CONTROL OF BIOPOLYMER POLY-β-HYDROXYBUTYRATE CHARACTERISTICS BY γ-IRRADIATION

I. Bibers and M. Kalnins

The influence of γ-irradiation on the structure and some mechanical properties of poly-β-hydroxybutyrate was studied. Specimens of PHB were γ-irradiated with various doses (1-20 Mrad) in air and vacuum. The molecular weight, structural characteristics of the crystalline and amorphous phases, characteristics of thermal degradation, and deformation of the polymer were determined. The crystallinity was found to increase with increase in the radiation dose and with the corresponding reduction in molecular weight. The increase of crystallinity was the greater the smaller the length of macromolecules (higher radiation dose). The melting temperature \( T_m \), which characterizes the crystalline order, decreased with decrease in the molecular weight. The results of calorimetric studies suggest that radiation-caused degradation, which occurs at a temperature at which "cold" crystallization (60°C) is possible, might also affect the crystalline part of the polymer.

Introduction

Poly-β-hydroxybutyrate (PHB) is a linear stereoregular aliphatic polyester synthesized and accumulated by some bacteria as a store of carbon and energy [1-3]. Because of its high biocompatibility and the ability to be fully biodegraded, PHB is of special interest in medicine (macrocapsules for monitoring the release of medicine substances, implants and fixing materials in orthopedics, dressings, etc.) [4, 5]. The thermoplasticity and the possibility of biological synthesis of PHB from the local recycled natural source materials are of particular importance.

PHB can prove to be a very promising polymer as a matrix of thermoplastic heterogeneous composites (filled and reinforced). One of the basic characteristics of linear polymers used for these purposes is its molecular weight. Control over the molecular weight of PHB during bacterial synthesis leads to a reduced output of the polymer, which raises the cost price of the product. Therefore, there is only one acceptable way of producing PHBs with different lengths of chains, namely the decomposition of a polymer with longer chains. Due to the presence of vulnerable ester groups in the aliphatic chain, the degradation of PHB proceeds easily. Among different methods of degradation (hydrolysis, thermal degradation, and biodegradation [1, 6-8]), γ-irradiation degradation is most reasonable from the viewpoint of output, reproducibility, possibility of precise dosing of outside action, and no necessity of contact between the polymer and foreign materials, which can contaminate the product obtained, etc.

In some studies, with the example of different polyhydroxyacids (lactic acid [9], β-hydroxybutyrate/hydroxyvalerate copolymers [10], etc. [11, 12]), it has been shown that even rather low doses of γ-irradiation cause a considerable decrease in the molecular weight.
It should be noted that, when using PHB or PHB-based composites as biomaterials in medicine, γ-irradiation with small doses is one of the most efficient methods of their sterilization, therefore, it is necessary to obtain information on the effect of small radiation doses on the basic characteristics of polymers.

The purpose of this study is to examine the influence of γ-irradiation on the molecular and on some structural and physicomechanical properties of a PHB distinguished by its unusually high molecular weight (≈ 2 \times 10^8), in comparison with another PHB having a tenfold smaller molecular weight. It is customary to investigate PHB with a molecular weight of just this order of magnitude [1, 13-15].

Materials and Methods of Investigation

We examined a PHB obtained during fermentation of glucose in the presence of a producer *Azotobacter chroococcum* 23 [2]. The polymer was extracted from the fermentation mass by chloroform with subsequent filtration, deposition by isopropanol, and drying of the powderous product (called now PHB-1). The average viscosimetric molecular weight of PHB-1 was 1.90 \times 10^6. For comparison, the product of the ICI corporation (producer of *Alcaligenes eutrophus*) with average viscometric molecular weight equal to 1.45 \times 10^5 (from here on referred to as PHB-2) was used.

Films 50±5 μm thick were obtained from a dilute (1%) solution of PHB in chloroform by slowly evaporating the solvent at room temperature. Films 100±10 μm thick were produced by hot molding at a temperature of 190±2°C for 1.5 min. Then, the specimens were either slowly cooled down to room temperature (in this case, crystallization with formation of an expressed structure with large-sized spherulites (100-200 μm) took place) or were quenched by immersing them in ice water. In the latter case, a high degree of supercooling of the polymer was achieved. To retain the nonequilibrium structure obtained, the supercooled specimens were kept at a temperature below the glass transition temperature of PHB (~10°C). The remaining specimens before further testing were held in a desiccator at room temperature for at least ten days to obtain an equilibrium structure.

The polymer specimens (powder and films) were irradiated with a constant dose of 0.78 Mrad/h in air and vacuum (10^{-3} torr) using Co^{60} as the γ-irradiation source. The temperature in the radiation chamber reached 60±1°C in 1 min after the beginning of irradiation and was then maintained constant. The radiation dose varied from 0.5 to 20 Mrad.

The molecular weight of PHB specimens (solutions of PHB in chloroform at 30°C) was determined by the method of viscometry. The viscosity was measured by an Ubbelohde capillary viscosimeter. The molecular weight is calculated according to the known Mark—Houwink expression [\eta] = K \cdot M^\alpha, where [\eta] is the characteristic viscosity and K and α are the constants for the given polymer—solvent system (K = 1.18 \times 10^{-4}, \alpha = 0.78) [16].

Specimens of PHB (flat dumbbells with width of the working part 5±0.05 mm and length 10±0.2 mm) were cut out from films and tested in tension. The ultimate strength σ_b [the stress corresponding to the maximum on the σ(ε) curve] and the relative tensile elongation ε_b (the value of ε corresponding to σ_b) were determined. The tests were carried out on a UTS-100 instrument at room temperature at a tension rate of 2 mm/min.

Calorimetric measurements were performed by differential scanning calorimetry using a Mettler DSC-30 instrument. The PHB specimens were scanned in nitrogen atmosphere at a heating rate of 10°C/min in the temperature range 25–220°C (the first cycle) when the melting parameters of the crystalline part of the specimen were registered. Then, the specimens were rapidly cooled (~100°C/min) and scanned again at a rate of 10°C/min in the temperature range from ~30 to 220°C (the second cycle). Upon quenching from the molten state, the polymer had time to crystallize only partially. Because of this, during a repeated cycle of heating we could more clearly register the transition of the amorphous part of the polymer from the glassy state, as well as subsequent “cold” crystallization of the polymer and melting of the crystalline phase formed. The calori-