Biomechanical and Histological Evaluation of Osteochondral Transplantation in a Rabbit Model

Ellis K. Nam,*† MD, Mohsen Makhsous,‡ PhD, Jason Koh,† MD, Mark Bowen,† MD, Gordon Nuber,† MD, and Li-Qun Zhang,‡ PhD

From the †Department of Orthopaedic Surgery, Northwestern Medical School, Chicago, Illinois, and the ‡Rehabilitation Institute of Chicago, Chicago, Illinois

Background: Biomechanical and histological properties of osteochondral transplantation have not been extensively examined.

Hypothesis: Osteochondral grafts have properties similar to native articular cartilage.

Study Design: Controlled laboratory study.

Methods: A 2.7 mm (diameter) × 4.0 mm (depth) osteochondral defect was created in 17 New Zealand white rabbit knees. An osteochondral graft, harvested from the contralateral knee, was transplanted into the defect. Eight rabbits were sacrificed each at 6 and 8 weeks.

Results: The 12-week grafts (1213.6 ± 309.0 N/mm) had significantly higher stiffness than the 6-week grafts (483.1 ± 229.1 N/mm; P < .001) and of normal cartilage (774.8 ± 117.1 N/mm; P < .003). Stiffness of the 6-week grafts was significantly lower than normal cartilage (P < .036). At all time points, full-thickness defects had significantly lower stiffness than normal cartilage (P < .001). Histologically, transplanted grafts scored significantly higher than the full-thickness defects (P < .001). The defects showed inconsistent, fibrocartilage healing. The grafts demonstrated cartilage viability, yet with a persistent cleft between the graft and host.

Conclusions: Osteochondral transplants undergo increased stiffness in the short term, with evidence of structurally intact grafts.

Clinical Relevance: Osteochondral transplantation may be a viable treatment option; however, long-term investigation on graft function is necessary.

Keywords: cartilage; osteochondral transplantation; biomechanic; histologic; knee

For centuries, it has been well known that damaged joint surfaces do not have the intrinsic ability to regenerate. In 1743, Hunter stated that cartilage “once destroyed, is not repaired.”39 Although much progress has been made in understanding the reasons behind this, we are incapable of regenerating true hyaline cartilage. Today, it is understood that cartilage lacks an intrinsic ability to heal completely secondary to the avascular environment, the immobility of chondrocytes, and the limited ability of mature chondrocytes to proliferate.9,29 The amount and quality of healing is dependent on the lesion size and depth of penetration.11,35 Furthermore, it has been suggested that in active individuals, untreated chondral defects may progress in size, leading to degenerative arthritis.4,20,24,37 Abrasion arthroplasty and drilling,28,33 although technically simple, promote a fibrocartilage healing response, which is biomechanically inferior to true hyaline cartilage. Chondrocyte transplantation, requiring 2 separate operative procedures, is demonstrating promise by replicating chondrocytes in culture and retransplanting them in the defect.5 Although some studies report a hyaline-like repair response,7 there are numerous treatment options for focal chondral defects, each having advantages and disadvantages.6,7 Abrasion arthroplasty and drilling,28,33 although technically simple, promote a fibrocartilage healing response, which is biomechanically inferior to true hyaline cartilage. Chondrocyte transplantation, requiring 2 separate operative procedures, is demonstrating promise by replicating chondrocytes in culture and retransplanting them in the defect.5 Although some studies report a hyaline-like repair response,7 one recent investigation reported a predominance of fibrocartilage healing with their technique.17 Transplantation of articular cartilage with allografts and autografts has received significant attention, as these techniques require one procedure with faster recovery than with chondrocyte transplantation.12,17 Osteochondral
Osteochondral transplantation in a rabbit model

allografts preclude donor site morbidity while allowing treatment of large defects. However, concern regarding chondrocyte viability as well as potential disease transmission has limited widespread use of this procedure. Osteochondral autograft transplantation, first described by Outerbridge et al., involves transferring an osteochondral graft (OG) from a nonweightbearing region to the chondral defect. This procedure has the theoretical advantage of increased cellular viability and no disease transmission relative to allografts. However, there is concern regarding donor site morbidity and limited availability of donor cartilage. Although osteochondral autograft transplantation is in clinical use, the actual weightbearing properties of transplanted grafts have not been determined. The purpose of this study was to determine how closely the biomechanical and histological properties of transplanted osteochondral autografts behave and resemble that of normal articular cartilage at different time intervals.

