Clinical, Radiographic, and Histological Outcomes After Cartilage Repair With Particulated Juvenile Articular Cartilage

A 2-Year Prospective Study

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Background: Biological repair of cartilage lesions remains a significant clinical challenge because of the lack of natural regeneration and limited treatment options.

Hypothesis: Treatment of articular cartilage lesions in the knee with particulated juvenile articular cartilage (PJAC) will result in an improvement in patient symptoms of pain and function and magnetic resonance imaging (MRI) findings at 2 years compared with baseline.

Study Design: Case series; Level of evidence, 4.

Methods: Patients with symptomatic articular cartilage lesions on the femoral condyles or trochlear groove of the knee were identified for treatment with PJAC. There were 25 patients with a mean age of 37.0 ± 11.1 years and a mean lesion size of 2.7 ± 0.8 cm^2. All patients were assessed preoperatively (baseline) with a knee examination and surveys including the International Knee Documentation Committee (IKDC) subjective knee form, 100-mm visual analog scale (VAS) for pain, and Knee injury and Osteoarthritis Outcome Score (KOOS). Patients were followed at predetermined time points postoperatively through 2 years. Also, MRI was performed at baseline and at 3, 6, 12, and 24 months. At 2 years, patients were given the option of undergoing voluntary diagnostic arthroscopic surgery with cartilage biopsy to assess the histological appearance of the cartilage repair including safranin O staining for proteoglycans and immunostaining for type I and II collagen.

Results: Clinical outcomes demonstrated statistically significant increases at 2 years after surgery compared with baseline, with improvements seen as early as 3 months. Over the 24-month follow-up period, the IKDC score increased from a mean of 45.7 to 73.6, KOOS-pain score from 64.1 to 83.7, KOOS-symptoms score from 64.6 to 81.4, KOOS–activities of daily living score from 73.8 to 91.5, KOOS–sports and recreation score from 44.6 to 68.3, and KOOS–quality of life score from 31.8 to 59.9. The MRI results suggested that T2-weighted scores were returning to a level approximating that of normal articular cartilage by 2 years. Histologically, the repair tissue in biopsy samples from 8 patients was composed of a mixture of hyaline and fibrocartilage; immunopositivity for type II collagen was generally higher than for type I collagen, and there appeared to be excellent integration of the transplanted tissue with the surrounding native articular cartilage. Other than elective biopsies, there were no reoperations, although 1 graft delamination was reported at 24 months.

Conclusion: This study demonstrates a rapid, safe, and effective treatment for cartilage defects. For the patient population investigated, the clinical outcomes of the PJAC technique showed a significant improvement over baseline, with histologically favorable repair tissue 2 years postoperatively.

Keywords: cartilage repair; juvenile cartilage; chondral repair; articular cartilage injury; DeNovo NT Natural Tissue Graft; particulated cartilage

It is well established that adult cartilage lesions have a severely impaired ability to heal, and as a result, surgical intervention is considered when these lesions are associated with pain and loss of function. Multiple authors have drawn the same conclusion regarding currently available cartilage treatments: they all have limitations. As a result, the search for new cartilage repair options continues. A recently introduced option is particulated juvenile articular cartilage (PJAC) (DeNovo NT Natural Tissue Graft, Zimmer Inc, Warsaw, Indiana, USA), which consists of allograft articular cartilage from donors younger than 13 years old that has been cut into approximately 1-mm cubes. It is applied to cartilage lesions in a monolayer and held in place with the use of

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fibrin sealant, as described in detail by Farr and Yao.[8] In vitro studies on the differences between juvenile and adult cartilage have demonstrated a good potential for repair.[1,11] While PJAC has been in clinical use since 2007, with over 7000 surgical implants, there have been little prospective clinical data available. Only a few cases have been reported (1 patellar lesion in Bonner et al[2] and 15 patellar treatments in 13 patients in Tompkins et al[12]), including a small cohort from the current study.[8] In light of the increased clinical adoption, prospective clinical studies such as this are important to aid clinicians in making proper treatment decisions. This report describes the first prospective study that evaluates patients at 2 years after PJAC implantation.

