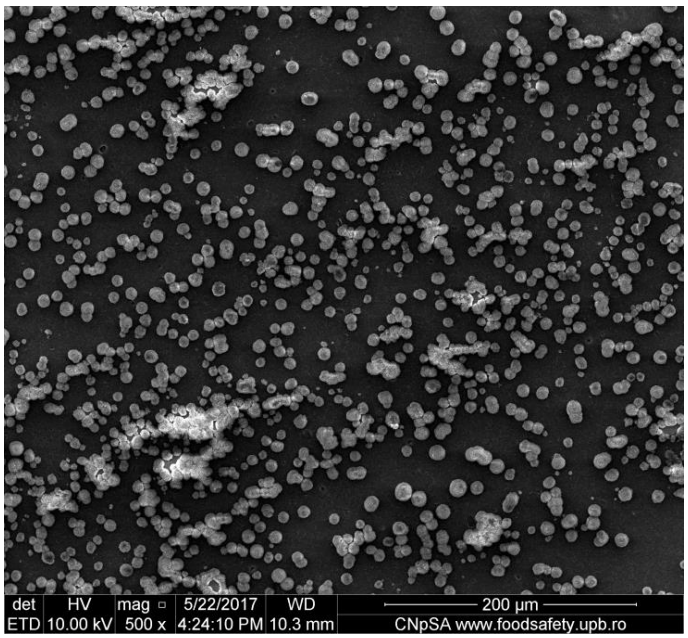


Fig. III-37. SEM image performed on the modified samples

FT-IR microscopy does not provide a very clear information about the presence of dicoumarol in the polymeric film, therefore the surface of the materials was analyzed using FT-IR spectroscopy in ATR mode after different periods of time. In order to be able to estimate the tendency of the active principle to be released from the deposited polymer film, the relative peak area corresponding to dicoumarol (~1650 cm^{-1}) from the FT-IR spectra was calculated.

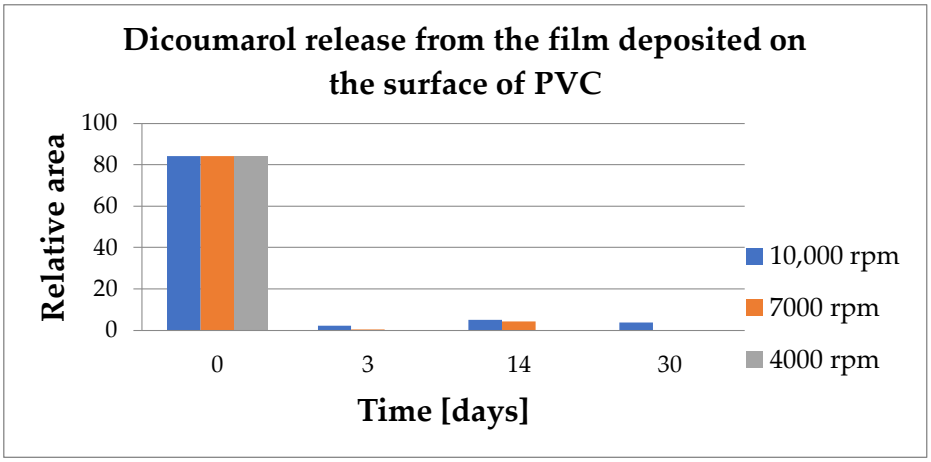


Fig. III-40. Schematic representation of the changes that occur over time in the case of the relative area of the peak corresponding to the dicoumarol incorporated on the surface of the sample as the release from the polymeric film occurs

b. Protein adsorbtion on the PVC surface

To study the behaviour of the material obtained by deposition of a thin film of PVC/diuoumarol at the interaction with the proteins from the biological fluids, the samples were immersed in a solution of SBF with a pH value of 7.4 containing 1% albumin (the main protein found in blood), at 36.5 °C. The samples were maintained in these conditions for 30 days.

To highlight the characteristic peaks of albumin and to observe whether they appear in the spectrum of samples immersed in the SBF / albumin solution, a PVC sample was coated with albumin by the drop-cast method. The spectrum corresponding to the sample obtained by the drop-cast method was therefore compared with the spectrum of samples (uncovered PVC, 4000 rpm, 7000 rpm, 10,000 rpm) after 7 days of immersion in the solution containing protein and the spectrum of PVC before immersion (Fig. III-41).

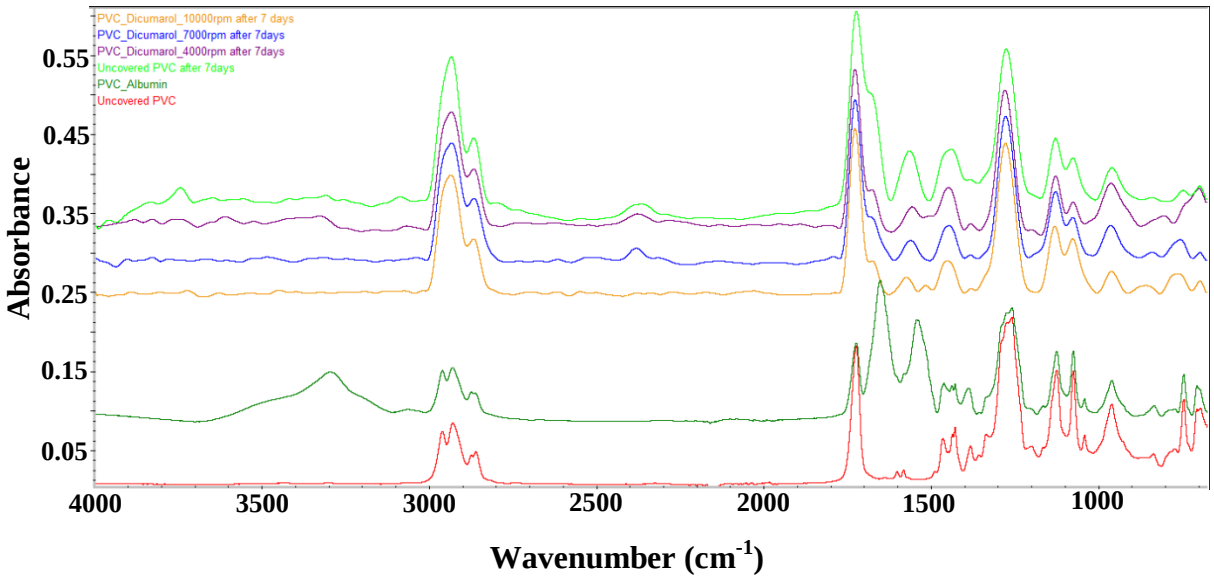


Fig. III-41. Comparison between the spectra of PVC/dicoumarol films after 7 days of immersion in the SBF/albumin solution, the spectrum of the uncovered PVC before immersion and respectively that of the sample obtained by drop-cast

In order to have a clearer picture of how the material reacts when it comes in contact with the albumin solution, to determine if this protein is adsorbed on the surface of the material, but also if its deposition leads to changes in morphology, the samples were analyzed. by FT-IR microscopy (Fig. III-42).

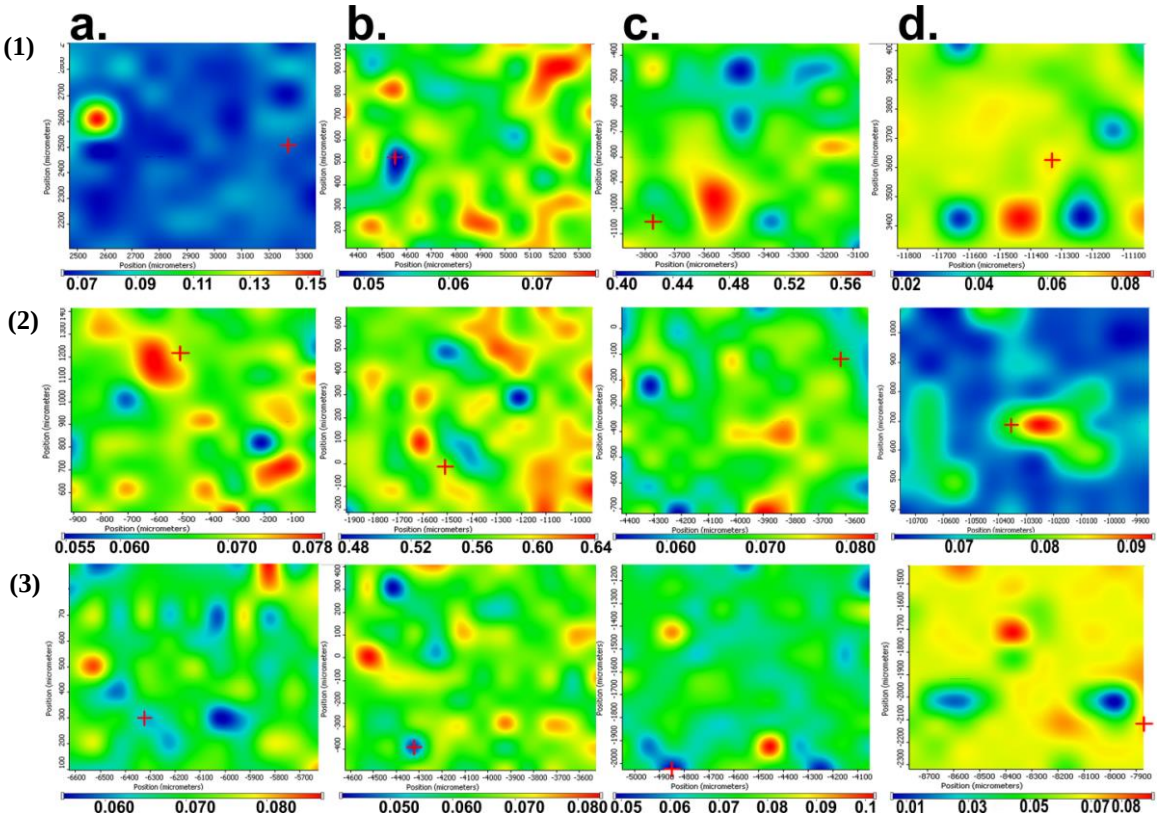


Fig. III-42. Contour maps of the samples obtained at 4000 rpm (a), 7000 rpm (b), 10,000 rpm (c) and respectively of the unmodified PVC (d), after immersion in SBF/albumin solution for 7 days (1), 21 days (2), and 30 days (3), obtained by FT-IR microscopy.

c. Bacterial adhesion

This study examined the resistance of the samples to a gram-positive strain (*S. aureus*) and a gram-negative strain (*P. aeruginosa*).