MATERIALS AND METHODS

Surgical Model

The following surgical protocol was approved by the Northwestern University Animal Care and Use Committee (Chicago, Illinois). Seventeen New Zealand white rabbits (Covance, Kalamazoo, Michigan) weighing between 3.2 and 3.6 kg underwent the following surgical procedure under anesthesia (ketamine 40 mg/kg IM; xylazine 5-7 mg/kg IM) supplemented with isoflurane (1%-2% inhalation) (Fig. 1). After shaving and steriley prepping both lower extremities, a 4-cm medial parapatellar arthrotomy was created in each knee, exposing the medial femoral condyle. Using a mosaicplasty harvester (Smith & Nephew, Memphis, Tennessee), a 2.7 mm (diameter) × 4.0 mm (depth) OG was harvested from the left medial femoral condyle, carefully minimizing disruption of the surrounding cartilage. The left knee served as the full-thickness defect (FD) group. The right medial femoral condyle was then debrided and dilated to the same dimensions of the harvested OG. The defect was carefully debrided of any remaining cartilaginous remnants, followed by careful placement of the OG into the defect. Special attention was made to apply the graft flush with the host cartilage surface. The capsule was closed with simple interrupted sutures (3-0 absorbable), followed by skin closure with a running subcuticular suture (4-0 absorbable). All subjects underwent a perioperative course of enterofloxin (5 mg/kg IM), with postoperative pain control using buprenorphine (0.05 mg/kg subcutaneously every 8-12 hours as needed × 7 days).

Figure 1. Surgical procedure: 2.7 mm (diameter) × 4.0 mm (depth) osteochondral graft harvested from medial femoral condyle of left knee (A), osteochondral graft (B), recipient site created on medial femoral condyle of right knee (C), and graft placed in recipient site (D).
Postoperatively, the subjects were allowed to ambulate freely without immobilization. One animal was sacrificed at 3 weeks due to a failure to thrive. There were no postoperative infections. Eight rabbits were sacrificed at 6 weeks, and 8 rabbits were sacrificed at 12 weeks using pentobarbital (100-150 mg/kg intravenously) with creation of bilateral pneumothoraces. In addition, 4 rabbits, which served as normal cartilage (NC), were sacrificed without operative intervention. All knees were harvested, wrapped with a moistened saline gauze, and stored at –20°C (4-7 days) until testing.

Biomechanical Testing

Biomechanical testing was performed on a total of 40 knees (8 NC, 8 OG each at 6 and 12 weeks, and 8 FD each at 6 and 12 weeks). The samples were slowly thawed overnight in an ice cooler, until room temperature equilibrium occurred. The femurs from each specimen were carefully stripped of all soft tissue, and the lateral femoral condyle was removed to prevent interference with the testing apparatus. The medial femoral condyle, containing the area of interest, was then potted in a cylindrical dowel using polymethylmethacrylate cement (Stryker/Howmedica, Allendale, New Jersey). The potted knee was rigidly mounted on the loading apparatus platform, which allowed motion in 3 orthogonal directions, permitting precise alignment of the cartilage surface perpendicular to the loading device. A force sensor and a linear potentiometer were attached to the testing apparatus (Fig. 2).

Standard indentation testing was performed, as previously described.16,26 Using a 1.9-mm diameter, cylindrical, plane-ended indenter fixed to the load assembly device, an applied force (up to 60 N) and displacement were recorded continuously for 30 seconds using a force sensor and potentiometer at a ramp speed of approximately 0.01 mm/s.