The purpose of this study was to evaluate the safety and clinical outcomes in a prospective cohort of patients. The study hypothesis was that patients would experience improvements in symptoms of pain and function at 2 years after surgery compared with baseline.

MATERIALS AND METHODS

Study Population and Study Design

A prospective series of 25 patients were enrolled at 3 clinical centers by 3 surgeons for elective treatment of symptomatic cartilage lesions in the knee (NCT00791245). Institutional review board approval was obtained at each center, and informed consent was obtained from each patient before any study-related activities were performed. Eighteen men and 7 women participated in the study. The inclusion criteria for the study were (1) symptomatic, focal, contained chondral lesions (International Cartilage Repair Society [ICRS] grade 3 or 4A) (osteochondritis dissecans) of the femur (condyles and trochlea), with defect areas ranging between 1 and 5 cm^2 after debridement; and (2) patient ages between 18 to 55 years with a willingness to participate in the study and sequential follow-up. Concomitant ligament or meniscal injuries were repaired simultaneously. Patients were excluded for a body mass index (BMI) >35 kg/m^2, the presence of bipolar lesions, prior total meniscectomy of either knee, failed microfracture within 12 months of surgery, or radiographic mechanical axis malalignment $>5°$. Additionally, patients with a disease diagnosis by clinical and/or radiographic assessment including osteoarthritis, rheumatoid arthritis, gout, or avascular necrosis were excluded; those receiving workers' compensation for their injury were also excluded.

Consenting patients were asked to carry out a preoperative visit that included the International Knee Documentation Committee (IKDC) knee examination performed by a qualified medical professional and the collection of data including demographics, knee and medical history, and medication usage. Patients also underwent preoperative magnetic resonance imaging (MRI) and radiographs and completed surveys. After surgery, patients returned for follow-up at 10 days and then at 3, 6, 12, 18, and 24 months, at which time the IKDC knee examination was performed and surveys collected. Postoperative MRI was performed at 3, 6, 12, and 24 months; radiographs were taken at all follow-up visits. Patients were queried at all follow-up visits regarding any complications or events that they may have experienced since the last visit, including events unrelated to the knee.

Surgical Technique

The surgical technique has been previously described by Farr and Yao.[8] Defects were surgically prepared after a medial or lateral parapatellar mini-arthrotomy by curettage to create a well-defined vertical defect perimeter. The calcified cartilage layer was carefully removed, taking care not to violate the subchondral cortical bone plate; that is, the defect was prepared the same as for marrow stimulation techniques before hole formation. After preparation of the lesion, the lesion size was measured with a graduated probe to calculate the number of vials of PJAC needed (1 vial per 2.5 cm^2). Sterile aluminum foil was pressed into the lesion to form a mold. The PJAC pieces were evenly spread in the foil mold in a monolayer with pieces within 1 to 2 mm of one another. Any residual (liquid) transport media were removed, and a thin layer of fibrin glue was applied on the pieces at the bottom of the mold. The fibrin PJAC construct did not exceed three quarters of the depth of the lesion. Fibrin was allowed to set for 5 to 10 minutes to form the implant. A thin layer of fibrin was then applied to the base of the patient's cartilage defect, and the cartilage/fibrin glue construct was placed into the defect and gently held in place until the glue cured to ensure good fill and adherence of the tissue within the defect. No additional membrane or patches were applied to the repair site. The final construct was shallower than the surrounding defect "shoulders."

Postoperative rehabilitation was conducted through a standardized protocol. For the first 2 postoperative weeks, patients were nonweightbearing in full extension using a knee immobilizer for condylar lesions and weightbearing as tolerated in full extension for isolated trochlear lesions. Continuous passive motion was performed for 6 to 8 hours per day during that time period. Foot-flat weightbearing was allowed for condylar lesions between 2 and 6 weeks, and progression to full weightbearing occurred between 6 and 12 weeks postoperatively.

