

ORIGINAL ARTICLE

Clostridium Infections Associated with Musculoskeletal-Tissue Allografts

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ABSTRACT

BACKGROUND

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Allografts are commonly used in orthopedic reconstructive surgery. In 2001, approximately 875,000 musculoskeletal allografts were distributed by U.S. tissue banks. After the death from *Clostridium sordellii* sepsis of a 23-year-old man who had received a contaminated allograft from a tissue bank (Tissue Bank A), the Centers for Disease Control and Prevention initiated an investigation, including enhanced case finding, of the methods used for the recovery, processing, and testing of tissue.

METHODS

A case of allograft-associated clostridium infection was defined as a culture-proven infection of a surgical site within one year after allograft implantation, from January 1998 to March 2002. We traced tissues to tissue banks that recovered and processed these tissues. We also estimated the rates of and risk ratios for clostridium infections for tissues processed by the implicated tissue bank and reviewed processing and testing methods used by various tissue banks.

RESULTS

Fourteen patients were identified, all of whom had received allografts processed by Tissue Bank A. The rates of clostridium infection were 0.12 percent among patients who received sports-medicine tissues (i.e., tendons, femoral condyles, menisci) from Tissue Bank A and 0.36 percent among those who received femoral condyles in particular. The risk-ratio estimates for clostridium infections from tissues processed by Tissue Bank A, as compared with those from other tissue banks, were infinite ($P < 0.001$) for musculoskeletal allografts, sports-medicine tissues, or tendons. Because Tissue Bank A cultured tissues only after treating them with a nonsporicidal antimicrobial solution, some test results were probably false negatives. Tissues from implicated donors were released despite the isolation of clostridium or bowel flora from other anatomical sites or reports of infections in other recipients.

CONCLUSIONS

Clostridium infections were traced to allograft implantation. We provide interim recommendations to enhance tissue-transplantation safety. Tissue banks should validate processes and culture methods. Sterilization methods that do not adversely affect the functioning of transplanted tissue are needed to prevent allograft-related infections.

TISSUE ALLOGRAFTS ARE COMMONLY used in orthopedic surgical procedures. In 2001, U.S. tissue banks distributed approximately 875,000 musculoskeletal allografts, as compared with 350,000 in 1990.¹ In contrast to blood banks, there is relatively little regulatory oversight of tissue banks, which can recover, process, and distribute tissue across multiple states.²

Processed tissue allografts are not necessarily sterile and may result in viral (hepatitis B,³ hepatitis C,⁴⁻⁶ or human immunodeficiency virus⁷) or bacterial (gram-negative organisms or enterococcus) infections.^{2,8-10} Clostridium septic arthritis remains a rare complication of orthopedic procedures.¹¹⁻¹⁷ After the death from *Clostridium sordellii* sepsis of a 23-year-old man who had received a cadaveric musculoskeletal allograft,⁸ the Centers for Disease Control and Prevention (CDC) enhanced its efforts to find cases of allograft-associated clostridium infections and conducted an investigation of the processing and testing methods used by the tissue bank (hereafter referred to as Tissue Bank A) that had processed this patient's allograft. Herein we report our findings and provide interim recommendations for enhanced tissue-transplantation safety.

METHODS

DEFINITION AND IDENTIFICATION OF CASES

A case was defined as a surgical-site infection¹⁸ within 12 months after an allograft transplantation in an otherwise healthy U.S. patient without known risk factors for surgical-site infections¹⁹ (e.g., diabetes) or clostridium infection (e.g., injection-drug use or hematologic or colon cancer) at the site of allograft implantation during January 1998 through March 2002. All cases were confirmed by culture of blood or knee aspirates.

We solicited case reports by examining published reports⁸⁻¹⁰ and electronic-mail lists and by contacting the Food and Drug Administration (FDA); state regulatory authorities in California, Florida, and New York; and tissue banks. Our investigation consisted of epidemiologic studies, a review of tissue-bank procedures, and laboratory investigations.

EPIDEMIOLOGIC STUDIES

We used standardized forms to record data from potential case patients' medical and surgical records. We contacted tissue banks that recovered tis-

sue from implicated donors and used standardized forms to record information, such as the time from the death of the donor to refrigeration of the body. We traced tissues from implicated donors to the tissue banks that recovered the tissue and to any other tissue banks that processed tissue from the same donor. We requested data on all available culture methods and results, tissue-processing methods, and patient outcomes.

Since reporting of clostridium infections was not mandatory and the number of allografts of specific types of tissue was unknown, we used available data to estimate the rates of clostridium infection. The numerator consisted of the number of cases reported to the CDC after allograft implantation in 2001. For the denominator, we obtained the total number of musculoskeletal allografts shipped by Tissue Bank A in 2001 using publicly available data filed with the Securities and Exchange Commission. Also, we used data from New York State's licensing program to obtain the number of so-called sports-medicine tissues (i.e., tendons used for reconstruction of the anterior cruciate ligament, fresh femoral condyles, and menisci) distributed nationally by Tissue Bank A in 2001. Next, we calculated the rates of clostridium infection according to the type of tissue transplanted.

