Principles of bone healing

Iain H. Kalfas, M.D., F.A.C.S.

Department of Neurosurgery, Section of Spinal Surgery, Cleveland Clinic Foundation, Cleveland, Ohio

Our contemporary understanding of bone healing has evolved due to knowledge gleaned from a continuous interaction between basic laboratory investigations and clinical observations following procedures to augment healing of fractures, osseous defects, and unstable joints. The stages of bone healing parallel the early stages of bone development. The bone healing process is greatly influenced by a variety of systemic and local factors. A thorough understanding of the basic science of bone healing as well as the many factors that can affect it is critical to the management of a variety of musculoskeletal disorders. In particular, the evolving management of spinal disorders can greatly benefit from the advancement of our understanding of the principles of bone healing.

Key Words • bone healing • spinal fusion • arthrodesis

Bone is a dynamic biological tissue composed of metabolically active cells that are integrated into a rigid framework. The healing potential of bone, whether in a fracture or fusion model, is influenced by a variety of biochemical, biomechanical, cellular, hormonal, and pathological mechanisms. A continuously occurring state of bone deposition, resorption, and remodeling facilitates the healing process.

The success of many spine operations depends on the restoration of long-term spinal stability. Whereas spinal instrumentation devices may provide temporary support, a solid osseous union must be achieved to provide permanent stability. The failure of fusion to occur may result in the fatigue and failure of supporting instrumentation and persistence or worsening of symptoms. Understanding the basic biological and physiological principles of bone transplantation and healing will aid the spine surgeon in selecting the most effective techniques to achieve successful fusions. In this paper the anatomical, histological and biological features of this process will be reviewed.

BONE ANATOMY AND HISTOLOGY

The cellular components of bone consist of osteogenic precursor cells, osteoblasts, osteoclasts, osteocytes, and the hematopoietic elements of bone marrow.[10,22] Osteoprogenitor cells are present on all nonresorptive bone surfaces, and they make up the deep layer of the periosteum, which invests the outer surface of bone, and the endosteum, which lines the internal medullary surfaces. The periosteum is a tough, vascular layer of connective tissue that covers the bone but not its articulating surfaces. The thick outer layer, termed the "fibrous layer," consists of irregular, dense connective tissue. A thinner, poorly defined inner layer called the "osteogenic layer" is made up of osteogenic cells. The endosteum is a single layer of osteogenic cells lacking a fibrous component.

Osteoblasts are mature, metabolically active, bone-forming cells. They secrete osteoid, the unmineralized organic matrix that subsequently undergoes mineralization, giving the bone its strength and rigidity. As their bone-forming activity nears completion, some osteoblasts are converted into osteocytes whereas others remain on the periosteal or endosteal surfaces of bone as lining cells. Osteoblasts also play a role in the activation of bone resorption by osteoclasts.

Osteocytes are mature osteoblasts trapped within the bone matrix. From each osteocyte a network of cytoplasmic processes extends through cylindrical canaliculi to blood vessels and other osteocytes. These cells are involved in the control of extracellular concentration of calcium and phosphorus, as well as in adaptive remodeling behavior via cell-to-cell interactions in response to local environment.

Osteoclasts are multinucleated, bone-resorbing cells controlled by hormonal and cellular mechanisms. These cells function in groups termed "cutting cones" that attach to bare bone surfaces and, by releasing hydrolytic enzymes, dissolve the inorganic and organic matrices of bone and calcified cartilage. This process results in the formation of shallow erosive pits on the bone surface called Howship lacunae.[12]

There are three primary types of bone: woven bone, cortical bone, and cancellous bone.[10,22] Woven bone is found during embryonic development, during fracture healing (callus formation), and in some pathological states such as hyperparathyroidism and Paget disease.[22] It is composed of randomly arranged collagen bundles and ir-

regularly shaped vascular spaces lined with osteoblasts. Woven bone is normally remodeled and replaced with cortical or cancellous bone.

Cortical bone, also called compact or lamellar bone, is remodeled from woven bone by means of vascular channels that invade the embryonic bone from its periosteal and endosteal surfaces. It forms the internal and external tables of flat bones and the external surfaces of long bones. The primary structural unit of cortical bone is an osteon, also known as a haversian system. Osteons consist of cylindrical shaped lamellar bone that surrounds longitudinally oriented vascular channels called haversian canals. Horizontally oriented canals (Volkmann canals) connect adjacent osteons. The mechanical strength of cortical bone depends on the tight packing of the osteons.

