Analysis of structure of hyperfine poly(3-hydroxybutyrate) fibers (PHB) for controlled drug delivery

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Abstract. Hyperfine fibers based on biodegradable poly (3-hydroxybutyrate) with encapsulated drug substance (dipyridamol) were obtained by using electrospinning method. Addition of dipyridamol has a significant effect on geometrical shape and structure of microfibers as well as total porosity of fibrous material. Observation of fibers using scanning electron microscopy (SEM) method showed that without or at lower dipyridamol content (<3%) fibers consisted of interleaved ellipsoid and cylindrical fragments. At higher dipyridamol content (3-5%) anomalous ellipsoid structures did not practically form, and fiber's shape became cylindrical. The totality of morphological and structural characteristics determined the rate of dipyridamol diffusive transports. The simplified model of drug desorption from fibrous matrix was presented. In current work it was showed that the rate-limiting stage of transport was the diffusion of dipyridamol in the bulk of cylindrical fibers.

1. Introduction

The development of polymer matrices for controlled and targeted drug delivery is the topical issue of modern therapy of many illnesses and traumas [1]. Generally for the development of such therapeutic systems the poly(3-hydroxybutyrate) (PHB) is used as a polymer matrix, which is characterized by high biocompatibility and optimal terms of biodegradation [2]. High physical and mechanical properties of PHB films and fibers allow consider this biopolymer as one of the most promising biopolymers in medicine [3].

In recent research the physical, chemical and dynamic properties of fibrous materials on PHB and solid nanoparticles basis were considered. The nanoparticles were as follows, such as modified titanium dioxide, nanocrystalline silicium in titanium dioxide cover and chitosan. It was concluded that physical, mechanical and diffusive properties of fibrous materials depend on degree of crystallinity and amorphous phase of PHB. It was found that addition of nanoparticles at low concentration (from 0.1 to 0.5%) into forming PHB solution causes the significant changes of polymer structure at supramolecular level. These data are in agreement with works [4-6], in which the gelatin, calcspar and L-lactide-ε-caprolactone copolymer were used as modifying additives. Reciprocal action
of crystalline and intercrystalline regions in biodegradable polymers and its blends still has been a challenging and poorly known topic of modern polymer material science.

2. Materials and Methods
In this work, polyhydroxybutyrate of the 16F series (BIOMER®, Germany) with a molecular weight $M_w = 2.06 \times 10^5$ g/mol was used to produce the investigated fibers. As a model drug substance for controlled release was used dipyridamol (2,2',2'',2'''-(4,8-di(piperidin-1-yl)pyrimido[5,4-d]pyrimidine-2,6-diyl)bis(azanetriyl)tetraethanol). Pharmacological class of dipyridamol (DPD) is «antiplatelet, angiotropeters and correctors of microcirculation, adenosyn synergistic agent». Hyperfine PHB fibers were obtained by the method of electrospinning from a solution. PHB and PHB with 1%, 3% and 5% DPD were soluted in chloroform.

The electron paramagnetic resonance spectrum (X-band) was recorded on an EPR-V automated spectrometer (Semenov Institute of Chemical Physics, RAS, Moscow) at microwave power of 7 mW and modulation amplitude of 0.5 gauss. As a probe, the stable nitroxyl radical 2,2,6,6-tetramethylpiperidin-1-oxyl (TEMPO) was used. The radical was incorporated into fibers from vapor at a temperature of 50°C to a concentration not exceeding $3 \times 10^{-1}$ mol/l. The differential scanning calorimetry (DSC) measurements were performed on a Netzsch DSC 204 F1 calorimeter in argon atmosphere at a heating rate of 10 K/min. The morphology of the fibers was studied via electron microscopy on Hitachi TM-1000 scanning electron microscope (Japan).

3. Results and Discussion
Analysis of morphology of obtained fibers (Figure 1) revealed that quantity of thickenings decreased with increase of DPD content, and thickenings practically were not observed at highest DPD content (5%). The emergence of thickenings may be explained by either insufficient conductivity of electrospinning solution or its nonoptimal viscosity.

Study of thermophysical characteristics using DSC method showed that specific melting enthalpy of PHB fibers increased with addition of DPD. With increasing DPD content in PHB fibers the formation of equilibrium and more superior structure was determined. Obtained results should be taken into consideration while studying the segmental mobility and diffusive kinetics. These two fundamental processes determine rate and mechanism of controlled drug release from polymeric therapeutic systems.

Structure and molecular dynamics of intercrystalline parts of samples were examined by using EPR method. Experimental EPR spectrum is presented in Figure 2. It was found that as degree of crystallinity in intercrystalline parts of samples increased, the radical rotation speed decreased, which can be explained by delaying of molecular mobility of segments. With increasing DPD content, the correlation time rose in following sequence: $6.6 \times 10^{-9}$ sec. (0% DPD) > $7.1 \times 10^{-9}$ sec. (1% DPD) > $8.8 \times 10^{-9}$ sec. (3% DPD) > $9.0 \times 10^{-9}$ sec. (5% DPD), which can be explained by increase of PHB degree of crystallinity in fibers.

Curves of DPD release from fibers are presented in Figure 3. All curves were characterized by the presence of initial non-linear and last linear fragments. At the linear fragment of the curves the concentration of desorpted DPD increased linearly.

Non-linear fragment of kinetic curves of DPD controlled release is determined by the diffusion process. Rapid change of values of diffusion coefficient was correlated with structural transition from spindle shaped fibers to cylindrical. Moreover, the rate of degradation process decreased exponentially with DPD increase, which was confirmed by the previous data. Exhibition of two release processes (diffusive and kinetic) allowed suppose that DPD existed in PHB fibers in two forms. The first form was presented by DPD in a free state, and what was highly important, only this form was capable of
desorption from PHB fibers according to diffusive mechanism. Another form of DPD existed in immobilized state and was characterized by extremely low diffusive mobility. Although, the release of such DPD form from the polymer matrix was explained by partial degradation of PHB (weight loss of PHB fibers including immobilized form of DPD) according to the zeroth-order equation.

![SEM photographs of fibrous materials obtained by electrospinning method from polymeric solutions: initial PHB (a), PHB +1% DPD (b), PHB+3% DPD (c), PHB+5% DPD (d).](image)

**Figure 1.** SEM photographs of fibrous materials obtained by electrospinning method from polymeric solutions: initial PHB (a), PHB +1% DPD (b), PHB+3% DPD (c), PHB+5% DPD (d).

### 4. Conclusion

Encapsulation of DPD within hyperfined PHB fibers had an influence on PHB geometrical shape, degree of crystallinity, and packing density in non-woven matrix. The totality of those characteristics determined the kinetics of diffusive release of DPD. Structural and thermophysical characteristics of fibrous materials and kinetic curves of DPD release allowed suppose that rate-limiting stage of general release process was the first stage i.e. DPD diffusion in the bulk of fibers.
Figure 2. EPR spectrum of nitroxy radical TEMPO in PHB fibers with 1% of DPD.

Figure 3. Kinetic curves of DPD controlled release from hyperfine PHB fibers.

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