Using New York State's licensing-program data, we calculated the proportion of musculoskeletal tissues processed and distributed by Tissue Bank A, as compared with that of other tissue banks in the period between January 1998 and March 2002 (i.e., the market share). We estimated risk ratios of clostridium infections associated with tissues distributed nationally by Tissue Bank A from January 1998 through March 2002. Tissue-specific denominator data for 1998 through 2001 were obtained from New York State. We estimated rates for January 2002 through March 2002, extrapolating 2001 data, assuming the same frequency of allograft distribution.

PROCEDURAL REVIEW

We visited three tissue banks, including Tissue Bank A, and observed how tissue was recovered from cadavers, processed, and cultured; reviewed operating-procedure manuals; and interviewed tissue-bank personnel. In addition, we reviewed inspection reports from New York State and the FDA, warning letters, and published regulations and standards.²⁰⁻²³

LABORATORY STUDIES

We obtained nonimplanted tissues from implicated donors from Tissue Bank A and cultured the specimens. We conducted in vitro experiments to evaluate the tissue bank's method of detecting clostridium spores. To determine whether residual antimicrobial agents on tissue previously suspended in antimicrobial solution could cause bacteriostasis,²⁴ we determined the expected antimicrobial concentrations in culture medium in anaerobic, standard blood-culture bottles that did not contain charcoal (BacT/Alert, Organon Teknika) used by Tissue Bank A. We conducted in vitro experiments to determine whether these antimicrobial concentrations caused bacteriostasis when exposed to low inoculums (100 spores per blood-culture bottle) of *C. sordellii* (American Type Culture Collection strain 14337); we used three different amounts of antimicrobial solution (0.5 ml, 1.0 ml, and 2.0 ml) to represent residual carryover. Blood-culture bottles were incubated at 35°C for 14 days; bottles showing growth underwent Gram's staining, were plated aerobically and anaerobically, and were confirmed as *C. sordellii*. Bottles having no growth after 14 days were held for an additional 7 days and then plated; plates were incubated for 10 days. We also examined whether the use of bottles containing activated charcoal helped in spore recovery.²⁵

STATISTICAL ANALYSIS

Data were analyzed with the use of Epi Info software (CDC, version 6.04). The chi-square or Fisher's exact test, where appropriate, was used to compare categorical variables. Risk ratios were calculated. All P values are two-sided.

RESULTS

EPIDEMIOLOGIC STUDIES*Characteristics of the Patients*

Between January 1998 and March 2003, 14 patients (including the index patient) met the case definition (Table 1). *C. septicum* was isolated in 12 (86 percent). The median age was 32 years (range, 15 to 52); 10 patients (71 percent) were male. Symptoms began a median of 4.5 days (range, 2 to 85) after allograft implantation. The sole death was that of the index patient, who had *C. sordellii* infection.⁹ All other patients required hospitalization, intravenous antimicrobial therapy, joint irrigation, and débridement. Ten patients (71 percent) required allograft removal; three were scheduled for knee-replacement sur-

gery for intractable pain, and three could not maintain full-time employment owing to postinfection disability.

Tissue Recovery

Tissues implanted into the 14 patients came from 9 donors by way of seven tissue banks. Donors had no overt signs of infection before death; six died from trauma. The median interval from death to refrigeration of the body was 5.9 hours (range, 4.1 to 19.0). Industry standards state that tissue may be recovered up to 24 hours after death as long as the interval between death and refrigeration of the body does not exceed 12 hours²³; this limit was exceeded in the case of two donors (15 hours and 19 hours).

Tissue Tracing

Allograft tissues from all 14 patients had been processed by Tissue Bank A. These included nine hemipatellar tendons used for reconstruction of the anterior cruciate ligament (64 percent), four femoral condyles (29 percent), and one meniscus (7 percent). The four femoral condyles were fresh; the other 10 allografts were frozen (71 percent). Tissues from three donors were processed by Tissue Bank A alone, whereas tissues from five donors were processed and distributed by Tissue Bank A as well as other tissue banks. Complete information on tissue processing and distribution was not available for one donor.

Some tissue allografts from the five donors that were sent to other tissue banks were processed with the use of sterilization methods. Gamma irradiation was used for 17 bone, 6 tendon, and 2 fascia lata allografts. Sixty-five bone allografts were processed with the use of a low-temperature chemical-sterilization method (BioCleanse, Regeneration Technologies). No reports of infection were identified among recipients of tissues processed by these other tissue banks with the use of sterilization with gamma irradiation or BioCleanse; tissues at Tissue Bank A did not undergo sterilization.

