3.

Bone Allograft Safety and Performance

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3.1 Introduction

Bone allograft transplantation is a common practice; in the United States 650,000 procedures were performed in 1999, a 186% increase from 1990 [3]. This increase can be attributed to morbidities associated with bone autografts [6, 18, 30, 35, 59], the increased availability of bone allografts, and the expansion of these applications [9, 16, 21, 22, 29, 31, 42, 66]. A variety of musculoskeletal allografts are available for different reconstructive applications. Bone allograft is an alternative to autograft because it has osteoconductive properties, acts as a scaffold for bone growth, and induces bone formation by providing osteogenic factors, in addition to mesenchymal precursor cells, osteoblasts, and osteocytes. Although these properties are advantageous, the potential for the transmission of infectious diseases remains a great concern [1, 2, 4, 10, 12, 24, 26, 27, 32, 38, 49, 53]. Because of the biological origin of bone allografts, the clinician must be educated about the effects of tissue preparation and processing on the immunogenic, osteoinductive, osteoconductive, and structural properties of allografts in order to make appropriate clinical decisions. This chapter discusses the safety of bone allografts and the effects of donor selection, harvesting, processing, and implantation on the performance of bone allograft in reconstructive surgery.

3.2 An Overview of Musculoskeletal Graft Harvesting and Processing

In the United States, the Food and Drug Administration (FDA) currently regulates organ and tissue transplants with mandated donor and tissue screening protocols for human immunodeficiency virus (HIV) types 1 and 2, hepatitis B virus (HBV), and hepatitis C virus (HCV) (Table 3.1). The FDA also requires documentation to accompany the donor graft to provide a medical history that precludes any recent infections or patient "social" habits, such as drug abuse, which would increase the risk of allograft infection. In addition, the American Association of Tissue Banks (AATB), a nonprofit organization, provides industry guidelines and recommendations for its accredited members beyond those of the FDA, which include testing for human T-lymphocytic virus (HTLV) types 1 and 2 and syphilis [67] (Table 3.1). However, there are no uniform industry standards for tissue processing, and not all tissue banks are AATB-accredited. Medical conditions contraindicated by the FDA and AATB for tissue and organ donation include benign tumors near the allograft excision sites, malignant tumors, autoimmune or inflammatory diseases, severe endocrine/metabolic disease, and collagen diseases [22, 29, 36, 42, 62, 63]. Additional contra-

indications for donations include deaths resulting from trauma with large resuscitation volumes, with or without blood or blood products, and deaths resulting from poisoning or related to toxic overdoses [67].

Upon identification and screening of an acceptable donor, appropriate consent must be obtained from the donor or nearest relative prior to tissue and/or organ procurement. Musculoskeletal allografts may be obtained from living donors, multiorgan donors, and cadavers. Harvesting of a musculoskeletal allograft from a living donor (such as a femoral head allograft harvested from a total hip replacement) is performed in a sterile operating room, as is harvesting from a multiorgan donor. Cadaveric musculoskeletal tissues must be procured within 24 hours of death, with the time interval between death and refrigeration not to exceed twelve hours. Harvesting of a musculoskeletal graft from a cadaver is performed in an approved, aseptic environment. Musculoskeletal allografts can be categorized as (1) bone with soft-tissue attachments (such as a bone-patellar tendon-bone allograft), (2) bone devoid of soft-tissue attachments (such as a femoral head), or (3) an isolated soft-tissue allograft (such as a meniscus). After the tissue is harvested, the donor serum and allograft are cultured for microbial contamination. The allograft is then cleaned, soaked in an antiseptic solution such as BioCleanse (Regeneration Technologies, Alachua, FL), and irrigated with or without pressurized lavage or by ultrasonic/mechanical cleansing techniques. The allograft is then frozen and may be terminally sterilized (described below). In some cases, freezing is replaced by cryopreservation techniques to retain cell viability and possible osteogenic ability.

Freezing cannot substitute for sterilization and at best may only prevent bacteria, fungi, spores, or viruses from growing. As a result, some tissue banks perform terminal bactericidal and virucidal sterilization that includes heating, gamma-irradiation, chemical sterilization, and lyophilization. These procedures further reduce the risk of infection and allogenic response by musculoskeletal tissues. Some tissue banks routinely "pasteurize" or autoclave allografts [36, 42]; the resulting increase in temperature eliminates the biological activity of the cells, but may decrease the strength of the grafts as a result of the denaturation of structural proteins [7]. In addition, heat sterilization may not inactivate bacterial spores [27]. Gamma-irradiation at the level of 1.5 to 2.5 megarads or above [8, 13, 36, 42, 45, 62] is believed to inactivate bacterial contaminants and HCV, but not HIV [20, 45, 47]. Gamma-irradiation, moreover, weakens musculoskeletal allografts [14, 44]. Lyophilization, i.e., freeze-drying, is a process by which water is removed from the tissue to the point where cellular activity is no longer supported. This process involves partially freezing the tissues to allow sublimation of water, followed by further drying with the aid of other techniques. As a result, HIV and HCV are inactivated and the risk of transmission is minimized in the infected blood products and bone marrow [53]. This technique may, however, reduce the strength of the musculoskeletal allografts [14, 44]. With proper storage, freeze-dried allografts retain biological activity for several years.