Histology

After biomechanical testing, the specimens were immediately fixed in 10% buffered neutral formalin and subsequently decalcified (RDO, hydrochloric acid). A total of 29 knees underwent histological analysis (7 OG at 6 weeks, 6 OG at 12 weeks, and 8 FD each at 6 and 12 weeks). Three osteochondral-transplanted knees were not included due to damage from the indentation studies. A sagittal section was made through the center of the repair site, and the 2 sides were embedded in paraffin. At least eight 5-µm sections were made, starting at the center of the defect, to minimize sampling error. The specimens were then stained with hematoxylin and eosin and examined under standard light microscopy (100×). Each specimen underwent analysis using a modified version of the O’Driscoll et al histological scoring system.30,31 Parameters used were the following (Table 1):

1. Nature of the predominant tissue: (1) cellular morphology (maximum, 4.0 points).
2. Structural characteristics: (1) surface regularity (maximum, 3.0 points), (2) structural integrity of the repair tissue (maximum, 2.0 points), (3) thickness of the repair tissue (maximum, 2.0 points), and (4) bonding of the repair tissue with the host cartilage (maximum, 2.0 points).
3. Freedom from cellular changes of degeneration: (1) degree of cellularity of the repair tissue (maximum, 3.0 points) and (2) chondrocyte clustering of the repair tissue (maximum, 2.0 points).
4. Freedom from degenerative changes in adjacent cartilage: (1) cellular characteristics in adjacent cartilage (maximum, 3.0 points) and (2) the presence of structural degenerative changes (modified), that is, fibrillation (maximum, 3.0 points).
5. Presence of fibrous tissue layer over OG: yes/no.
6. Presence of subchondral bone healing with host: yes/no.

The presence of fibrous tissue (5) and subchondral bone healing (6) were additional parameters we used in our evaluation of osteochondral transplantation. Subchondral bone healing was determined by noting the presence of bony trabecular interdigitation between the OG and the surrounding host subchondral bone.
A total indices of healing score was derived by the summation of categories 1 through 4. The maximum value was 24.0 points. Our analysis did not include evaluation of safranin-o staining, as originally described by O’Driscoll et al.\textsuperscript{30,31}

**TABLE 1**

Modified O’Driscoll Histological Score (maximum, 24.0 points)