Rates and Estimated Risk of Clostridium Infection for Tissues Processed by Tissue Bank A

The rates of clostridium infection in 2001 among patients receiving tissues processed by Tissue Bank A were 0.12 percent for all sports-medicine tissues, 0.09 percent for tendons, 0.15 percent for menisci, and 0.36 percent for femoral condyles. Tissue Bank A was estimated to have 0.1 percent of the market share for all musculoskeletal tissue processed for

Table 1. Characteristics of 14 Patients with Allograft-Associated Clostridium Infections in the United States, 1998 through 2002.

Patient No.	Year	Age (yr)/ Sex	Tissue	Clostridium Species Isolated from Blood or Surgical Site	Donor No.	Common Donor*	Culture Results of Tissue	
							Before Implantation†	After Processing‡
1	1998	50/M	Hemipatellar tendon	<i>C. septicum</i>	1	Yes	<i>C. sordellii</i>	Unknown
2	1998	50/M	Hemipatellar tendon	<i>C. septicum</i>	1	Yes	<i>C. sordellii</i>	Unknown
3	1998	16/F	Hemipatellar tendon	<i>C. septicum</i>	2	No	Unknown	Unknown
4	1999	40/F	Hemipatellar tendon	<i>C. septicum</i>	3	No	<i>C. septicum</i>	<i>C. septicum</i>
5	2001	23/M	Femoral condyle	<i>C. sordellii</i>	4	Yes	<i>C. sordellii</i>	Negative
6	2001	15/F	Femoral condyle	<i>C. septicum</i>	5	No	Unknown	Unknown
7	2001	35/M	Meniscus	<i>C. septicum</i>	6	Yes	<i>C. septicum</i>	Unknown
8	2001	28/M	Femoral condyle	<i>C. septicum</i> , <i>C. butyricum</i>	6	Yes	<i>C. septicum</i>	Unknown
9	2001	17/M	Hemipatellar tendon	<i>C. septicum</i>	7	Yes	<i>C. septicum</i> , <i>C. parapatrificum</i>	<i>C. septicum</i> , <i>C. parapatrificum</i>
10	2001	35/M	Hemipatellar tendon	<i>C. septicum</i>	7	Yes	<i>C. septicum</i> , <i>C. parapatrificum</i>	<i>C. septicum</i> , <i>C. parapatrificum</i>
11	2001	41/F	Hemipatellar tendon	<i>C. septicum</i>	7	Yes	<i>C. septicum</i> , <i>C. parapatrificum</i>	<i>C. septicum</i> , <i>C. parapatrificum</i>
12	2001	29/M	Hemipatellar tendon	<i>C. septicum</i>	8	Yes	<i>C. septicum</i> , <i>C. subterminale</i> , <i>B. fragilis</i>	Unknown
13	2001	52/M	Hemipatellar tendon	<i>C. septicum</i>	8	Yes	<i>C. septicum</i> , <i>C. subterminale</i> , <i>B. fragilis</i>	Unknown
14	2002	16/M	Femoral condyle	<i>C. bifermentans</i>	9	No	<i>Escherichia coli</i> , Enterococcus species	Gram-negative organism; otherwise not specified

* A common donor was one from whom two or more patients received tissue and subsequently had surgical-site infections.

† These cultures may have been performed at tissue recovery, before exposure to antimicrobial solution (by Tissue Bank A or other tissue banks), after processing (by Tissue Bank A or other tissue banks), or by the surgical team in the operating room before implantation into the recipient.

‡ Results shown are of cultures of tissue performed by Tissue Bank A as part of quality control. "Unknown" indicates that this information was not provided by Tissue Bank A.

transplantation, 25 percent for sports-medicine tissues, 23 percent for tendons, 78 percent for femoral condyles, and 94 percent for menisci in the period between January 1998 and March 2002. The risk of clostridium infection was significantly higher after the receipt of musculoskeletal tissues processed by Tissue Bank A than after the receipt of tissue from other tissue banks ($P < 0.001$) (Table 2).

PROCEDURAL REVIEW OF TISSUE BANK A

Tissue-Processing Methods

Tissue Bank A used aseptic techniques during the processing of recovered tissues. We observed no cross-contamination between tissues from different donors. Tissue was decontaminated by suspension in a proprietary solution containing imipenem, vancomycin, amikacin, amphotericin B, and flucon-

Table 2. Estimated Relative Risk of Clostridium Infection from Allografts Processed by Tissue Bank A, as Compared with Other Tissue Banks, January 1998 through March 2002.

Type of Tissue	No. of Clostridium Infections/ Total No. of Allografts (%)	Risk Ratio*	P Value
Musculoskeletal tissue			
Tissue Bank A	14/20,152 (0.07)	2898	<0.001
Other tissue banks	0/2,015,428		
Sports-medicine tissues			
Tissue Bank A	14/20,152 (0.07)	85	<0.001
Other tissue banks	0/59,284		
Tendons			
Tissue Bank A	9/16,354 (0.06)	64	<0.001
Other tissue banks	0/54,750		
Femoral condyles			
Tissue Bank A	4/1197 (0.33)	2.6	0.58
Other tissue banks	0/347		
Menisci			
Tissue Bank A	1/2600 (0.04)	—	1.00
Other tissue banks	0/173		

* When the risk ratios were infinite, we obtained Woolf's estimates by adding 0.5 to each cell.

azole. Tissue Bank A did not provide validation of the efficacy of this solution in killing spore-forming organisms, such as clostridium.

