Injectable Polymer/β-TCP Biocomposite Delivery Systems for rhBMP-2

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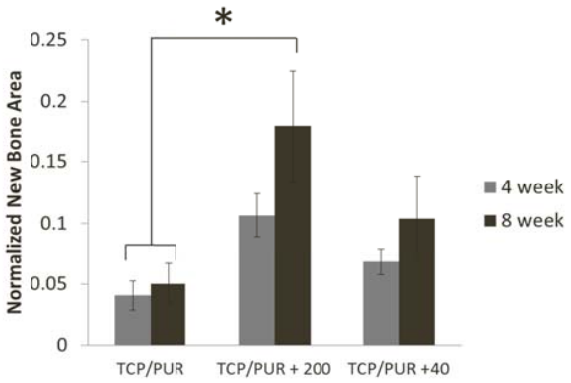
Statement of Purpose: Growth factors incorporated into scaffolds for tissue engineering promote the infiltration of cells and tissue. Recombinant human bone morphogenetic growth factor-2 (rhBMP-2) stimulates osteoblast differentiation and new bone formation. Biodegradable polyurethane (PUR) biocomposites incorporating allograft bone particles have been reported to be effective carriers for rhBMP-2 and support new bone growth[1]. However, allograft presents both biological and regulatory challenges. Delivery of even a low dose of rhBMP-2 combined with allograft can result in transient resorption.[2] Additionally, the combination of a recombinant human growth factor with allograft bone may cause regulatory concerns. β-Tricalcium phosphate (β -TCP) is a biocompatible, resorbable ceramic that has been used effectively as a substitute for allograft bone.[3] In the present study, we investigated the ability of an injectable PUR/β-TCP composite with two doses of rhBMP-2 to heal 8-mm critical-size calvarial bone defects in rats.

Methods: The biodegradable polyurethane (PUR) was synthesized from a lysine triisocyanate (LTI) prepolymer and polyethylene glycol (PEG), a polyester triol (900 g/mol), and triethylene diamine catalyst. The lyophilized rhBMP-2 was hand mixed into the PUR and injected into 8-mm critical-size calvarial defects in rats. Animals were sacrificed at 4 or 8 weeks and new bone formation evaluated by radiographs, μCT, histology, and histomorphometry. Treatment groups included the biocomposite containing 45% β-TCP with no rhBMP-2 (negative control), a recommended dose of 200 μg/mL rhBMP-2, or a low dose of 40 μg/mL rhBMP-2 (n=13/group). During histomorphometry, new bone area was normalized to the area occupied by the biocomposite, as the material expanded outside the thickness of the defect in some cases. Statistics were determined using one-way ANOVA and Tukey's test at ($p<0.05$).

Results: Bone growth within the biocomposite was evident for all groups as determined by histomorphometry (Fig. 1). The 8 week PUR/TCP group with 200 μg/mL rhBMP-2 had significantly more new bone than the group without rhBMP-2. Both rhBMP-2 doses showed increased new bone formation as compared to the negative control. The new bone was uniformly distributed throughout the interior of the biocomposite with 40 μg/mL dose (Fig. 2A), but the bone formation was primarily in a superior and inferior callus for the 200 μg/mL dose with the inside of the defect devoid of new bone(Fig. 2B). In both doses, β-TCP particles were still evident at 8 weeks (Fig. 2), as noted by the bright white particles in the μCT images. Histological sections showed voids due to rapid polymer degradation in the interior of the PUR/β-TCP graft with 200 μg/mL at 8 weeks.



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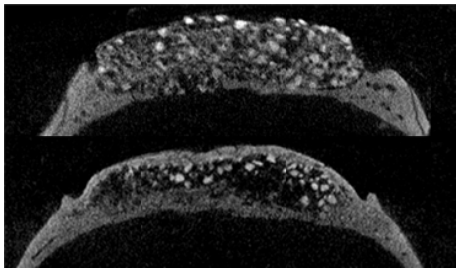

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Conclusions: Injectable PUR/β-TCP biocomposites support new bone formation and remodeling in a rat calvarial defect model, which is enhanced by the addition of rhBMP-2. The samples with the recommended dose experienced increased PUR degradation that may have been caused by the increase in cellular activity due to the rhBMP-2. The low dose of rhBMP-2 could allow for the optimal rate of polymer degradation and new bone formation so that no voids form in the interior of the biocomposite Additionally, a significant amount of β-TCP remained at 8 weeks which could hinder new bone formation. In ongoing experiments, we are investigating alternatives to β-TCP to test the effects of matrix resorption rate on new bone growth. We also aim to tailor the degradation rate of the PUR by changing the molecular weight and composition of the polyesterol triol to match the rate of cellular infiltration.

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