

SURFACE CHARACTERIZATION OF THE ELECTROSPUN CHITOSAN NANOFIBERS AFTER METHANE PLASMA TREATMENT

ZBIGNIEW ROŻEK^{1*}, DANIELA LUBASOVÁ², JINDRICH MATOUSEK³, PETR LOUDA⁴, PIOTR NIEDZIEMSKI⁵, STANISLAW MITURA⁶

^{1,4} INSTITUTE OF MATERIALS SCIENCE AND ENGINEERING, TECHNICAL UNIVERSITY OF LIBEREC, 2 STUDENSKÁ ROAD, 461 17 LIBEREC 1, CZECH REPUBLIC

² FACULTY OF TEXTILE ENGINEERING, DEPARTMENT OF NONWOVENS, TECHNICAL UNIVERSITY OF LIBEREC, 2 STUDENSKÁ ROAD, 461 17 LIBEREC 1, CZECH REPUBLIC

³ DEPARTMENT OF PHYSICS, FACULTY OF SCIENCE, J.E. PURKINJE UNIVERSITY, 8 CESKE MLADZEZE ROAD, 400 96 USTI NAD LABEM, CZECH REPUBLIC

^{5,6} INSTITUTE OF MATERIALS SCIENCE AND ENGINEERING, TECHNICAL UNIVERSITY OF LODZ, 1 STEFANOWSKIEGO ROAD, 90-924 LODZ, POLAND

*E-MAIL: ZBYNEKROZEK@SEZNAM.CZ

Abstract

Present nanofiber technology is one of the most important objects in the recent research topics. Electrospinning is a unique technology that can produce non-woven fibrous materials with interesting characteristics such as diameters ranging from sub-micron to several nanometers, high surface to volume ratio, high porosity and small interfibrous pore size. Polymer nanofibres have great potential for technical applications in filtration, composites and electronics. Nanofibers are also of importance in many different applications as the drug delivery, biomaterials and tissue engineering. For these applications there is a great need for polymer nanofibers with well defined surface properties. In this field, plasma surface treatment has been applied in the textile industry for the modification of polymer nanofibers.

In this study, chitosan nanofibers were prepared by modified electrospinning method called Nanospider™ and treated with plasma in the presence of methane gas. The surface characteristics of the nanofibers after plasma treatment were examined using contact angle measurements, SEM and XPS analysis.

Keywords: electrospinning, electrospun nanofibers, plasma treatment, hydrophobicity

[*Engineering of Biomaterials*, 94, (2010), 2-6]

Introduction

Nanoscale materials have attracted great academic and industrial interest in recent years. In this field, electrospinning is a straightforward method to produce fibers with nano/micro scaled diameters [1,2]. This is a simple and highly effective technique for the preparation of polymer fibers in the form of individual fibers or of non-woven fiber mats [3]. Microstructural properties of this material include high porosity with very small pore size, interconnectivity and controllable mesh thickness. All these properties, together with the large surface area to volume ratio make the nonwoven fiber mats a suitable material for different applications such as

biosensor/chemosensor [4,5], reinforced nanocomposites [6] electronic and semi-conductive materials [7,8] and filters [9]. Moreover, electrospun fiber mats can be a promising material for many important biomedical applications, including artificial blood vessels [10], wound dressings [11], the drug delivery and scaffolds for tissue engineering [12].

