

## Strategies for Segmental Bone Repair

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**Background:** The management of large bone defects caused by trauma, degenerative disease, or tumor resection is one of the most challenging problems faced by orthopaedic surgeons (Rommens *et al.*, 1989). Autogenous bone grafting is currently the clinical standard for osseous reconstruction. However, the use of autograft bone has several disadvantages including limited available tissue for transplantation, lack of structural integrity to withstand functional loads, and increased patient morbidity at the site of harvest. Because of these limitations, structural allografts are increasingly being used clinically. Allografts provide structural integrity in the short-term but are associated with a high rate of complications and late fractures. The 25 – 35% failure rate of allografts due to nonunions and fractures 1-2 years after implantation is directly associated with their recognized lack of ability to revascularize and remodel (Berrey *et al.*, 1990; Kofoed *et al.*, 2005). These problems have led to the search for improved methods for stimulating bone repair. We recently collaborated on a study led by Drs. Regis O'Keefe and Xinping Zhang that showed the reduced healing potential of allografts is due to the absence of cellular activity and that revitalization of allografts via incorporation of an engineered periosteum containing BMP-2 expressing mesenchymal progenitor cells significantly improves allograft incorporation (Zhang *et al.*, 2005).

Tissue engineering strategies may therefore be used to improve graft repair or alternatively to develop bone graft substitutes. The basic elements required for successful bone repair include an extracellular matrix scaffold, cells, a vascular supply, and osteoinductive factors (Bruder and Fox, 1999). If these elements are not available from tissues surrounding the injury site, they may need to be provided in some combination within an implantable tissue-engineered construct. The transformation from bone grafting to bone tissue engineering began with the introduction of osteoconductive bone graft substitutes or scaffolds and is now evolving to include local delivery of

osteogenic cells and bioactive factors. Osteoconductive scaffolds facilitate invasion of capillaries, attachment of osteoprogenitor cells and subsequent appositional mineralized matrix formation in large bone defects. In addition to the scaffold material, microarchitectural parameters, such as porosity, pore size, interconnectivity, surface morphology, and anisotropy, strongly influence the mechanical properties of, and biological responses, to porous scaffolds (Lin *et al.*, 2003). Scaffolds may be used to locally deliver osteoinductive or angiogenic proteins or genes. A major remaining barrier to the use of bioactive factors for tissue regeneration has been the identification of effective and safe doses and delivery methods. However, sophisticated temporal and spatial release strategies may ultimately overcome these limitations (Rose *et al.*, 2004). Cell-based strategies for engineering bone regeneration involve the implantation of differentiated osteoblasts or osteoprogenitor cell populations, derived from a growing number of tissue sources. Cellular augmentation may be especially important for difficult clinical cases involving older patients, smokers, patients receiving chemotherapy or radiation, and patients with severely damaged wound beds or metabolic diseases in which the endogenous cellular supply may be diminished (Bruder and Fox, 1999).

### Strategies to Augment Bone Repair Scaffolds:

We have established a challenging 8 mm rat segmental defect model to test different strategies for inducing functional bone repair. The model facilitates use of micro-CT imaging to quantify 3D ingrowth of bone and vascularity and mechanical testing to evaluate functional integration. Using these models and methods, three distinct strategies are being investigated involving the delivery of proteins, cells, or genes. For protein delivery, our goal is to identify combinations of growth factors and/or sustained delivery strategies that induce functional bone repair at lower and therefore safer and less costly doses. Studies involving protein release from RGD alginate and nanoparticles will be presented. For cell delivery, we seek to

quantitatively evaluate the ability of different cell sources to promote bone repair, with and without in vitro predifferentiation. In addition to marrow-derived cells, we are evaluating the osteogenic potential of amniotic fluid derived cells both in vitro and in vivo. Finally, our gene delivery strategy is to use an rAAV coating approach developed by Dr. Edward Schwarz at the University of Rochester to augment the bioactivity of bone repair scaffolds. This approach has recently been shown to improve structural allograft healing (Koefoed *et al.*, 2005).

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