

GRAFTING OF POLY(ETHYLENE OXIDE)/POLY(BUTYLENE TEREPHTHALATE) BLOCK COPOLYMERS ONTO HYDROXYAPATITE PARTICLES

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Summary

Composite materials based on hydroxyapatite (HA) particles and poly(ethylene oxide)/poly(butylene terephthalate) copolymers, PEOT/PBT, were prepared by blending methods and by grafting. Grafting of 1000PEOT70PBT30 onto HA filler particles (38–53 μm) was accomplished by directly adding HA particles to the polycondensation reaction mixture.

Introduction

Biodegradable HA composite systems based on PEOT/PBT have attracted much attention [1]. Making use of the reactivity of surface hydroxyl groups, PEOT/PBT [2] copolymers have been grafted to HA particles using diisocyanate coupling agents. This resulted in improved mechanical properties of composites. In this study, we set out to covalently bind 1000PEOT70PBT30 to HA by direct surface grafting via polycondensation, avoiding the use of potentially harmful coupling agents or additional modifications of the particle surface. We aim at preparing porous PEOT/PBT scaffolds for bone tissue engineering that can be seeded and cultured with bone marrow stromal cells before implantation in the bone defect.

Materials and methods

HA-composites prepared by blending

A 1000PEOT70PBT30 copolymer was prepared as previously described [3] from PEG of molecular weight 1000, 1,4-butanediol, dimethyl terephthalate and vitamin E as antioxidant. The soft segment to hard segment weight ratio was 70 to 30. To 20% (w/v) copolymer solutions in chloroform, sintered HA with a particle size of 38–53 μm was added; the suspension was precipitated in ethanol and dried.

HA-composites prepared by grafting

The HA particles were immediately added to the reaction mixture, which was allowed to polymerize by polycondensation for 8 h.

Films prepared by compression molding

Films of HA-composites prepared by blending were prepared by compression molding at 140 °C. HA-composites prepared by grafting were compression molded at 180 °C, since compression molding at 140 °C did not result in homogeneous films. The film thickness for both the composites prepared by blending and by grafting was approximately 500 μm .

Water uptake

Composite films were swollen in milliQ water at 37 °C for 48 h.

Tensile testing

Strips of 5×100 mm (HA-composites prepared by blending) or 5×50 mm (HA-composites prepared by grafting) were cut from compression-molded films and subjected to tensile testing at 50 mm/min using a Zwick Z020.

Extraction of the soluble fraction of HA-composites.

Compression molded HA-composite films were extracted for 48 h with chloroform using a Soxhlet setup.

Thermogravimetric analysis (TGA)

TGA measurements were performed using a Perkin Elmer TGA 7 from 50 to 700 °C at a heating rate of 10 °C/min under a nitrogen flow.

Infrared spectroscopy

IR spectra were recorded using a Biorad FTS-60. Extracted HA-composites, HA and 1000PEOT70PBT30 were mixed with KBr and pressed into a pellet.

Results and discussion

IR-spectra of the residues after extraction of the HA-composites prepared by blending and grafting were compared to the spectra of 1000PEOT70PBT30 and HA. An example (residues from HA-composites prepared with approximately 17.5 vol.% HA) is shown in Fig. 1. After extraction, the residue of the HA-composite prepared by grafting clearly shows characteristics of the absorption spectrum of the copolymer, indicating a strong bond (most likely chemical) between the copolymer and the HA.

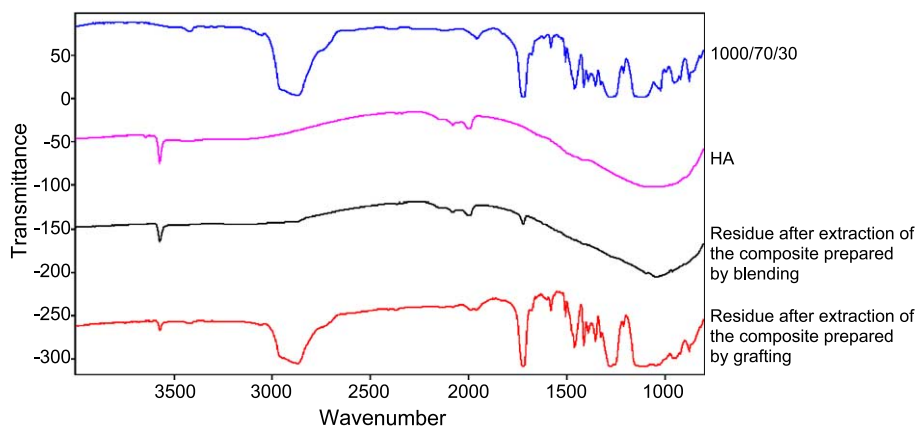


Fig. 1. IR-spectra of 1000PEOT70PBT30, HA and the extracted residues of HA-composites prepared by blending and grafting (17.5 vol.% HA).

The residues of the extracted HA-composite films were analyzed using TGA. The results are summarized in Table 1.