Testing Methods

Companion tissue (e.g., a cartilage sliver from a femoral condyle) was processed in parallel with the allograft. After suspension of allografts and companion tissue in antimicrobial solution, companion tissue was placed in cell-culture medium and briskly agitated for 30 seconds. Aliquots of this medium were then cultured aerobically with the use of BacT/Alert blood-culture bottles containing activated charcoal and anaerobically with the use of standard anaerobic bottles (Organon Teknika) and on non-prereduced blood agar.

All cultures were incubated at 35°C; the bottles were incubated for seven days in a BacT/Alert 3D automated system introduced in September 2001. Nonautomated broth-culture methods were used before September 2001. Identification of isolates was performed by Tissue Bank A; not all isolates were fully identified. Tissue Bank A did not routinely measure the microbial burden (quantitative cultures) of incoming tissues and, in 1995, had stopped performing qualitative cultures of tissue before exposing them to antimicrobial solution. When Tissue Bank A identified a positive culture, only tissue asso-

ciated with the culture was discarded, not other tissues from the same donor. Also, tissues from implicated donors were released for implantation despite reports of infections in other recipients. Tissue Bank A had not conducted bacteriostasis or fungistasis studies to evaluate their culture methods.

LABORATORY STUDIES

Cultures of Non-Implanted Tissues from Tissue Bank A
Tissue Bank A provided 27 processed allografts that had not been implanted from four of nine implicated donors (44 percent). We isolated clostridium (*C. sordellii* or *C. septicum*) in cultures from 4 of 17 musculoskeletal tissues (24 percent; from three of four implicated donors). We grew *C. sordellii* from 2 of 11 musculoskeletal tissues from Donor 4, although all 12 specimens of companion tissue from Donor 4 cultured after antimicrobial treatment by Tissue Bank A for the purpose of quality assurance were negative. Tissue Bank A did not provide corresponding culture results for Donors 3 and 6.

Bacteriostasis

The antimicrobial solution used for processing tissue at Tissue Bank A included two agents with potential activity against clostridium — imipenem and vancomycin. Approximately 1 ml of fluid was carried over with each companion tissue, according to reports obtained from personnel at the tissue bank. Our in vitro experiment demonstrates the potential effect on bacterial growth, especially with a low inoculum, of carrying over antimicrobial solution (Table 3). The use of anaerobic blood-culture bottles that contained charcoal improved the ability to detect *C. sordellii*, especially with increasing volumes of antimicrobial carryover.

DISCUSSION

Allografts can substantially improve the quality of life.²⁶ Our investigation, however, demonstrates that infection acquired through bacterial contamination of allografts may result in substantial complications or death. Furthermore, our findings suggest that current federal regulations and industry standards for processing and testing allograft tissue need to be enhanced to prevent allograft-associated infections.^{2,10}

Our investigation highlighted several factors that contributed to clostridium infections. First, implanted tissues were not processed with the use of methods that achieved sterility or that were spori-

Table 3. Effect of Carryover of Antimicrobial Solution on the Recovery of *Clostridium sordellii* from Standard and Charcoal-Containing Anaerobic Blood-Culture Bottles.*

Antimicrobial Solution Carried Over	Positive Standard Blood-Culture Bottles	Positive Charcoal-Containing Blood-Culture Bottles
<i>ml</i>	<i>no./total no. (%)</i>	
0.0	3/3 (100)	3/3 (100)
0.5	1/3 (33)	2/3 (67)
1.0	0/0	1/3 (33)
2.0	0/0	1/3 (33)

* Low inocula (100 spores of *C. sordellii*) were used.

cidal. However, current regulations do not require tissue banks to eliminate bacteria present on tissues at the time of recovery or to use processing methods that guarantee tissue sterility. Second, the concentration of bacteria (“bioburden”) before processing was unknown, because no tissues were cultured before being exposed to antimicrobial agents. Third, cultures of specimens obtained after processing were probably false negative as a result of carryover of antimicrobial solution. Fourth, evidence of clostridium or bowel flora at other anatomical sites or reports of infections in other allograft recipients were not used as criteria for determining the suitability of donor tissues for transplantation.

At the CDC, cultures of two nonimplanted tissues from one donor yielded clostridium. In contrast, cultures of all 12 companion tissues from the donor were reported to be negative by Tissue Bank A. There are at least two potential reasons for this discrepancy. Because tissues were cultured only after their suspension in antimicrobial solution, residual antimicrobial agents on the tissues may have caused false negative culture results through bacteriostasis. Furthermore, companion tissues used for quality-assurance testing were small and had a larger surface-area-to-volume ratio than the allografts themselves, permitting better antimicrobial penetration of companion tissue.