Chemical sterilization with proprietary solutions or ethylene oxide has also been used for terminal sterilization. Adverse reactions, such as moderate inflammation from residual ethylene oxide in the allograft, have been reported [8, 54, 60, 62]. Proprietary solutions may contain particular bactericidal, virucidal, and fungicidal agents, but there is no industry-wide standard for their usage.

Allogenic bone can be machined and separated into cortical, corticocancellous, and cancellous preparations. Cortical and corticocancellous allografts are used for structural support and have limited osteoconductive capability, with no osteoinductive properties. Cortical and corticocancellous bone grafts undergo slow resorption in the host secondary to limited vascular invasion; this decreases the structural properties of the graft. The cortical/corticocancellous allograft is incorporated by the host through creeping substitution in conjunction with slow bone remodeling. These grafts are available in several forms: morsellized "bone chips," short segments of diaphyseal rings from femora or tibiae, iliac crest bone wedges, cortical struts, and whole bones en bloc, such as a fibula. Large areas of nonincorporated necrotic bone often remain in a patient for years after implantation. Cancellous allografts provide limited structural support and osteoconductivity that can be enhanced with demineralization. In the course of bone remodeling, cancellous allografts

Table 3.1. Graft donor infectious pathogens screened

Mandated by FDA
Human immunodeficiency virus (HIV) types 1 and 2
Hepatitis B virus (HBV)
Hepatitis C virus (HCV)
Additional AATB screening
Human T-lymphocytic virus (HTLV) types 1 and 2
Syphilis

are resorbed more quickly than cortical grafts and are typically available as small, porous, spongy blocks that are used to fill segmental bone defects.

After terminal sterilization, bone allografts can be demineralized to make osteoinductive biological molecules, such as bone morphogenetic proteins (BMPs), more readily available to augment new bone formation [17, 37, 50]. The demineralization process is thought to destroy the antigenic surface of the bone graft, which reduces the host immune response. Like gamma-irradiation and lyophilization, the demineralization process weakens musculoskeletal allografts [14, 44]. Thus, choosing an appropriate allograft becomes critical when the primary requirement is structural augmentation.

Quality control of tissue banks is maintained through documentation and periodic audits of stored allografts. Some tissue banks routinely test stored tissues as new laboratory methods become available [12]. These periodic audits increase the chance of detection of potential cases of HIV transmission and/or epidemiological exposures to other previously undetected infections.

3.3 Infection from Musculoskeletal Transplants

Musculoskeletal transplantation is a safe, comprehensively regulated practice with a low incidence of infections, especially in light of its substantial usage in reconstructive procedures. However, the risk of potentially fatal complications from infectious transmission does exist. The literature describes many cases of contamination with HIV [2, 32, 53], HCV [1, 12], *Clostridium* species [4, 27, 38], and other bacteria [4, 24, 26, 65], and viruses in transplants procured from acceptable donors.

HIV infection is one of the most serious risks associated with allograft transplantation. There is currently no cure or vaccine for this lifelong, disabling disease. With proper donor screening and HIV antibody and antigen testing, the estimated risk of HIV transmission in musculoskeletal transplantation is 1 in 150,471 and can be reduced to 1 in 1.67 million with lymph node testing, serology, and checking for complications associated with grafts from the same donor [10]. The risk of infection following allograft transplantation is comparable to the risk of HIV infection from screened whole red blood cell transfusion; it is thought to be between 1 in 250,000 and 1 in 2,000,000 [10]. Between 1988 and 1992, four cases of HIV transmission were reported resulting from procedures that utilized fresh-frozen bone allografts in 1984 and 1985, which were traced to two donors [2, 53]. These investigations were initiated after the allograft recipients, whose only risk for HIV was transplantation, were found to be positive for HIV several years later. Other infected allograft recipients were then identified through analysis of banked tissue. The donors of these tissues were screened for HIV and tested negative. It is believed that the infection occurred during an early stage when HIV antibodies were not yet detectable. In another case, a fresh-frozen bone allograft was implanted that had been subjected to extensive intramedullary reaming prior to implantation and did not test positive, yet became the source of the HIV infection. Conceivably, the removal of blood and bone marrow from the allograft prior to implantation cleared infectious cells from the tissue and thus led to a negative test result [53]. To date, there have been no reports of HIV transmission from musculoskeletal allografts obtained from seronegative donors that were subjected to freeze-drying or other terminal sterilization methods [8, 32, 47, 48, 53, 62, 63, 64]. Since then, tests have been developed for other markers of HIV, including the p24 antigen assay and the use of the polymerase chain reaction (PCR) [20, 32, 47, 48, 53, 62, 63].