Table 1
TGA data of extracted composites films, prepared by compression molding

Composite	vol.% HA	Onset of mass loss	mass% 1000PEOT70PBT30 remaining after extraction
Pure HA	98.9	No onset	–
10 vol.% HA blended [§]	95.6	385 °C	1.7
17.5 vol.% HA blended [§]	96.8	378 °C	1.2
25 vol.% HA blended [§]	97.2	378 °C	1.1
10 vol.% HA grafted [‡]	11.7±0.1	411±2 °C	73.9±0.2
17.5 vol.% HA grafted [‡]	71.5±3.0	401±1 °C	13.1±1.7
25 vol.% HA grafted [‡]	95.5±0.1	397±1 °C	1.7±0.1

§: Single measurement.

‡: Average result of a duplicate measurement.

The residues of the extracted composites prepared by blending show only small amounts of copolymer (up to 1.7 mass%), this is most likely due to physical adsorption. The table shows that the residues of the extracted composites with 10 and 17.5 vol.% HA, prepared by grafting, contain high amounts of copolymer.

Model calculations, assuming spherical and non-porous HA particles with a diameter of 38–53 μm , covered with polymer chains with a cross-sectional area of 0.13 nm^2 (minimal area that a single ethylene oxide unit occupies [4] and a molecular weight of 150,000 g/mol (which is the maximum chain length obtained during polymerization in the absence of HA), result in a maximum value of 6.4–8.7 mass% of copolymer that can be expected on the HA particles.

The higher amounts of copolymer observed in the extracted residues of HA-composites prepared by grafting with 10 and 17.5 vol.% HA, could be the result of a subsequent cross-linking reaction of bound (to HA) and unbound copolymer chains during the polycondensation reaction upon evaporation of the vitamin E antioxidant. The HA-composite with 10 vol.% HA prepared by grafting only showed swelling in CHCl_3 and in HFIP. Gel formation was not observed for the HA-composites prepared by grafting containing 17.5 and 25 vol.% HA, although the presence of cross-linked copolymer chains cannot be excluded.

Composite films were subjected to tensile testing, both in the dry and water-swollen state. Data are presented in Table 2.

Table 2
Tensile properties of 1000PEOT70PBT30-HA composites.

Composite	E -modulus [†] (N/mm ²)	σ_{max} (N/mm ²)	ϵ_{break} (%)	σ_{break} (N/mm ²)	Energy up to break (Nmm)
1000/70/30	27±3 (23±1)	11.1±0.5 (6.5±0.2)	981±153 (144±33)	10.9±0.7 (6.3±0.2)	6380±1237 (1084±334)
10 vol.% HA blended	45±4 (20±2)	7.0±0.2 (3.1±0.3)	214±32 (32±4)	6.9±0.2 (3.0±0.3)	960±188 (96±22)
17.5 vol.% HA blended	54±5 (15±1)	6.2±0.4 (1.2±0.3)	62±15 (11±3)	5.9±0.3 (1.1±0.3)	264±79 (13.4±6.4)
25 vol.% HA blended	78±1 (12±2)	6.8±0.1 (0.67±0.08)	34±2 (6.5±0.7)	6.7±0.1 (0.66±0.09)	133±10 (3.8±0.7)
10 vol.% HA grafted	37±2 (16±1)	6.9±1.5 (3.7±0.7)	87±39 (29±6)	6.9±1.4 (3.4±0.9)	396±258 (113±48)
17.5 vol.% HA grafted	47±2 (10±1)	7.2±0.6 [§] (3.3±0.2) [‡]	271±93 [§] (58±11) [‡]	7.1±0.6 [§] (3.2±0.3) [‡]	1467±587 [§] (205±56) [‡]
25 vol.% HA grafted	61±3 (12±1)	6.1±0.2 [§] (2.0±0.2) [‡]	179±20 [§] (28±5) [‡]	5.8±0.1 [§] (1.9±0.2) [‡]	813±104 [§] (57±17) [‡]

Results after 48 h of water uptake are shown in parentheses. Experiments in fourfold (\pm S.D.).

[†] The sample deformation was derived from the grip-to-grip separation; therefore the presented E -moduli are only an indication of the stiffness.

[§] Significantly different from the corresponding values of the blended composite in the dry state.

[‡] Significantly different from the corresponding values of the blended composite in the water-swollen state.

In the water-swollen state, these hydrophilic composites show substantially deteriorated mechanical properties. Significant differences are observed between the HA-composites prepared by grafting and the HA-composites prepared by blending. The HA-composites with 17.5 and 25 vol.% HA, prepared by grafting, show a higher elongation at break and a higher energy up to break both in the dry and the water-swollen state than the corresponding HA-composites prepared by blending. In contrast to our expectations, HA-composites prepared by blending and those prepared by grafting show a decrease in E -modulus in the water-swollen state with an increase in HA vol.%.