We hypothesized that donor tissue became hematogenously contaminated by bowel flora, including clostridium and spores, at or before tissue recovery. Factors that might contribute to such contamination include a prolonged interval between the donor’s death and tissue recovery, delays in refrigeration, or death from trauma.^{27,28} With respect

to 2 of the 14 patients with clostridium infections (14 percent, or 2 of 9 donors [22 percent]), the interval between the donor’s death and refrigeration of the body exceeded the limit recommended by voluntary industry standards.²³

Aseptic processing of tissue minimizes bacterial contamination but will not eradicate contamination with organisms or spores, especially in tissue that is heavily contaminated at the time of recovery.² To reduce bacterial contamination of allografts, some tissue banks, including Tissue Bank A, suspend tissue in antimicrobial solutions. However, these solutions may not eradicate spores, as demonstrated by our in vitro studies (CDC: unpublished data). Two sterilization methods that would eliminate spores — gamma irradiation and treatment with ethylene oxide — have technical problems, limiting their use in tissue processing.² High doses of gamma irradiation may adversely affect the biomechanical properties of allografts.²⁹⁻³² Ethylene oxide has a limited ability to penetrate tissue and has been associated with adverse outcomes such as synovitis³³ or damage to musculoskeletal tissue, resulting in an unacceptably high rate of mechanical failure.³⁴

One tissue bank has developed and implemented a low-temperature chemical-sterilization approach (BioCleanse) that kills spores but preserves the biomechanical integrity and function of some allografts.³⁵⁻³⁷ The efficacy of this sterilization method is supported by the absence of reports of bacterial or viral allograft-associated infections in tissue processed by this method (CDC: unpublished data). In contrast, tissues processed with all other disinfection and sterilization methods, including gamma irradiation, have been associated with reports of allograft-associated infections (CDC: unpublished data). Currently, BioCleanse and other sterilization methods cannot be used to process fresh femoral condyles, since it is thought that chondrocytes must be viable to maintain articular cartilage function.

There were several limitations to our investigation. First, we identified a relatively small number of cases. To meet the case definition, cases had to be culture-positive. However, according to a recent survey of U.S. infectious-disease specialists, only 22 percent of respondents always cultured joint aspirates for anaerobic microorganisms; 39 percent rarely or never did.³⁸ Second, determination of the true rate of clostridium infection is challenging, because patients who become symptomatic may not present to the institution (often ambulatory surgical centers) where the surgery was performed.

Table 4. Recommendations to Reduce the Risk of Allograft-Associated Infections.

<p>Tissue banks should process tissue using a method that can kill bacterial spores. Existing sterilization techniques used for tissue allografts, such as gamma irradiation, or new techniques effective against bacterial spores can be used.</p> <p>Unless a sporicidal method is used, tissue should not be considered sterile. Health care providers and patients should be informed of the possible risk of bacterial infection from these tissues.</p> <p>If no sporicidal method is available (e.g., for fresh femoral condyles), tissue banks should minimize the potential for release of contaminated tissue.</p> <p>Allograft tissues should be cultured before suspension in antimicrobial solutions, and if clostridium or other bowel flora are isolated (i.e., if the presence of enteric pathogens suggests that clostridium spores may be present), all donor tissue that cannot be sterilized should be discarded.</p> <p>Tissue banks should consider performing both destructive testing and swab cultures of tissue to increase sensitivity for detecting bacterial contamination.</p> <p>Recommended time limits for recovery of tissue, from the time of donor death or asystole to the time of tissue recovery, should be followed²³; research should be performed on the effect of such restrictions on tissue procurement and tissue safety.</p> <p>Tissue banks should validate all quality-assurance methods used for tissue culture to ensure that carryover of residual antimicrobial agents does not result in false negative culture results.²⁴</p> <p>After a tissue bank receives a report of potential allograft-associated infection, any remaining tissue from the implicated donor should not be released until it is determined that the allograft is not the source of infection. In the event of a reported allograft-associated infection, tissue-bank personnel should notify health care providers of other recipients of tissue from the same donor. A sample of nonimplanted tissues that was processed in the same way as the tissue from an allograft-associated infection should be cultured by an independent laboratory using a validated method.</p> <p>Tissue banks with identified tissue-processing problems that resulted in a contaminated end product should perform a one-time audit of their unreleased tissue inventory to estimate the proportion of unreleased tissue that may be contaminated with microorganisms or spores.</p>

Furthermore, because clinicians presume that allografts are sterile, allografts are not usually considered a potential source of infection.