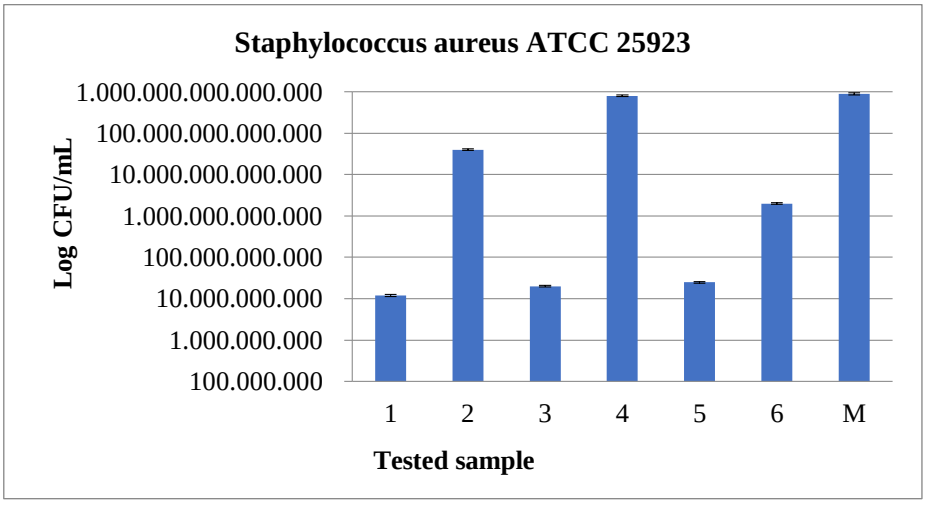


Fig. III-43. Graphical representation of the CFU/mL values for *Staphylococcus aureus* ATCC 25923 strain

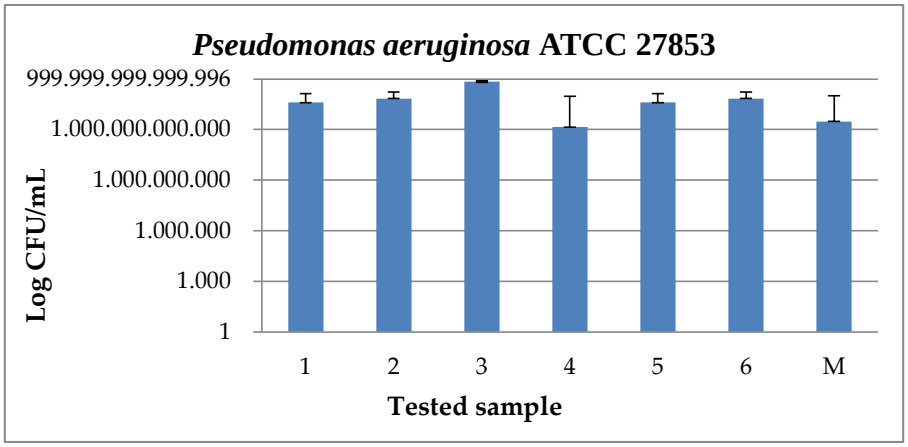


Fig. III-44. . Graphical representation of the CFU/mL values for *Pseudomonas aeruginosa* ATCC 27853 strain

III.5.1.4.*Conclusions*

The PVC surface can be successfully modified by the spin coating method. The obtained samples were analyzed on both the covered and the uncovered surface. According to FT-IR microscopy it can be concluded that the surface morphology is significantly changed when this coating method is applied. The initial surface was rough, uneven, with "hill-valley" defects that were covered with dicoumarol-doped PVC polymer film, resulting in a smooth, uniform surface.

After comparing the results obtained from the analyzes performed on all three samples, it can be concluded that the best results related to the surface morphology, the distribution of dicoumarol on the surface were observed for the sample obtained at 10,000 rpm. The release was also slower and more constant in the case of this sample and showed a good ability to prevent salts deposition and reduced protein adhesion. The proposed methodology has led to an improved anti-adherent activity of the PVC surface, especially against gram-positive strains such as *S. aureus*.

III.5.2. Silver nanoparticles deposition on the surface of PVC

The aim of this study was to modify the surface of PVC so that the bacterial adhesion is as low as possible, thus avoiding the discomfort caused by the side effects that occur after biofilm formation.

III.5.2.1. *Materials and methods*

Bovine Serum Albumin (Fluka), silver nitrate (Carl Roth), Trisodium Citrate (), standard flat samples of polyvinyl chloride and cyclohexanone (Silal) were used without prior purification. SBF (simulated body fluid) was prepared as described by Oyane *et al.* [359]. The samples were characterised using a Thermo Scientific Nicolet iN10 Infrared Microscope in order to analyze the surface morphology. The Thermo Fischer Nicolet iN10 Attenuated Total Reflection Infrared spectroscope was used to determine the surface composition of the samples obtained. Inductively Coupled Plasma (ICP) was performed to determine the degree of migration of silver ions from the surface. Scanning electron microscopy (SEM) was used to examine the morphology, microstructure and homogeneity of the samples as well as to visualize the distribution of silver nanoparticles on the surface.

III.5.2.2. *Experimental*

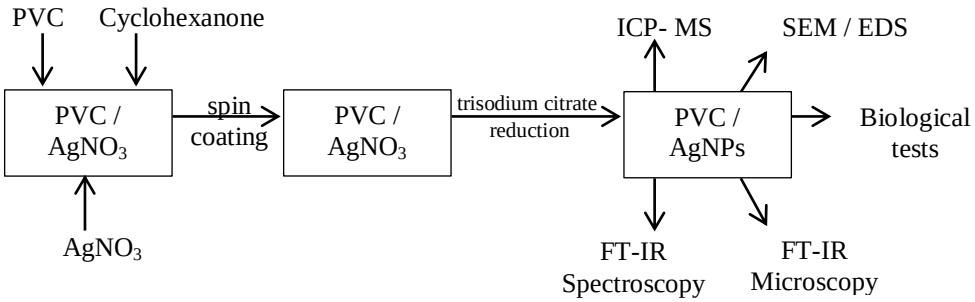


Fig. III-45. Technological flow scheme for the polymer film deposition

Table III-1. Parameters used in the spin coating method

Sample	$AgNO_3$	Dispenser	Spread	EBR	Dry
P1 **P4** **P7** **M1**	1% 5% 10% -	Acc. spin: 1000rpm Spin time: 5s Spin speed: 100rpm	Spin speed: 500rpm Spin accel: 1000rpm Spin time: 10s	Spin speed: 2000rpm Spin acc.: 100rpm Spin time: 20s	Spin speed: 4000rpm Spin accel: 1000rpm Spin time: 20s
P2 **P5** **P8** **M2**	1% 5% 10% -	Acc. spin: 1000rpm Spin time: 5s Spin speed: 100rpm	Spin speed: 500rpm Spin accel: 1000rpm Spin time: 10s	Spin speed: 2000rpm Spin acc.: 100rpm Spin time: 20s	Spin speed: 7000rpm Spin accel: 1000rpm Spin time: 20s
P3 **P6** **P9** **M3**	1% 5% 10% -	Acc. spin: 1000rpm Spin time: 5s Spin speed: 100rpm	Spin speed: 500rpm Spin accel: 1000rpm Spin time: 10s	Spin speed: 2000rpm Spin acc.: 100rpm Spin time: 20s	Spin speed: 10000rpm Spin accel: 1000rpm Spin time: 20s

III.5.2.3. *Results and discutions*

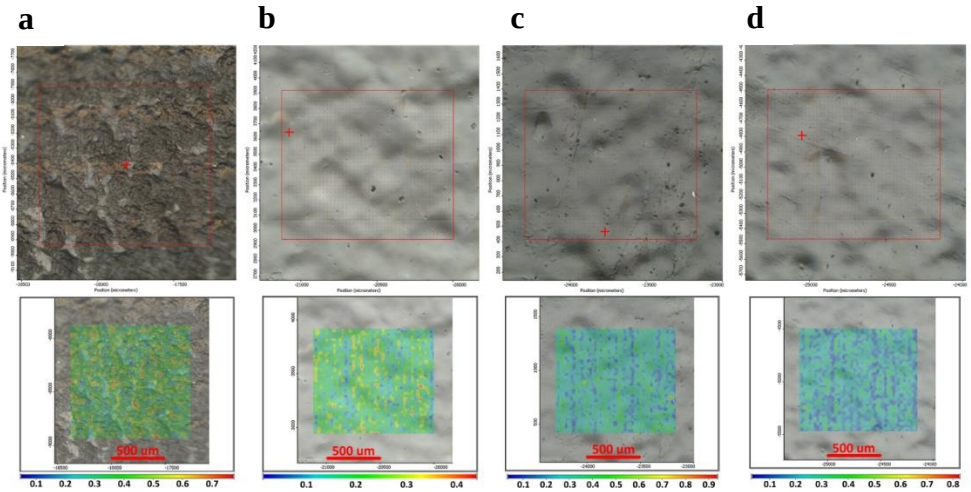


Fig. III-47. FT-IR microscopy images coresponding to the uncovered PVC (a) and to samples M1 (b), M2 (c), M3 (d)