Especially from the point of view of the biomedical applications, there is a significant need and demand for the polymer nanofiber mats with well defined surface properties. In this case, it seems that the most important aspect is hydrophilicity/hydrophobicity balance. Currently gas plasma treatment processes are extensively used to increase the hydrophilicity of hydrophobic nonwoven fiber mats. It has been reported that the hydrophilicity of silk fibroin nanofibers or nanofibers made of poly(ϵ -caprolactone) was increased meaningfully after an O₂ plasma treatment. Such modification of these materials resulted in an improved fibroblasts adhesion and proliferation [13,14]. On the other hand, CF₄ or CH₄ plasma treatment increased the hydrophobicity of the nanofibers made of poly(3-hydroxybutyrate-co-3-hydroxyvalerate) or poly[bis(2,2,2-trifluoroethoxy)phosphazene], respectively [15,16]. Hydrophobicity is an important property of a surface of different materials for practical applications [17]. Superhydrophobic surfaces have great attention due to their interesting properties, such as their self-cleaning, and antifouling properties. Kitahara et al. modified the cotton fibres with plasma in the presence of hydrocarbon gas and investigated the antibacterial activities of the modified samples. It has been established that plasma modified samples had antibacterial property in contact with *Staphylococcus aureus* and *Klebsiella pneumoniae* [18].

Many of the scientific facts mentioned above have persuaded the authors of this paper to modify the chitosan nonwoven nanofiber mesh with plasma in the presence of methane gas. Chitosan is a biosynthetic polysaccharide comprising of deacetylated chitin. Chitin is a naturally occurring β -1,4 linked polymer of 2-acetamido-2-deoxy- β -D-glucopyranose polysaccharide that can be extracted from crustacean exoskeletons or generated via a fungal fermentation process. Chitosan is a β -1,4 linked polymer of 2-amino-2-deoxy- β -D-glucopyranose that is soluble in an aqueous solution of acids. Past research has shown that chitosan is biocompatible and biodegradable and does not induce any strong immune response. Additional positive features are its low cost due to abundance and diverse methods that enable the chemical processing of this polymer [19].

In this study the chitosan nanofiber mats with a hydrophilic character were prepared by modified electrospinning method called Nanospider™ and treated with plasma in the presence of methane gas. The surface properties of the plasma-treated nanofibers were characterized by water contact angle (WCA), scanning electron microscopy (SEM) and X-ray photoelectron spectroscopy (XPS).

Materials and methods

Materials and preparation of solution for electrospinning

Chitosan-5 was purchased from Wako Pure Chemical Industries. PEO with a molecular weight (M_w) 900 kD and surfactant Triton[®]X-100 were obtained from Aldrich. Deionized water with sodium chloride was used to prepare PEO solution. Sodium chloride was used by adding salt together with a water solution of 5 wt% PEO solution in concentrations 0.24 mol/L. Chitosan was dissolved in 10 wt% citric acid to achieve a polymer concentration of 8 wt%. PEO was dissolved in water with sodium chloride to achieve a polymer concentration of 5 wt%. Chitosan solutions and PEO solutions were prepared separately.

Both solutions were then mixed and stirred again at room temperature overnight in the volume ratios 9/1 (chitosan/PEO). The surfactant Triton®X-100 was added in concentration 2 wt% into solution.

Electrospinning of nanofibers

Chitosan nanofibers have been prepared by modified electrospinning method called Nanospider™. The scheme of Nanospider™ system is shown in FIG. 1.

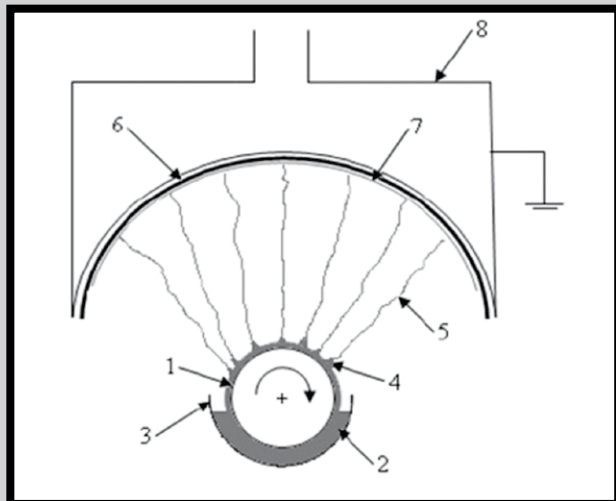


FIG. 1. The scheme of Nanospider™ system.