<table>
<thead>
<tr>
<th>Category</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Nature of the predominant tissue</td>
<td></td>
</tr>
<tr>
<td>Cellular morphology</td>
<td></td>
</tr>
<tr>
<td>Hyaline articular cartilage</td>
<td>4</td>
</tr>
<tr>
<td>Incompletely differentiated mesenchyme</td>
<td>2</td>
</tr>
<tr>
<td>Fibrous tissue or bone</td>
<td>0</td>
</tr>
<tr>
<td>2. Structural characteristics</td>
<td></td>
</tr>
<tr>
<td>Surface regularity</td>
<td></td>
</tr>
<tr>
<td>Smooth and intact</td>
<td>3</td>
</tr>
<tr>
<td>Superficial horizontal lamination</td>
<td>2</td>
</tr>
<tr>
<td>Fissures: 25% to 100% of thickness</td>
<td>1</td>
</tr>
<tr>
<td>Severe disruption, including fibrillation</td>
<td>0</td>
</tr>
<tr>
<td>Structural integrity</td>
<td></td>
</tr>
<tr>
<td>Normal</td>
<td>2</td>
</tr>
<tr>
<td>Slight disruption, including cysts</td>
<td>1</td>
</tr>
<tr>
<td>Severe disintegration</td>
<td>0</td>
</tr>
<tr>
<td>Thickness</td>
<td></td>
</tr>
<tr>
<td>100% of normal adjacent cartilage</td>
<td>2</td>
</tr>
<tr>
<td>50% to 100% of normal cartilage</td>
<td>1</td>
</tr>
<tr>
<td>0% to 50% of normal cartilage</td>
<td>0</td>
</tr>
<tr>
<td>Bonding to the adjacent cartilage</td>
<td></td>
</tr>
<tr>
<td>Bonded at both ends of graft</td>
<td>2</td>
</tr>
<tr>
<td>Bonded at one end or partially at both ends</td>
<td>1</td>
</tr>
<tr>
<td>Not bonded</td>
<td>0</td>
</tr>
<tr>
<td>3. Freedom from cellular changes of degeneration</td>
<td></td>
</tr>
<tr>
<td>Hypocellularity</td>
<td></td>
</tr>
<tr>
<td>Normal cellularity</td>
<td>3</td>
</tr>
<tr>
<td>Slight hypocellularity</td>
<td>2</td>
</tr>
<tr>
<td>Moderate hypocellularity</td>
<td>1</td>
</tr>
<tr>
<td>Severe hypocellularity</td>
<td>0</td>
</tr>
<tr>
<td>Chondrocyte clustering</td>
<td></td>
</tr>
<tr>
<td>No clusters</td>
<td>2</td>
</tr>
<tr>
<td>&lt;25% of the cells</td>
<td>1</td>
</tr>
<tr>
<td>25% to 100% of the cells</td>
<td>0</td>
</tr>
<tr>
<td>4. Freedom from degenerative changes in adjacent cartilage</td>
<td></td>
</tr>
<tr>
<td>Cellular characteristics</td>
<td></td>
</tr>
<tr>
<td>Normal cellularity, no clusters</td>
<td>3</td>
</tr>
<tr>
<td>Normal cellularity, mild clusters</td>
<td>2</td>
</tr>
<tr>
<td>Mild or moderate hypocellularity</td>
<td>1</td>
</tr>
<tr>
<td>Severe hypocellularity</td>
<td>0</td>
</tr>
<tr>
<td>Fibrillation</td>
<td></td>
</tr>
<tr>
<td>No fibrillation</td>
<td>3</td>
</tr>
<tr>
<td>&lt;25% of articular thickness</td>
<td>2</td>
</tr>
<tr>
<td>25% to 50% of articular thickness</td>
<td>1</td>
</tr>
<tr>
<td>&gt;50% of articular thickness</td>
<td>0</td>
</tr>
<tr>
<td>5. Presence of fibrous tissue layer over osteochondral graft</td>
<td>yes/no</td>
</tr>
<tr>
<td>6. Presence of subchondral bone healing with host</td>
<td>yes/no</td>
</tr>
</tbody>
</table>

**Data Analysis**

Stiffness of the cartilage, defined as the ratio of the force required to displace the cartilage over a range of translation ($K = \Delta F/\Delta T$), was calculated. Analysis of variance (ANOVA) was used to test the null hypothesis that there was no difference in surface stiffness between normal articular cartilage, transplanted OGs, and FTDs at 6 and 12 weeks using a multiple comparisons procedure (SAS Institute, Cary, North Carolina). Furthermore, this procedure was used to determine whether there was any difference in the histological scores between the transplanted and untreated groups. The significance level was set at .05.

**RESULTS**

**Biomechanical Testing**

Stiffness values were reported within the linear portion of the force-displacement curve for all samples (overall $R^2 = 0.93 \pm 0.12; P < .05$). The stiffness of the transplanted OG group and FD group at 6 and 12 weeks, as well as the stiffness of the NC group, are given in Figure 3. The stiffness of the OG group at 12 weeks ($1213.6 \pm 309.0 \text{ N/mm}$) was significantly higher than the OG group at 6 weeks ($483.1 \pm 229.1 \text{ N/mm}; P < .001$) and that of NC ($774.8 \pm 117.1 \text{ N/mm}; P < .003$). The stiffness of the 6-week OG group was significantly lower than NC ($P < .001$).

**Gross Morphology**

There were no significant joint contractures in either group (OG and FD) for both time periods. On gross examination, the OG group at 6 and 12 weeks appeared to have a uniform appearance, with the grafts having a similar color and consistency to the surrounding articular cartilage (Fig. 4). The grafts were firm to palpation, similar to the surrounding host cartilage. No grafts lost fixation in this study, with the grafts remaining flush with the recipient cartilage surface for both time periods. There was no gross evidence of degenerative changes surrounding the transplanted region or the articulating tibial plateau. It was more difficult to localize the defect site in the 12-week OG group, as the graft margin tended to blend with the host site.