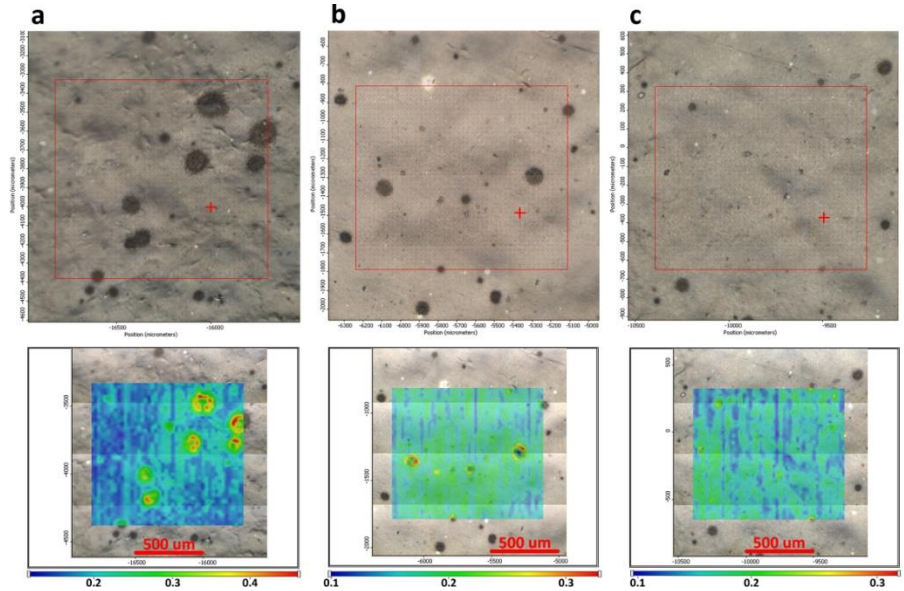


Fig. III-48. FT-IR microscopy images coresponding to samples P7(a), P8(b) and respectively P9(c)

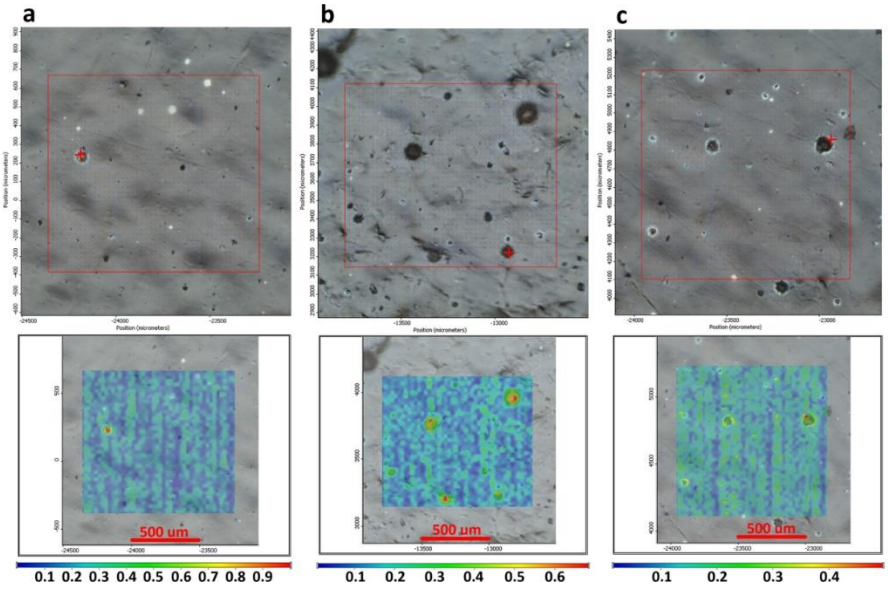


Fig. III-49. FT-IR microscopy images coresponding to samples P7 (a), P8 (b) and P9 (c) after the reduction was performed

Silver nanoparticles present on the surface of the thin films deposited by spin coating was confirmed by SEM (Fig. III-50) and showed a uniform dispersion of silver nanoparticles on the surface.

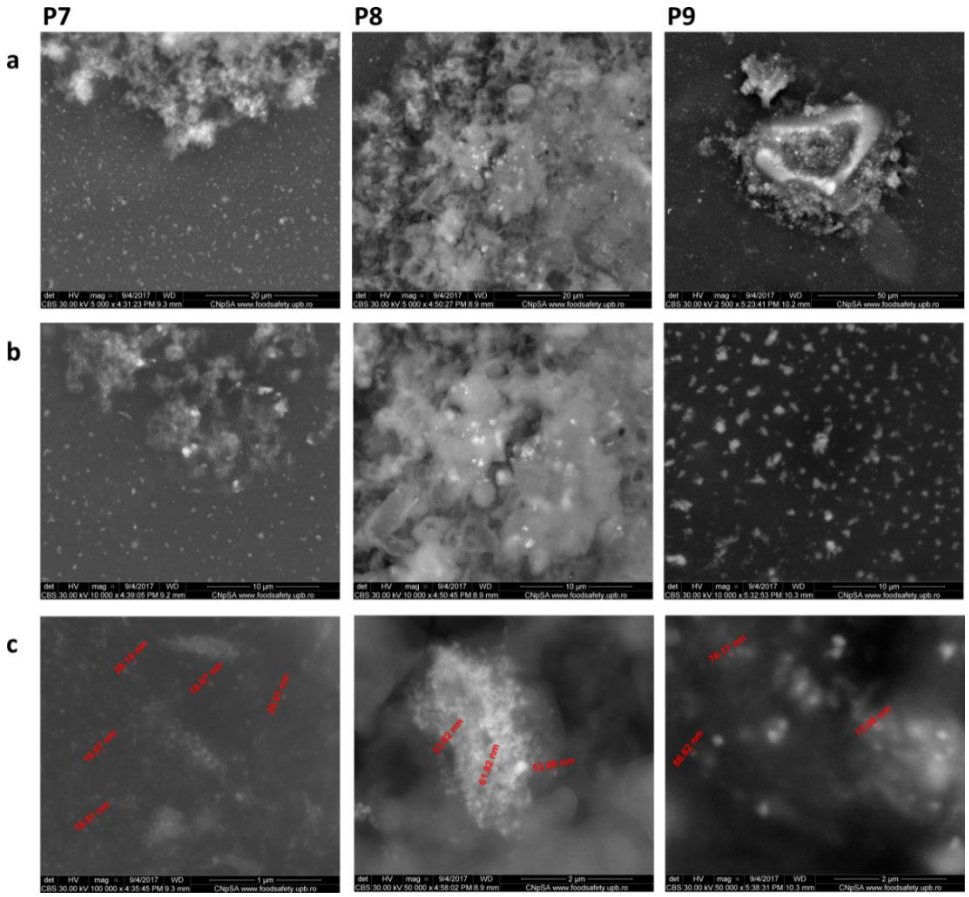


Fig. III-50. Imaginile SEM images recorded at a magnification of 5000 x (a), 10,000 x (b) and respectively 50,000 x for the samples P7, P8, P9

EDS spectra of the samples characterised after the reduction was performed, also confirmed the presence of silver and reveals significant differences in the amount deposited.

a. Silver ions migration

A very important aspect to consider when using silver nanoparticle coatings for medical devices is related to the concentration of ions released in biological fluids because it can present cytotoxicity at certain concentrations. In order to monitor the migration behavior of silver ions in the body fluids, the samples were immersed in SBF with pH = 7.4, maintaining the temperature at 36.5 ºC, thus mimicking the conditions in the body. These were kept in solution for up to 24 days, periodically analyzing the SBF solution (1h, 2h, 6h, 1 day, 2 days, 3 days, 9 days, 13 days and 23 days, respectively), using ICP-MS analysis (Fig. III-52).

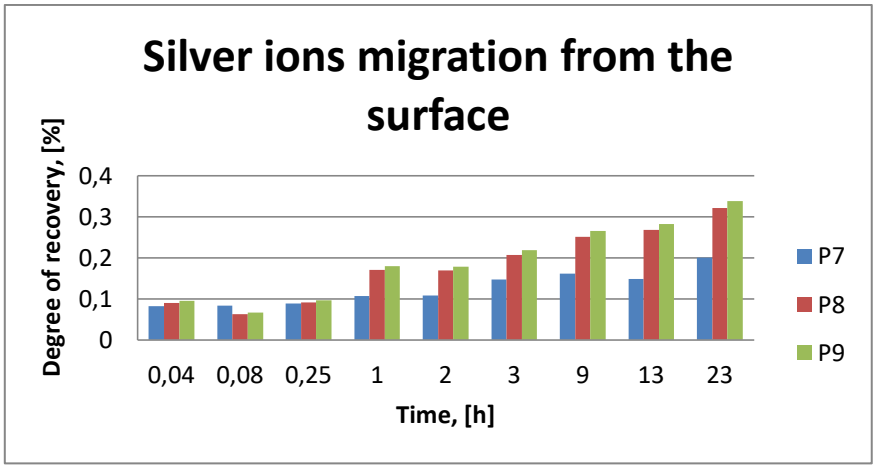


Fig. III-52. Graphical representation of the recovery degree of silver ions fron the SBF solution, in time

b. Albumin adsorbtion on the surface

Adhesion of proteins to the surface of the material can lead to the formation of thrombosis, calcifications, bacterial adhesion or biofilm formation, which can lead to blockage of the tubular devices. In order to observe the behavior of the surfaces obtained by spin coating at the interaction with blood proteins, the samples were immersed in a solution of SBF pH = 7, at 36.5 ºC with albumin content, the main protein found in the blood. The samples were maintained under these conditions for 24 days and were analyzed by FT-IR microscopy after 1 day, 6 days, 14 days and 24 days, respectively.