Conclusions

TGA and IR-data of extracted composites prepared by grafting show the presence of relatively high amounts of copolymer compared to the extracted composites prepared by blending.

Both in the dry and water-swollen state, the composites prepared by grafting (with 17.5 and 25 vol.% HA) were significantly stronger and tougher than those prepared by blending. The combined IR, TGA and tensile data suggest that the copolymer is grafted onto the surface of HA particles. The incorporation of HA (either by grafting or by blending) does not result in composite materials, however, with improved mechanical properties compared to the original 1000PEOT70PBT30 copolymer, especially in the wet state.

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PEG–PLA HYDROGELS BY STEREOCOMPLEXATION FOR TISSUE ENGINEERING OF CARTILAGE

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Summary

PEG–PLA hydrogels were prepared by stereocomplexation of poly(lactide) (PLA) blocks of opposite chirality. Gelation occurred upon mixing aqueous solutions of both enantiomers of PEG6500–(PLA)₂, PEG12500–(PLA)₂ and PEG21800–(PLA)₈ block copolymers. Stereocomplexed hydrogels from PEG6500–(PLA)₂ polymers were found to be stable up to ~37 °C, while those from PEG12500–(PLA)₂ and PEG21800–(PLA)₈ block copolymers were stable up to ~50 °C.

Introduction

Due to their similarity with the extracellular matrix and their high biocompatibility, hydrogels have found wide interest as matrices for tissue engineering, in particular for soft tissues. Thermosensitive hydrogels composed of block copolymers of poly(ethylene glycol) (PEG) and aliphatic polyesters are promising materials for drug delivery applications and tissue engineering. PEG is known to have excellent antifouling properties and biocompatibility and is excreted by the kidney at molecular weights up to ca. 30,000 [1]. Aliphatic polyesters, such as poly(lactide) (PLA) are known to be biocompatible and render the hydrogel biodegradable. More recently, Li et al. [2], Fujiwara et al. [3] and Lee et al. [4] have shown that hydrogels can be prepared from PEG–PLLA and PEG–PDLA triblock copolymers, where the crosslinks in the hydrogel are provided by stereocomplexation between D- and L-lactide blocks. Stereocomplex-based oligolactide grafted dextran hydrogels have been developed by de Jong et al. [5]. These hydrogels provide a full preservation of enzymatic activity combined with a quantitative release and full degradation in 1 to 7 days. In this paper, we describe the solubility of a series of PEG–PLA triblock copolymers and the effect of stereocomplexation on gelation behaviour. PEG–PLA star-block copolymers with opposite chirality of PLA blocks were investigated to study the effect of multiple interaction sites on gelation and gel strength.

Experimental methods

PEG–(PLA)₂ block copolymers were prepared by the Sn(Oct)₂ catalysed ring opening polymerisation of D- or L-lactide initiated by hydroxyl groups of PEG–(OH)₂ (Fig. 1) at 105 °C in toluene for 4 h. PEG–(PLA)₈ block copolymers were prepared similarly at room temperature in dichloromethane for 4 h using the single site Zn catalyst Zn(Et)[S(C₆H₆–CH(Me)NC₅H₁₀)]-2.

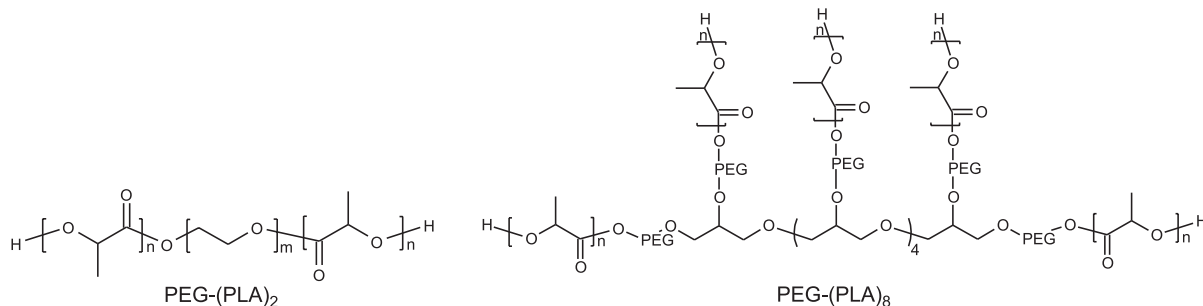


Fig. 1. Molecular structures of PEG–(PLA)₂ and PEG–(PLA)₈.

Aqueous polymer solutions were prepared by dissolving the appropriate amount of polymer in distilled water at room temperature. In case of hydrogels by stereocomplexation, polymer solutions of both D- and L-enantiomers were mixed and the sample was stirred vigorously for ~2 min. For both single enantiomer polymer solutions and for polymer solutions containing both D- and L-enantiomers, temperature-dependant gelation was tested at temperatures between 20 and 60 °C with intervals of 10 °C using a water bath. At each temperature, the samples were allowed to equilibrate for at least 20 min. No flow within 20 s while inverting the vial was regarded as a gel state.