Third, most clostridium isolates obtained at the time of tissue recovery, before processing, packaging, or implantation, were not available to confirm species identification. This may partially explain the discrepancy between the clostridium species isolated from blood or surgical sites and culture results of tissues obtained before implantation. However, the most likely reason for this discrepancy is that agonal or postmortem bacteremia is frequently polymicrobial, and tissue contamination is not uniform.²

Fourth, there are no reliable data on the number of specific types of tissue distributed by all U.S. tissue banks for use in estimating the incidence of

clostridium infections. We used data collected by New York State, which licensed 17 of the largest U.S. tissue banks that processed musculoskeletal tissue in 2001, including Tissue Bank A. Thus, we may have overestimated the market share of Tissue Bank A and consequently underestimated the risk ratio of clostridium infections associated with this tissue bank.

On the basis of our investigation, we made recommendations to tissue banks, the American Association of Tissue Banks, and the FDA to reduce the risk of allograft-associated infection (Table 4). The use of a validated sporicidal process will confer the greatest protection for patients; the other proposed measures are less effective. For example, the sensitivity of cultures of specimens obtained before processing is low (10 to 22 percent)^{39,40}; thus, relying on the results of cultures alone to identify and discard tissues potentially contaminated with clostridium spores is problematic. However, for tissues that are not amenable to sterilization (e.g., fresh femoral condyles), the use of donor screening and preprocessing cultures will most likely reduce, but not eliminate, the risk of infection.

According to FDA regulations, each tissue bank is required to have written procedures for the prevention of microbial contamination or cross-contamination of tissues during processing.²¹ In March 2002, in response to our investigation, the FDA released for immediate implementation a guidance document for tissue banks (available at www.fda.gov/cber/guidelines.htm#tissval). This document highlighted that existing regulations require tissue processors to validate processing and testing methods. Furthermore, in August 2002, a recall of all tissues, with the exception of heart valves, processed by Tissue Bank A was issued. Product-liability cases and claims were filed against the tissue bank by patients.

In conclusion, improved guidelines for tissue processing and testing, together with monitoring of allograft-associated adverse events, should enhance tissue-transplantation safety. However, the best way to reduce the risk of pathogen transmission is to develop and use sterilization methods that do not adversely affect the functioning of the tissue after transplantation.

We are indebted to the clinicians, patients, and public health staff who reported cases, and to tissue-bank personnel who assisted in performing trace-backs and trace-forwards of tissues from implicated donors and shared data on microbiologic cultures and processing methods.