Hepatitis C is a chronic hepatic disease that for several years after infection may exhibit no clinical signs or symptoms, yet ultimately lead to severe morbidity and mortality. There is no cure or vaccine for hepatitis C. Nine cases

of HCV transmission by musculoskeletal allografts from three donors were reported in the United States between 1995 and 2003 [1, 12]. These donors had negative medical and social histories and initially tested negative for HCV when subjected to an anti-HCV immunoassay. In these cases, previously undetected HCV was identified from retrospective testing of tissue and sera with newer anti-HCV immunoassays and PCR analysis. After the donor tissues had been identified, a protocol was initiated to inform and test all recipients of tissues or organs from these donors. Interestingly, this study reported that when the high-risk seroconverted individual was excluded, all recipients of minimally processed allografts seroconverted for HCV. However, recipients of irradiated tissue that had been freeze-dried, frozen or cryopreserved did not test positive for HCV infection [12]. In 2002, the Centers for Disease Control (CDC) reported four cases of HCV transmission that resulted from a screened donor of bone-patellar-bone and tendon allografts [1]. The CDC investigation was prompted by the fact that acute hepatitis C was diagnosed 6 weeks after a recipient received a bone-patellar-bone allograft. Further testing with an anti-HCV immunoassay showed that the donor serum was negative for the HCV protein, but PCR analysis showed a positive result for HCV mRNA. Testing of the other recipients of the infected allografts revealed no cases of HCV transmission if the bone allografts had undergone gamma irradiation [1, 12]. Gamma-irradiation of musculoskeletal allografts would therefore appear to reduce the risk of HCV transmission from infected tissues.

Studies of bacterial infection or contamination of musculoskeletal allografts have shown that most of the allograft contamination is due to *Staphylococcus* and other mixed skin flora [4, 8, 24, 26, 33, 65]. Kainer et al. [27] identified 14 cases of infection by *Clostridium* species that they traced to nine donors. The time between death and tissue procurement in two of the nine donors exceeded industry standards. The 14 infected patients had received nine frozen bone-patellar tendon-bone allografts, four fresh femoral condyles, and one meniscus graft. All of the processed allograft tissues from the 14 identified cases came from one tissue bank, and the unprocessed donor tissues originated from seven other tissue banks. The tissue banks that provided the allografts to the recipients had procured the tissues using aseptic techniques that included decontamination by suspension in a proprietary antibiotic solution, but did they did not employ terminal sterilization. However, when terminal sterilization was performed, whether by gamma-irradiation or by low temperature, or if chemical sterilization had been employed at other tissue banks, the resulting allografts from five of the nine identified donors did not induce infection. Even though the overall rate of *Clostridium* infection was less than 0.5% among recipients of allografts from the tissue bank that reported *Clostridium* infection, this rate was still significantly higher than the rate among recipients of allografts from the tissue banks with no *Clostridium* infection [27].

An additional means of reducing the risk of contamination involves harvesting the tissues in an operating room with sterile techniques [26, 65]. The degree of bacterial contamination

Table 3.2. Factors influencing allograft performance

Factor	Implication
Graft donor age	Osteoinductive potential is greater from donors aged 42 years and younger. Mechanical properties of allograft bone are inversely proportional to donor age after the fifth decade.
Presence of osteoporosis or osteopenia	Osteoporotic and osteopenic bone have decreased mechanical properties. According to histologic appearance, the incidence of osteoporosis is higher in donors after the fifth decade of life.
Graft anatomic origin	Fibular strut grafts are stronger than femoral ring or tibial grafts. Iliac crest grafts from the anterior iliac spine are stronger than those from the posterior iliac spine.
Tissue processing	Gamma-irradiation of ≥3.0 megarads (virucidal levels) reduces mechanical properties. Lyophilization can also weaken allografts. Pasteurization may also decrease the mechanical strength of allograft bone.