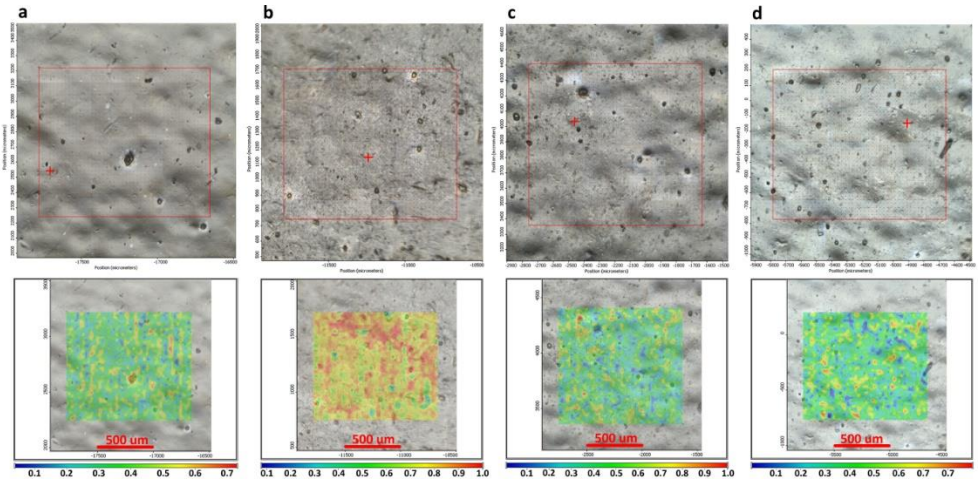


Fig. III-53. FT-IR microscopy images obtained for sample P7 after: a - 1 day, b - 6 days, c – 14 days, d – 24 days of immersion in SBF/albumin solution

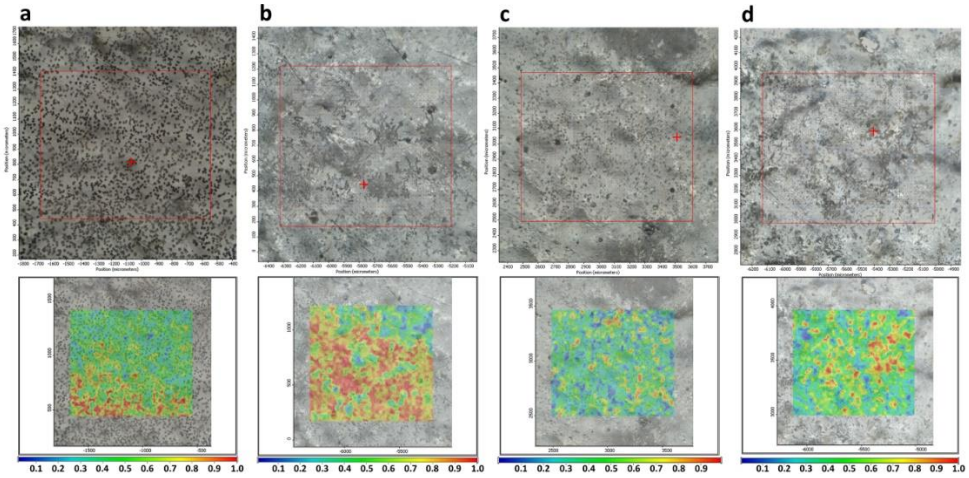


Fig. III-54. FT-IR microscopy images obtained for sample M1 after: a - 1 day, b - 6 days, c – 14 days, d – 24 days of immersion in SBF/albumin solution

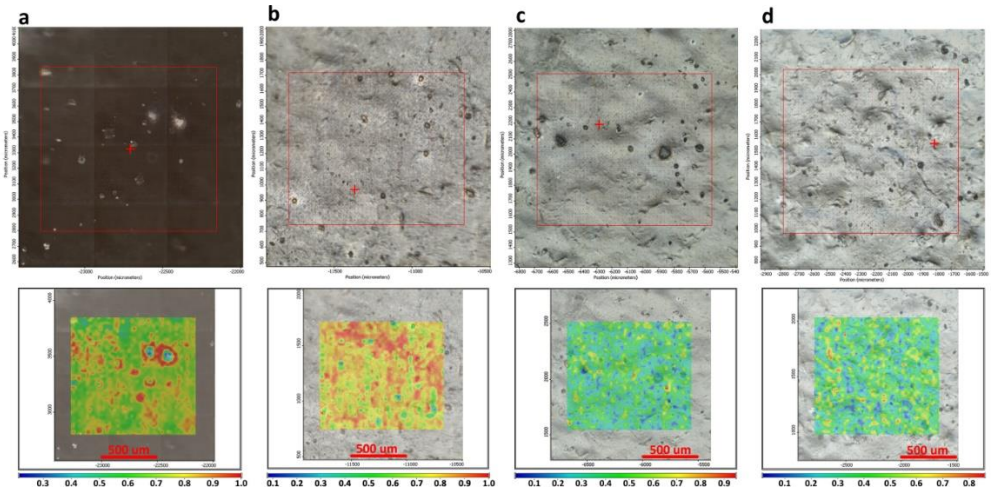


Fig. III-55. FT-IR microscopy images obtained for sample P8 after: a - 1 day, b - 6 days, c – 14 days, d – 24 days of immersion in SBF/albumin solution

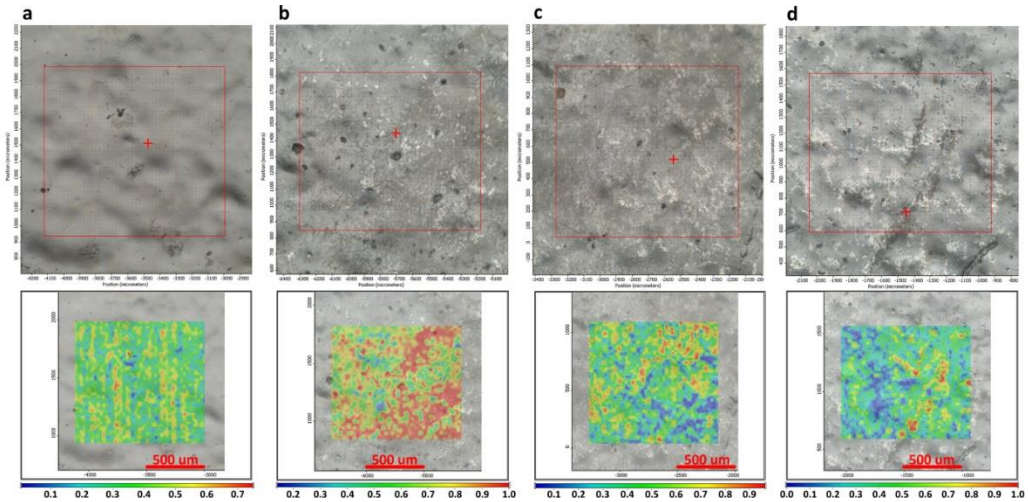


Fig. III-56. FT-IR microscopy images obtained for sample M2 after: a - 1 day, b - 6 days, c – 14 days, d – 24 days of immersion in SBF/albumin solution

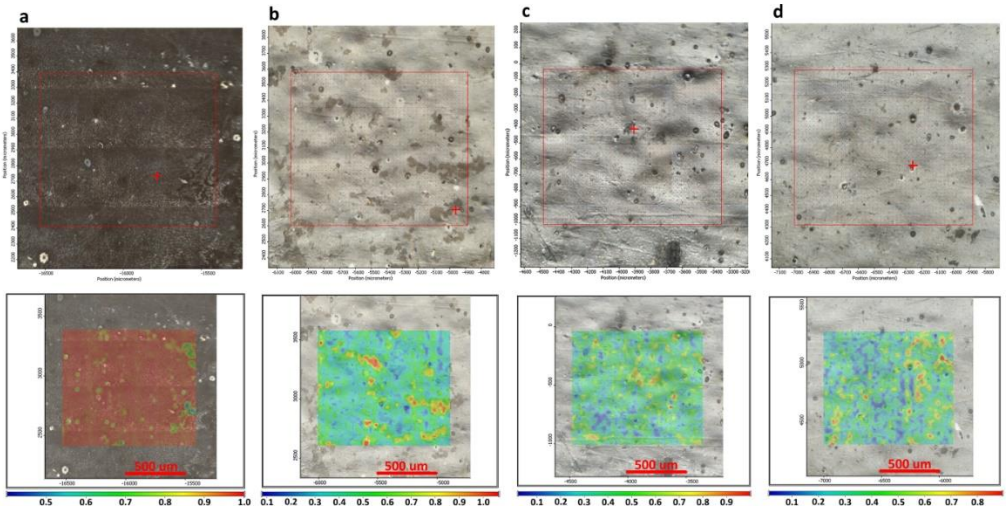


Fig. III-57. FT-IR microscopy images obtained for sample P9 after: a - 1 day, b - 6 days, c – 14 days, d – 24 days of immersion in SBF/albumin solution