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Clinical Assessment

Safety and efficacy outcomes included the Knee injury and Osteoarthritis Outcome Score (KOOS, including all subdomains of pain, symptoms, activities of daily living [ADL], function in sports and recreation, and knee-related quality of life [QoL]), IKDC knee physical examination, 2000 IKDC subjective knee evaluation, IKDC current health assessment (which includes the Short Form–36 [SF-36]), Marx Activity Scale, and pain intensity measured on a 100-mm visual analog scale (VAS). In addition to these measures, the incidence of postoperative complications and adverse events was collected at each follow-up visit. All patients were approached and asked whether they would undergo optional, voluntary diagnostic arthroscopic surgery at 2 years. All patients who consented for voluntary arthroscopic surgery underwent the procedure within 2 months of their 24-month follow-up time point. During the voluntary arthroscopic procedure, the ICRS cartilage repair assessment was performed by the principal investigator at that site, and then a 2 mm– to 3 mm–diameter biopsy specimen was taken from the center of the area of the cartilage repair, including the entire depth of the articular cartilage as well as several millimeters of subchondral bone.

Histology and Immunochemistry

The biopsy specimens were fixed in 10% neutral buffered formalin, briefly decalcified in 10% ethylenediaminetetraacetic acid (EDTA), processed into paraffin, and embedded. Serial 5 μm–thick sections were stained with hematoxylin and eosin (H&E), Masson trichrome, and safranin O. Immunohistochemistry sections were also prepared using antibodies against type I and type II collagen. Briefly, sections were deparaffinized, rehydrated to distilled water, treated with proteinase K for type I collagen staining or pepsin for type II collagen staining, blocked with undiluted protein block (DAKO Envision, DAKO, Glostrup, Denmark) at room temperature, and then treated with mouse monoclonal COL-1 (Sigma Catalog #c2456, Sigma-Aldrich, St Louis, Missouri, USA) at 1:100 for 30 minutes at room temperature or mouse monoclonal CIICI (Hybridoma Bank, Iowa City, Iowa, USA) at 1:10 for 60 minutes at room temperature. Sections were treated with secondary antibody (DAKO Rabbit Envision +) for 30 minutes, followed by horseradish peroxidase for 30 minutes. The reaction product was detected with 3,3′-diaminobenzidine (DAB), and slides were counterstained in Mayer hematoxylin (DAKO). In negative control sections, normal rabbit serum was substituted for the primary antibody. Positive control sections included sections of normal human articular cartilage (for type II collagen) and subchondral bone (for type I collagen).

Each section was carefully evaluated, and the following articular cartilage parameters were graded on a scale of 0 to 4: fibrillation, cellularity, and chondrocyte necrosis/matrix degeneration (H&E); fibrillar versus hyaline character of the matrix (Masson trichrome); loss of safranin O staining (safranin O); and immunopositivity for type I or II collagen in the respective immunostained sections. For each parameter, a score of 0 represents the situation most similar to native cartilage (eg, lack of fibrillation), with the exception of immunopositivity, which is graded from 0 (no immunopositivity) to 4 (marked immunopositivity). Histological grading was performed by an experienced board-certified (American College of Veterinary Pathologists) veterinary pathologist according to previously published methods.[10]

MRI Assessment

Patients were assessed by MRI using a standardized MRI protocol at all study time points using a minimum 1.5-T MRI machine (Toshiba, Irvine, California, USA). The MRI protocol consisted of sagittal and coronal fast spin echo imaging with and without fat suppression. The matrix size was 512 × 256 pixels, with a field of view of 14 to 16 cm and a slice thickness of 3.5 mm. A sagittal 3-dimensional spoiled gradient echo imaging sequence was also acquired as well as a sagittal 4-echo T2 spin echo for calculation of T2-weighted maps. The total MRI examination time was about 40 minutes.

Morphological evaluation, qualitative scoring of the defect site, and T2 mapping of the repair site were undertaken by an experienced radiologist. The percentage of lesion fill was calculated by measuring the volume of fill tissue divided by the volume of the debrided cartilage defect on the 3-month MRI scans. The volume of the defect or fill tissue was calculated by measuring the area on individual MRI slices and multiplying by slice thickness to calculate the volume per slice. These volumes were summed across the defect or tissue to obtain the total volume. Measurements were performed twice by an experienced independent radiologist, once in the coronal plane and once in the sagittal plane, and the mean volume taken. Slice gaps were calculated by interpolation. The T2 relaxation times in the fill tissue were calculated by fitting the data from a region of interest to a single exponential. Qualitative image scoring for the extent of high T2 signal areas in the fill tissue, degree of graft hypertrophy, and bone marrow edema was assessed on a scale of 0 to 3, where 0 = none, 1 = mild, 2 = moderate, and 3 = severe.