The FD groups, however, had wide variability in healing, both qualitatively and quantitatively, ranging from incomplete healing of the defect to an apparent exaggerated healing response with regenerate tissue expanding over the normal host articular cartilage (Fig. 4). The quality of the tissue was readily different in appearance and consistency to that of the surrounding articular cartilage. It was softer to palpation and had color ranging from a reddish-
brown hue to a whitish appearance. The surface also had an irregular, nonsmooth texture. Several of the specimens displayed evidence of degenerative changes surrounding the defect, which was more apparent in the 12-week group.

HISTOLOGY

Nature of the Predominant Tissue

Cellular Morphology. The nature of the predominant tissue was hyaline cartilage in 100% (13/13) of the OG groups at 6 and 12 weeks, whereas none of the FD specimens (0/16) healed with hyaline cartilage (Fig. 5). Eighty-eight percent (7/8) of the specimens from the 6-week FD group and 75% (6/8) of the specimens from the 12-week FD group healed with fibrocartilage. The remainder (12% and 25%, respectively) healed with fibrous tissue and bone only. The mean cellular morphology scores (mean ± SD) out of a maximum possible score of 4.0 from each group were the following: 6-week OG, 4.0 ± 0.0; 12-week OG, 4.0 ± 0.0; 6-week FD, 1.8 ± 0.7; and 12-week FD, 1.5 ± 0.9. At both time periods, the transplanted OG groups scored significantly higher than the FD groups (P < .001).

Structural Characteristics

Surface Regularity. The surface was smooth and intact (score, 3.0 points) in 100% (13/13) of the transplanted OG groups from both time periods. None (0/8) of the FD specimens from the 6-week group and 25% (2/8) from the 12-week FD group had an intact and smooth surface. Superficial lamination (score, 2.0 points) or fissures (score, 1.0 point) was seen in 88% (7/8) and 63% (5/8) of the FD groups at 6 and 12 weeks, respectively. Severe disruption (score, 0.0 points) was noted in 13% (2/16) of the FD groups for both time periods. Of a maximum possible score of 3.0 for surface regularity, the mean scores were the following: 6-week OG, 3.0 ± 0.0; 12-week OG, 3.0 ± 0.0; 6-week FD, 1.4 ± 0.7; and 12-week FD, 1.5 ± 1.1. The transplanted OG groups scored significantly higher than the FD groups for both time periods (P < .001).

Structural Integrity. The structural integrity of the tissue was normal (score, 2.0 points) in 100% (7/7) of the 6-week OG group, in 83% (5/6) of the 12-week OG group, in none (0/8) of the 6-week FD group, and in 13% (1/8) of the 12-week FD group. Slight disruption (score, 1.0 point) was noted in 17% (1/6) of the 12-week OG group, 75% (6/8) of the 6-week FD group, and 63% (5/8) of the 12-week FD group. Severe disintegration was noted in 25% (4/16) of the FD groups for both time periods. The mean scores, out of a maximum possible score of 2.0 from each group, were the following: 6-week OG, 2.0 ± 0.0; 12-week OG, 1.8 ± 0.4; 6-week FD, 0.6 ± 0.7; and 12-week FD, 0.9 ± 0.6. The transplanted OG groups scored significantly higher than the FD groups for both time periods (P < .001).

Cartilage Thickness. The transplanted OG group maintained the immediate post-transplant thickness in 100% (7/7) of the transplanted grafts at 6 weeks and in 83% (5/6) of the transplanted grafts at 12 weeks. The newly formed fibrocartilage in the FD group reconstituted the original cartilage thickness in 13% (1/8) and 25% (2/8) at 6 and 12 weeks, respectively. The mean scores, out of a maximum score of 2.0, were the following: 6-week OG, 2.0 ± 0.0; 12-week OG, 1.8 ± 0.4; 6-week FD, 0.6 ± 0.7; and 12-week FD,
0.6 ± 0.9. The transplanted OG groups scored significantly higher than the FD groups for both time periods (P < .003).

