Tissue engineering strategies in spinal arthrodesis: the clinical imperative and challenges to clinical translation

Skeletal disorders requiring the regeneration or *de novo* production of bone present considerable reconstructive challenges and are one of the main driving forces for the development of skeletal tissue engineering strategies. The skeletal or mesenchymal stem cell is a fundamental requirement for osteogenesis and plays a pivotal role in the design and application of these strategies. Research activity has focused on incorporating the biological role of the mesenchymal stem cell with the developing fields of material science and gene therapy in order to create a construct that is not only capable of inducing host osteoblasts to produce bone, but is also osteogenic in its own right. This review explores the clinical need for reparative approaches in spinal arthrodesis, identifying recent tissue engineering strategies employed to promote spinal fusion, and considers the ongoing challenges to successful clinical translation.

KEYWORDS: autograft ■ bone graft substitute ■ clinical translation ■ fusion ■ mesenchymal stem cell ■ osteogenesis ■ spinal arthrodesis ■ tissue engineering

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Spinal arthrodesis is a common surgical procedure with over 250,000 cases performed annually in the USA alone [1]. It is used to relieve pain and restore motion segment stability in the treatment of spinal trauma, degenerative disc disease, spondylolisthesis, deformity, tumor and infection, but nonunion rates of between 5 and 35% have been reported [2,3], causing significant patient morbidity and placing a considerable financial burden on the healthcare system. With its inherent biological and mechanical properties, autograft is regarded as the 'gold standard' in spinal fusion procedures as it contains the necessary components for achieving a solid fusion. Integral to bone formation is a pool of mesenchymal stem cells (MSCs) with considerable osteogenic potential. In addition, the porous trabecular structure of cancellous bone provides an osteoconductive/osteoinductive environment that facilitates the attachment and migration of osteogenic cells and aids vascular ingrowth, while osteoinductive growth factors – both inherent to autologous bone but also synthesized by the MSCs themselves – provide the necessary stimulus for the differentiation and proliferation of osteoprogenitor cells. The iliac crest is the predominant site of autograft harvest, but with donor site morbidity reported in up to 39% of cases [4], an increase in operative and rehabilitation time and limited availability, alternative materials to promote spinal fusion have been sought.

Autologous bone graft substitutes & their problems

Allograft

The use of allograft overcomes some of the problems associated with iliac crest bone graft harvest, but while exhibiting good osteoconductive properties, allograft has minimal osteogenic and only weak osteoinductive activity, resulting in slower graft incorporation and inferior fusion results compared with autograft [5,6]. Furthermore, there are concerns regarding host immunogenicity and the risk of disease transmission [7]. Demineralized bone matrix (DBM), a processed form of allograft in which the inorganic mineral content has been removed leaving the organic collagen matrix and exposed BMPs, has enhanced osteoinductivity and reduced immunogenicity in comparison with untreated allograft [8,9]. However, studies have found considerable variation in the BMP content not only between different DBM preparations, but also in DBM preparations from the same source [10,11]. Furthermore, the efficacy of DBM to induce bone formation *in vivo* has been shown to be significantly influenced by the host environment [12–15]. These findings may explain the disparate results reported in clinical spinal arthrodesis studies using DBM [5,16,17].

Synthetic scaffolds

Synthetic scaffolds have been developed for use in spinal arthrodesis to overcome the

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complications associated with bone graft use and are available in various preparatory forms (powder, pellets, paste, gel, putty, cement, granules, blocks, strips and sponge) depending on their clinical application and role as either bone graft extenders, enhancers or substitutes. Ceramic composites (β-tricalcium phosphate [β-TCP], hydroxyapatite and calcium sulfate), polymers (poly-glycolic acid, poly-L-lactic acid, poly-lactic-co-glycolic acid and polyether ether ketone), extracellular matrix proteins (collagen, laminin, fibronectin and glycosaminoglycans) and trabecular metal (tantalum) have all been used in the fabrication of these scaffolds given their outstanding osteoconductive properties, providing not only a framework to facilitate vascular ingrowth and cell migration, but also an environment that protects the osteogenic cells and encourages bone formation [18–25]. Despite the osteoconductivity of these materials, they too display limited osteogenic and osteoinductive potential when used in isolation.

Osteoinductive agents

Bone formation is regulated by the complex interaction of numerous growth factors and cytokines (e.g., TGF-β, FGF, IGF, PDGF and IL-1 and -6). This cellular interaction is instrumental in coordinating the chemo-attractive, migratory, differentiation and proliferative responses of individual cells as new bone is formed. BMPs form part of the TGF-β family that provide a biological stimulus to bone formation through stimulation of the host osteoblasts, and as such have been incorporated into scaffolds for their osteoinductive potential. The ability of BMP-2 and -7 to promote spinal fusion has been demonstrated both *in vivo* [26,27] and in clinical translational studies [28,29]. Recombinant technology has enabled the development of tailored BMPs, specifically modified at a molecular level to improve their osteoinductivity. Furthermore, clinical outcome studies have shown comparability between rhBMP-2 and autograft in achieving spinal fusion [29,30]. However, there are a number of potential limitations associated with the use of BMPs. First, the effectiveness of recombinant growth factors is reliant on the existence of an adequate osteogenic cell population. Second, a carrier is needed that controls and maintains their release so as to prevent their diffusion from the fusion site. Third, the doses required in humans to promote a fusion are considerably higher (and thus more expensive) than those used in animal studies, thereby prohibiting the widespread use of these products [31,32]. A recent systematic review by Mroz *et al.* has also highlighted a number of complications associated with the use of rhBMP-2, including dysphagia following anterior cervical interbody surgery and excessive vertebral body resorption, interbody space subsidence, graft migration and ectopic bone formation following posterior lumbar interbody surgery [33]. Thus, only rhBMP-2 has current US FDA approval for clinical use, and even then, rhBMP-2 is only licensed for single-level anterior lumbar interbody fusions between L4 and S1 in patients with degenerative disc disease unresponsive to 6 months of conservative management.

Biological constructs, incorporating allograft or bone graft substitutes with agents that provide a biological stimulus for bone formation, have been developed with variable clinical success, but the absence of any inherent osteogenic activity remains a fundamental problem, with complete reliance on the hosts own osteoblasts to initiate bone formation. Theoretically, the abundance of cancellous bone within the vertebral body and posterior elements should provide a rich supply of osteogenic cells. However, even with good surgical technique, where the vertebral endplates or posterior elements are decorticated to expose bleeding cancellous bone, Romih *et al.* have demonstrated that the number of potential osteoblast progenitor cells released is significantly less than from an iliac crest or vertebral body aspirate [34]. Building on the existing knowledge of scaffold design, tissue engineering strategies have been employed in an attempt to create a construct that not only overcomes this osteogenic deficiency, but also minimizes the morbidity associated with autograft harvest.

Tissue engineering strategies

Bone marrow-derived MSCs

Iliac crest bone marrow aspiration provides one of the quickest and simplest means of obtaining an osteogenic pool of osteoprogenitor and skeletal stem cells – commonly referred to as MSCs. By directly applying bone marrow aspirate (BMA) to a scaffold, the ability of MSCs to promote spinal fusion has been investigated *in vivo* in both small and large animal studies (Table 1) [35–42]. There appears to be a clear osteogenic and mechanical advantage conferred to scaffolds seeded with BMA over unseeded scaffolds, with a number of studies demonstrating improved spinal fusion rates compared with autograft controls [35,37,40,42].