Safety Assessment

Adverse events, including adverse reactions, were collected throughout the study follow-up period. Patients were queried at every visit regarding any complications that they may have experienced since their last visit. Additionally, physical examinations, during surgery and follow-up, and any laboratory tests ordered by the investigator were used to collect adverse event information. Treatment site complications and adverse events were assessed with regard to event severity (mild, moderate, or severe), relationship to the procedure, relationship to the study implant, and any anesthetic agents.

Statistical Analysis

Continuous variables (age, BMI, KOOS subscore, etc) were summarized by descriptive statistics. Categorical variables (sex, defect location, ICRS grade, etc) were summarized by

TABLE 1
Baseline Patient Characteristics (N = 25 Patients)[a]

Characteristics	Values
Age, mean ± SD, y	37.0 ± 11.1
Height, mean ± SD, inches	69.0 ± 3.9
Weight, mean ± SD, lb	178.4 ± 37.1
BMI, mean ± SD, kg/m^2	25.6 ± 3.4
Sex	
Male	18 (72)
Female	7 (28)
Injury onset	
Nontraumatic	
Gradual	11 (44)
Acute	7 (28)
Traumatic	
Contact	1 (4)
Noncontact	6 (24)
Duration since onset, mean ± SD, y	2.6 ± 4.5
Previous knee surgeries	
0	10 (40)
1	9 (36)
≥2	6 (24)
Cartilage resurface/reconstruction	13 (52)

[a]Values are expressed as n (%) unless otherwise indicated. BMI, body mass index.

TABLE 2
Lesion Characteristics (N = 25 Patients, 29 Lesions)[a]

Characteristics	Values
Defect size after debridement, mean ± SD, cm^2	2.7 ± 0.8
No. of patients with 2 lesions	4
Defect location	
Femoral condyle	18 (62.1)
Trochlea	11 (37.9)
ICRS grade	
3A	2 (6.9)
3B	3 (10.3)
3C	16 (55.2)
3D	2 (6.9)
4A	6 (20.7)

[a]Values are expressed as n (%) unless otherwise indicated. ICRS, International Cartilage Repair Society.

counts and percentages per nonmissing categories. The Wilcoxon signed-rank test was used to determine the statistical significance for change from baseline values. *P* values were not adjusted for multiplicity.

RESULTS

Patient Demographics

A total of 29 lesions were treated in 25 patients (Table 1), with 4 patients having 2 lesions treated in the index knee. The mean patient age at the time of surgery was 37.0 years (range, 18.0-56.0 years), and the overall mean lesion size was 2.7 ± 0.8 cm^2 (range, 1.2-4.6 cm^2). Forty-eight percent of the lesions were in the medial femoral condyle (14/29), 13.8% in the lateral femoral condyle (4/29), and the remainder (37.9%; 11/29) in the trochlea (Table 2). Four patients underwent concomitant partial meniscectomies. There were no concomitant ligament repairs or replacement procedures. The most commonly reported symptom onset was atraumatic and gradual (44%; 11/25), followed by nontraumatic acute onset (28%; 7/25), traumatic noncontact (24%; 6/25), and traumatic contact (4%; 1/25). The mean time from symptom onset to surgery was 2.6 ± 4.5 years (range, 0.12-20.7 years). The mean number of prior surgeries performed on the index knee was 1.1, with 10 patients reporting debridement and 8 reporting microfracture procedures. Ten patients underwent no prior surgical procedures on the index knee.