Cancellous bone (trabecular bone) lies between cortical bone surfaces and consists of a network of honeycombed interstices containing hematopoietic elements and bony trabeculae. The trabeculae are predominantly oriented perpendicular to external forces to provide structural support.[16,29] Cancellous bone is continually undergoing remodeling on the internal endosteal surfaces.

BONE BIOCHEMISTRY

Bone is composed of organic and inorganic elements. By weight, bone is approximately 20% water.[22] The weight of dry bone is made up of inorganic calcium phosphate (65–70% of the weight) and an organic matrix of fibrous protein and collagen (30–35% of the weight).[10,19,21,22]

Osteoid is the unmineralized organic matrix secreted by osteoblasts. It is composed of 90% type I collagen and 10% ground substance, which consists of noncollagenous proteins, glycoproteins, proteoglycans, peptides, carbohydrates, and lipids.[20,22] The mineralization of osteoid by inorganic mineral salts provides bone with its strength and rigidity.

The inorganic content of bone consists primarily of calcium phosphate and calcium carbonate, with small quantities of magnesium, fluoride, and sodium. The mineral crystals form hydroxyapatite, which precipitates in an orderly arrangement around the collagen fibers of the osteoid. The initial calcification of osteoid typically occurs within a few days of secretion but is completed over the course of several months.

REGULATORS OF BONE METABOLISM

Bone metabolism is under constant regulation by a host of hormonal and local factors. Three of the calcitropic hormones that most affect bone metabolism are parathyroid hormone, vitamin D, and calcitonin. Parathyroid hormone increases the flow of calcium into the calcium pool and maintains the body's extracelluar calcium levels at a relatively constant level. Osteoblasts are the only bone cells that have parathyroid hormone receptors. This hormone can induce cytoskeletal changes in osteoblasts. Vitamin D stimulates intestinal and renal calcium-binding proteins and facilitates active calcium transport. Calcitonin is secreted by the parafollicular cells of the thyroid gland in response to an acutely rising plasma calcium level. Calcitonin serves to inhibit calcium-dependent cellular metabolic activity.

Bone metabolism is also affected by a series of proteins, or growth factors, released from platelets, macrophages, and fibroblasts. These proteins cause healing bone to vascularize, solidify, incorporate, and function mechanically. They can induce mesenchymal-derived cells, such as monocytes and fibroblasts, to migrate, proliferate, and differentiate into bone cells. The proteins that enhance bone healing include the BMPs, insulin-like growth factors, transforming growth factors, platelet derived growth factor, and fibroblast growth factor among others.[18,32]

The most well known of these proteins are the BMPs, a family of glycoproteins derived from bone matrix. Bone morphogenetic proteins induce mesenchymal cells to differentiate into bone cells. Although typically present in only minute quantities in the body, several BMPs have been synthesized using recombinant DNA technology and are currently undergoing clinical trials to assess their potential to facilitate bone fusion in humans.[26–28]

Other proteins influence bone healing in different ways. Transforming growth factor–β regulates angiogenesis, bone formation, extracellular matrix synthesis, and controls cell-mediated activities. Osteonectin, fibronectin, osteonectin, and osteocalcin promote cell attachment, facilitate cell migration, and activate cells.[15,20,22]

PHYSIOLOGY OF BONE REPAIR AND FUSION

The use of a bone graft for purposes of achieving arthrodesis is affected by each of the aforementioned anatomical, histological, and biochemical principles. Additionally, several physiological properties of bone grafts directly affect the success or failure of graft incorporation. These properties are osteogenesis, osteoinduction, and osteoconduction.[20]

Osteogenesis is the ability of the graft to produce new bone, and this process is dependent on the presence of live bone cells in the graft. Osteogenic graft materials contain viable cells with the ability to form bone (osteoprogenitor cells) or the potential to differentiate into bone-forming cells (inducible osteogenic precursor cells). These cells, which participate in the early stages of the healing process to unite the graft with the host bone, must be protected during the grafting procedure to ensure viability. Osteogenesis is a property found only in fresh autogenous bone and in bone marrow cells, although the authors of radiolabeling studies of graft cells have shown that very few of these transplanted cells survive.[19]

