The Investigation of the Structure and Properties of Nonwoven Fibrous Materials Based on Polyhydroxybutyrate Biopolymer

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Abstract. Currently, the development and research of nonwoven medical fibrous materials based on biopolymers is an area of great practical interest. One of the most promising methods for producing nonwoven materials with a highly developed surface is electrospinning (ES). In the article, the possibility of effective sterilization of ultrathin fibers based on polyhydroxybutyrate (PHB) by the ozone treatment was considered. The purpose of this work was to establish the interrelation between changes in the ES process and accompanying changes in the supramolecular structure, morphology, physical and mechanical properties of the fibrous materials based on PHB.

1. Introduction

Highly porous polymer carriers of biologically active substances are widely used in biology and medicine as long-acting matrices, matrices for cell engineering, antibacterial therapeutic systems, controlled release matrices of medical substances, etc. [1]. Fibrous structures and fibrillous materials based on such structures can be obtained in various ways, but the greatest attention should be paid to the method of electrospinning (ES) from a solution of biopolymers. ES has already proven itself as an effective way to produce continuous fibers with a large variation in diameters [2]. Different technical solution of the ES process allow modifying of the fiber surface and controlling of a wide range of properties of the resulting materials [3]. In this work, a number of regularities were established that allow us to vary the properties of fibrous materials based on the biopolymer polyhydroxybutyrate (PHB).

2. Experimental

In the work, nonwoven fibrous materials obtained by the ES method on a single-capillary laboratory unit with the capillary diameter 0.1 mm were used. Molding of the materials was carried out from the forming solutions of the polymer PHB. PHB was produced by the bacteriological synthesis by BIOMER (Germany), grade 16F, viscosity-average molecular weight was 2.6x10^5 Da. In the work, four methods for obtaining the material were used. Methods differed in the formulations of molding solutions with a
content of 7-15% PHB and in the conditions of the ES process. The choice of the conditions varied depending on the properties of the molding solutions: conductivity, viscosity, homogeneity. The voltage was 17-22 kV, the distance between the electrodes was 150-200 mm, the gas pressure on the solution was 10-30 kgf/cm². Samples of the nonwoven materials with different densities were studied by the electron paramagnetic resonance (EPR) method on the EPR spectrometer EPR-B (Russia). The crystallinity of the materials was studied by differential scanning calorimetry (DSC) on the DSC 214 Polyma of Netzsch (Germany) with a heating rate of 10° K/min. The morphology and the mutual arrangement of fibers in the structure of material was investigated using the optical polarizing microscope Micromed polar-3 (Russia). Microphotographs were prepared in the reflected light at a magnification of 200 times. Mechanical tests were performed on a machine for mechanical analysis DEVOTRANS (Turkey) in compliance with the ASTM D5035-11.

3. Results and discussion
The supramolecular structure of biopolymer fibers obtained by the ES method is highly influenced not only by the molecular characteristics of the polymer, but also by the conditions of its production into the fiber, the curing process, and the stages of subsequent processing [4]. The porosity, geometry, surface topology of each individual fiber, as well as their mutual orientation, make a significant contribution to the formation of physical, mechanical, diffusion and thermal properties of the whole material. It was established, that the supramolecular structure of PHB largely determines the biodegradation rate of the material, its functional quality, which can be improved and controlled in several effective ways: the introduction of modifying additives, chemical processing of fibrous materials, variation of technological parameters of the electroforming process at various stages of fiber production [5, 6].

In the article, two technological solutions for several formulations of forming solutions of the PHB are presented. The considered technological solutions allow obtaining of the fibrillary composites with different fiber morphology, which cause differences in the physical and mechanical characteristics of non-woven materials. This study makes it possible to vary the properties of biopolymer fibrous material, which can be used in products for medicine, veterinary, hygiene, biology, capable of biodegradation after use.

Formulations 1 and 2 allowed obtaining fragile, voluminous, highly porous materials (Figure 1, 1 and 2). It should be noted a large number of defects, thickening, irregularity and heterogeneous of the fibers. Microphotography clearly shows ordered formations in the structure of fibers, which can be obtained because of orientation extraction of the polymer, which occurs during ES. Formulations 3 and 4 allowed getting a fundamentally different structure of the material. The fibrous material could be characterized as a thin and flexible polymer film. At the same time, microphotographs (Figure 1, 3 and 4) show highly developed fibrous structure. It was mentioned, that the average diameter of the fibers decreases and the inter-fiber space increases. Formula 4 was characterized by the highest straightening of fibers and the smallest number of defects.

Figure 1. Microphotographs of the nonwoven materials obtained by the ES from the forming solutions based on PHB: 1, 3 – 7% of PHB, 2, 4 – 15% of PHB.
Thus, from Figure 1 it can be seen that the content of PHB in the molding solution did not principally affect on the structure of the whole material. The main factor, which specified the structure type, was based on the conditions of the ES process. In any case, the nature of the dispersion of the diameters of the formed fibers, the number of defects, which include thickening, drops, gluing, breaks, and other current structural features were depending on the formulations. Moreover, these characteristics significantly affected the physical and mechanical properties of the material, which are shown in the Table 1.