The metal roller (1), which serves as the positive electrode, raises a thin film of polymer solution (2). The so-called Taylor cones (4) are then formed due to the effect of applied high voltage. The method is based on the possibility of creating Taylor cones from a thin layer of polymer solution. The cones cleave into fibers (5), which are carried onto the negative electrode (8) and retained on the polypropylene non-woven fabric (6). During this process, the solvent evaporates and the fibers become stretched at either ambient or elevated temperature. The reservoir (3) is filled with 20 ml of polymer solution and rotation of the metal roller (1) ensures its perfect coating with the solution. The Taylor streams are formed next to each other, throughout the entire length of the metal roller, resulting in the high production capacity of nanofiber sheet (7). Electrospinning of mixture of chitosan/PEO solutions is in this study carried out at a voltage of 50-55 kV, distance of electrodes 10 cm and air temperature 22°C.

Optimal conditions for heat treatment of nanofiber sheet prepared from chitosan/PEO solution after electrospinning were investigated [20]. A temperature of 130-140°C applied for 30 min was found to be optimal. In other case, nanofiber sheet was dissolved in water.

Plasma treatment

For the plasma treatment a laboratory scale plasma system was used. The vacuum chamber was 345 mm in diameter and 360 mm in height. The system was equipped with a radio frequency generator operating at 13.56 MHz, connected with the inner electrode through an impedance matching unit. The substrates were mounted on the water cooled RF electrode, plasma reactor was evacuated, and methane gas was introduced into the reactor. In order to remove any other gases from inside the reactor, chamber was flushed with methane gas for 5 min. The negative self-bias voltage was varied in the range from 100 to 600 V.

Characterization

Contact angles of all samples were measured at room temperature using a home-made apparatus equipped with a digital camera, which is connected to a computer. Measurements were performed by the sessile drop method with distilled water as test liquid. The volume of the water droplet for each measurement was kept at $3 \times 10^{-9} \text{ m}^3$. For each sample, six different measurements were taken and the average values for contact angles were calculated.

The morphology of the electrospun chitosan nanofibers was observed on a scanning electron microscope (SEM) (TESCAN – VEGA XMU) after gold coating.

The XPS measurements were performed with the hemispherical analyzer operated in FAT mode (Phoibos 100 from Specs). The photoelectron spectra were referenced to the aliphatic C1s peak at 285 eV. The elemental composition was calculated from survey spectra. The high resolution spectra were acquired to understand the chemical bonds on the surface of the samples.

Results and Discussions

Changes in contact angles of chitosan nanofibers treated with the methane plasma as a function of negative self-bias voltage are shown in FIG. 2. The plasma treatment leads to hydrophobization of the originally hydrophilic chitosan nanofiber mats. The untreated film has water contact angle of 0° (the water drop was immediately absorbed into the sample) which increases and reaches a maximum value of 126° for the plasma treated sample at 400 V of negative bias. Further voltage increase resulted in the decrease of contact angle of the nanofiber mats. Hydrophobization of the chitosan nanofiber mats can be explained by the formation of hydrophobic moieties on nanofibers surfaces after the methane plasma treatment.

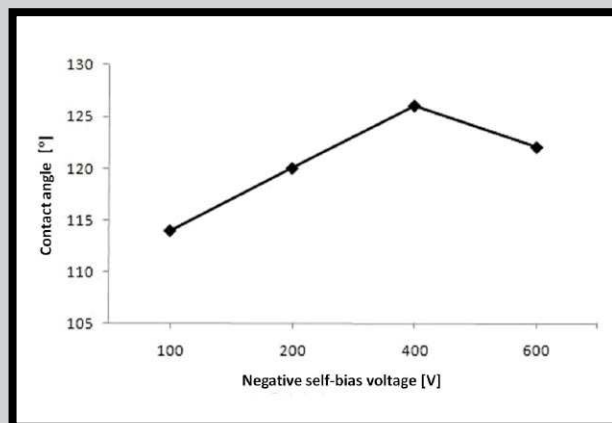


FIG. 2. Water contact angle of chitosan nanofibers versus negative self-bias voltage.

