Injectable Allograft Bone/Polymer Composite Bone Void Filler with Recombinant Human Bone Morphogenetic Protein

JE Dumas[1], KJ Zienkiewicz[1], PB Baer[2], and SA Guelcher[1]
1. Department of Chemical Engineering, Vanderbilt University, Nashville, TN
2. US Army Institute of Surgical Research, San Antonio, TX

Statement of Purpose. The treatment of craniofacial defects is a challenging problem due to the requirements for both biological and mechanical properties. Autograft bone, which is both osteoconductive and osteoinductive, is an attractive choice for treating craniofacial bone defects. However, autograft bone is limited in supply and can be difficult to fabricate into shapes conforming to irregularly shaped defects. Injectable biomaterials eliminate this problem with the ability to cure and conform to wound geometry *in situ*. We have developed an injectable allograft/polymer composite bone void filler that incorporates recombinant human bone morphogenetic protein (rhBMP-2). Polyurethane scaffolds have been shown to degrade into non-toxic compounds[1] and have tunable porosities, degradation rates, mechanical properties, and working times. Compressive modulus and strength values range from 173-444.1 MPa and 4.38-9.47 MPa, respectively, which are in the range required to withstand pulsatile forces from the dura.[2] MBP/PUR composite void fillers have shown remodeling potential in a rabbit calvarial model as shown in Figure 1.

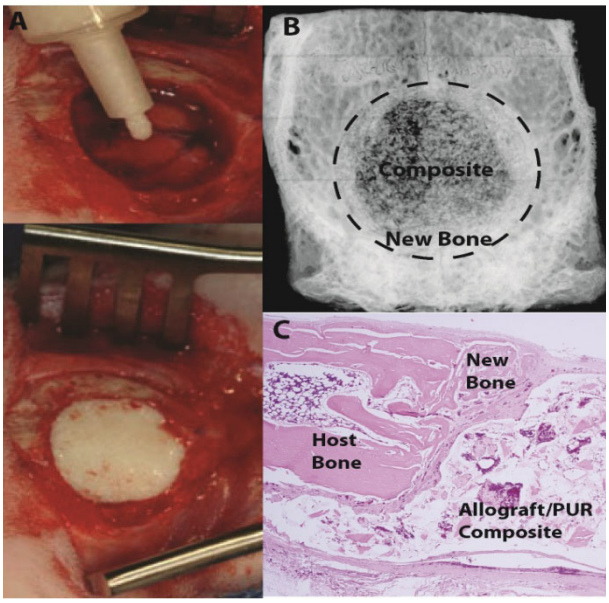

Figure 1. Injectable MBP/PUR composites. (A) Injection of composites into a 15-mm rabbit calvarial defect, where the material conforms to the wound and cures to form a solid. (B) Faxitron image after 6 weeks shows new bone formation around the perimeter of the defect (dashed line). (C) Histological section (H&E stain) of the defect taken from a rabbit that died on day 12 due to causes unrelated to surgery. No adverse tissue response was observed, and there was evidence of new bone formation at the edge of the defect

Materials and Methods. Injectable MBP/PUR composite void fillers are composed of lysine triisocyanate (LTI), poly(ethylene glycol) (PEG, 200 g/mol), polyester polyol (60% caprolactone, 30% glycolide, 10% lactide, 900 g/mol) (6C3G1L900), rabbit mineralized bone particles (RMBP, 100-500 μm), and Infuse rhBMP-2(provided as lyophilized powder). LTI-PEG prepolymer was synthesized from the reaction between LIT and PEG. The rhBMP-2 was reconstituted in sterile water and aliquoted in glass vials to yield a mass of 210 μg in each vial. The appropriate amounts of 6C3G1L900, MBP, and LTI-PEG prepolymer were added to a 10 mL cup and hand-mixed for 90 seconds. A 0.25 mL scoop was used to transfer approximately 0.38 g of the mixture into the vial of rhBMP-2, and the appropriate amount of catalyst solution (5% triethylene diamine in dipropylene glycol) was added to the vial. The components were mixed for 1 minute followed by loading and injection from a 1 mL syringe. The target bone content was 47 wt%, and the target porosity was 40%. To quantify the release of the rhBMP-2, MBP/PUR composite discs were placed in complete α-MEM media with the addition of 0 .75% BSA. A critical-sized rabbit calvarial defect study was designed to study the enhanced remodeling capability of the MBP/PUR composites with the incorporation of rhBMP-2. A 15-mm circular defect was cut in the calvaria of New Zealand white rabbits as shown in Figure 1A. The volume of the defects was measured to be ~0.5 mL. Thus, a volume of 0.25 mL of MBP/PUR/rhBMP-2 composites was injected into the defect to allow for expansion.

Results. The MBP/PUR/rhBMP-2 composites expanded to fill the entire defect volume. After 10 min, the foams had cured and become tack-free, completely dampening the pulsation of the dura. The wounds were subsequently closed and the rabbits were closely monitored until all vital signs were normal. Sustained release of biologically active from rhBMP-2 from PUR scaffolds has been demonstrated in rat femoral plug and segmental defect models.[3] Radiographs of MBP/PUR scaffolds without rhBMP2 showed ~2 – 4 mm of new bone ingrowth after 6 weeks implantation time *in vivo*.

Conclusions. MBP/PUR composites exhibit suitable mechanical properties and remodeling for repair of calvarial defects.

References. 1.) Guelcher S. Biodegradable polyurethanes: synthesis and applications in regenerative medicine. Tissue Eng B.14:3-17. 2008, 2.) Dumas J, et al. Synthesis and Characterization of an Injectable Allograft Bone/polymer Composite Bone Void Filler with Tunable Mechanical Properties. (Submitted) 3.) Li B, et al. The effects of rhBMP-2 released from biodegradable polyurethane/microsphere composite scaffolds on new bone formation in rat femora. Biomaterials. 2009 Dec;30(35):6768-79

Acknowledgements. This work was supported by the Armed Forces Institute of Regenerative Medicine: Rutgers - Cleveland Clinic Consortium (DOD/USAMRAA contract # W81XWH-08-2-0034).