of a musculoskeletal allograft is a direct function of the time that elapses between death and refrigeration [38, 65]. Tissues obtained from living donors have lower rates of bacterial contamination than tissues harvested from cadavers at autopsy [26, 65]. Musculoskeletal allografts from donors who suffered multiple trauma, with or without resuscitation, had higher rates of bacterial contamination than allografts from organ donors [65]. These observations are best explained by the fact that as postmortem time increases, the risk of infection by intestinal flora such as *Clostridium* and *Escherichia* species also increases [27]. This is particularly true for spore-forming bacteria such as *Clostridium* that are capable of long dormancy. As with surgical infection rates, the rate of allograft contamination is directly proportional to the number of persons present in the operating room during procurement [65]. The order in which tissues are harvested also affects the rate of bacterial contamination; the rate is higher in specimens from the hemipelvis than in specimens from the femur or tibia [26], probably because the hemipelvis is typically the last large structural bone to be harvested. Prolonged handling of the skin also increases the risk of contamination [26]. The risk of contamination can be reduced by antiseptic soaking, irrigation, and terminal sterilization [24].

3.4 Donor Selection Factors Affecting Musculoskeletal Allograft Performance

All potential allograft transplant donors are screened for a variety of factors, including but not limited to sex, age, cause of death, and past medical and social history; the results of serological tests for medical diseases; and, most importantly, the presence of bacterial and viral pathogens. The most commonly reported exclusion factors for tissue donors include a medical history of infection at the excision sites, benign or malignant tumors at the excision sites, autoimmune diseases, severe endocrine/metabolic diseases, collagen diseases, and infection by HIV, HCV, and/or HBV. Age, sex, medical history, and the type of bone harvested from screened tissue donors have been evaluated for their effects on osteoinductive potential and structural support of the allografts (Table 3.2).

Increased donor age may be inversely related to the osteoinductive potential of bone allografts. Using an in vivo nude murine model, Schwartz et al. [50] reported that an increase in donor age decreased the osteoinductivity of the demineralized, freeze-dried bone allograft (DFDBA). Areas of new bone formation, new cortical bone function, and new bone-marrow production were smaller in allografts obtained from older donors (>50 years) than in allografts obtained from younger donors (<29 years). Osteoconductivity was not affected by donor gender. Lohman et al. [37] confirmed the age-dependent effect by noting that allograft osteoinductive potential was significantly greater for donors under 42 than for donors over 70 years of age.

Several studies have shown that the mechanical properties of bone decline with age. Burnstein et al., using cadaveric human specimens, observed a highly significant negative correlation between age and femoral yield stress, ultimate stress, elastic modulus, and ultimate strain [11]. Smith et al. [55] observed a negative correlation between the tensile stress of bone and age in vivo. McCalden et al. demonstrated that there is an inverse relationship between the mechanical properties of cortical bone and age, and theorized that the decrease in bone strength is the result of an age-dependent increase in bone porosity [39].

Allografts from donors with osteoporosis or osteopenia, conditions that are not contraindicated for bone transplant donation, may have less strength and stiffness [17]. Dickenson et al. [15] reported a significant decrease in the modulus of elasticity, the ultimate tensile strength, and the amount of plastic and elastic energy absorbed in osteoporotic bone in comparison with nonosteoporotic bone in vitro. They also theorized that the decrease in strength and stiffness in osteoporotic bone grafts was due to greater porosity. In vivo, Lill et al. observed a significant reduction in the bending stiffness of intact osteoporotic tibiae in comparison with normal tibiae, as well as delayed fracture healing in osteoporotic bone [34].

Histologic evaluation of bone allografts has shown that osteoporosis and osteopenia affect bone allograft performance [43, 51, 58]. Histo-

Table 3.3. Effects of graft tissue processing on allograft mechanical performance

Processing technique	Study	Observations
Lyophilization	Brantigan et al. 1993 [9]	Fresh frozen cancellous bone is 219% stronger than lyophilized cancellous bone.
	Simonian et al. 1994 [54]	Lyophilization significantly decreases screw pullout strength.
	Kang and Kim 1995 [28]	In vivo lyophilized graft had decreases of 30.1% in bending strength and 41.3% in compressive strength.
	Thoren and Aspenberg 1995 [60]	Lyophilization decreased mechanical stiffness by 19%, yield by 16%, and energy to failure by 31%.
	Nather et al. 2004 [41]	Lyophilized allografts significantly weaker than deep-frozen grafts.
Gamma-irradiation	Anderson et al. 1992 [5]	Failure stress and elastic moduli of cancellous bone significantly decreased after 6.0 megarads but not after 2.5 megarads.
	Rasmussen et al. 1994 [45]	12% decrease in stiffness and 26% decrease in maximum force after 4.0 megarads.
	Zhang et al. 1994 [68]	No significant difference in mechanical properties of iliac crest wedge grafts after 2.0 to 2.5 megarads.
	Fideler et al. 1995 [19]	Mechanical properties of fresh-frozen bone-patella-bone graft reduced by 15% after 2.0 megarads, with further reduction of 46% after 4.0 megarads.
	Hamer et al. 1996 [23]	Dose-dependent decreases of up to 46% in mechanical strength after irradiation.
	Currey et al. 1997 [13]	Virucidal irradiation levels decreased bending strength by 52% to 67%, work to fracture by 74% to 96%, and impact energy by 37% to 75%.
Pasteurization	Borcher et al. 1995 [7]	Boiling and autoclaving decreased allograft strength by 26% and 58%, respectively. Freezing did not compromise allograft strength.
Ethylene oxide	Wittenberg et al. 1990 [66]	Ethylene oxide had no significant effect on immediate compression strength of grafts.