Osteoconduction is the physical property of the graft to serve as a scaffold for viable bone healing. Osteoconduction allows for the ingrowth of neovasculature and the infiltration of osteogenic precursor cells into the graft site. Osteoconductive properties are found in cancellous autografts and allografts, demineralized bone matrix, hydroxyapatite, collagen, and calcium phosphate.[19]

Osteoinduction is the ability of graft material to induce stem cells to differentiate into mature bone cells. This process is typically associated with the presence of bone growth factors within the graft material or as a supplement to the bone graft. Bone morphogenic proteins and demineralized bone matrix are the principal osteoinductive materials. To a much lesser degree, autograft and allograft bone also have some osteoinductive properties.[19]

BONE HEALING PROCESS

The process of bone graft incorporation in a spinal fusion model is similar to the bone healing process that occurs in fractured long bones.[4] Fracture healing restores the tissue to its original physical and mechanical properties and is influenced by a variety of systemic and local factors. Healing occurs in three distinct but overlapping stages: 1) the early inflammatory stage; 2) the repair stage; and 3) the late remodeling stage.[9,13]

In the inflammatory stage, a hematoma develops within the fracture site during the first few hours and days. Inflammatory cells (macrophages, monocytes, lymphocytes, and polymorphonuclear cells) and fibroblasts infiltrate the bone under prostaglandin mediation. This results in the formation of granulation tissue, ingrowth of vascular tissue, and migration of mesenchymal cells. The primary nutrient and oxygen supply of this early process is provided by the exposed cancellous bone and muscle. The use of antiinflammatory or cytotoxic medication during this 1st week may alter the inflammatory response and inhibit bone healing.

During the repair stage, fibroblasts begin to lay down a stroma that helps support vascular ingrowth. It is during this stage that the presence of nicotine in the system can inhibit this capillary ingrowth.[11,23–25] A significantly decreased union rate had been consistently demonstrated in tobacco abusers.[2,3,6]

As vascular ingrowth progresses, a collagen matrix is laid down while osteoid is secreted and subsequently mineralized, which leads to the formation of a soft callus around the repair site. In terms of resistance to movement, this callus is very weak in the first 4 to 6 weeks of the healing process and requires adequate protection in the form of bracing or internal fixation. Eventually, the callus ossifies, forming a bridge of woven bone between the fracture fragments. Alternatively, if proper immobilization is not used, ossification of the callus may not occur, and an unstable fibrous union may develop instead.

Fracture healing is completed during the remodeling stage in which the healing bone is restored to its original shape, structure, and mechanical strength. Remodeling of the bone occurs slowly over months to years and is facilitated by mechanical stress placed on the bone. As the fracture site is exposed to an axial loading force, bone is generally laid down where it is needed and resorbed from where it is not needed. Adequate strength is typically achieved in 3 to 6 months.

Although the physiological stages of bone repair in the spinal fusion model are similar to those that occur in long bone fractures, there are some differences. Unlike long bone fractures, bone grafts are used in spinal fusion procedures. During the spinal fusion healing process, bone grafts are incorporated by an integrated process in which old necrotic bone is slowly resorbed and simultaneously replaced with new viable bone. This incorporation process is termed "creeping substitution."[17,20] Primitive mesenchymal cells differentiate into osteoblasts that deposit osteoid around cores of necrotic bone. This process of bone deposition and remodeling eventually results in the replacement of necrotic bone within the graft.

The most critical period of bone healing is the first 1 to 2 weeks in which inflammation and revascularization occur. The incorporation and remodeling of a bone graft require that mesenchymal cells have vascular access to the graft to differentiate into osteoblasts and osteoclasts. A variety of systemic factors can inhibit bone healing, including cigarette smoking, malnutrition, diabetes, rheumatoid arthritis, and osteoporosis. In particular, during the 1st week of bone healing, steroid medications, cytotoxic agents, and nonsteroidal antiinflammatory medications can have harmful effects. Irradiation of the fusion site within the first 2 to 3 weeks can inhibit cell proliferation and induce an acute vasculitis that significantly compromises bone healing (SE Emery, unpublished data).

Bone grafts are also strongly influenced by local mechanical forces during the remodeling stage. The density, geometry, thickness, and trabecular orientation of bone can change depending on the mechanical demands of the graft. In 1892, Wolff first popularized the concept of structural adaptation of bone, noting that bone placed under compressive or tensile stress is remodeled. Bone is formed where stresses require its presence and resorbed where stresses do not require it.[22,31] This serves to optimize the structural strength of the graft. Conversely, if the graft is significantly shielded from mechanical stresses, as in the case of rigid spinal implants, excessive bone resorption can potentially occur and result in a weakening of the graft. This potential disadvantage of instrumentation needs to be balanced with the beneficial effects that spinal fixation has on the fusion process.