Bonding to Adjacent Cartilage. Bonding was defined as the histological appearance of perfect apposition between the OG and the host cartilage surface. No specimens from the transplanted OG groups demonstrated complete bonding with the host articular cartilage for both time periods. Twenty-nine percent (2/7) and 50% (3/6) of the specimens from the 6-week and 12-week transplanted OG groups, respectively, had bonding at 1 end of the graft. Out of a maximum possible score of 2.0, indicating complete apposition of the repair tissue, the mean scores were the following: 6-week OG, 0.3 ± 0.5; 12-week OG, 0.5 ± 0.5; 6-week FD, 1.1 ± 0.6; and 12-week FD, 0.8 ± 0.5. There was no significant increase in the amount of bonding from 6 to 12 weeks in the transplanted OG groups (P = .483).

Combined Scoring for Structural Characteristics. The maximum possible score for the quality of the graft or repair tissue was 9.0 points. The mean scores were the following: 6-week OG, 7.3 ± 0.5; 12-week OG, 7.2 ± 0.8; 6-week FD, 3.9 ± 1.1; and 12-week FD, 3.8 ± 2.0. The transplanted OG scored significantly higher than the FD for both time periods (P < .001).

Freedom From Cellular Changes of Degeneration

Cellularity. The degree of chondrocyte cellularity was normal in 86% (6/7) of the specimens from the 6-week OG group and in 67% (4/6) of the specimens from the 12-week OG group. Although there was a trend toward increased cellularity in the transplanted OG groups as compared to the FD groups, with our sample size, there was a significantly better score (P < .047) in only the 6-week OG group (2.9 ± 0.4) relative to the 12-week FD group (1.9 ± 1.2). However, a direct comparison between the OG and the FD groups was difficult due to the difference in cell types between the two groups (ie, chondrocytes versus fibrocytes). There was no significant difference (P = .709) from the 6-week OG group (2.9 ± 0.4) to the 12-week OG group (2.7 ± 0.5).

Clustering. The presence of chondrocyte clustering is a typical feature of osteoarthritic cartilage.1 Chondrocyte clustering was absent in 71% (5/7) of the specimens in the 6-week OG group and in 67% (4/6) of the specimens in the 12-week OG group. However, no specimens from the FD groups lacked chondrocyte clustering. For a best possible score of 2.0, the mean scores were the following: 6-week OG group, 1.7 ± 0.5; 12-week OG group, 1.7 ± 0.5; 6-week FD group, 0.4 ± 0.5; and 12-week FD group, 0.6 ± 0.5. The OG groups scored significantly better than the FD groups for both time periods (P < .001). Furthermore, there was no significant increase in chondrocyte clustering from the 6-week OG group to the 12-week OG group (P = .868).

Combined Score for Freedom From Degenerative Changes. Out of a best possible score of 5.0, the mean combined scores for the degree of degenerative changes in each group were the following: 6-week OG group, 4.6 ± 0.5; 12-week OG group, 4.3 ± 1.0; 6-week FD group, 2.6 ± 1.2; and 12-week FD group, 2.5 ± 1.7. The OG groups scored significantly better than the FD groups for both time periods (P < .015). Furthermore, there was no significant progression in the degree of degenerative changes with time in the OG groups (P = .728).

Freedom From Degenerative Changes in Adjacent Cartilage

Cellular Characteristics. Histological grading of the host cartilage was performed adjacent to the defect. The host cartilage from the OG group was normal in 86% (6/7) of the 6-week specimens and in 67% (4/6) of the 12-week OG group. However, only 13% (2/16) of the FD groups had normal-appearing cartilage adjacent to the defect for both time periods. Out of a best possible score of 3.0 for the degree of cellular degenerative changes, the mean scores were the following: 6-week OG, 2.9 ± 0.4; 12-week OG, 2.7 ± 0.5; 6-week FD, 1.8 ± 0.7; and 12-week FD, 1.6 ± 0.7. The OG groups scored significantly higher than the FD groups for both time periods (P < .011). In addition, the OG groups demonstrated no significant progression in cellular degeneration of the adjacent cartilage with time (P = .585).