SEM images of both the modified and the non-modified chitosan fiber mesh were shown in FIG. 3. It can be seen that morphology of the nanofibers was not changed by methane plasma as the used negative self-bias voltage was varied in the range from 100 to 200 V. When voltage was changed from 200 to 600 V, the surface roughness of the nanofibers increased. This observation can be attributed to the etching effect of the plasma treatment and deposition of carbon from methane plasma on the surface of chitosan nanofibers. Strong interaction between the plasma and the surface of nanofibers can be observed in FIG. 3e.

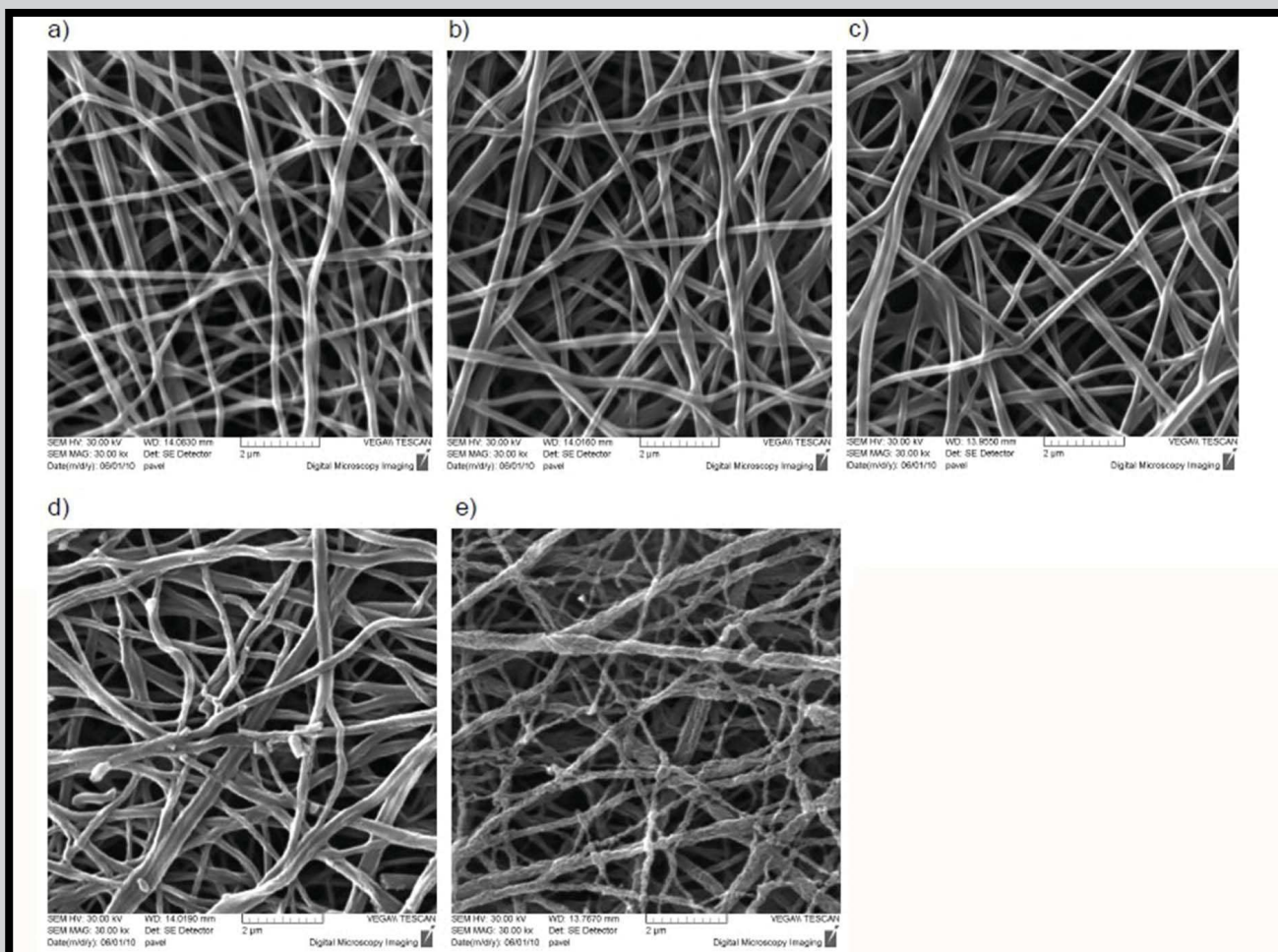


FIG. 3. SEM images of the chitosan nanofibers: a) non-modified and modified with methane plasma at b) - 100 V, c) - 200 V, d) - 400 V, e) - 600 V.

XPS survey spectra for non-modified and methane plasma-modified (with the negative self-bias voltage 600 V) chitosan nanofiber mats are shown in FIG. 4. The only elements detected were carbon, oxygen and nitrogen. The 1s (C1s, O1s and N1s) peaks were used for