Clinical Outcomes

The IKDC score increased from a preoperative mean of 45.7 ± 15.9 to 73.6 ± 14.1 at 24 months (Figure 1). All

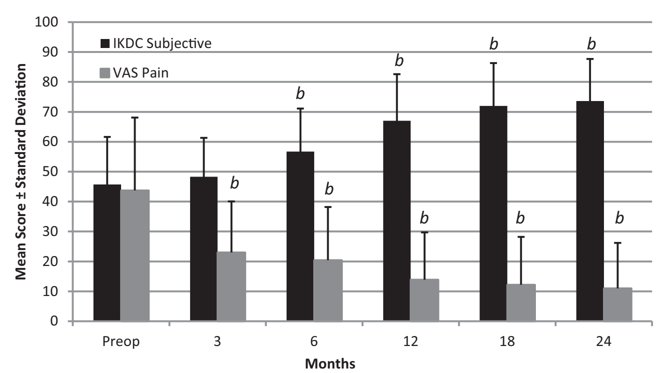

Figure 1. International Knee Documentation Committee (IKDC) subjective knee evaluation scores and visual analog scale (VAS) scores for pain for patients over a 24-month period after graft implantation. A gradual, consistent improvement in knee function and a decrease in pain are seen for patients treated with DeNovo NT. [b]Mean change from the preoperative evaluation was statistically significant ($P < .001$).

intermediate assessment time points for the IDKC showed a statistical difference ($P < .001$) with the exception of the 3-month time point. The mean change from baseline increased from 2.6 ± 18.9 at the 3-month time point to 27.0 ± 21.0 at 24 months ($P < .001$). The IKDC frequency of pain decreased from a preoperative mean value of 6.3 ± 2.7 to 2.7 ± 2.1 at 24 months ($P < .001$), and severity of pain decreased from 5.5 ± 2.6 to 3.0 ± 1.9 at 24 months ($P < .05$). The VAS pain score (Figure 1) decreased from a preoperative mean of 43.7 ± 24.4 to 11.1 ± 15.2 by 24 months ($P < .001$). The IKDC/SF-36 physical and mental component scores and Marx Activity Scale results did not show statistically significant changes over the course of the study (see the Appendix, available in the online version of this article at http://ajsm.sagepub.com/supplemental).

The KOOS-pain score increased from a preoperative mean of 64.1 ± 16.4 to 83.7 ± 10.5 by 24 months ($P < .001$), KOOS-symptoms score from a preoperative

TABLE 3
Unadjusted Patient-Reported KOOS Subscores[a]

Domain	Baseline	3 mo	12 mo	24 mo
Pain	64.1 ± 16.4	76.6 ± 11.6[b]	82.8 ± 9.4[b]	83.7 ± 10.5[b]
Symptoms	64.6 ± 17.2	73.7 ± 13.6[b]	79.9 ± 10.4[b]	81.4 ± 11.3[b]
ADL	73.8 ± 16.2	83.7 ± 10.7[b]	91.0 ± 8.4[b]	91.5 ± 10.6[b]
Sports/recreation	44.6 ± 25.9	42.5 ± 29.8	64.9 ± 18.6[b]	68.3 ± 20.5[b]
QoL	31.8 ± 19.2	43.5 ± 16.7[b]	57.3 ± 20.5[b]	59.9 ± 20.7[b]

[a]Values are expressed as mean ± standard deviation. ADL, activities of daily living; KOOS, Knee injury and Osteoarthritis Outcome Score; QoL, knee-related quality of life.
[b]Mean change from baseline is statistically significant ($P < .05$).

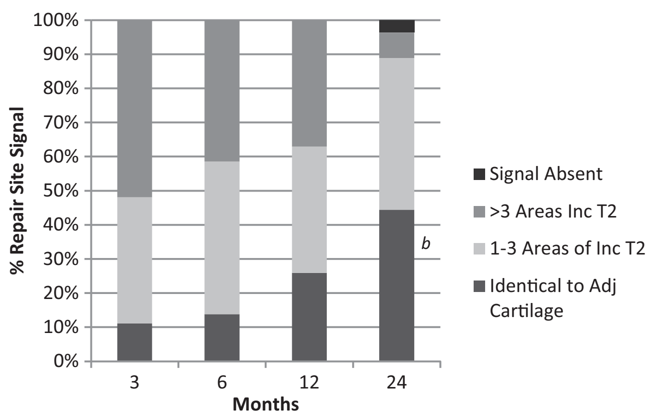

Figure 2. T2 signal grading of magnetic resonance imaging. [b]Statistically significant compared with 3 months ($P < .05$).

mean of 64.6 ± 17.2 to 81.4 ± 11.3 ($P < .001$), KOOS-ADL score from 73.8 ± 16.2 to 91.5 ± 10.6 ($P < .001$), KOOS–sports and recreation score from 44.6 ± 25.9 to 68.3 ± 20.5 ($P < .001$), and KOOS-QoL score from 31.8 ± 19.2 to 59.9 ± 20.7 ($P < .001$). The increase in the KOOS–sports and recreation score was statistically significant at all time points except the 3-month time point (Table 3).