Fibrillation. Of the OG groups, 86% (6/7) and 83% (5/6) of the specimens from the 6- and 12-week groups displayed no evidence of degenerative changes adjacent to the transplanted grafts. However, there were moderate or severe degenerative changes in 50% (8/16) of the FD groups. Out of a best possible score of 3.0, the mean scores were the following: 6-week OG, 2.9 ± 0.4; 12-week OG, 2.8 ± 0.4; 6-week FD, 1.5 ± 0.9; and 12-week FD, 1.4 ± 0.7. The OG groups scored significantly better than the FD groups for both time periods (P < .002). Furthermore, the OG groups demonstrated no significant changes in fibrillation of the surrounding cartilage with time (P = .950).

Total Degree of Degenerative Changes in Adjacent Cartilage. Out of a best possible score of 6.0, the mean scores assessing the degree of degenerative changes in the adjacent cartilage were the following: 6-week OG group, 5.7 ± 0.8; 12-week OG group, 5.5 ± 0.5; 6-week FD group, 3.3 ± 1.5; and 12-week FD group, 3.0 ± 1.2. At both time periods, the OG groups scored significantly better than the FD groups in the total degree of adjacent cartilage degeneration (P < .001).

Presence of Fibrous Tissue Layer

Thirty-three percent (2/6) of the specimens from the 12-week OG group had a thin, smooth, fibrous tissue layer overlying the intact graft. However, this layer was not intimately attached to the otherwise normal-appearing hyaline chondral surface of the transplanted grafts. Although none (0/7) of the 6-week transplanted OG group had a fibrous tissue layer, there was no significant difference (P < .057) between these 2 groups.

Subchondral Bone Healing

An additional parameter we chose to examine was the presence of subchondral bone healing with the host bone.
Combined histological scores between the two OG groups. Furthermore, there was no significant change in the total periods (P < .001).

For both time periods, all of the transplanted OGs (13/13) for both time periods demonstrated excellent trabecular interdigitation with the host subchondral bone.

**TOTAL INDICES OF HEALING**

Out of a maximum possible score of 24.0, the mean total indices of healing scores were the following: 6-week OG, 21.6 ± 1.3; 12-week OG, 21.0 ± 1.8; 6-week FD, 11.5 ± 2.8; and 12-week FD, 10.8 ± 4.4 (Fig. 6). The total combined histological scores were significantly better in the OG groups relative to the FD groups for both time periods (P < .001). Furthermore, there was no significant change in the total combined histological scores between the two OG groups (P = .729).

**DISCUSSION**

Resurfacing chondral defects is a great challenge to modern orthopaedics. Although the natural history of untreated defects is unknown, there appears to be evidence of arthritic progression for significant sized lesions. Osteochondral transplantation, although in clinical use today, has not undergone thorough biomechanical and histological analysis. In our study, there were consistently better results both morphologically and biomechanically in the transplanted OG groups as compared to the full-thickness osteochondral defect groups. Combined histological scores were significantly better for both time periods in the transplanted groups, as compared to the FTD groups (P < .001). There was incorporation of the OGs, as demonstrated by bony interdigitation with the host subchondral bone in all transplanted grafts. Although there was successful articular cartilage bonding with the host in several specimens, none of the transplanted grafts were completely bonded on all sides. These results are in agreement with a previous investigation. The original architecture of the grafts did not appear to deteriorate with time, maintaining their original hyaline cartilage appearance. In addition, there was no progressive degeneration in the surrounding host cartilage from 6 to 12 weeks. Thirty-three percent (2/6) of the transplanted grafts displayed a thin, fibrous membrane over the chondral surface in the 12-week group, which was not evident in the 6-week group (P < .057). Although this may represent a sign of degeneration, we do not feel this is the case as the underlying hyaline plug maintained its native smooth architecture without fibrillation, and the cellular morphology and distribution was unaltered.