the quantification. The C1s high resolution spectra were acquired to provide more detailed information about the bonding on the nanofiber mats surface. The unmodified chitosan shows elemental composition, which could be expected from the chitosan chemical structure. Slightly higher carbon content can be explained by the small amount of PEO in the nanofiber network or by the carbon contaminants from the ambient atmosphere.

The plasma treatment results in a significant decrease of the oxygen amount. Possibly the OH groups are detached from the polymer chain due to interaction with the plasma active species (this can also be the reason for the higher hydrophobicity of the treated samples) or the CH_x thin film can be deposited on the surface (the information depth of XPS is 10 nm at the most). Probably both processes take place. As the negative bias voltage changes to 600 V, the C/O ratio increases.

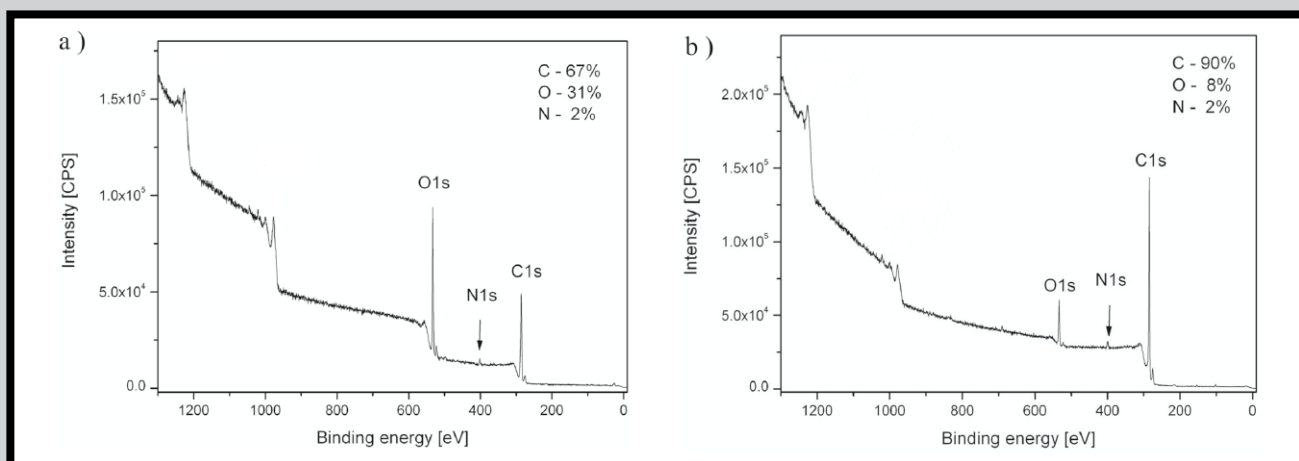


FIG. 4. XPS survey spectra of chitosan nanofibers: a) unmodified, b) plasma-modified with methane at - 600 V.