REFERENCES

1. Organ transplants and grafts, 1990 to 2000. No. 161. In: Statistical abstracts of the United States: 2003. Washington, D.C.: Census Bureau, 2002:113 (table).
2. Septic arthritis following anterior cruciate ligament reconstruction using tendon allografts — Florida and Louisiana, 2000. *MMWR Morb Mortal Wkly Rep* 2001;50:1081-3.
3. Shutkin NM. Homologous-serum hepatitis following the use of refrigerated bone-bank bone. *J Bone Joint Surg Am* 1954;36:160-2.
4. Hepatitis C virus transmission from an antibody-negative organ and tissue donor — United States, 2000–2002. *MMWR Morb Mortal Wkly Rep* 2003;52:273-4, 276.
5. Eggen BM, Nordbø SA. Transmission of HCV by organ transplantation. *N Engl J Med* 1992;326:411.
6. Conrad EU, Gretch DR, Obermeyer KR, et al. Transmission of the hepatitis-C virus by tissue transplantation. *J Bone Joint Surg Am* 1995;77:214-24.
7. Simonds RJ, Holmberg SD, Hurwitz RL, et al. Transmission of human immunodeficiency virus type 1 from a seronegative organ and tissue donor. *N Engl J Med* 1992;326:726-32.
8. Unexplained deaths following knee surgery — Minnesota, November 2001. *MMWR Morb Mortal Wkly Rep* 2001;50:1035-6.
9. Update: unexplained deaths following knee surgery — Minnesota, 2001. *MMWR Morb Mortal Wkly Rep* 2001;50:1080.
10. Update: allograft-associated bacterial infections — United States, 2002. *MMWR Morb Mortal Wkly Rep* 2002;51:207-10.
11. Brook I. Anaerobic bacterial bacteremia: 12-year experience in two military hospitals. *J Infect Dis* 1989;160:1071-5.
12. D'Angelo GL, Ogilvie-Harris DJ. Septic arthritis following arthroscopy, with cost/benefit analysis of antibiotic prophylaxis. *Arthroscopy* 1988;4:10-4.
13. Small NC. Complications in arthroscopic surgery of the knee and shoulder. *Orthopedics* 1993;16:985-8.
14. Ryan MJ, Kavanagh R, Wall PG, Hazleman BL. Bacterial joint infections in England and Wales: analysis of bacterial isolates over a four year period. *Br J Rheumatol* 1997;36:370-3.
15. Finegold SM, George WL, Mulligan ME. Anaerobic infections. *Dis Mon* 1985;31:1-97.
16. Ketterl R, Beckurts T, Kovacs J, Stubinger B, Hipp R, Claudi B. Gas-gangrene following arthroscopic surgery. *Arthroscopy* 1989;5:79-83.
17. Fitzgerald RH Jr, Rosenblatt JE, Tenney JH, Bourgault AM. Anaerobic septic arthritis. *Clin Orthop* 1982;164:141-8.
18. Horan TC, Gaynes RP, Martone WJ, Jarvis WR, Emori TG. CDC definitions of nosocomial surgical site infections, 1992: a modification of CDC definitions of surgical wound infections. *Infect Control Hosp Epidemiol* 1992;13:606-8.
19. Mangram AJ, Horan TC, Pearson ML, Silver LC, Jarvis WR. Guideline for prevention of surgical site infection, 1999. *Infect Control Hosp Epidemiol* 1999;20:250-78.
20. N.Y. State Department of Health, part 52 of title (Health) of the Official Compilation of Codes, Rules and Regulations of the State of New York, 1991 (amended 1993 and 2000).
21. Food and Drug Administration. 21 CFR part 1270: human tissue intended for transplantation: final rule. *Fed Regist* 1997;62(145):40429-47.
22. *Idem*. 21 CFR part 1271: current good tissue practice for manufacturers of human cellular and tissue-based products: inspection and enforcement: proposed rule. *Fed Regist* 2001;66(5):1507-59.
23. Woll JE, Kasprisin D. Standards for tissue banking. McLean, Va.: American Association of Tissue Banks, 2001.
24. Sterility tests. In: United States pharmacopeia, national formulary 2002 (USP 25–NF 20). Rockville, Md.: Pharmacopeial Convention, 2001:1878-83.
25. Whaley DN, Kainer MA, Archibald LK, Jernigan DB, Hageman J, Holmes HT. Allograft-associated *Clostridium sordellii* infection: enhanced detection using anaerobic blood culture bottles containing charcoal. Presented at the 103rd General Meeting of the American Society of Microbiology, Washington, D.C., May 18–22, 2003:117. abstract.
26. Harner CD, Olson E, Irrgang JJ, Silverstein S, Fu FH, Silbey M. Allograft versus autograft anterior cruciate ligament reconstruction: 3- to 5-year outcome. *Clin Orthop Relat Res* 1996;324:134-44.
27. Martinez OV, Malinin TI. The effect of postmortem interval and manner of death on blood and bone marrow cultures from non-septic cadaver donors of tissues for transplantation. In: Abstracts of the 96th General Meeting of the American Society for Microbiology, New Orleans, May 19–23, 1996:16.
28. Deijkers RL, Bloem RM, Petit PL, Brand R, Vehmeyer SB, Veen MR. Contamination of bone allografts: analysis of incidence and predisposing factors. *J Bone Joint Surg Br* 1997;79:161-6.
29. Gibbons MJ, Butler DL, Grood ES, Bylski-Austrow DI, Levy MS, Noyes FR. Effects of gamma irradiation on the initial mechanical and material properties of goat bone-patellar tendon-bone allografts. *J Orthop Res* 1991;9:209-18.
30. Rasmussen TJ, Feder SM, Butler DL, Noyes FR. The effects of 4 Mrad of gamma irradiation on the initial mechanical properties of bone-patellar tendon-bone grafts. *Arthroscopy* 1994;10:188-97.
31. Fideler BM, Vangness CT Jr, Lu B, Orlando C, Moore T. Gamma irradiation: effects on biomechanical properties of human bone-patellar tendon-bone allografts. *Am J Sports Med* 1995;23:643-6.
32. Loty B, Courpied JP, Tomeno B, Postel M, Forest M, Abelanet R. Bone allografts sterilized by irradiation: biological properties, procurement and results of 150 massive allografts. *Int Orthop* 1990;14:237-42.
33. Jackson DW, Windler GE, Simon TM. Intraarticular reaction associated with the use of freeze-dried, ethylene oxide-sterilized bone-patella tendon-bone allografts in the reconstruction of the anterior cruciate ligament. *Am J Sports Med* 1990;18:1-11.
34. Roberts TS, Drez D Jr, McCarthy W, Paine R. Anterior cruciate ligament reconstruction using freeze-dried, ethylene oxide-sterilized, bone-patellar tendon-bone allografts: two year results in thirty-six patients. *Am J Sports Med* 1991;19:35-41. [Erratum, *Am J Sports Med* 1991;19:272.]
35. Bianchi JR, Ross K, James E, Keesling J, Mills CR. The effect of preservation/sterilization processes on the shear strength of cortical bone. In: Vol. 42 of Proceedings of the 1999 Bioengineering Conference, Big Sky, Montana, June 16–20, 1999:407. abstract.
36. Summitt MC, Bianchi JR, Keesling JE, Roberts M, Mills CR. Biomechanical testing of bone treated through a new tissue cleaning process. In: Proceedings of the 25th Annual Meeting of the American Association of Tissue Banks, Washington, D.C., August 25–29, 2001:55. abstract.
37. *Idem*. Mechanical evaluation of soft tissue treated through a new tissue cleaning process. In: Proceedings of the 25th Annual Meeting of the American Association of Tissue Banks, Washington, D.C., August 25–29, 2001:54. abstract.
38. Kainer MA, Strausbaugh LJ, Liedtke LA, Jernigan DB, Archibald LK, IDSA Emerging Infections Network. An overview of allograft-associated infections (AAI) in the U.S. — 1998-2002. In: Proceedings of the 40th Annual Infectious Diseases Society of America (IDSA) Meeting, Chicago, October 24–27, 2002:46. abstract.
39. Mills AR, Roberts MR. Evaluation of culturing methods at predicting allograft sterility for aseptically processed tissue. In: Proceedings of the 25th Annual Meeting of the American Association of Tissue Banks, Washington, D.C., August 25–29, 2001:49. abstract.
40. Veen MR, Bloem RM, Petit PL. Sensitivity and negative predictive value of swab cultures in musculoskeletal allograft procurement. *Clin Orthop* 1994;300:259-63.