MRI Findings

Mean lesion fill increased through 24 months of the study to 43.5% ± 48.5% by month 3 and reached 109.7% ± 62.9% at 24 months (Table 4). Mild graft hypertrophy was noted in 6 lesions in 5 patients at 24 months (20.7%). T2 relaxation time decreased from a mean of 64.8 ms ± 14.6 ms at 3 months to 47.4 ms ± 10.4 ms. The site of the original lesion was scored for the number of areas where the T2 signal was similar to adjacent cartilage: the percentage of lesion found to be identical to the surrounding cartilage rose from 11.1% at 3 months postoperatively to 44.4% by 24 months ($P < .05$). Similarly, the percentage of area having more than 3 regions of increased T2 signals dropped over time from 51.9% at 3 months to 7.4% by 24 months (Figure 2). At 24 months, 3.7% had no T2 signal, which reflected the single reported delamination in the study.

TABLE 4
MRI Fill and T2 Values[a]

Time	Percentage of Fill	T2 Relaxation Time, ms
Preoperative	0	N/A
Postoperative		
3 mo	43.5 ± 48.5	64.8 ± 14.6
6 mo	91.4 ± 20.8[b]	62.1 ± 15.3
12 mo	82.6 ± 27.3[b]	56.0 ± 10.9[b]
24 mo	109.7 ± 62.9[b]	47.4 ± 10.4[b]

[a]Values are expressed as mean ± standard deviation. MRI, magnetic resonance imaging; N/A, not applicable.
[b]Statistically significant compared with 3 months ($P < .001$).

Elective Arthroscopic Surgery

Of the 11 elective arthroscopic surgeries performed at 24 months, 1 partial (10% of lesion area) delamination occurred in a patient with effusion and pain. About 10% of the graft required debridement, followed by microfracture of the exposed area. One patient had a full delamination that was asymptomatic. The graft was found as 2 loose bodies, which were surgically removed during the procedure. No reoperations were performed outside the elective procedures.

The ICRS repair score was noted for the 11 elective reoperations with an overall mean of 9.5 ± 3.6. Nine of the 11 lesions (82%) were graded at ≥9 (nearly normal), of which 4 lesions (36%) were graded as 12 (normal). The 2 lesions that scored below 9 involved the above-mentioned partial delamination (score of 6 = abnormal, treated with a microfracture procedure) and the full delamination (score of 0).

Histological/Immunochemistry Findings

Histological evaluation was performed on 8 of the 11 biopsy samples; technical difficulties with sample handling resulted in a loss of the first 3 collected samples. Sections from all samples included full-thickness articular cartilage, the chondro-osseous junction, and at least 5 mm of subchondral bone. Results are summarized in Table 5. The mean score for fibrillation was 1, representing minimal or no superficial fibrillation, with 1 sample demonstrating moderate (40%-80%) fibrillation. The mean articular cartilage cellularity score was 2 (mixed