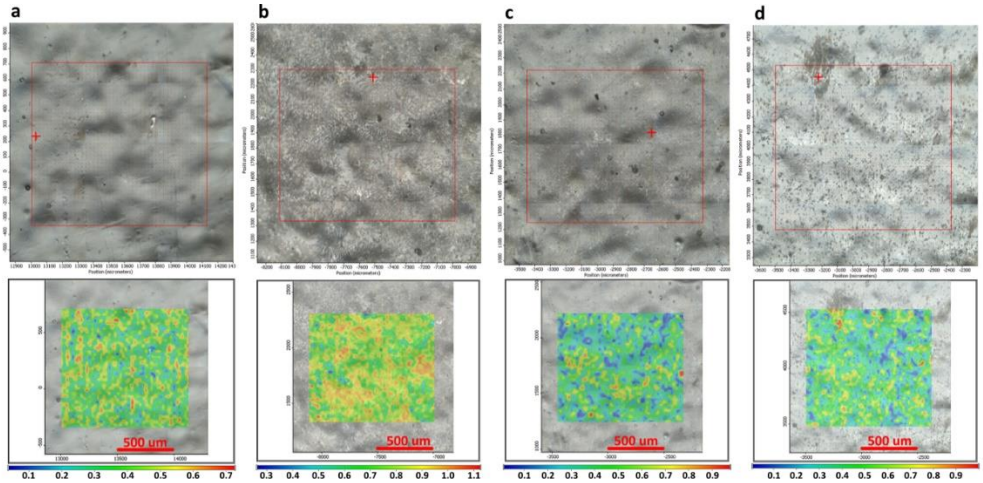


Fig. III-58. FT-IR microscopy images obtained for sample M3 after: a - 1 day, b - 6 days, c – 14 days, d – 24 days of immersion in SBF/albumin solution

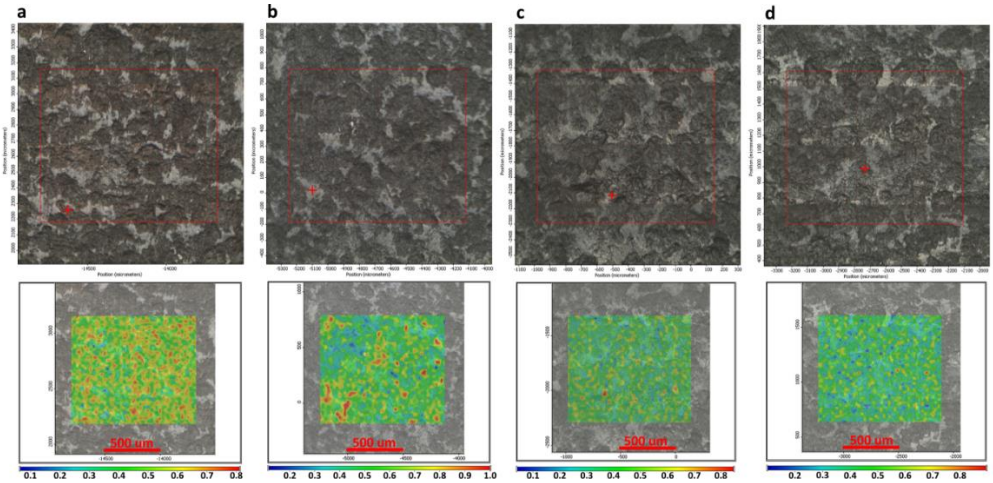


Fig. III-59. FT-IR microscopy images obtained for the initial, uncovered PVC after: a - 1 day, b - 6 days, c – 14 days, d – 24 days of immersion in SBF/albumin solution

The FT-IR spectrum (Fig. III-60) confirms the presence of albumin on the sample surface by the appearance of additional peaks after immersion in the albumin solution. These peaks occur at wavelengths of 1680 cm^{-1}, the band corresponding to amide I, consisting mostly of vibrations of the υ bond (C = O) and at ~1580 cm^{-1} the band corresponding to amide II, consisting of the most of the vibrations of the υ (C - N) bond [387-389].

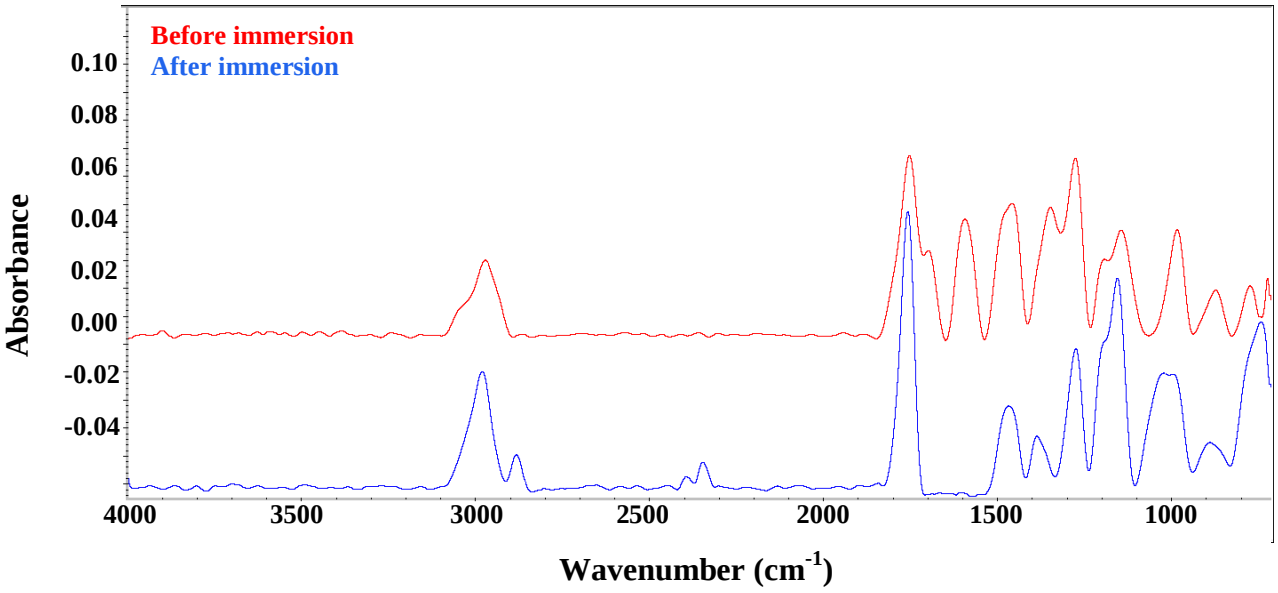


Fig. III-60. Graphical representation of the FT-IR spectra of the surface before and after immersion in the SBF/albumin solution

c. Biological tests

To test the resistance to colonization of the modified surfaces, but also of the initial, uncovered surface, the bacterial strain *S. aureus* ATCC 6538 was used, the final results being expressed in CFU/ml after different time intervals from the contact of these surfaces with the bacterial suspension. (Fig. III-61).

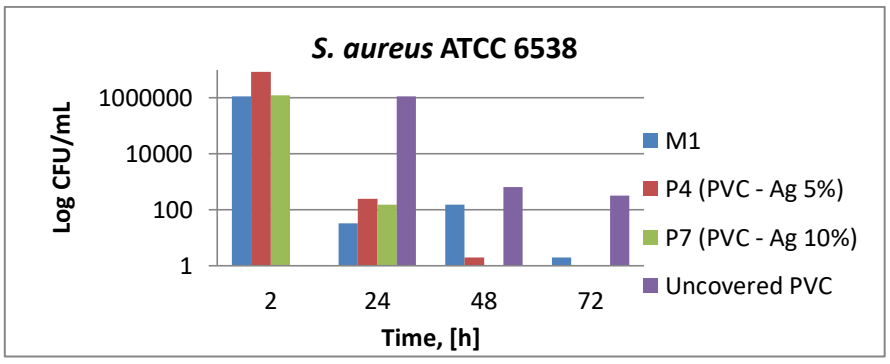


Fig. III-61. Quantitative evaluation of the degree of development of the monospecific microbial biofilm developed by *Staphylococcus aureus* ATCC 6538 at the initial PVC surface and respectively to the modified surfaces.

E. coli is the most representative bacterial species among Gram-negative species, capable of developing biofilms, therefore the ability of the samples to form a monospecific biofilm on contact with the standard strain *Escherichia coli* ATCC 25922 was tested. (Fig. III-62).

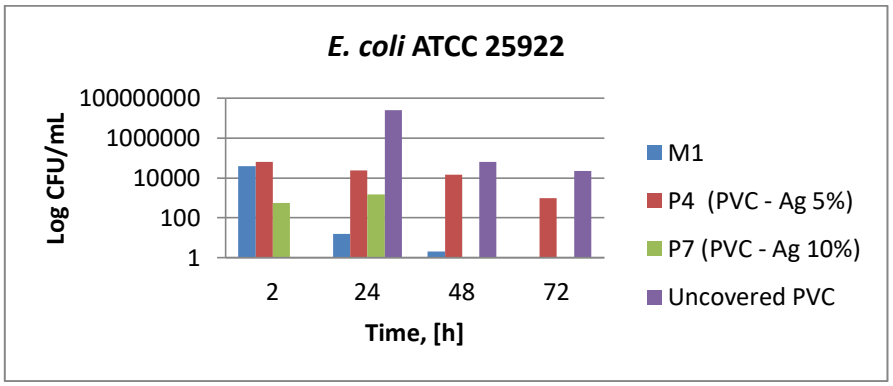


Fig. III-62. Quantitative evaluation of the degree of development of the monospecific microbial biofilm developed by *E. coli* ATCC 25922 at the initial PVC surface and respectively to the modified surfaces

Nosocomial infections associated with venous catheters are frequently caused by Candida species. Induction of biofilm formation on the surface of functionalized biomaterials used in the present study allowed the evaluation of their behavior in contact with planktonic *C. albicans* cells ATCC 26790 (Fig. III-63).

