Intra-Operative Culturing of Donor Allograft Bone: A Lack of Clinical Utility

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Abstract

A common procedure for donor allograft bone includes intraoperative sampling of the allograft for bacterial culture. The goal of this study was assess the clinical effectiveness of intra-operative cultures in identifying microorganisms in donor bone allografts. Retrospective data were collected from 205 patients (230 allografts) to assess the clinical effectiveness of intra-operative cultures in identifying microorganisms in donor bone allografts. Eight of 230 allografts were positive for growth of microorganisms. Seven patients with a positive allograft culture exhibited no signs of surgical site infection during their hospitalization or clinical follow-up. One patient (positive for Staphylococcus similans) developed a clinical post-operative infection which cultured Enterococci. No association between positive intra-operative allograft cultures and the development of post-operative clinical infection was identified. These results fail to support routine intra-operative culturing of commercially supplied sterile allograft bone.

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Introduction

Allograft bone is used frequently to augment bone loss by providing a structural framework for host bone osteoconduction in orthopaedic procedures. There is a risk for disease transmission if the donor bone is contaminated; however, allograft bone used for many orthopedic procedures today undergoes a rigorous evaluation for the presence of microorganisms by the commercial supplier prior to acquisition and use by the hospital.[2-5,11] Previously reported contamination rates during the time between harvest and commercial sale can be as high as 22-24%, with *Staphylococcus epidermidis* being the most common organism.[4,12] However, multiple screening cultures (during processing and packaging) and the use of radiation and peroxide treatment prior to sale and distribution to hospitals are used today to effectively sterilize bone bank allografts.

The benefit of utilizing sterile versus aseptic allografts has been observed by a reduction in post-operative infections when using sterile allografts for patients undergoing anterior cruciate ligament reconstructive surgery.[6,8] However, even with rigorous commercial testing, allograft tissue may still undergo additional intra-operative sampling and culturing by hospital laboratories, the results of which would be unavailable for several days following surgery. This practice was most likely established as an internal check on the sterility of allograft processed within each institution and additional cultures have continued only as a double check to verify sterility. The present study was designed to examine the clinical effectiveness of intra-operative cultures in identifying microorganisms in commercially supplied, sterile allograft bone.

Materials and Methods

This study was conducted at an affiliated 450-bed community teaching hospital. Patients who underwent an orthopaedic surgical procedure requiring allograft bone use during surgery were

identified from a microbiology database. As per hospital standard, when a commercially acquired allograft was used or placed on the operative field during surgery, a sample was taken and placed in a sterile container. This sample was subsequently sent for (in-house) culture and sensitivity testing. The allograft was used during the operative procedure which always occurred prior to any knowledge of the culture results. In the microbiology laboratory, the allograft specimen was placed in thioglycollate broth only; no initial Gram stain was performed. The broth was examined visually each day for 7 days. If turbidity was not detected, the culture was reported as negative. If turbidity were detected, a Gram stain was performed and subcultures were initiated to identify the specific microorganism(s).

Retrospective data were collected from hospital records and included demographic data, type of procedure, surgeon, allograft type, pre-operative wound status, any culture results, and the presence of clinical infection during hospitalization and treatment. The positive culture results were recorded in reference to organism and time of growth post-operatively. Any patient identified as having a positive culture had their outpatient clinical charts reviewed for the presence of post-operative infection and had their identifying hospital number searched for readmissions to this institution within 225 days after surgery. Medical records identified for those readmissions within 225 days were reviewed to assess whether any readmission was due to a post-operative or surgical-site infection.

Data were tabulated and basic descriptive statistics performed using Microsoft Excel 2003 (Microsoft Corporation, Redmond, WA).

Results

Twenty-two orthopaedic surgeons and 5 neurosurgeons operated on 205 patients and used 230 allografts (crushed cancellous chips, 105; cortical strut, 42; femoral head, 39; bone-patellar tendon-bone, 27, tricortical iliac crest, 8; proximal femoral allograft, 2; distal femoral allograft, 2, achilles tendon allograft, 5). A minimum of one culture per allograft was obtained and multiple cultures were sent on several allografts (282 cultures).