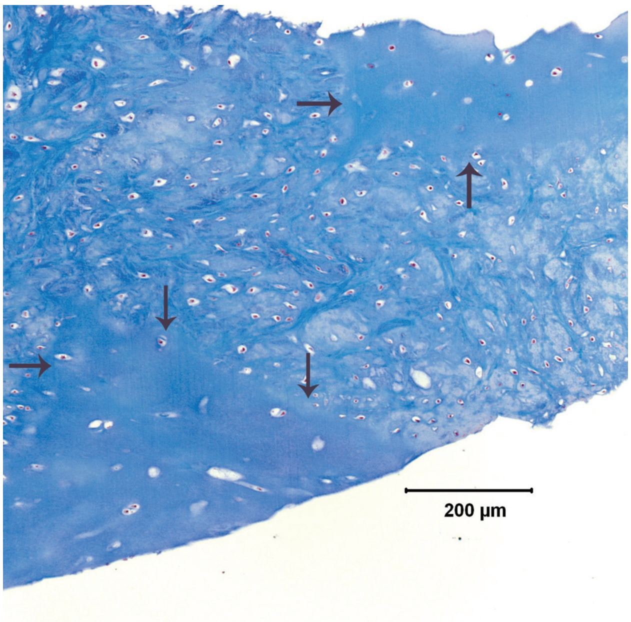

Figure 3. Trichrome-stained biopsy section from 24 months demonstrating the integration of hyaline cartilage (arrows) and fibrocartilage (remainder of tissue in section). The patient had a 2.4-cm^2 lesion in the medial femoral condyle.

TABLE 5
Summary of Histological Findings

Patient	Fibrillation Score	Cellularity Score	Chondrocyte Necrosis Score	Loss of Safranin O Score	Masson Trichrome Score
1	1	0	0	1	4
2	1	4	2	2	4
3	1	0	0	3	4
4	0	0	1	0	4
5	1	4	0	1	0
6	1	1	2	1	4
7	0	2	0	1	2
8	3	4	0	3	3
Mean	1	2	1	2	3

hypocellular/hypercellular), with a relatively high degree of variability between the biopsy specimens and 3 of the sections demonstrating some chondrocyte cloning. The mean score for chondrocyte necrosis was 1 (minimal necrosis), indicating that all chondrocytes were viable in the majority of the sections. The mean score for loss of safranin O staining was 2 (loss of staining in 20%-40% of section). In 2 sections, the loss of staining extended up to 40% of the cartilage depth. Masson trichrome–stained sections (mean score of 3; fibrillar matrix in 40%-80%) revealed that the areas of hyaline cartilage were variable across the samples, with 1 section having complete hyaline cartilage and others with ≤40%. In sections containing both hyaline cartilage and fibrocartilage, these tissues were usually very well integrated (Figure 3).

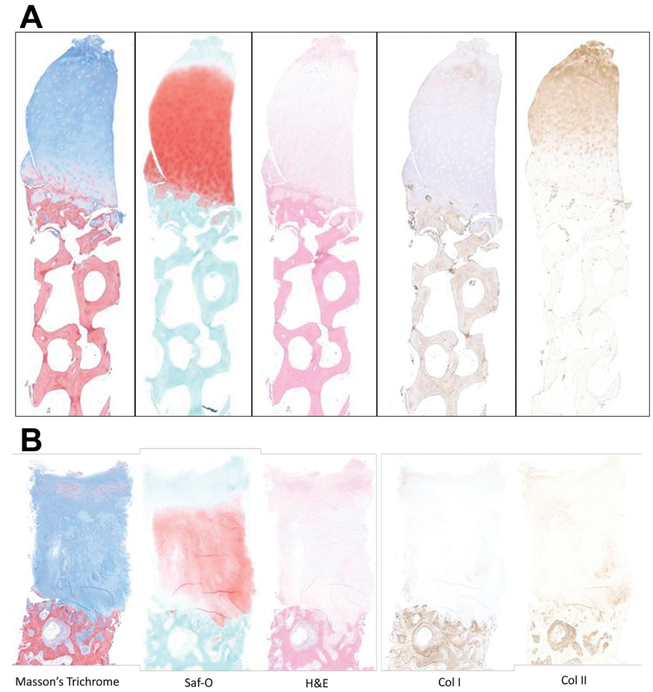

Figure 4. Serial sections from 2 patients representing (A) a sample that scored well and (B) a sample that scored poorly at 2 years postoperatively. (A) The patient was a 35-year-old man with a 3.3-cm^2 defect in the medial femoral condyle (MFC). (B) The patient was a 39-year-old woman with a 3.2-cm^2 lesion in the MFC. Stains used were Masson trichrome, safranin O (Saf-O), hematoxylin and eosin (H&E), and immunohistochemistry preparations for type I collagen (Col I) and type II collagen (Col II).

All sections contained areas of articular cartilage that were immunopositive for type II collagen, and 6 of 8 sections contained areas with moderate to marked immunopositivity. For type I collagen, 1 section contained articular cartilage that completely lacked immunopositivity, and 5 of 8 sections contained areas with minimal or mild immunopositivity. For the 8 biopsy samples analyzed, maximum immunopositivity scores were higher for type II collagen than type I collagen in 6 samples and were equal in the remaining 2 samples. Representative histology/immunohistochemistry results from a sample that scored well and from a sample that scored poorly are shown in Figure 4.

