Chapter 9

Bioabsorbable Polymer and Bone Growth Factor Composites

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Summary

Bone growth factors are small proteins contained by bone tissue. The most intensively investigated bone growth factors are the bone morphogenetic proteins (BMPs) and the transforming growth factor-bβ (TGF-βs). Their local application induces bone formation in experimental fractures and bone defects. Possible clinical applications for bone growth factors include impaired bone healing, inadequate bone stock, and incomplete incorporation of prosthetic implants into bone. In therapeutic use the bone growth factors need a carrier. The carrier should assure local, sustained release of the growth factors which may otherwise be rapidly absorbed before instituting their effect. Several biodegradable materials have been investigated as carriers for bone growth factors, including organic materials, ceramics, and synthetic polymers such as polylactide and polylactide-polyglycolide co-polymer. Bioabsorbable polymers possess several attractive features as delivery systems. They can be engineered to allow release of accurate dosage of bone growth factors and they also allow combining several growth factors to be delivered concomitantly or at different points of time. They can also be manufactured or sculpted into a variety of forms to fit a given tissue defect or to fix a fracture. Moreover, they are both sterilizable and can be resorbed completely. An increasing number of positive results from experimental studies exist which could indicate a possible future success for growth factors within clinical orthopaedic practice. Recently, the first human studies using recombinant human BMP-2 or BMP-7 (osteogenic protein-1, OP-1) have been published.

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Bone growth factors are small proteins contained by bone tissue. The most intensively investigated bone growth factors are the bone morphogenetic proteins (BMPs) and the transforming growth factor-βs (TGF-βs). Their local application induces bone formation in experimental fractures (1,2) and bone defects (3,4). A simplistic summary may be that TGF-β, released by platelets at the fracture haematoma, stimulates cells to make and to react to BMPs and other growth factors in a synergistic cascade. This cascade, given the right mechanical conditions and the presence of new blood supply, leads to regenerative bone repair.

Possible clinical applications for bone growth factors include impaired bone healing, inadequate bone stock, and incomplete incorporation of prosthetic implants into bone. Complicated fractures with extensive soft tissue damage often heal insufficiently, resulting in delayed union or nonunion. Large amounts of bone grafts are needed in bone tumour surgery, endoprosthetic surgery, and reconstructive and maxillofacial surgery. Today, the above-mentioned clinical problems are solved by bone grafting. Autogeneic bone graft from the iliac crest is the main source of trabecular bone. It has a good osteoinductive capacity, but the sources are limited and the harvest procedure causes postoperative discomfort to the patient. Allogeneic bone is also widely used but provides mainly osteoconductive properties. Moreover, despite of extensive testing, there are still potential risks for transmitting diseases.

Bone-derived growth factors are produced by osteoblasts and incorporated into the extracellular matrix during bone formation. Trauma or remodeling causes solubilization of the proteins (5). After release, the growth factors may initiate and control a healing response after bone trauma and they are able to regulate osteoblast and osteoclast metabolism during bone remodeling. They exhibit their effects only in the local environment. Most growth factors are released as high molecular weight precursors which are split by proteolysis to produce active factors which are generally of a low molecular weight (6). They exhibit their effects by binding to membrane bound receptors on the surface of the target cells. This leads to a cascade of intracellular events which affect the expression of genes that encode for such functions as protein synthesis and cell devision. The total number of identified growth factors able to affect proliferation, differentation, and secretive functions of bone-related cells is continually expanding as a result of research.

The following bone growth factors are the most important in the bone tissue. Transforming growth factor-beta (TGF-β) exists in five different subtypes (TGFβ1-5). TGF-β is probably the most potent multifunctional regulator of bone cell metabolism. Fifteen bone morphogenetic proteins (BMPs) have, so far, been identified (7). They are the only growth factors known to stimulate the mesenchymal stem cells to differentiate into osteoblastic and chondroblastic lineage. Platelet-derived growth factors (PDGFs) exist in three isotypes and are released from the platelets at the site of injury. They are potent mitogens for cells of connective tissue origin and are thought to play a role in soft tissue and fracture healing (8). Insulin-like growth factors (IGF I and II) are produced by osteoblasts. They have been suggested to have important functions for skeletal growth by mediating the growth-promoting actions of groth hormone (9). Fibroblast growth factors (FGFs) are secreted by osteoblasts. The prototype members of the fibroblast growth factor family are the acidic fibroblast growth factor (aFGF, FGF-1) and the basic fibroblast growth factor (bFGF, FGF-2) (10). They are best known for their effects on endothelial cell replication and neovascularization (11). PDGFs, IGFs, and FGFs are all mitogenic factors produced in early phases of fracture repair and localized in the bone matrix (12). However, their true therapeutic potential for fracture healing has not yet been revealed.

In therapeutic use the bone growth factors need a carrier. The carrier should assure local, sustained release of the growth factors which may otherwise be rapidly absorbed before instituting their effect.