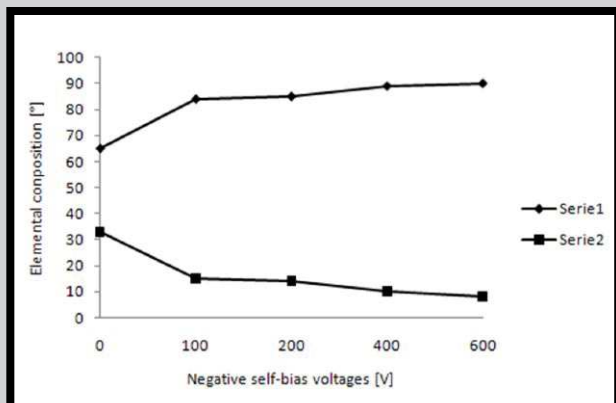


FIG. 5. XPS elemental composition of plasma-treated chitosan nanofibers surface at different applied negative self-bias voltages.

The decrease in the oxygen amount can be also related to the disposal of oxygen by atomic hydrogen via chemical reaction [21]. The elemental composition after plasma treatment identified by the XPS spectra is presented in FIG. 5.

The changes in the chitosan structure are also observed in the C1s high resolution spectra (FIG. 6). The untreated chitosan shows three main components. The component A is attributed to C-C or C-H bonds. Based on [22-24] the component B can be related to C-O, C-OH or C-N bonds. This component significantly decreases after plasma treatment and almost disappears for higher negative bias voltages. As it had been already proposed – this is probably related to cleavage of the OH groups from the polymer chain or to covering of the nanofiber surface by the CH_x thin film. The last component is usually attributed to carboxylic groups or to O-C-O and O=C-N groups. The O-C-O is the most probable of them as it is present in the chitosan structure; also the O-C-OH should be taken into account (as it is present at the ends of the chitosan polymer chains). This component again disappears with the plasma treatment.

