Novel Polymers and Nanocomposites as Injectable Bone Tissue Engineering Materials

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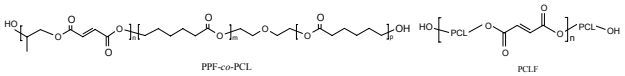
Introduction: Biodegradable and crosslinkable copolymers poly(propylene fumarate-*co*-caprolactone) (PPF-*co*-PCL) and polycaprolactone fumarate (PCLF) have been developed to obtain controllable physical properties that can meet the design requirements of tissue engineering scaffolds for use in bone and nerve regenerations.[1,2] Hydroxyapatite (HA) has been used for various applications such as implant coating. Its advantages include the composition similarity to bone mineral, bioactivity and promotion of cellular function, and osteoconductivity. In this study, nano-HA has been used as a filler material to the crosslinkable polymer matrix to fabricate biodegradable and bioactive nanocomposites for orthopedic applications. Both *in vitro* and *in vivo* biological evaluations of the polymers and their nanocomposites with HA have been performed.



Methods: Two PPF-*co*-PCL copolymers and two PCLFs (PCLF530 and 2000 as labeled using their PCL precursor molecular weights[2]) with distinct mechanical properties were selected for making nanocomposites. Nano-HA whiskers with long and short axis of ~100 and ~20 nm, respectively, were purchased from Berkeley Advanced Biomaterials. Polymer and nano-HA were mixed well in CH_2Cl_2 at 37°C before crosslinking. In chemical crosslinking, benzoyl peroxide and *N*-dimethyl toluidine were used as the initiator and accelerator, respectively. The polymerizing mixture was transferred into molds and the molds were placed in a convection oven overnight to facilitate crosslinking. In photo-crosslinking, polymer/HA mixture with a certain amount of photoinitiator bisacylphosphine oxide was poured in a mold formed by two glass plates and a Teflon spacer. The mold was then placed directly under UV light to allow crosslinking.

The chemical structures of both polymers and nanocomposites before and after crosslinking were examined by FTIR spectra. The thermal properties were measured by DSC and TGA. The mechanical properties including compression and tensile moduli have been tested. The dynamic frequency sweep measurements of the polymer nanocomposites as well as the pure polymers were performed on a rheometer. Cell attachment and proliferation were tested by seeding rat bone marrow stromal cells (BMSCs) directly onto polymer disks. A colorimetric MTS assay was used to determine the cell number. Porous polymer and nanocomposite scaffolds were implanted in rats subcutaneously or in femoral bone defects. After 8 weeks, biocompatibility and bone tissue ingrowth was examined by a variety of methods such as micro-CT and histological analysis.

Results/Discussion: Selected results are presented below. In Fig.1a, it can be seen that the nano-HA particles were well dispersed in the polymer matrix. As shown in Fig.1b, crosslinked PCLF2000 had a higher surface modulus than crosslinked PCLF530 due to a higher crystallinity. For crosslinked PCLF530, the compression modulus was enhanced by increasing the composition of HA whereas only a slight increase was observed for crosslinked PCLF2000. As shown in Fig.2, with similar surface chemistry, topology, and protein adsorption, PCLF2000 supported cell adhesion and proliferation much better than PCLF530 as the result of stiffer surface modulus. With increasing HA composition, PCLF530/HA series showed a significant improvement in cell adhesion and proliferation while there was not much enhancement for PCLF2000/HA series. The role of mechanical factor in regulating cell responses[3] can be reflected in both pure polymers and nanocomposites. Porous 3D polymer and nanocomposite scaffold were found to be biocompatible and osteoconductive for bone ingrowth. Microspheres containing bone morphogenetic protein (BMP-2) can be incorporated in the scaffold to induce bone ingrowth.

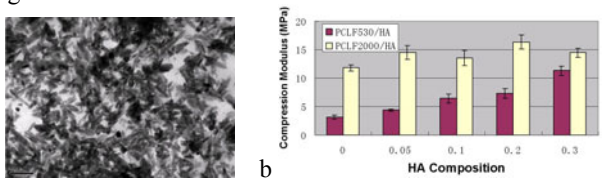


Fig. 1: (a) TEM picture of PCLF2000/HA with 30% HA; (b) Compression moduli of PCLF/HA nanocomposites at different HA compositions.

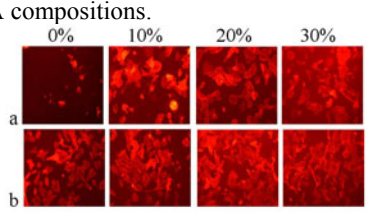


Fig. 2: Day 1 BMSC (stained with rhodamine phalloidin) attachment and proliferation on the photocrosslinked PCLF/HA (a: PCLF530/HA; b: PCLF2000/HA) nanocomposite disks at different HA compositions (0 – 30%).

Conclusions: Novel crosslinkable polymer and nanocomposite scaffolds have been prepared and characterized in detail. These materials are promising as injectable scaffolds for bone tissue engineering.

References:

1. Wang SF et al. Macromolecules 2005;38:7358.
2. Wang SF et al. Biomaterials 2006;27:832.
3. Discher DE et al. Science 2005;310:1139.

Acknowledgments

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