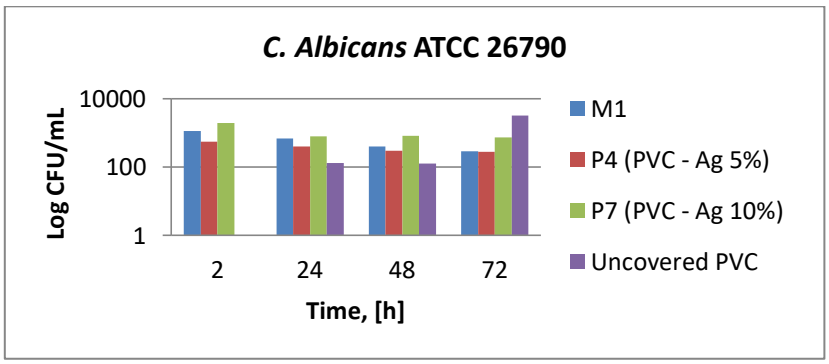


Fig. III-63. Quantitative evaluation of the degree of development of the monospecific microbial biofilm developed by *C. albicans* ATCC 26790 at the initial PVC surface and respectively to the modified surfaces

III.6. SURFACE EVALUATION OF OXYNITRIDE COATINGS (TIOxNy) USED FOR OBTAINING LAYERED CARDIOVASCULAR STENTS

In this paper were studied TiOxNy coatings with different N / O ratios deposited by magnetron sputtering deposition, in order to determine how the properties of this type of surface are influenced by the concentration of N or O.

III.6.1. Materials and methods

The studied TiOxNy coatings were deposited both on flat samples, in the form of discs, and also on stainless steel stents. The coatings were performed using various concentrations of nitrogen (ratio O: N = 1: 2, 1: 5, 1:10) in the gas supply flow. The depositions were performed using magnetron sputtering technique, using a UVN-200 MI system at medium frequency.

The surface morphology was analyzed before and after exposure to SBF by Scanning Electron Microscopy (SEM) using a Philips ESEM-FEG XL 30 microscope, 3.0 kV. Chemical and elemental analysis was performed using FT-IR spectroscopy and EDS (Energy-dispersive X-ray spectroscopy). To determine the surface hydrophilicity, the flat samples were

analyzed using the OCA 20 Contact Angle System. The interaction of the samples with blood proteins was studied by immersion in an SBF/albumin solution. The samples were analyzed at different time intervals by FT-IR microscopy. The biocompatibility of the obtained coatings was analyzed using Human Umbilical Vein Endothelial Cells (HUVEC).

III.6.2. Results and discutions

The samples obtained were analyzed by SEM and as it can be seen from Fig. III-63, the characteristics of the films obtained differ depending on the O/N ratio used.

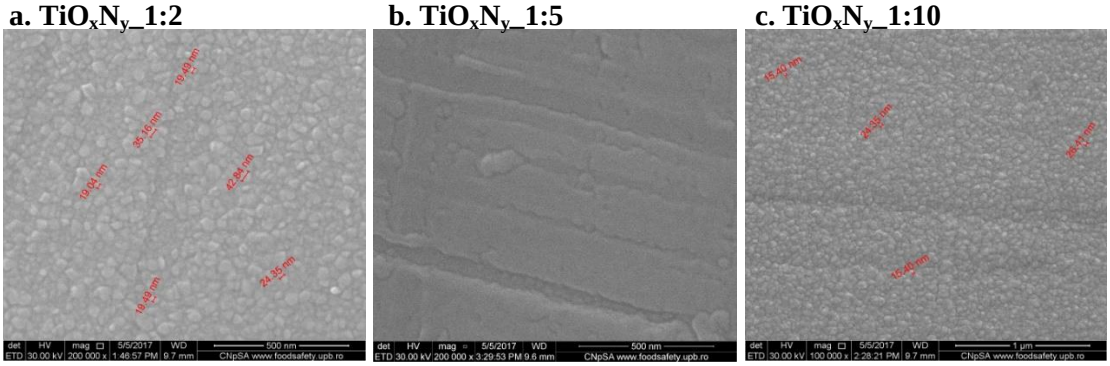


Fig. III-64. SEM images corresponding to samples covered with TiO_xN_y (c).

To analyze the uniformity of the depositions, the flat samples were analyzed by FT-IR microscopy (Fig.III-65).

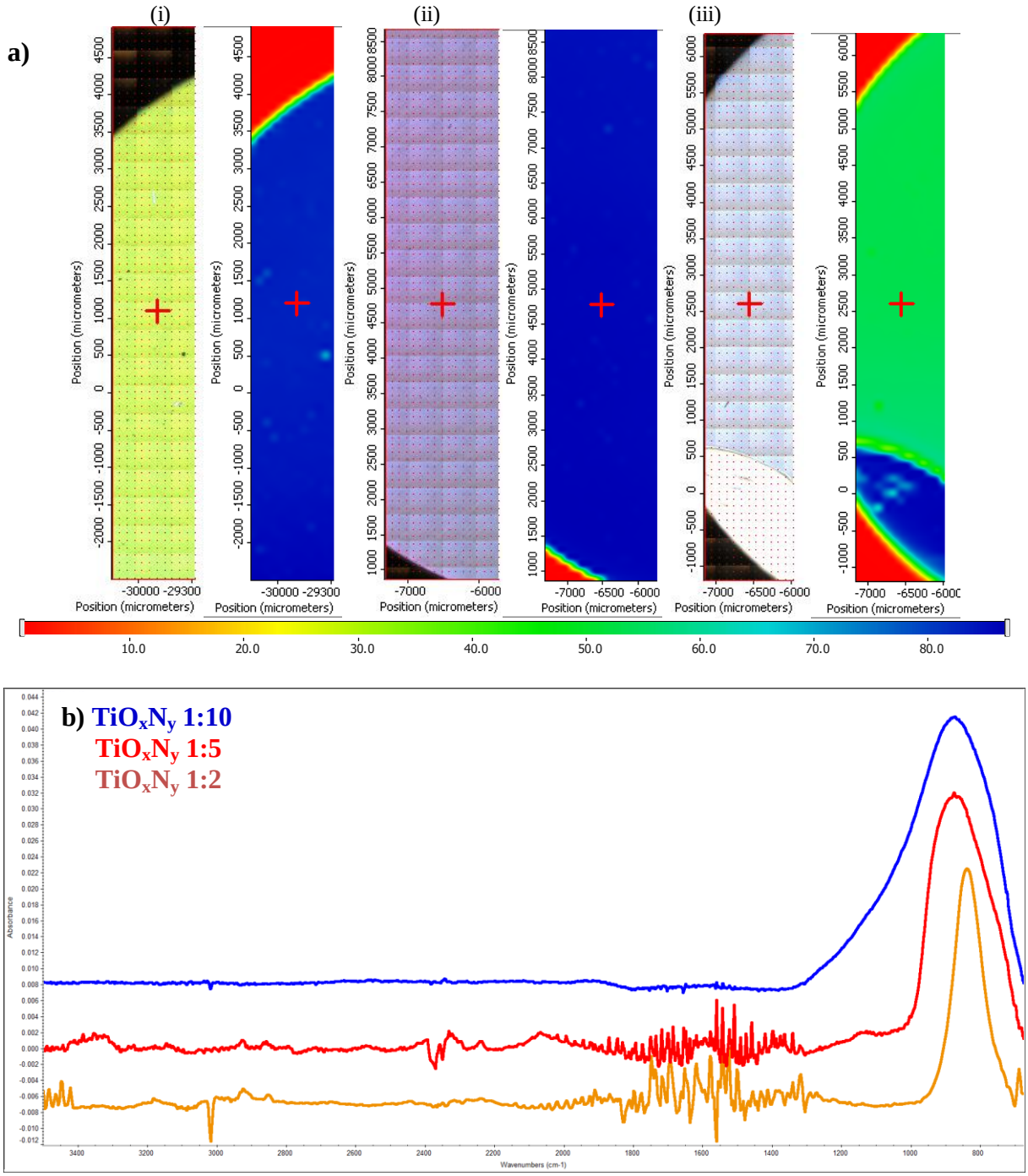


Fig. III-65. FT-IR microscopy images corresponding to the flat samples covered with TiO_xN_y (a.i – O_2/N_2 1/2; a.ii - O_2/N_2 1/5; a.iii - O_2/N_2 1/10), recorded at 840-884 cm^{-1} (b –FT-IR spectra corresponding to the modified surfaces, highlighting the peak at which the recording was performed).

The surfaces of the stents coated with TiO_xN_y by magnetron sputtering were analyzed both at micro and nano scale. (Fig.III-66).

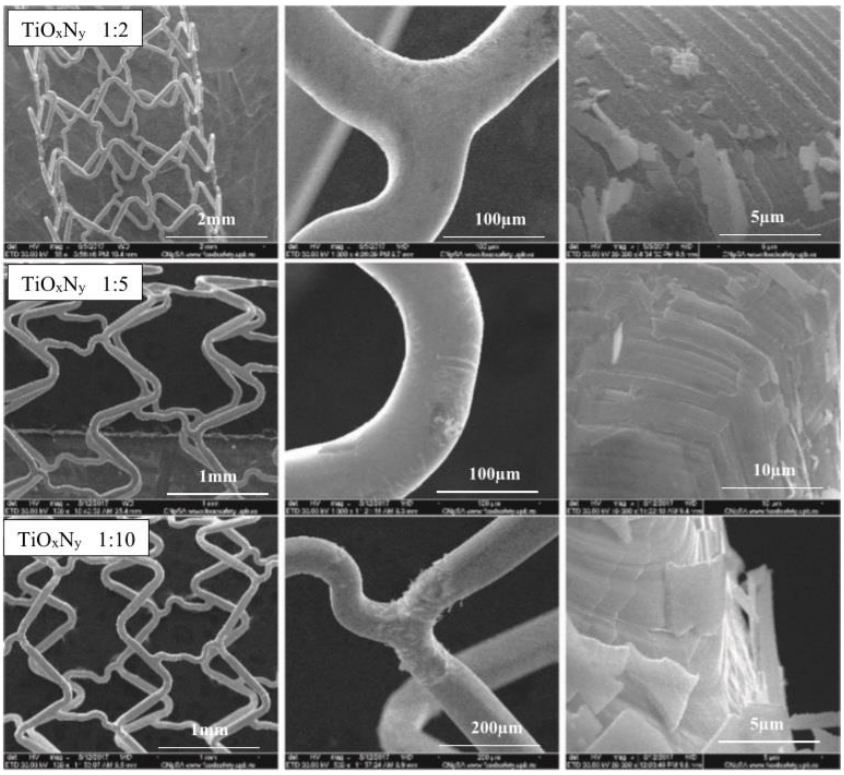


Fig. III-66. SEM images corresponding to samples covered with TiO_xN_y , at magnifications of 55x (a), 2000x (b) and respectively 200,000x (c).