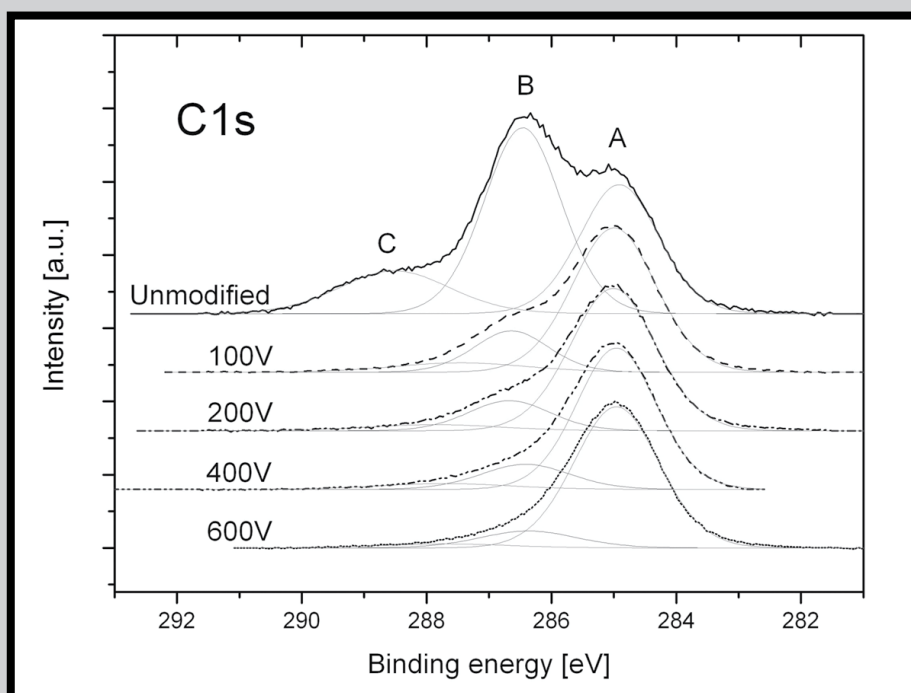


FIG. 6. XPS high resolution spectra of plasma-treated chitosan nanofibers surface at different applied negative self-bias voltages.

Conclusions

In this study electrospun chitosan nanofibers were modified with plasma in the presence of methane gas and the surface characteristics of the modified and unmodified nanofibers were investigated. It was found that the chitosan nanofibers have hydrophobic properties after methane plasma treatment. SEM analyses of the unmodified and plasma-modified chitosan nanofibers indicate that plasma treatment can induce physical changes on the surface of the nanofibers. The changes in the chitosan surface chemistry caused by the plasma treatment are documented by the XPS measurements. The amount of oxygen on the nanofiber surface decreases with the plasma treatment. The oxygen content further drops with increased bias voltage. The high resolution spectra deconvolution also supports the possible explanation of the hydrophobicity changes (less OH group – higher hydrophobicity).

Acknowledgments

This work was supported by MSM4674788501 and ME10145 project.