logical evaluation was performed on 27% of the 1,146 osteoarthritic femoral heads donated by patients undergoing elective total hip arthroplasty. More than 30% of the samples exhibited osteopenia on radiographic examination. Marked, generalized osteopenia with thinning of the cortical and cancellous bone was found in 3% of the samples [43]. The increased incidence of osteopenia clearly affects the bone quality, but the effects of metabolic and inflammatory diseases noted in some specimens are not known [43]. Siddiqui et al. [51] observed that 12% of 40 allografts from screened donors in their fifties had osteoporosis. They suggested that these allografts would not be suitable in cases where graft strength is required.

Bone allografts can be used in either orthotopic or heterotopic transplantations. In orthotopic transplantation, cortical bone allografts are placed in an anatomically appropriate site, as in an area of large bone loss. In heterotopic transplantation, bone allografts are placed in an anatomically abnormal location, such as a fibular strut allograft used adjunctively during a vertebral fusion [57]. In general, cortical bone allografts are stronger than cancellous bone allografts. Cortical bone graft strength varies according to anatomical location, with fibular struts being stronger than femoral rings, which in turn are stronger than tricortical iliac bone crest [21, 46, 52, 66]. Additionally, iliac bone grafts harvested close to the anterior superior iliac spine are stronger than those harvested near the posterior iliac spine [31]. Various combinations of cortical and cancellous bone allografts can augment reconstructive procedures, but terminal sterilization, though recommended, may reduce their strength.

3.5 Effects of Processing on Biomechanical Properties of Musculoskeletal Allografts

Gamma-irradiation and lyophilization (freeze-drying), two commonly used techniques for terminal sterilization of allografts, lead to weakening of grafts (Table 3.3). Gamma-irradiation of at least 3.0 megarads is required to inactivate viruses, whereas 1.5 to 2.5 megarads can inactivate bacteria [20, 45, 47]. Radiation dose weakens the biomechanical properties of musculoskeletal allografts. Gamma-irradiation at virucidal levels significantly increases fatigue while decreasing failure strength, failure energy, and stiffness [5, 13, 19, 23, 45, 54, 68]. Lyophilization reduces screw pullout strength and the maximum limits of strength, torque, and torsional stiffness and diminishes absorption energy [28, 41, 54]. Lyophilized allografts need to be rehydrated before transplantation; the quality of rehydration can also affect the mechanical parameters [28, 41, 61]. Terminal sterilization with ethylene oxide does not significantly weaken screw pullout strength [54]. Musculoskeletal tissues subjected to boiling or autoclaving exhibit significant reductions in strength, but freezing does not reduce their strength [7].

3.6 Conclusions

Musculoskeletal allografts are an alternative to autografts without the associated morbidities [6, 18, 30, 35, 59]. Allografts are widely available in a variety of preparations, and their transplantation is a safe, comprehensively regulated practice with a low incidence of HIV [2, 32, 53], HCV, and bacterial infection. The risk of infection is further decreased when musculoskeletal allografts are obtained from AATB-accredited tissue banks that practice comprehensive donor screening and tighter tissue procurement and employ more testing than required by the FDA [67]. The use of tissue banks that perform PCR analysis and/or histomorphometric testing of donor tissues further minimizes the risk of viral or bacterial transmission [47, 48, 51, 58]. The risk of HIV infection from fresh-frozen, non-terminally-sterilized musculoskeletal allografts is comparable to that from blood transfusion [10]. Terminal sterilization by gamma-irradiation or lyophilization of musculoskeletal tissues can further diminish HIV and HCV infection rates, but at the cost of a decrease in the mechanical properties of the allograft. Until more tissue-screening tests become available, active surveillance and audit of stored nonimplanted allografts will provide further assurance for the quality control of musculoskeletal transplants.

Using fresh, fibular strut or femoral ring cortical bone allografts from younger, nonosteoporotic donors permits the surgeon to maximize the structural integrity of reconstructive procedures [11, 15, 17, 39, 46, 52, 55]. The osteoinductive ability of bone allografts can be maximized by selecting demineralized, freeze-dried grafts from younger donors [37, 50].