Although the surface of the stent presented only limited areas showing detachments of the coating, the deposition technology must be improved because these coatings are performed in order to be used at the manufacture of the stent, and degradation and peeling of the surface film must be removed.

The measurement of the contact angle was performed on all flat samples. It was determined that all three coatings made induce a slightly hydrophobic behavior of the surfaces, the contact angle being greater than 90º. The determined value of the contact angle corresponding to the stainless steel is approximately 47º and is in accordance with the value in the literature [398].

III.6.2.1. *In vitro stability*

To determine the way the modified stents covered with TiO_xN_y interacts with the body fluids, the samples were immersed in SBF for several days. SEM images corresponding to the samples after 7 days of immersion in SBF are presented in Fig. III-67. To verify whether Ti-OH groups can indeed induce the nucleation and crystallization of apatite, but also to determine the chemical nature of the precipitate deposited on the surface of the stents after immersion in SBF [400], elemental mapping using EDS was performed on the flat samples.

The distribution of the elements on the surface is shown in Fig. III-67 and confirms the presence of phosphates, calcium, magnesium.

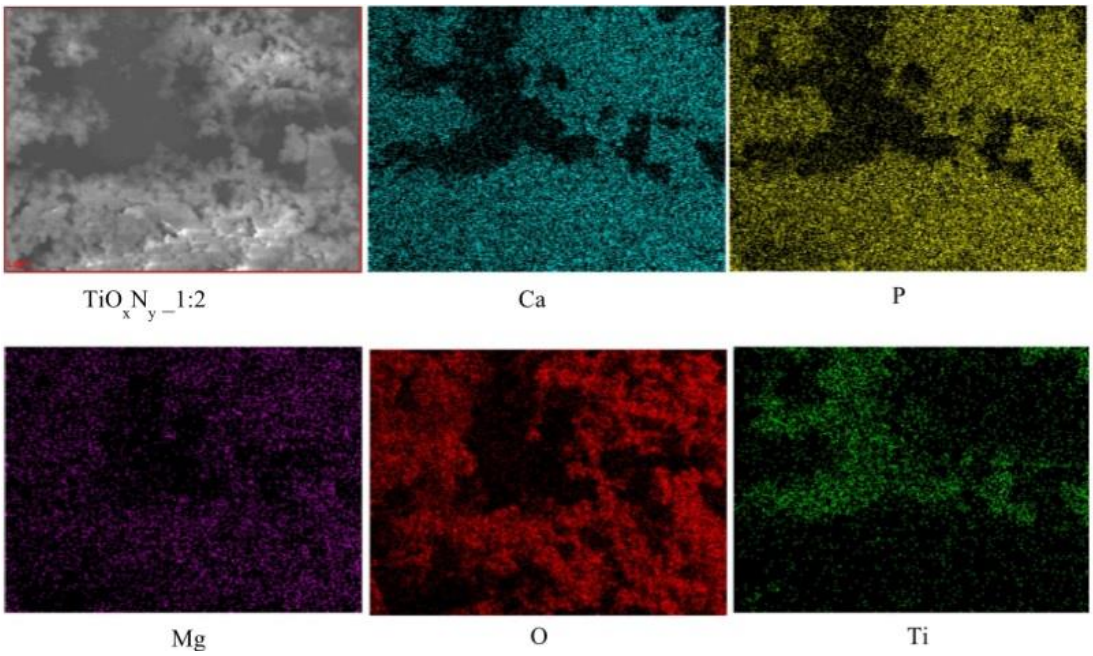


Fig. III-68. Elemental mapping of the precipitate formed on the surface of TiOxNy coatings after 7 days of immersion in SBF

III.6.2.2. *Protein adsorbtion (albumin)*

To determine the behavior of the samples in the presence of proteins in the blood, they were immersed in a solution of albumin in SBF for 1, 3, 8, 14 and 28 days. They were then carefully washed with water and analyzed by FT-IR microscopy.

After 14 days, the albumin adsorption started to be present on the surface of the samples O2 / N2 = 1/5 (Fig. III-73) and O2 / N2 = 1/10 (Fig. III-74). The spectra corresponding to the areas where the wires meet are different from the other areas of the stents, because the adsorption of albumin takes place predominantly on the areas where the TiOxNy deposition was degraded. Defects arising from the deposition detachment favor the anchoring of proteins on the surface.

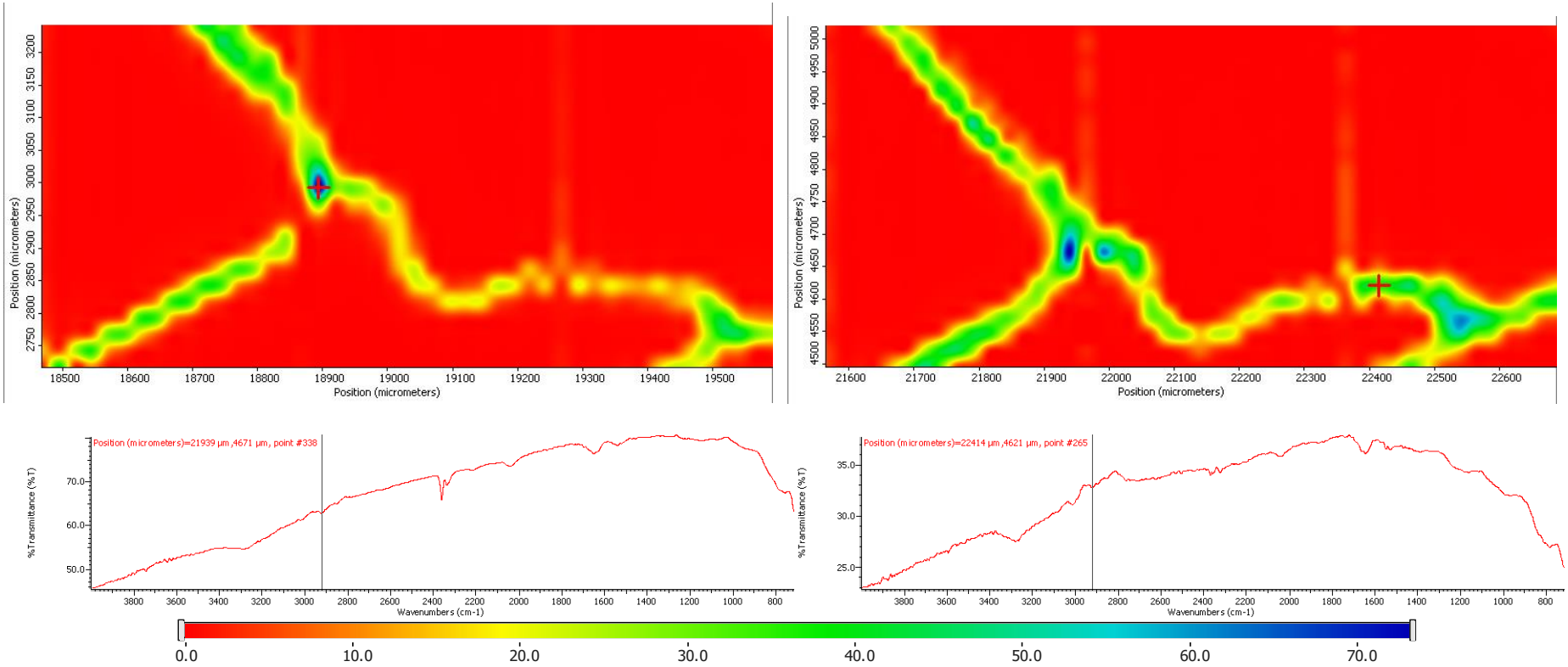


Fig. III-73. Contour maps and FT-IR spectra corresponding to sample TiO_xN_y O_2/N_2 = 1/5, at the site of wire conjunction, and also on another site on the wire surface, after 14 days of immersion in albumin.