- [1] Fujihara, K., Kotaki, M., Ramakrishna, S., „Guided bone regeneration membrane made of polycaprolactone/calcium carbonate composite nano-fibers“, *Biomaterials*, Vol. 26, No.19., 2005, pp 4139-4147.
- [2] Demir, M.M., Yilgor, I., Yilgor, E., Erman, B., „Electrospinning of polyurethane fibers“, *Polymer*, Vol. 43 No.11., 2002, pp 3303-3309.
- [3] Verdonk, P., Caloïpe, P.B., Del Moral Hernandez, E., Da Silva, A.N.R., „Plasma etching of electrospun polymeric nanofibers“, *Thin Solid Films*, Vol. 515, No.2., 2006, pp 831-834.
- [4] Kowalczyk, T., Nowicka, A., Elbaum, D., Kowalewski, T.A., „Electrospinning of Bovine Serum Albumin. Optimization and the Use for Production of Biosensors“, *Biomacromolecules*, Vol. 9, No.7., 2008, pp 2087-2090.
- [5] Virji, S., Kaner, R.B., Weiller, B.H., „Hydrogen Sensors Based on Conductivity Changes in Polyaniline Nanofibers“, *Journal of Physical Chemistry B*, Vol. 110, No.44., 2006, pp 22266-22270.
- [6] Huang, Z.-M., Zhang, Y.-Z., Kotaki, M., Ramakrishna S., „A review on polymer nanofibers by electrospinning and their applications in nanocomposites“, *Composites Science and Technology*, Vol. 63, No.15., 2003, pp 2223-2253.
- [7] Norris, I.D., Shaker, M.M., Ko, F.K., MacDiarmid, A.G., „Electrostatic fabrication of ultrafine conducting fibers: polyaniline/polyethylene oxide blends“, *Synthetic Metals*, Vol. 114, No.2., 2000, pp 109-114.
- [8] Dong, F., Li, Z., Huang, H., Yang, F., Zheng, W., Wang, C., „Fabrication of semiconductor nanostructures on the outer surfaces of polyacrylonitrile nanofibers by in-situ electrospinning“, *Materials Letters*, Vol. 61, No.11-12., 2007, pp 2556-2559.
- [9] Yun, K. M., Jr. Hogan, Ch.J., Matsubayashi, Y., Kawabe, M., Iskandar, F., Okuyama, K., „Nanoparticle filtration by electrospun polymer fibers“, *Chemical Engineering Science*, Vol. 62, No.17., 2007, pp 4751-4759.
- [10] Ma, Z., Kotaki, M., Yong, T., Heb, W., Ramakrishna, S., „Surface engineering of electrospun polyethylene terephthalate (PET) nanofibers towards development of a new material for blood vessel engineering“, *Biomaterials*, Vol. 26, No.15., 2005, pp 2527-2536.
- [11] Torres Vargas, E.A., do Vale Baracho, N.C., de Brito, J., de Queiroz, A.A., „Hyperbranched polyglycerol electrospun nanofibers for wound dressing applications“, *Acta Biomaterialia*, Vol. 6, No.3., 2010, pp 1069-1078.
- [12] Sill, T.J., von Recum, H.A., „Electrospinning: Applications in drug delivery and tissue engineering“, *Biomaterials*, Vol. 29, No.13., 2008, pp 1989-2006.
- [13] Jeong, L., Yeo, I.-S., Kim, H.N., Yoon, Y.I., Jang, D.H., Jung, S.Y., Min, B.-M., Park, W.H., „Plasma-treated silk fibroin nanofibers for skin regeneration“, *International Journal of Biological Macromolecules*, Vol. 44, No.3., 2009, pp 222-228.
- [14] Jia, J., Duan, Y.-Y., Yu, J., Lu, J.W., „Preparation and immobilization of soluble eggshell membrane protein on the electrospun nanofibers to enhance cell adhesion and growth“, *Journal of Biomedical Materials Research Part A*, Vol. 86A, No.2., pp 364-373.
- [15] Yoon, Y.I., Moon, H.S., Lyoo, W.S., Lee T.S., Park, W.H., „Superhydrophobicity of PHBV fibrous surface with bead-on-string structure“, *Journal of Colloid and Interface Science*, Vol. 320, No.1., 2008, pp 91-95.
- [16] Allcock, H.R., Steely, L.B., Kim, S.H., Kim, J.H., Kang, B.-K., „Plasma Surface Functionalization of Poly[bis(2,2,2-trifluoroethoxy)phosphazene] Films and Nanofibers“, *Langmuir*, Vol. 23, No.15., 2007, 8103-8107.
- [17] Sun, T., Feng, L., Gao, X., Jiang, L., „Bioinspired Surfaces with Special Wettability“, *Accounts of Chemical Research*, Vol. 38, No.8., 2005, pp 644-652.
- [18] Kitahara, N., Sato, T., Isogawa, H., Ohgoe, Y., Masuko, S., Shizuku, F., Hirakuri, K.K., „Antibacterial property of DLC film coated on textile material“, *Diamond and Related Materials*, Vol. 19, No.7-9., 2010, 690-694.
- [19] Haider, S., Park, S.Y., Lee, S.H., „Preparation, swelling and electro-mechanochemical behaviours of gelatin-chitosan blend-membrane“, *Soft Matter*, Vol. 4, 2008, 485-492.
- [20] L. Martinová and D. Lubasová, „Electrospun Chitosan based Nanofibers“, *Research Journal of Textile and Apparel*, Vol. 12, No.2, 2008, pp 72-79.
- [21] Ogino, A., Kral, M., Yamashita, M., Nagatsu, M., „Effects of low-temperature surface-wave plasma treatment with various gases on surface modification of chitosan“, *Applied Surface Science*, Vol. 255, No.5., 2008, pp 2347-2352.
- [22] Kang, J., Liu, H., Zheng, Y.-M., Qua, J., Paul Chen, J.P., „Systematic study of synergistic and antagonistic effects on adsorption of tetracycline and copper onto a chitosan“, *Journal of Colloid and Interface Science*, Vol. 344, No.1., 2010, 117-225.
- [23] <http://www.lasurface.com>
- [24] Briggs, D., Grant, J., „Surface Analysis by Auger and X-Ray Photoelectron Spectroscopy“, IM Publication, 2003.