Several biodegradable materials have been investigated as carriers for bone growth factors, including (1) organic materials such as inactive demineralized bone proteins (13,14), collagen (15,16), fibrin (17,18), squalene (19,20), and coral (21,22); (2) ceramics, including tricalcium phosphate (23,24), hydroxyapatite (25,26), calcium-sulphate composites (27,28), and bioactive glasses (29,30); and (3) synthetic polymers such as polylactide (31,32,33), polylactide-polyglycolide co-polymer (34,13,35,36), polyanhydride (37), and polyorthoester (38,39).

Bioabsorbable polymers possess several attractive features as delivery systems. They can be engineered to allow release of accurate dosage of bone growth factors and they also allow combining several growth factors to be delivered concomitantly or at different points of time. They can also be manufactured or sculpted into a variety of forms to fit a given tissue defect or to fix a fracture.

Moreover, they are both sterilizable and can be resorbed completely. The disadvantages of the bioabsorbable polymers include the foreign body reactions due to the degradation products (40,41,42,43). The porosity of the polymer material is often insufficient but can be increased by developing new materials or combining them to porous materials, such as demineralized bone matrix, to improve their osteoconductive properties. Polylactide (44,45,46,47,48,49) and the copolymer of polylactide and polyglycolide (50,13,51,52,53) are the most frequently used components of bioabsorbable carriers for bone growth factors in experimental studies.

Polyglycolide (PGA) and polylactide (PLA) are members of a large family of poly-α-hydroxy acids. These polymers can be manufactured by polymer processing methods into various shapes such as films, rods, plates and screws. Polyglycolide (PGA) or polyglycolic acid can be synthetized from glycolide (cyclic diester of glycolic acid l. hydroxyacetic acid) under the influence of an inorganic metal salt catalyst at low concentrations by a ring-opening polymerization. Polylactide (PLA) or polylactic acid is a synthetic polymer of lactide (cyclic diester of lactic acid). It can also by synthetized by the direct condensation polymerization of the monomer lactic acid. Polylactic acid has two enantiomeric forms, optically active stereoisomers, poly-L-lactic acid (PLLA) and poly-D-lactic acid (PDLA), with similar intrinsic chemical properties but opposite congurational structures (54). The physical properties of the co-polymers of L-lactic acid and D-lactic acid (PDLLA) are dependent on the relative amounts of L-and D-monomers in the polymer chain. The degradation process of PGA and PLA begins with molecular degradation; random hydrolysis starts in aqueous environment. The high surface area/weight ratio, porosity of the implant, low initial Mw, and high monomer concentration all speed up the molecular degradation process (54,55,56). The events are followed by a decrease of strength and mass of the implant, microscopic degradation in tissue, and transfer of the monomer lactic acid or glycolic acid to the metabolism of cells (57,58).

Our research group used a blend of an L-lactic acid oligomer and a co-polymer of epsilon caprolactone and DL-lactic acid as a carrier for TGF-β1 (59). This polymer was in paste form. The paste containing the growth factor TGF-β1 was used to fill the grooves on the surface of a bioabsorbable fracture fixation pin. The self-reinforced pin was made of poly-LD-lactic acid. Prior to starting the study we made an in vitro assay in which sustained release of TGF-β1 was demonstrated. A rat model was used to study the effect of TF-β1 combined to the bioabsorbable

carrier. A rat distal femoral osteotomy was made and stabilized with the bioabsorbable fracture fixation pin containing 0, 5, or 50 μg of TGF-β1. The study consisted of alltogether 60 rats and the follow-up times were 1, 3, and 6 weeks. After sacrifice the femurs were examined radiographically, histologically, histomorphometrically, microradiographically, and with oxytetracycline labelling studies. We report enhanced bone formation around the osteotomy in the TGF-β1-treated rats as compared to controls treated without the growth factor.

At the present, mostly preclinical, experimental data exists concerning the in vivo growth factor effects in clinically related animal models. An increasing number of positive results from experimental studies, such as ours, exist which could indicate a possible future success for growth factors within clinical orthopaedic practice. Nevertheless, it is not known whether or not the results from the animal studies can be transferred to humans with equal success. Recently, the first human studies using recombinant human BMP-2 (60,61,62) or BMP-7 (osteogenic protein-1, OP-1) (15,16,63,64) have been published. The results of these studies show a large variation among the responses of individual patients. These inconsistent results suggest that modulating factors known from animal studies affect the bone growth factor dependent bone induction process in humans as well. Such factors are the growth factor concentration, carrier properties and influence of other local and systemic growth factors and hormones. For better performance of bone growth factor containing bone-graft substitutes these factors need to be elucidated.

The use of bone growth factors will open a new set of treatment options. These will, ideally, enable us to obtain improved bone healing and bone formation in situations where the natural healing capacity of bone tissue is inadequate.

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