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Future Treatment Strategies for Delayed Bone Healing: An Osteoimmunologic Approach

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Bone is a remarkable organ because it retains the capacity to regenerate.[1] When successful, the healing of a fractured bone results in complete reconstitution of form and function. However, the healing process itself is quite complex and thus prone to failure. Even today, >10% of fracture patients in industrialized countries experience delayed or compromised healing. A high percentage of these patients are elderly persons prone to fracture and compromised healing. Specific treatment options to prevent delayed bone healing in such patients are still unavailable. Currently, patient fracture fixation and treatment are widely managed without specialized concepts for the elderly or for individual patient immune profiles.

Growing evidence on the interplay of the immune and skeletal systems during bone healing might offer new possibilities to identify patients at risk earlier and suggest new approaches aimed at personalized treatment, depending on the patient's immune status. Our understanding of the process of regenerative healing has shown that bone repair is a postnatal process that recapitulates embryologic processes of skeletal development.[2] Following injury, the healing process is initiated by an inflammatory reaction, with both the innate and adaptive systems as central mediators. The essential role of the immune system also has recently been recognized in all subsequent healing phases, catabolic and anabolic.[3,4]

The essential role of immune cells in bone healing predestined them as therapeutic targets. Because of the plethora of immune cells with differing functions during specific healing phases, the first priority was the definition of possible targets. Evaluation of immune cell composition in a small cohort of successfully healed versus delayed-healing patients (ranked by delayed return to full weight bearing and bone bridging, identified radiographically and on CT) revealed elevated levels of terminally differentiated CD8+ T cells (T_{EMRA}, or effector memory RA T cells). This immune cell subpopulation negatively influenced the bone healing process through cytokines, delaying the osteogenic differentiation of

mesenchymal stromal cells (in vitro).[5] Depletion of CD8+ T cells in a clinically relevant animal model led to a better healing outcome. The question is whether the adaptive immune system and its memory capacity is a naturally occurring compromise providing a strong defense against pathogens while simultaneously reducing the regenerative healing capacity, particularly because the immunologic memory is developed only in higher vertebrate organisms, and in these organisms, the regenerative capacity is lower—the opposite of that, for example, in a limb-regenerating newt. Aging significantly delays bone fracture healing, but the underlying mechanisms remain unknown. However, inflammaging (ie, chronic inflammation) has been proposed as underlying many of the changes associated with human aging.[6]

Using lethal irradiation followed by bone marrow transplantation, we replaced the immune system of elderly mice with that of juvenile mice. This rejuvenated the inflammatory system and accelerated the rate of bone fracture healing.[7] To assess the mechanism underlying the accelerated healing, we focused on the role of macrophages in bone healing. In young animals, blocking the ability of macrophages to go to the injury delays healing.[8] However, in old animals, blocking recruitment of inflammatory macrophages to sites of bone fracture stimulates repair. This apparent paradox suggests that removing the deleterious effects of proinflammatory macrophages may overcome some of the age-related decline in healing potential.

Immunomodulatory intervention to enhance bone healing could be achieved by either downregulating the immune cells that hinder regeneration or enhancing the immune cells that further the healing. Currently under discussion as candidates to accelerate healing are regulatory T cells, T helper 17 cells, or innate macrophages with an M2 phenotype. At the Orthopaedic Research Society symposium on osteoimmunology in bone regeneration this past March, we presented results illustrating the potential of these cells to enhance bone healing.[9] During the early stages of bone healing, using cytokines that direct the immune cells toward a regulatory phenotype, such as interleukin (IL)-4 and IL-3, successfully enhanced the healing.[3] These results not only confirm the feasibility of immunomodulatory strategies to enhance bone healing but also highlight the importance of understanding the inflammatory potential of an individual patient.

Age-related alterations on the tissue, matrix, cellular, subcellular, and signaling molecule levels, as well as their influence on the interaction of the immune and skeletal systems, will require further research to realize treatment approaches of immunomodulation to benefit patients in the future.[7]

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References

1. Schmidt-Bleek K, Petersen A, Dienelt A, Schwarz C, Duda GN. Initiation and early control of tissue regeneration – bone healing as a model system for tissue regeneration. Expert Opin Biol Ther. 2014; 14(2):247–259. [PubMed: 24397854]
2. Gerstenfeld LC, Cullinane DM, Barnes GL, Graves DT, Einhorn TA. Fracture healing as a post-natal developmental process: Molecular, spatial, and temporal aspects of its regulation. J Cell Biochem. 2003; 88(5):873–884. [PubMed: 12616527]
3. Schlundt C, El Khassawna T, Serra A, et al. Macrophages in bone fracture healing: Their essential role in endochondral ossification. Bone. 2015 S8756-3282(15)00392-0.
4. Könnecke I, Serra A, El Khassawna T, et al. T and B cells participate in bone repair by infiltrating the fracture callus in a two-wave fashion. Bone. 2014; 64:155–165. [PubMed: 24721700]
5. Reinke S, Geissler S, Taylor WR, et al. Terminally differentiated $CD8^+$ T cells negatively affect bone regeneration in humans. Sci Transl Med. 2013; 5(177):177ra36.
6. Lu C, Miclau T, Hu D, et al. Cellular basis for age-related changes in fracture repair. J Orthop Res. 2005; 23(6):1300–1307. [PubMed: 15936915]
7. Xing Z, Lu C, Hu D, Miclau T III, Marcucio RS. Rejuvenation of the inflammatory system stimulates fracture repair in aged mice. J Orthop Res. 2010; 28(8):1000–1006. [PubMed: 20108320]
8. Xing Z, Lu C, Hu D, et al. Multiple roles for CCR2 during fracture healing. Dis Model Mech. 2010; 3(7–8):451–458. [PubMed: 20354109]
9. Duda, GN.; Schmidt-Bleek, K. The good and the bad: Interdependency of bone and immune cells. Possibilities for future treatments?. http://www.ors.org/wp-content/uploads/2016/02/ORS-2016-PROGRAM-BOOK_FOR_WEB.pdf Accessed August 8, 2016