The influence of process conditions on the type of structure was established: texturized highly porous or film-like. It was found that voltage, pressure, distance between the electrodes, type of coating of the lower electrode were the key factors. Identical conditions for formulations 1 and 2 contributed to an intensive molding, low jet injection speed, and a smaller diameter of the fibrous trajectory. As a result, the jet was already cured in the material layer. The conditions for formulations 3 and 4 contributed to a less intensive molding. The trajectory of the fiber had a larger diameter and was more stable. The fibers within the layer were already well cured, the surface of the electrode contributed to the denser packing of fibers. All this properties of material samples gives the possibility to vary the average diameters, their physical and mechanical properties and etc. These conditions allowed effective controlling of the quality of the material. Depending on the purpose and field of application, it is possible to obtain a material with different values of porosity, density, surface area and therefore permeability.

Table 1. Results of the physical and mechanical tests of the samples with different structure parameters.

| Number of the compound | Physical and mechanical characteristics | Structural characteristics |
|------------------------|----------------------------------------|----------------------------|
|                        | Maximum Strength [N] (Δ±0.5) | Relative Deformation [%] (Δ±0.2) | Average fiber diameter [µm] | Average thickness of the sample [mm] |
| 1                      | 2.4                              | 4.4                        | 13                          | 0.3                        |
| 2                      | 0.9                              | 3.3                        | 13                          | 0.2                        |
| 3                      | 7.8                              | 1.3                        | 8.2                         | 0.16                       |
| 4                      | 2.3                              | 5.0                        | 9.5                         | less than 0.1              |

In view of the fact that the issue of sterilization preparation of these materials before use in medical devices is acute. The highly developed surface makes it difficult to sterilize nonwoven materials by UV treatment. In addition, various studies have shown an uncontrolled decrease in the physical and mechanical properties of the materials based on the PHB after irradiation at a wavelength of 254 nm [7]. PHB is characterized by the temperature of the beginning of destruction of 140-150 ºC, which is 40-30 ºC below the recommended temperature required for the death of pathogenic microorganisms. This fact makes high-temperature annealing inapplicable for nonwoven materials. The problem of sterilization can be solved by an effective method of disinfection of medical devices, which is the ozone treatment [8]. The effect of the ozone on the supramolecular structure of PHB fibers obtained by different formulations was investigated in this work.

Table 2. Physical-mechanical properties of nonwoven materials with high fibre dispersity before and after ozone treatment (duration of the ozone treatment was 5 minutes long).

| Material                  | Maximum Strength [N] | Relative Deformation [%] (Δ±0.2) |
|---------------------------|----------------------|----------------------------------|
| PHB Initial               | 1,7                  | 3,4                              |
| PHB Ozonized              | 3,5                  | 7,6                              |

It was found that ozone could not only provide effective sterilization of the material, but also could significantly improve the physical and mechanical parameters. Moreover, the increase in breaking stress
and elongation, compared to the initial one, is fixed at the first minutes of ozonation and reaches a maximum in the range of 3-5 minutes of material processing. Table 2 shows the results of the study for one of the formulations. It is important to note that this effect applies to all formulations of PHB-based fibrous materials.

In the work, the reasons for the observed effects are suggested and they consist in the fracture of macromolecules in the amorphous phase followed by more regular laying of macrocycles of PHB. It is important to stress, that at the moment of fiber formation, polymer macromolecules are subjected to orientation and extraction under the action of a complex of physical forces. This leads to structural stresses, as well as disequilibrium in the crystalline and amorphous phases of the PHB. The EPR method allowed seeing the effect of ozone on the amorphous phase of the polymer, where the reaction is quite intense. In the first minutes of ozonation, the correlation time of the probe increased from $30 \times 10^{-10}$ to $60 \times 10^{-10}$ s, which indicated changes in the amorphous phase of the PHB and its densification. The DSC method showed accompanying increases in the degree of crystallinity, as well as an increase in the melting temperature in the samples as they were ozonized during 3-5 minutes. This may indicate that the most stressed sections of the polymer chain are broken, as well as the reorganization of the amorphous phase sections, which contribute to a more orderly arrangement of macromolecules in the crystal regions. These phenomena in the macromolecular structure, most likely, lead to an increase in physical and mechanical properties in the first minutes of ozonation. Moreover, if the ozonation time exceeds 7-8 minutes, then the destruction of the material is visible, observed in a uniform drop in the correlation time below the initial one.

4. Conclusions
Different types of materials could be obtained within different conditions of the electrospinning process. The study of the physical and chemical properties of such nonwoven materials obtained from polymer solutions after their effective sterilization is an area of great interest. The article established and showed an example of an effective method of sterilization that can improve the physical and mechanical characteristics of the material, and also considered the reasons for this process, due to the influence of ozone on the supramolecular structure of the material.

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