BONE GRAFTS

The two types of bone grafts frequently used in spinal fusion are autografts and allografts. Autograft bone is transplanted from another part of the recipient's body. Allograft bone is transplanted from genetically nonidentical members of the same species. Both types of bone grafts are commonly used in spine surgery.

The ideal bone graft should be: 1) osteoinductive and conductive; 2) biomechanically stable; 3) disease free; and 4) contain minimal antigenic factors. These features are all present with autograft bone. The disadvantages of autografts include the need for a separate incision for harvesting, increased operating time and blood loss, the risk of donor-site complications, and the frequent insufficient quantity of bone graft.[13,30]

The advantage of allograft bone is that it avoids the morbidity associated with donor-site complications and is readily available in the desired configuration and quantity. The disadvantages of allograft include delayed vascular penetration, slow bone formation, accelerated bone resorption, and delayed or incomplete graft incorporation.[1,5,14,16] In general, allograft bone has a higher incidence of nonunion or delayed union than autograft.[1,7–9,15,33] Allografts are osteoconductive but are only weakly osteoinductive. Although transmission of infection and lack of histocompatibility are potential problems with allograft bone, improved tissue-banking standards have greatly reduced their incidence.

Bone grafts can also be classified according to their structural anatomy: cortical or cancellous. Cortical bone has fewer osteoblasts and osteocytes, less surface area per unit weight, and contributes a barrier to vascular ingrowth

and remodeling compared with cancellous bone. The advantage of cortical bone is its superior structural strength.

The initial remodeling response to cortical bone is resorptive as osteoclastic activity predominates. Cortical grafts progressively weaken with time because of this bone resorption as well as slow, incomplete remodeling. Conversely, cancellous bone becomes progressively stronger because of its ability to induce early, rapid, new bone formation.

When selecting a bone graft, the spine surgeon needs to consider the specific structural and biological demands that will be placed on the graft. If the graft is placed anteriorly in a compressive mode, cortical bone, either autogenic or allogenic, will be required. If placed posteriorly as a graft under tension with lower demands for structural support but also a lower probability of early vascular ingrowth, a cancellous autograft is preferred.

CONCLUSIONS

An understanding of the basic science of bone healing is critical to the consistent success of spinal fusion surgery. Although great advances have been made in the field of spinal instrumentation, it is only a solid osseous union that will ensure long-term spinal stability. Selection of the most appropriate bone graft material as well as careful attention to the principles of bone healing can greatly facilitate the potential for clinical success.