Of the 230 allografts used in operative treatment of 205 patients, 8 (3.5%) were found to have bacterial growth and 1 of these 8 also had fungal growth (Table 1). No statistical association between the type of graft used and infection was identified. Six different surgeons each had 1 patient with a positive allograft culture and 1 surgeon had 2 patients with positive allograft cultures. Other than peri-operative prophylactic antibiotics, none of the 8

patients received antibiotics as a result of the positive allograft culture results. Seven of the 8 patients with positive allograft cultures had no clinical signs of hospital infection, no record of infection during their outpatient clinic follow-up, and no hospital readmissions related to infection of the surgical site were identified within a 225 day post-operative evaluation period. Only one patient out of 8 with positive allograft cultures, had a clinically recognized post-operative infection. This patient presented with a post-operative wound infection 21 days after surgery and underwent surgical site debridement. Cultures from tissue taken at time of debridement grew *Enterococcus faecalis*. The intra-operative allograft bone culture taken at the time of the original surgery on this patient grew *Staphylococcus simulans*.

Discussion

Our findings suggest that the use of intra-operative cultures of commercially-supplied allograft bone is a poor predictor for the clinical development of a post-operative infection. It seems logical that culturing the allograft intra-operatively would give the physician knowledge about a potentially infecting organism prior to the development of a post-operative infection. However, in this study there was only one post-operative infection among a small number of patients with a positive culture and the causative organism was different from the isolate cultured at the time of surgery. This finding is consistent with previous data indicating the information obtained from intra-operative allograft cultures would be of little clinical value in the assessment and management of this type of patient.[9,12,14] It is possible, that the absence of an observed positive culture with a corresponding allograft induced infection was due to a sample size not large enough to produce such an occurrence or antibiotic coverage in these cases.

Using current charges, the total amount billed to patients for these cultures would be approximately $36,800.00. If the money saved in early identification of a post-operative infective organism exceeds the total cost of all cultures performed in the screening, then culturing would be supported from a cost analysis perspective alone. However, in our study culturing allografts at the time of surgery was not predictive of post-operative wound infection.

Multiple studies have illustrated that during the process of bone graft harvest, there is a relatively high contamination rate.[5,7,12] Removal of this contamination has been achieved during the processing and preparation of bone allografts by

Pt	Allograft Type	Culture Growth	Clinical Infection
1	Crushed cancellous chips	Coagulase negative *Staphylococcus*, day 4	None
2	Cortical strut	Coagulase negative *Staphylococcus*, day 4	None
3	Femoral head	*Staphylococcus simulans*, day 7	Surgical-site, *Enterococcus faecalis*, ~21 days post-op
4	Femoral head	*S.* epidemeridis, day 3; diptheroids, day 5; *Alternaria*, day 7	None
5	Crushed chips	*Propionibacterium*, day 6	None
6	Crushed chips	*Propionibacterium*, day 6	None
7	Crushed chips	Diptheroids, day 5	None
8	Patellar tendon-bone ACL	*Enterococcus faecium*, day 2	None

vendors. They commonly utilize high pressure washes and peroxide soaks for bone graft that grew normal skin flora; irradiation, and automatic discard of any graft which grew *Clostridium* or fungus. In addition, all grafts are discarded in the event that they are positive for human immunodeficiency virus, hepatitis B, hepatitis C, human T-lymphocytic virus, and syphilis. A second culture is commonly taken during packaging process and, if positive, the tissue is reworked, irradiated, and repackaged.[4,5,9,13] A final culture is also taken of the allograft prior to the release to the hospital.[4,5,9] No product is released for distribution to hospitals until the final culturing shows no positive growth. The banking and allograft release procedures described by Liu et al., 2002 differ slightly from this methodology.[10] That study was a report from Taiwan and, as described, the bank providing allograft material did not perform a culture prior to release of a stored sample for use and the sample was implanted prior to any culture result.[10] They performed intraoperative cultures on 262 specimens taken at the time of allograft implant and identified a 4.6% positive finding.[10] Of the 12 patients implanted with allograft bone positive for culture, 9 developed postoperative allograft bone infection.[10]

Recent failures by vendors to sterilize implanted allografts have caused serious (even fatal) infections due to a variety of organisms, including Group B *Streptococcus, Clostridium* species*,* and a variety of Gram negative bacilli[1,3,11]. The vast majority of these cases were traced to a small number of donors and/or tissue banks. Nearly all of the cases involved soft tissue allografts (most commonly for ACL reconstruction), which were processed aseptically, but were not sterilized, in order to prevent damage to the tissue via the sterilization processes. In spite of these concerns, the recommendations from the Center for Disease Control were not changed to include the practice of culturing donor bone at the time of surgery.[3,9,14]

The current study was unable to identify any association between positive intra-operative allograft cultures and the development of post-operative clinical infection. These results fail to support continued, routine intra-operative culturing of allograft bone and such culturing likely leads to unnecessary costs and overall unwarranted clinical concern. Within our institution, these findings have been presented to the orthopaedic, neurosurgical, and infection control departments and our current policy was changed such that routine culturing is no longer performed on donor allograft bone.

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