In the future, new methods or modifications of existing tissue-processing techniques may be developed to maximize the osteoinductive and osteoconductive properties of bone allografts. Possible approaches to augment bone allograft performance may include the use of bone morphogenetic proteins (BMPs) or other local or systemic mediators of growth and inflammation. For example, the use of structural cortical bone allografts with osteoconductive and structural capabilities could add to the osteoinductive ability of the graft [25, 40, 56]. Although promising, the use of BMPs in conjunction with allografts needs further study.

References

1. Hepatitis C virus transmission from an antibody-negative organ and tissue donor-United States, 2000–2002 (2003) MMWR 53:273–276.
2. Transmission of HIV through bone transplantation: case report and public health recommendations (1988) MMWR 37:597–599.
3. United States Census Bureau, Statistical Abstract of the United States (2001) No. 168 Organ Transplants and Grafts, 1990 to 2000.
4. Update: allograft-associated bacterial infections-United States (2002) MMWR 51:207–210.
5. Anderson MJ, Keyak JH, Skinner HB (1992) Compressive mechanical properties of human cancellous bone after gamma irradiation. J Bone Joint Surg Am 74:747–752.
6. Banwart JC, Asher MA, Hassanein RS (1995) Iliac crest bone graft harvest donor site morbidity. A statistical evaluation. Spine 20:1055–1060.