The FTD group consistently displayed evidence of degenerative changes not only of the regenerate tissue but also of the articular cartilage adjacent to the created defect. Caution should be warranted, however, in interpreting our findings to the clinical situation. FTDs rarely involve deep penetration into the underlying subchondral bone as were in our study group. As we harvested the OGs from the FTD group, by necessity, significant subchondral bone was procured. Careful attention was made not to disrupt the remaining cartilage during graft procurement, however, it is likely that surrounding cartilage was damaged in several specimens, contributing to our histologically observed degenerative changes in the FTD group. However, as seen in multiple previous investigations, our results are in agreement with the natural history of fibrocartilaginous healing in untreated defects. There was a highly variable healing response, as many specimens demonstrated incomplete healing, whereas others had an exaggerated healing response, invading the adjacent normal articular milieu.

A recent biomechanical investigation on osteochondral transplantation in a goat model concurs with our findings of increased stiffness of transplanted grafts relative to normal hyaline cartilage. Previous explanations to this phenomenon have been possible mismatch between donor and recipient cartilage thickness and a compaction of the cartilage as a result of the implantation process. To evaluate whether cartilage thickness mismatch is a cause of this increased stiffness, we harvested our grafts from the same location as our host site, the medial femoral condyle, and still generated increased stiffness. Furthermore, our histological analysis did not detect any change in cartilage thickness as a result of the surgical technique. Thus, differences in neither cartilage thickness nor the transplantation process appear to account for this discrepancy.

In a rabbit model, Makino et al performed histological evaluation of osteochondral autografts that were harvested and reinserted in the donor site without changing graft orientation. Interestingly, they found increased thickness in their OGs, relative to the surrounding cartilage up to 24 weeks postoperatively. Although they did not perform biomechanical evaluation, they theorized that progressive increases in cartilage thickness may account for stiffness alterations. We propose that increased stiffness in osteochondral transplants may be attributed in part to bony callus formation analogous to fracture healing. During fracture repair, an abundance of callus is initially formed, followed over time with resorption and remodeling.
et al, in a rat model, demonstrated that callus undergoes a progressive increase in stiffness with time. Although there was a significant increase in stiffness with time, our maximal time period was 12 weeks; thus, we could not determine whether stiffness values normalize. Longer-term studies of osteochondral transplantation may help answer this finding. Trabecular interdigitation of the transplanted OGs with host subchondral bone was evident; however, quantitative subchondral bone density measurements were not performed; thus, we were unable to provide conclusive evidence, histologically.

One weakness of our biomechanical testing methodology was the large diameter probe (1.9 mm) used in our measurements. As the OGs were 2.7 mm in diameter, potential remains for inaccurate representation of the biomechanical variability in the properties between the center and edges of the graft. Furthermore, as stiffness is dependent on sample geometry, reporting our results in terms of modulus would have more accurately created a valid assessment between the OGs and the FTDs. However, as our OGs were geometrically equivalent, we feel our comparison of the OGs from 6 to 12 weeks is valid. Finally, stiffness, as opposed to modulus, was used to compare our results with the reported biomechanical study of osteochondral grafting in a goat model.

As mentioned, another weakness of this study is the short time period (12 weeks). Although we found significant findings, that is, increased stiffness in the transplanted OGs as well as graft viability, the ultimate mechanical and structural fate of transplanted grafts can only be determined with longer-term studies. Furthermore, with our limited time point, the effects of a persistent cleft between the OG and host cartilage, as well as potential cartilage degeneration, cannot be determined. Horas et al recently compared their results of autologous chondrocyte implantation with osteochondral transplantation. With a 2-year follow-up, they found similar clinical results using the Myers score and the Tegner activity score. Histologically, biopsy specimens of the autologous chondrocyte implantation group were mainly fibrocartilage, with locally, biopsy specimens of the autologous chondrocyte implantation group were mainly fibrocartilage, with locally, biopsy specimens of the autologous chondrocyte implantation group were mainly fibrocartilage. Additionally, the osteochondral transplants demonstrated hyaline cartilage viability with no degeneration; however, there remained a persistent cleft between the graft and the surrounding cartilage.

CONCLUSION

Our biomechanical and histological results suggest that osteochondral transplantation is a promising method of resurfacing cartilage defects. Long-term studies are necessary to determine whether normalization of OG stiffness occurs and the effects on graft functioning. Although short-term data suggest graft viability, further investigation is necessary to exclude long-term degeneration.

REFERENCES


