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Extracellular signaling molecules to promote fracture healing and bone regeneration☆

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ABSTRACT

To date, the delivery of signaling molecules for bone regeneration has focused primarily on factors that directly affect the bone formation pathways (osteoinduction) or that serve to increase the number of bone forming progenitor cells. The first commercialized growth factors approved for bone regeneration, Bone Morphogenetic Protein 2 and 7 (BMP2 and BMP7), are direct inducers of osteoblast differentiation. As well, newer generations of potential therapeutics that target the Wnt signaling pathway are also direct osteoinducers. On the other hand, some signaling molecules may play a role as mitogens and serve to increase the number of bone producing cells or may increase vascularization. This is true for factors such as Platelet Derived Growth Factor (PDGF) or Fibroblast Growth Factor (FGF). Vascular Endothelial Growth Factor (VEGF) likely has a special role. Not only does it induce new blood vessel formation, it also has direct effects on osteoblasts through endothelial cell-based BMP production. In addition to these pathways that classically have targeted bone production, there are also opportunities to target other aspects of the bone healing process such as inflammation, vascularization, and cell ingress to the fracture site. Bone regeneration is highly complex with defined, yet overlapping stages of healing. We will review established and novel extracellular signaling factors associated with various stages of fracture healing that could be targeted to promote enhanced bone regeneration. Importantly, multiple potential cell and tissues could be targeted to enhance healing in addition to focusing solely on osteoinductive therapeutics.

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1. Introduction

Bone heals robustly, and in most cases is capable of complete regeneration of normal structure and function; however, there are a various clinical scenarios where being able to accelerate or enhance bone formation would be advantageous. Approximately 10% of fractures do not heal, termed non-union, and many others show compromised regeneration in which there is delayed union or improper tissue restoration. The likelihood of delayed union and non-union is increased with severity of trauma, age, and underlying metabolic conditions.

Despite the clinical need for therapeutics to increase bone healing and regeneration, few signaling molecules have received regulatory approval for the promotion of bone regeneration. In part this paucity of biologics that are clinically available may reflect a lack of in-depth

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understanding of the cellular and molecular regulation of bone healing. However, over the past 10 years, our understanding of bone healing has greatly expanded due to an increased use of genetically modified mouse models and novel analysis techniques. With this new knowledge base, there is great opportunity for the development of a new generation of signaling molecules that can be used clinically to increase bone healing.

When considering the therapeutic targeting of bone regeneration, it is imperative to consider the different phases of healing (Fig. 1). Bone healing is a highly complex, temporally coordinated process (reviewed in Hankenson) [1]. When bone is injured, vascular disruption results in clot formation to provide hemostasis. Clotting is followed by a non-infectious inflammatory response in closed fractures. This inflammatory period is then followed by a fibrovascular phase of healing where blood vessels and mesenchymal stem/progenitor cells (MSCs) are recruited to the provisional callus. Many of these same recruited progenitors then develop into bone forming osteoblasts and chondrocytes, which will eventually lead to bone formation via endochondral and intramembranous ossification. Finally, the bone that has formed through the dual-mechanisms of endochondral and intramembranous ossification is remodeled through the process of osteoclast recruitment and formation of new mature intramembranous bone. This end stage of remodeling can be lengthy, and ultimately leads to restoration of prior structure and function, termed regeneration. A primary factor guiding this regeneration process is the mechanical load on the bone. It is feasible that targeting any of these temporal stages could lead to enhanced healing.

This review will consider both novel potential pathways for therapeutic targeting that have recently emerged and consider extracellular signaling factors that are currently being pursued for clinical application through translational and pre-clinical animal studies. Potential factors will be considered sequentially based on the temporal phase of fracture repair and bone regeneration (Fig. 1). Table 1 provides a reference guide to translational studies that target various extracellular signaling factors.

2. Extracellular signaling molecules during the inflammatory phase

Inflammation is the initial stage in fracture repair (Fig. 1). During this phase of healing, inflammatory cells are either deposited within the clot

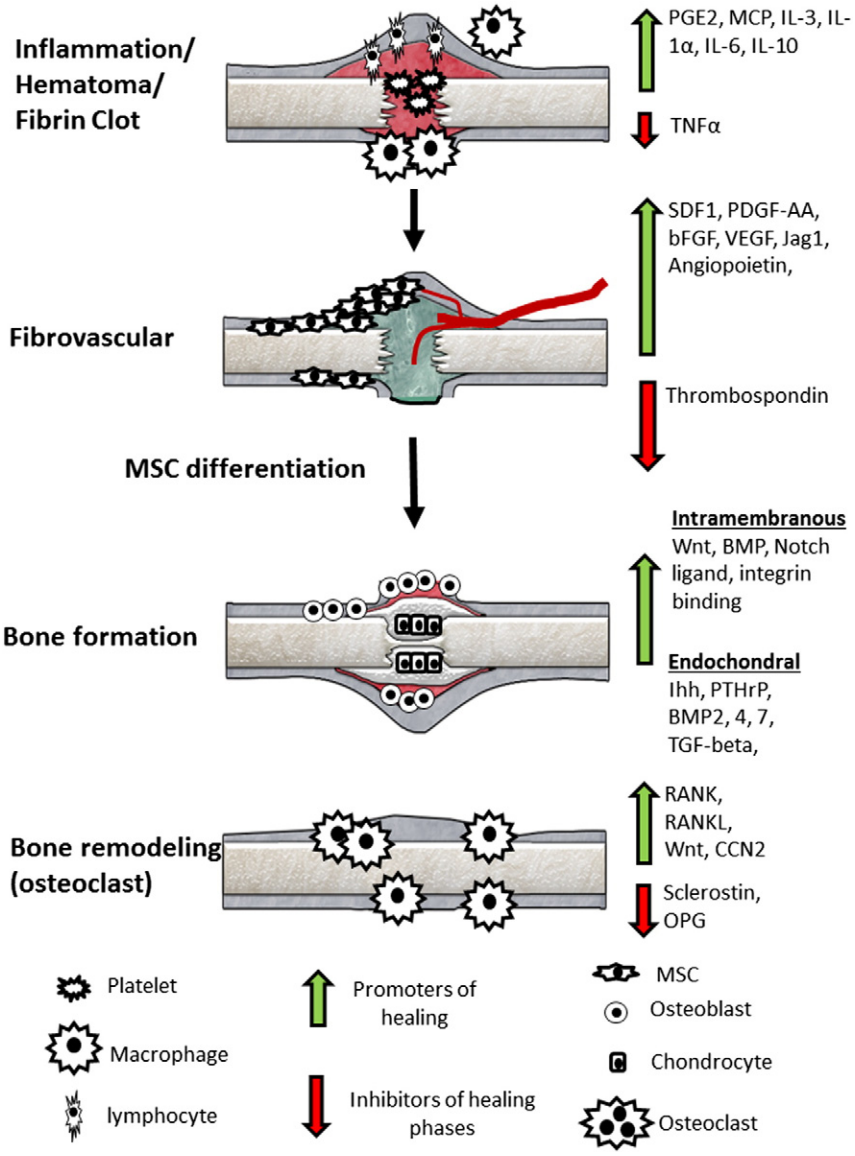

Fig. 1. Phases of fracture healing and positive and negative acting signaling factors during each phase. Arrows, green (positive acting) and red (negative acting), indicate signaling factors that are known to influence fracture healing or that could have a potential role on bone healing.