Poly(Butylene Fumerate) and Poly(Butylene Fumerate)-*co*-(Butylene Maleate) as Biodegradable Materials for Tissue Engineering

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INTRODUCTION: Two new synthetic biodegradable polymers, poly(butylene fumerate) (PBF) and poly(butylene fumerate)-*co*-(butylene maleate) (PBFcBM), have been examined as tissue engineering scaffold materials. The polymers have been synthesized from the ring opening and condensation reactions of maleic anhydride and 1,3-butanediol. The cytotoxicity, cell attachment and viability, degradation as well as PBF and PBFcBM polymer processibility were all examined.

METHODS: Polymer Synthesis: Polymers of PBF and PBFcBM were synthesized following previously established procedures.[1]
Cell Culture: MC3T3-E1 cells were cultured in MEM alpha modification media supplemented with 10% (v/v) fetal bovine serum and 1% (v/v) penicillin/streptomycin at 37°C, 5% CO_2, and 95% RH. At 80% confluence, cells were lifted with 0.25% (w/v) trypsin.
Cytotoxicity: Monomers, polymers, and crosslinked networks were evaluated using a procedure established for evaluating poly(propylene fumarate) (PPF).[2]
Cell Attachment and Proliferation: Crosslinked PBF and PBFcBM substrates (d= 6mm) were used to evaluate attachment and viability using the MC3T3-E1cell line. Cells were seeded at 25,000 cells/cm^2 onto solid crosslinked polymer samples (n=5) which had been fitted in a 96 well non-TCPS plates. Attachment and viability were assessed using LIVE/DEAD Cell Viability for Mammalian Cells (Invitrogen®, L3224) per manufacturer's specifications followed by imaging using a Nikon Eclipse TS-100 –microscope.
Degradation: PBFcBM containing 33% maleate was evaluated. A polymer/BAPO (3wt%) solution was poured into glass vials followed by centrifugation (3 minutes at 5000rpm). PBFcBM was than crosslinked at $\lambda = 365$ nm for 3 hrs. Molds were broken and samples were cut to a height: diameter of 12:6 (mm). Samples were placed in glass vials with ~20mL of 1X phosphate buffer solution at pH 7.4 and incubated at 37°C with gentle agitation (75 rpm). Samples (n= 5) were removed at various time points to perform mechanical testing (ASTM standard D695) using an Instron 6564.
Electrospinning: A modified electrospinning technique was employed following a previously reported procedure.[3]

RESULTS: PBF and PBFcBM monomers were incubated with media for 24 hrs. The media, "extraction media," was removed from the samples and added to MC3T3-E1 cells at 80% confluence. In all cases, cells were viable independent of the extraction media concentration, indicating that the monomers and polymers have no toxic effect on the cells (data not shown). Films of PBF and PBFcBM were used to examine cell attachment and be proliferation to 72 hours. Qualitative assessment with LIVE/DEAD® fluorescence staining confirmed that cells were attached and proliferated when seeded directly onto crosslinked networks (data not shown). The compressive modulus of PBFcBM at several time points (1, 3, 5, 7, 10, 15 and 24 weeks) was measured (Figure 1). PBFcBM's loss of mechanical properties is faster than that reported for the well studied PPF, indicating and increased rate of degradation for the PBFcBM networks.[4] The increased degradation is potentially due to reduced steric hindrance to interactions between the ester groups of the polymer backbone and water. The liquid (low T_g) polyester PBF was also processed using electrospinning coupled with *in situ* photochemical crosslinking in order to produce nonwoven fiber mats with fiber diameters in the micron range.

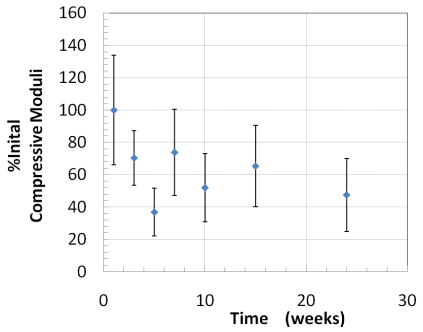

Figure 1- Normalized compressive moduli of PBFcBM

CONCLUSIONS: Synthesis of new polyesters for potential use in tissue engineering applications is presented. The polymers are non-cytotoxic and have the ability to form mats via an altered electrospinning method. Initial degradation results indicate that the introduction of the maleate functionality influences the degradation rate of the copolymer versus the fumarate homopolymer.

REFRENCES:

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