7. Borchers RE, Gibson LJ, Burchardt H, Hayes WC (1995) Effects of selected thermal variables on the mechanical properties of trabecular bone. Biomaterials 16: 545–551.
8. Boyce T, Edwards J, Scarborough N (1999) Allograft bone. The influence of processing on safety and performance. Orthop Clin North Am 30:571–581.
9. Brantigan JW, Cunningham BW, Warden K, McAfee PC, Steffee AD (1993) Compression strength of donor bone for posterior lumbar interbody fusion. Spine 18:1213–1221.
10. Buck BE, Malinin TI, Brown MD (1989) Bone transplantation and human immunodeficiency virus. An estimate of risk of acquired immunodeficiency syndrome (AIDS). Clin Orthop 240:129–136.
11. Burstein AH, Reilly DT, Martens M (1976) Aging of bone tissue: mechanical properties. J Bone Joint Surg Am 58:82–86.
12. Conrad EU, Gretch DR, Obermeyer KR, Moogk MS, Sayers M, Wilson JJ, Strong DM (1995) Transmission of the hepatitis-C virus by tissue transplantation. J Bone Joint Surg Am 77:214–224.
13. Currey JD, Foreman J, Laketic I, Mitchell J, Pegg DE, Reilly GC (1997) Effects of ionizing radiation on the mechanical properties of human bone. J Orthop Res 15:111–117.
14. Davy DT (1999) Biomechanical issues in bone transplantation. Orthop Clin North Am 30:553–563.
15. Dickenson RP, Hutton WC, Stott JR (1981) The mechanical properties of bone in osteoporosis. J Bone Joint Surg Br 63B:233–238.
16. Ehrler DM, Vaccaro AR (2000) The use of allograft bone in lumbar spine surgery. Clin Orthop 371:38–45.
17. Einhorn TA (2003) The structural properties of normal and osteoporotic bone. Instr Course Lect 52: 533–539.
18. Fernyhough JC, Schimandle JJ, Weigel MC, Edwards CC, Levine AM (1992) Chronic donor site pain complicating bone graft harvesting from the posterior iliac crest for spinal fusion. Spine 17:1474–1480.
19. Fideler BM, Vangsness CT Jr, Lu B, Orlando C, Moore T (1995) Gamma irradiation: effects on biomechanical properties of human bone-patellar tendon-bone allografts. Am J Sports Med 23:643–646.
20. Fideler BM, Vangsness CT Jr, Moore T, Li Z, Rasheed S (1994) Effects of gamma irradiation on the human immunodeficiency virus. A study in frozen human bone-patellar ligament-bone grafts obtained from infected cadavera. J Bone Joint Surg Am 76:1032–1035.
21. Glazer PA, Colliou O, Lotz JC, Bradford DS (1996) Biomechanical analysis of lumbosacral fixation. Spine 21:1211–1222.
22. Goldberg VM (2000) Selection of bone grafts for revision total hip arthroplasty. Clin Orthop 381:68–76.
23. Hamer AJ, Strachan JR, Black MM, Ibbotson CJ, Stockley I, Elson RA (1996) Biochemical properties of cortical allograft bone using a new method of bone strength measurement. A comparison of fresh, fresh-frozen and irradiated bone. J Bone Joint Surg Br 78: 363–368.
24. Hirn M, Laitinen M, Pirkkalainen S, Vuento R (2004) Cefuroxime, rifampicin and pulse lavage in decontamination of allograft bone. J Hosp Infect 56:198–201.
25. Jensen TB, Overgaard S, Lind M, Rahbek O, Bunger C, Soballe K (2002) Osteogenic protein 1 device increases bone formation and bone graft resorption around cementless implants. Acta Orthop Scand 73:31–39.
26. Journeaux SF, Johnson N, Bryce SL, Friedman SJ, Sommerville SM, Morgan DA (1999) Bacterial contamination rates during bone allograft retrieval J Arthroplasty 14:677–681.
27. Kainer MA, Linden JV, Whaley DN, Holmes HT, Jarvis WR, Jernigan DB, Archibald LK (2004) Clostridium infections associated with musculoskeletal-tissue allografts. N Engl J Med 350:2564–2571.
28. Kang JS, Kim NH (1995) The biomechanical properties of deep freezing and freeze drying bones and their biomechanical changes after in vivo allograft. Yonsei Med J 36:332–335.
29. Komiya K, Nasuno S, Uchiyama K, Takahira N, Kobayashi N, Minehara H, Watanabe S, Itoman M (2003) Status of bone allografting in japan—nationwide survey of bone grafting performed from 1995 through 1999. Cell Tissue Bank 4:217–220.
30. Kreibich DN, Scott IR, Wells JM, Saleh M (1994) Donor site morbidity at the iliac crest: comparison of percutaneous and open methods. J Bone Joint Surg Br 76:847–848.
31. Kummer FJ, Chen D, Spivak JM (1998) Optimal selection and preparation of fresh frozen corticocancellous allografts for cervical interbody spinal fusion. Spine 23:2295–2298.
32. Li CM, Ho YR, Liu YC (2001) Transmission of human immunodeficiency virus through bone transplantation: a case report. J Formos Med Assoc 100:350–351.
33. Lietman SA, Tomford WW, Gebhardt MC, Springfield DS, Mankin HJ (2000) Complications of irradiated allografts in orthopaedic tumor surgery. Clin Orthop 375:214–217.
34. Lill CA, Hesseln J, Schlegel U, Eckhardt C, Goldhahn J, Schneider E (2003) Biomechanical evaluation of healing in a non-critical defect in a large animal model of osteoporosis. J Orthop Res 21:836–842.
35. Lim EV, Lavadia WT, Roberts JM (1996) Superior gluteal artery injury during iliac bone grafting for spinal fusion. A case report and literature review. Spine 21:2376–2378.
36. Lobo Gajiwala A (2003) Tissue banking in India: gamma-irradiated allografts. Cell Tissue Bank 4:203–211.
37. Lohmann CH, Andreacchio D, Koster G, Carnes DL Jr, Cochran DL, Dean DD, Boyan BD, Schwartz Z (2001) Tissue response and osteoinduction of human bone grafts in vivo Arch Orthop Trauma Surg 121:583–590.
38. Malinin TI, Buck BE, Temple HT, Martinez OV, Fox WP (2003) Incidence of clostridial contamination in donors' musculoskeletal tissue. J Bone Joint Surg Br 85:1051–1054.
39. McCalden RW, McGeough JA, Barker MB, Court-Brown CM (1993) Age-related changes in the tensile properties of cortical bone. The relative importance of changes in porosity, mineralization, and microstructure. J Bone Joint Surg Am 75:1193–1205.
40. McGee MA, Findlay DM, Howie DW, Carbone A, Ward P, Stamenkov R, Page TT, Bruce WJ, Wildenauer CI, Toth C (2004) The use of OP-1 in femoral impaction grafting in a sheep model. J Orthop Res 22:1008–1015.