References

1. Aurori BF, Weierman RJ, Lowell HA, et al: Pseudoarthrosis after spinal fusion for scoliosis. A comparison of autogenic and allogenic bone grafts. **Clin Orthop 199:**153–158, 1985
2. Bishop RC, Moore KA, Hadley MN: Anterior cervical interbody fusion using autogeneic and allogeneic bone graft substrate: a prospective comparative analysis. **J Neurosurg 85:**206–210, 1996
3. Blumenthal SL, Baker J, Dossett A, et al: The role of anterior lumbar fusion for internal disc disruption. **Spine 13:**566–569, 1988
4. Boden SD, Schimandle JH, Hutton WC: An experimental intertransverse process spinal fusion model. Radiographic, histologic, and biomechanical healing characteristics. **Spine 20:**412–420, 1995
5. Brooks DB, Heiple KG, Herndon CH, et al: Immunological factors in homogenous bone transplantation. IV. The effect of various methods of preparation and irradiation on antigenicity. **J Bone Joint Surg Am 45:**1617–1626, 1963
6. Brown CW, Orme TJ, Richardson HD: The rate of pseudarthrosis (surgical nonunion) in patients who are smokers and patients who are nonsmokers: a comparison study. **Spine 11:**942–943, 1986
7. Bucholz RW, Carlton A, Holmes RE: Hydroxyapatite and tricalcium phosphate bone graft substitutes. **Orthop Clin North Am 18:**323–334, 1987
8. Burchardt H: Biology of bone transplantation. **Orthop Clin North Am 18:**187–196, 1987
9. Burchardt H, Enneking WF: Transplantation of bone. **Surg Clin North Am 58:**403–427, 1978
10. Copenhaver WM, Kelly DE, Wood RL: The connective tissues: cartilage and bone, in Copenhaver WM, Kelly DE, Wood RL (eds): **Bailey's Textbook of Histology, ed 17.** Baltimore: Williams & Wilkins, 1978, pp 170–205
11. Daftari TK, Whitesides TE Jr, Heller JG, et al: Nicotine on the revascularization of bone graft. An experimental study in rabbits. **Spine 19:**904–911, 1994
12. Dee R: Bone healing, in Dee R, Mango E, Hurst E, (eds): **Principles of Orthopaedic Practice.** New York: McGraw-Hill, 1988, pp 68–73
13. DePalma AF, Rothman RH, Lewinnek GE, et al: Anterior interbody fusion for severe cervical disc degeneration. **Surg Gynecol Obstet 134:**755–758, 1972
14. Friedlaender GE, Strong DM, Sell KW: Studies on the antigenicity of bone. II. Donor-specific anti-HLA antibodies in human recipients of freeze-dried allografts. **J Bone Joint Surg Am 66:**107–112, 1984
15. Habal MB: Different forms of bone grafts, in Habal MB, Reddi AH (eds): **Bone Grafts and Bone Substitutes.** Philadelphia: WB Saunders, 1992, pp 6–8
16. Herron LD, Newman MH: The failure of ethylene oxide gas-sterilized freeze-dried bone graft for thoracic and lumbar spinal fusion. **Spine 14:**496–500, 1989
17. Kaufman HH, Jones E: The principles of bony spinal fusion. **Neurosurgery 24:**264–270, 1989
18. Lane JM, Muschler GF, Kurz LT, et al: Spinal fusion, in Rothman RH, Simeone FA (eds): **The Spine.** Philadelphia: WB Saunders, 1992, pp 1739–1755
19. Muschler GF, Lane JM, Dawson EG: The biology of spinal fusion, in Cotler JM, Cotler HP (eds): **Spinal Fusion Science and Technique.** Berlin: Springer-Verlag, 1990, pp 9–21
20. Prolo DJ: Biology of bone fusion. **Clin Neurosurg 36:**135–146, 1990
21. Prolo DJ, Pedrotti PW, White DH: Ethylene oxide sterilization of bone, dura mater, and fascia lata for human transplantation. **Neurosurgery 6:**529–539, 1980
22. Recker RR: Embryology, anatomy, and microstructure of bone, in Coe FL, Favus MJ (eds): **Disorders of Bone and Mineral Metabolism.** New York: Raven, 1992, pp 219–240
23. Riebel ED, Boden SD, Whitesides TE, et al: The effect of nicotine on incorporation of cancellous bone graft in an animal model. **Spine 20:**2198–2202, 1995
24. Rubenstein I, Yong T, Rennard SI, et al: Cigarette smoke extract attenuates endothelium-dependent arteriolar dilation in vivo. **Am J Physiol 261:**H1913–H1918, 1991
25. Silcox DH III, Daftari T, Boden SD, et al: The effects of nicotine on spinal fusion. **Spine 20:**1549–1553, 1995
26. Urist MR: Bone transplants and implants, in Urist MR (ed): **Fundamentals and Clinical Bone Physiology.** Philadelphia: JB Lippincott, 1980, pp 331–368
27. Urist MR, Dawson E: Intertransverse process fusion with the aid of chemosterilized autolyzed antigen–extracted allogeneic (AAA) bone. **Clin Orthop 154:**97–113, 1981
28. Urist MR, Mikulski A, Lietze A: Solubilized and insolubilized bone morphogenetic protein. **Proc Natl Acad Sci USA 76:**1828–1832, 1979
29. White AA III, Hirsch C: An experimental study of the immediate load bearing capacity of some commonly used iliac bone grafts. **Acta Orthop Scand 42:**482–490, 1971.
30. Whitecloud TS III. Complications of anterior cervical fusion. **Instr Course Lect 27:**223–227, 1976
31. Wolff J: **The Law of Bone Remodelling.** Translated by Maquet P, Furlong R. Berlin: Springer-Verlag, 1986
32. Wozney JM, Rosen V, Celeste AJ, et al: Novel regulators of bone formation: molecular clones and activities. **Science 242:**1528–1534, 1988
33. Zdeblick TA, Ducker TB: The use of freeze-dried allograft bone for anterior cervical fusions. **Spine 16:**726–729, 1991