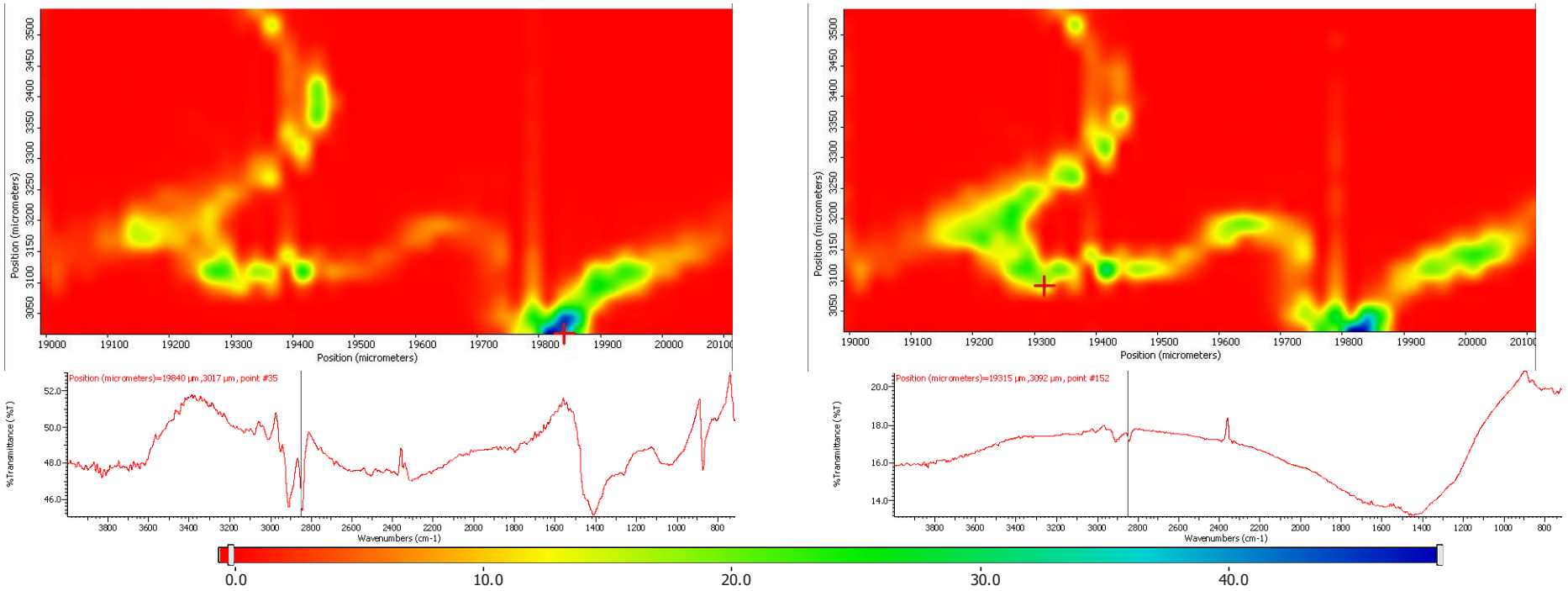


Fig. III-74. Contour maps and FT-IR spectra corresponding to sample TiO_xN_y O_2/N_2 = 1/10, at the site of wire conjunction, and also on another site on the wire surface, after 14 days of immersion in albumin.

The analyzes performed revealed that the TiO_xN_y coating improves the inhibitory properties of albumin deposition on the surface of stainless steel stents.

III.6.2.3. *Biological tests*

The metabolic activity of HUVEC after 24 h and 48 h of incubation on TiO_xN_y-coated flat disks, was compared with the stainless steel control sample. As no changes in the metabolic activity of the cells were observed, it can be concluded that the coatings performed do not show toxicity for HUVEC cells (Fig. III-75). After 48 hours all samples coated with TiOxNy showed slightly better metabolic activity compared to the control sample of 316L stainless steel.

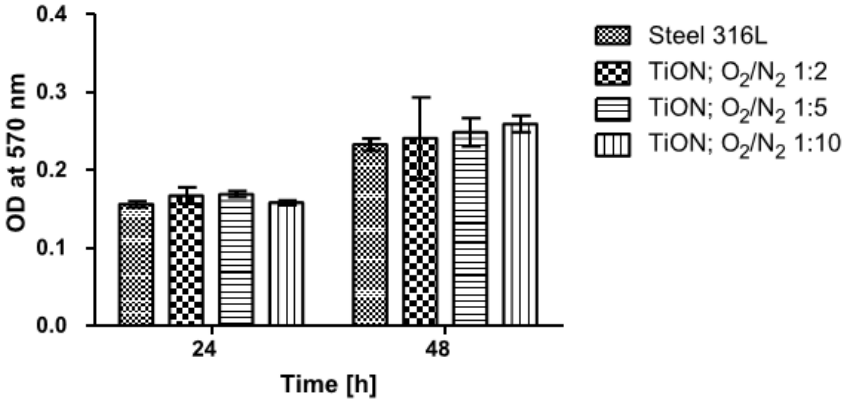


Fig. III-75. The metabolic activity of HUVEC cells incubated on the samples TiO_xN_y : O_2/N_2 = 1/2, O_2/N_2 = 1/5, O2/N2 = 1/10 compared with the control sample of stainless steel.

CONCLUSIONS

C1. GENERAL CONCLUSIONS AND ORIGINAL CONTRIBUTIONS

Hydrophilicity of the PVC surface can be successfully improved by introducing ester groups on the surface by chemical methods, using both AcOAg and LAg. The presence of these groups was confirmed by analyzing the samples obtained by infrared spectroscopy. IN the spectra corresponding to the modified surface, the appearance of the ester bond (1516 cm^{-1}) and the decrease of the intensity of C-Cl bond (746 cm^{-1}) cand be observed. The reaction time needed to obtain better yeld is lower when the reaction is performed using silver acetate. The longer the polymer is in contact with the solvent, the greater the risk of changing the properties of PVC.

The PVC material resulting from the synthesis have a rough surface, with flaws. The surface of the PVC can be physically modified by immersing it in a suitable solvent that does not dissolve the polymer, but only to inflate the surface so that the unevenness resulting from the synthesis process is removed and a smooth surface is obtained. This modification was successfully performed by immersion in acetone and 50% aqueous acetone solution, respectively.

Another method that has proven to be effective to change the surface of PVC without changing the properties of the bulk material is the spin coating method. This technique was used in this work for the deposition of a film also composed of PVC, in order to cover the surface defects arising from the synthesis process. Thus, smooth, uniform surfaces were obtained, containing active principles that have an anticoagulant effect (dicoumarol and warfarin, respectively), but also surfaces that incorporates silver nanoparticles. The PVC samples modified by coating with a thin film of PVC/Dicoumarol, using spin coating,

presented smooth surfaces with a uniform distribution of the activ principle. Dicoumarol was released gradually, in time, on a long period of time, thus the material can present antithrombogenic activity on an extended period of time, sufficient to prevent blood clotting, thrombosis and thus clogging of the catheter. The morphology of the thin film resulted after performing coating using the spin coating is influenced by the spin speed value in the drying step. Albumin adherence takes place on all the studied samples, but the presence of dicoumarol and the surface smoothness obtained after performing the deposition at 10,000 rpm, resulted in the reduction of protein adhesion. Is important to be mentioned that the proposed methodology resulted in an improved anti-adherent activity of the modified PVC, especially against Gram-positive bacteria as for example *S. aureus*. Bacterial adhesion resistance has been improved by incorporating dicoumarol on the surface due to its antibacterial properties and its protein binding capacity.

The PVC surface can be successfully modified by coating with films incorporating AgNPs. This deposition can be performed by the spin coating method at various working parameters. The resulting films are compact enough to prevent the migration of silver ions into biological fluids (the degree of recovery being <0.4%). The nanoparticle size is between ~16 - ~27 nm for dispersed nanoparticles and reaches up to ~80 nm in agglomerations. All samples modified by spin coating in this paper showed anti-biofilm activity against *E. coli, S. aureus* strains, but not against *C. albicans* strain.

In the second part of the paper was performed the characterisation of the surfaces of stainless steel flat samples, but also of stents made from the same material, after the coating with TiO_xN_y. was conducted. The coating was made by magnetron sputtering using a mixture of oxygen and nitrogen as a reactive gas. It was observed that depending on the ratio between the two gases, the surface properties differ. The best results were observed in the case of an O_2 / N_2 = 1/5 ratio in the gas supply flow, followed by O_2 / N_2 = 1/10. These samples showed the lowest degree of albumin adsorption and minimal salts depositions due to exposure to SBF. As the nitrogen content in the surface composition increases, the adsorption of albumin decreases significantly. The morphology of the surface is also influenced by the amount of nitrogen, the higher the amount of nitrogen, the smoother the surface is. Although the results obtained for this type of coating are promising in terms of surface properties, they do not show a sufficiently good adhesion to the substrate so that the joint areas do not show damage when a mechanical stress such as stent expansion is applied.

C2. PERSPECTIVES FOR FURTHER DEVELOPMENT

The methods of modifying the PVC surface described in this doctoral thesis can be used together to obtain a surface with improved properties, which can be used in medical applications to obtain various medical devices. By using the physical method of modification, respectively of a solvent that induce surface swelling of the polymer, it favors the contacting of the C-Cl bonds and can thus be modified by introducing ester groups that increase the hydrophilicity of the surface. The spin coating method can be used to incorporate in the polymer film both active principles and silver nanoparticles, thus obtaining smooth, uniform surfaces with anticoagulant effect (release of the active principle taking place slowly in the case of dicoumarol due to very low solubility in water), but also antibacterial. The spin coating method cannot be applied to medical devices such as catheters, but the principles used can be applied using an alternative method that can mimic the deposition conditions studied in the spin coating method and that can be used for tubular devices. The study of such a method is an objective for future studies, and the goal is to develop a method that consists in circulating a solution (prepared in the same way as the one used in this study) through a tubular device, using a certain flow, using a peristaltic pump, so that the results obtained can be reproduced in the case of devices such as catheters.

Regarding the study on stents, it can be said that the TiO_xN_y coating is promising, but the method of film deposition must be studied in more detail and improved so that the flaws that appear at the joints due to mechanical stress are eliminated. Establishing the optimal ratio between oxygen and nitrogen is also very important, because depending on it, the properties and morphology of the surface changes.

DISSEMINATION OF RESULTS

Published articles

1. O. C. Duţă, A. Ficai, D. Ficai, R. D. Truşcă, E. Grosu, L. M. Ditu, G. Mihăescu, M. C. Chifiriuc, E. Andronescu, *PVC Modification by incorporating silver nanoparticles on the surface*, manuscris acceptat, University Polittehnica of Bucharest Scientific Bulletin, Series B: Chemistry and Materials Science, 2020, 82 (3), 85-100.

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4. O. C. Duta, M. Maximov, R. Trusca, A. Ficai, D. Ficai, C.-I. Ilie, L.-M. Ditu, E. Andronescu, *Advanced Drug-Eluting Poly (Vinyl Chloride) Surfaces Deposited by Spin Coating*, Medicina 2019,55, 421; doi:10.3390/medicina55080421

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2. O. C. Duta, D. Ficai, A. Ficai, L. M. Ditu, M. C. Chifiruc, E. Grosu, E. Andronescu, Physical and chemical surface modification of PVC based catheters, 19th Romanian International Conference on Chemistry and Chemical Engineering, 2015.

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