41. Nather A, Thambyah A, Goh JC (2004) Biomechanical strength of deep-frozen versus lyophilized large cortical allografts. Clin Biomech 19:526–533.
42. Navas J, Soto C (2003) The Colombian experience in tissue banking: the bone and tissue bank of the Cosmos and Damian Foundation, Bogota. Cell Tissue Bank 4:157–161.
43. Palmer SH, Gibbons CL, Athanasou NA (1999) The pathology of bone allograft. J Bone Joint Surg Br 81:333–335.
44. Pelker RR, Friedlaender GE (1987) Biomechanical aspects of bone autografts and allografts. Orthop Clin North Am 18:235–239.
45. Rasmussen TJ, Feder SM, Butler DL, Noyes FR (1994) The effects of 4 Mrad of gamma irradiation on the initial mechanical properties of bone-patellar tendon-bone grafts. Arthroscopy 10:188–197.
46. Reilly DT, Burstein AH (1974) The mechanical properties of cortical bone. J Bone Joint Surg Am 56:1001–1022.
47. Roder W, Muller H, Muller WE, Merz H (1992) HIV infection in human bone. J Bone Joint Surg Br 74:179–180.
48. Salzman NP, Psallidopoulos M, Prewett AB, O'Leary R (1993) Detection of HIV in bone allografts prepared from AIDS autopsy tissue. Clin Orthop Relat Res 292: 384–390.
49. Sanzen L, Carlsson A (1997) Transmission of human T-cell lymphotrophic virus type 1 by a deep-frozen bone allograft. Acta Orthop Scand 68:72–74.
50. Schwartz Z, Somers A, Mellonig JT, Carnes DL Jr, Dean DD, Cochran DL, Boyan BD (1998) Ability of commercial demineralized freeze-dried bone allograft to induce new bone formation is dependent on donor age but not gender. J Periodontol 69:470–478.
51. Siddiqui SA, Lipton JF, Vigorita VJ, Evangelista J, Bryk E (2004) Bone biopsy as a screening technique for bone bank allograft donation. Am J Orthop 33: 123–126.
52. Siff TE, Kamaric E, Noble PC, Esses SI (1999) Femoral ring versus fibular strut allografts in anterior lumbar interbody arthrodesis. A biomechanical analysis. Spine 24:659–665.
53. Simonds RJ, Holmberg SD, Hurwitz RL, Coleman TR, Bottenfield S, Conley LJ, Kohlenberg SH, Castro KG, Dahan BA, Schable CA, et al (1992) Transmission of human immunodeficiency virus type 1 from a seronegative organ and tissue donor. N Engl J Med 326:726–732.
54. Simonian PT, Conrad EU, Chapman JR, Harrington RM, Chansky HA (1994) Effect of sterilization and storage treatments on screw pullout strength in human allograft bone Clin Orthop 302:290–296.
55. Smith CB, Smith DA (1976) Relations between age, mineral density and mechanical properties of human femoral compacta. Acta Orthop Scand 47: 496–502.
56. Soballe K, Jensen TB, Mouzin O, Kidder L, Bechtold JE (2004) Differential effect of a bone morphogenetic protein-7 (OP-1) on primary and revision loaded, stable implants with allograft. J Biomed Mater Res 71A:569–576.
57. Stevenson S (1999) Biology of bone grafts. Orthop Clin North Am 30:543–552.
58. Sugihara S, van Ginkel AD, Jiya TU, van Royen BJ, van Diest PJ, Wuisman PI (1999) Histopathology of retrieved allografts of the femoral head. J Bone Joint Surg Br 81:336–341.
59. Summers BN, Eisenstein SM (1989) Donor site pain from the ilium. A complication of lumbar spine fusion. J Bone Joint Surg Br 71:677–680.
60. Thoren K, Aspenberg P (1995) Ethylene oxide sterilization impairs allograft incorporation in a conduction chamber. Clin Orthop 114:259–264.
61. Thoren K, Aspenberg P, Thorngren KG (1995) Lipid extracted bank bone. Bone conductive and mechanical properties. Clin Orthop 311:232–246.
62. Tomford WW, Mankin HJ (1999) Bone banking. Update on methods and materials. Orthop Clin North Am 30:565–570.
63. Tomford WW, Mankin HJ, Friedlaender GE, Doppelt SH, Gebhardt MC (1987) Methods of banking bone and cartilage for allograft transplantation. Orthop Clin North Am 18:241–247.
64. Tomford WW, Thongphasuk J, Mankin HJ, Ferraro MJ (1990) Frozen musculoskeletal allografts. A study of the clinical incidence and causes of infection associated with their use. J Bone Joint Surg Am 72:1137–143.
65. Vehmeyer S, Wolkenfelt J, Deijkers R, Petit P, Brand R, Bloem R (2002) Bacterial contamination in postmortem bone donors. Acta Orthop Scand 73:678–683.
66. Wittenberg RH, Moeller J, Shea M, White AA 3rd, Hayes WC (1990) Compressive strength of autologous and allogenous bone grafts for thoracolumbar and cervical spine fusion. Spine 15:1073–1078.
67. Woll JE, Kasprisin D (2001) Standards for Tissue Banking. McLean, Virginia: American Association of Tissue Banks.
68. Zhang Y, Homsi D, Gates K, Oakes K, Sutherland V, Wolfinbarger L Jr (1994) A comprehensive study of physical parameters, biomechanical properties, and statistical correlations of iliac crest bone wedges used in spinal fusion surgery. IV. Effect of gamma irradiation on mechanical and material properties. Spine 19:304–308.