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CORRECTION

**Clostridium Infections Associated with
Musculoskeletal-Tissue Allografts**

Clostridium Infections Associated with Musculoskeletal-Tissue Allografts . On page 2566, under Results, the first line should have read, "Between January 1998 and March 2002," rather than "March 2003," as printed. On page 2569, in Table 3, the last two rows in the second column (under Positive Standard Blood-Culture Bottles) should have read, "0/3 (0)," rather than "0/0," as printed. And on page 2571, reference 1 should have read, "Organ transplants and grafts, 1990 to 2001," rather than "1990 to 2000," as printed.

CORRECTION

Correction: Infections and Musculoskeletal-Tissue Allografts

To the Editor: Our article on clostridium infections associated with musculoskeletal-tissue allografts (June 17 issue)¹ describes an epidemiologic investigation that was initiated by the Division of Healthcare Quality Promotion, Centers for Disease Control and Prevention (CDC), in November 2001 and concluded in April 2002. Dr. Marion A. Kainer carried out the investigation and was supervised by Dr. Lennox K. Archibald. A manuscript describing the investigation was completed in August 2002. After clearance by the director of the Division of Healthcare Quality Promotion, the manuscript was submitted to the *Journal* for publication in December 2002. There were no changes to the discussion relating to the BioCleanse process, which was mentioned in the article, between the time the manuscript was submitted and publication.

On January 20, 2003, Dr. Archibald became an employee of Regeneration Technologies, the manufacturer of BioCleanse. Stock options were granted to Dr. Archibald subsequent to his employment at Regeneration Technologies, subject to a vesting program over a period of five years.

The article was accepted for publication, pending revision, by the *Journal* in March 2004. Dr. Archibald signed a financial-disclosure form on August 1, 2003, attesting in good faith that the investigation had been conducted and completed while he was employed by the CDC. Dr. Archibald did not indicate on that financial-disclosure form that he was now employed by Regeneration Technologies. In March 2004, Dr. Archibald orally discussed his new affiliation with staff at the *Journal*. It was his understanding that no further revision of his financial disclosure was required. Although Dr. Kainer noted in revised manuscripts that Dr. Archibald was no longer affiliated with the CDC and was now working for Regeneration Technologies, this point was not separately addressed in an accompanying letter to the editor, and Dr. Archibald's new affiliation failed to appear in the publication.

We regret any perception of impropriety that might have resulted from Dr. Archibald's subsequent employment with Regeneration Technologies after his tenure with the CDC.

At no stage has Dr. Kainer been a paid expert witness on behalf of any tissue bank. However, after having left the CDC she was retained late in 2002 as an expert witness on behalf of patients affected by Tissue Bank A and shareholders who are filing a class-action lawsuit. In the article, per CDC policy, the use of trade names and commercial sources is for identification only and does not imply endorsement by the Department of Health and Human Services.

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Editor's note: When we publish a research report, our policy is to disclose to readers any relevant financial ties of the authors. To accomplish that, we rely on disclosure forms that all authors complete and sign. It is essential that we receive this information in writing; we cannot rely on telephone communication. In this case, Dr. Archibald's disclosure form, completed on August 1, 2003, stated that he had no relevant financial associations. Specifically, his form did not indicate that after the research was completed, he became an employee of Regeneration Technologies, an association that is relevant because Regeneration Technologies makes BioCleanse, a product that is mentioned in the article. It is our policy that disclosure forms must reflect the most current information. If this author's new affiliation had been indicated on the disclosure form, it would have been printed in the article according to our policy. The above letter with the financial disclosure has been linked permanently to the article as a correction, both on the *Journal* Web site and in the Medline database.

References

1. Kainer MA, Linden JV, Whaley DN, et al. Clostridium infections associated with musculoskeletal-tissue allografts. *N Engl J Med* 2004;350:2564-2571.