Controlled Release of bFGF from Elastomeric Biodegradable Microporous Sheets

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Introduction

For wound healing applications in which the underlying traumatized tissue will experience substantial volumetric changes due to edema, it would be attractive to have a barrier dressing that could be sutured to the healthy wound periphery, but readily distend and contract to meet the mechanical demands imposed by the underlying tissue. Controlled release of a growth factor to facilitate wound healing from this material could provide added benefit. In an effort to meet such requirements we report here on the development of a thin, microporous elastic sheet with high tensile strength made from a biodegradable poly(ester urethane)urea (PEUU) loaded with the angiogenic basic fibroblast growth factor (bFGF) and processed with electrospinning. Characterization included assessing morphology, bFGF release and bioactivity, cell adhesiveness, and tensile properties.

Materials and Methods

PEUU was synthesized from polycaprolactone diol and 1,4-diisocyanatobutane with chain extension by putrescine as previously reported.[1] Bovine serum albumin (BSA) and bFGF (100:1) were dissolved in buffer and the solution was lyophilized. PEUU was dissolved at 6wt% in hexafluoroisopropanol and bFGF/BSA was added to the polymer solution at 500ng bFGF / mg PEUU. After solubilization the solution was electrospun over a 15-cm distance using a 1mL/min feed rate and by charging the solution at 10kV and the aluminum target at -10kV in a manner similar to that previously reported.[2]

Scanning electron microscopy (SEM) was utilized to characterize fiber morphologies in the sheets. Strips of material were placed in DMEM with 0.5% fetal bovine serum and 1% penicillin /streptomycin at 37°C for bFGF release studies. Immunoassay quantified bFGF concentration in the medium. Released bFGF bioactivity was measured by mitogenic assay. Briefly, rat vascular smooth muscle cells (SMCs) were seeded at 1.5 x 10^5 in a 96-well plate. Media was replaced at 4h with the appropriate bFGF containing degradation media and cell number was measured 48 h later using the MTT mitochondrial activity assay. For cell adhesion, SMCs were seeded onto sheets at a density of 2.0 x 10^5 cells/mL and cell adhesion was quantified 1 day after seeding using MTT. Tensile testing was completed according to ASTM D638-98.

Results

Electrospun sheets of PEUU and PEUU with bFGF consisted of continuous, bead-free sub-micron diameter fibers as observed by SEM. bFGF release from sheets consisted of an initial 25% burst followed by slower release up to 50% by 3 wks **(Figure 1)**. Bioactivity

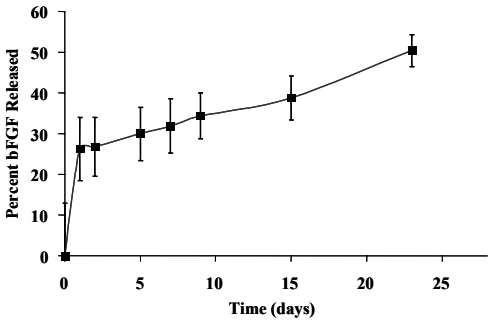

Figure 1. *Release of bFGF from electrospun PEUU.*

results from 0-7 days showed a statistically similar SMC mitogenic effect from PEUU loaded with bFGF compared to a control group directly receiving 1 ng/mL bFGF **(Figure 2)**. Significantly higher SMC adhesion at 148 ± 5% of tissue culture polystyrene (TCPS) was observed on PEUU loaded with bFGF relative to both TCPS and electrospun PEUU alone ($p<0.05$). No significant differences in tensile properties were found between PEUU and PEUU loaded with growth factor.

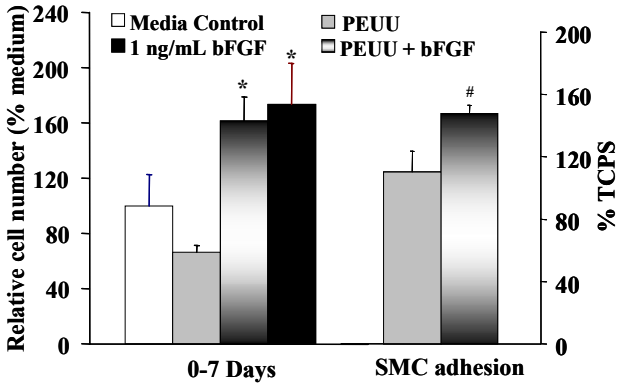

Figure 2. *Released bFGF bioactivity at 1wk using a SMC mitogenic assay (left) and SMC adhesion on PEUU loaded with bFGF (*$p < 0.05$ vs. medium control, $^{\#}p < 0.05$ greater than PEUU control).*

Conclusions

A biodegradable polyurethane was loaded with bFGF and processed into microporous sheets comprised of sub-micron scale fibers. These elastomeric sheets were capable of bioactive bFGF release for at least 1 week and facilitated increased SMC adhesion. In wound healing applications where material flexibility and growth factor release from a barrier would be desirable, these growth factor loaded sheets may provide attractive functionality.

References

1. Guan J et al. *J Biomed Mater Res* 61:493 (2002).
2. Stankus JJ et al. *J Biomed Mater Res* 70:603 (2004).