Safety and AE Profile

Adverse events (Table 6) were noted and were found to be in line with previously reported profiles of similar procedures.[3] The most common adverse event was joint effusion (31 occurrences in 21 patients), followed by reduced range of motion (24 in 20 patients) and swelling/bruising (14 in 10 patients). The full and partial delaminations noted

TABLE 6
Adverse Events[a]

Adverse Event Category	No. of Reports	No. of Patients
Joint effusion	31	21
Reduced range of motion	24	20
Swelling and bruising	14	10
Knee pain	13	10
Crepitus	7	6
Chondromalacia	5	5
Quadriceps weakness	4	4
Knee instability	3	3
Arthritis	2	2
Decreased sensation at incision site	2	2
Osteophyte	2	2
Partial graft failure/delamination	2	2
Subchondral bone marrow edema	2	2
Superficial infection	1	1
Adhesions	1	1
Complete graft failure/delamination	1	1
Inflammation	1	1
Plica formation	1	1
Hematoma	1	1
Deep vein thrombosis	1	1

[a]Events not related to the index knee include contralateral knee pain (n = 6), postoperative nausea and vomiting (n = 4), chondromalacia of the contralateral knee (n = 2), fever (n = 2), upper respiratory infection (n = 2), anesthesia reaction (n = 2), back pain (n = 1), epicondylitis (n = 1), fall (n = 1), laceration due to motor vehicle accident (n = 1), hypertriglyceridemia (n = 1), and tachycardia (n = 1).

during elective arthroscopic surgery were reported as adverse events and included in Table 6.

DISCUSSION

A previous report has detailed outcomes of patients with grade 4 patellar lesions treated with PJAC.[12] This is the first report of a prospective patient cohort whose trochlea and femoral condyle cartilage lesions were treated with PJAC and were followed for 2 years postoperatively. The positive clinical outcomes represent statistically significant and meaningful clinical improvements over baseline (preoperative) levels for multiple measures of pain, symptoms, ADL, and sports and recreation.

Comparison with other treatment regimens can be problematic, owing to the multifactorial nature of the repair mechanism (eg, including lesion size, patient age, and BMI) and also the widely differing outcome scores that have been reported. Nevertheless, some conclusions can be drawn concerning the relative efficacy and safety profile of PJAC. Ebert et al[6] have reported on the 2-year outcomes of matrix-induced autologous chondrocyte implantation (MACI) in a single-arm case evaluation and showed similar outcomes to the current study. The KOOS subscores improved from 58.06 to 86.81 for pain, 59.46 to 85.94 for symptoms, 73.24 to 94.61 for ADL, 27.88 to 67.19 for sports and recreation, and 24.86 to 56.25 for QoL. Comparable increases from baseline scores were recorded in the current study. Rates of graft failure (5%) and hypertrophy (20%) were also similar.

In a separate study[7] comparing different rehabilitation protocols for MACI at up to 5 years postoperatively, the KOOS subscores were similar to those scores reported above, with a graft failure rate of 8.6% and a hypertrophy rate of up to 27%. Corpus et al[4] have reported on long-term outcomes of autologous chondrocyte implantation (ACI), which included 2-year follow-up time points. Similar to other studies, the mean change in KOOS outcomes from baseline is broadly similar to those in the current study, with, for example, the KOOS pain score increasing from 56.0 to 78.7 at 2 years compared with 64.1 to 83.7 in the current study.

The IKDC outcomes were all reasonably similar to those in the current study as well. Corpus et al[4] demonstrated a large increase in the IKDC score (approximately 20 points) from preoperatively to 24 months (a very similar change from baseline as the current study), which subsequently declined at 4 years and further declined at or beyond 7 years. Longer term follow-up will therefore be needed.

Other clinical outcomes demonstrate continued improvement in pain and function throughout the course of the 2-year follow-up: MRI showed that there was about 50% fill by 3 months, reflecting the fairly rapid improvement in clinical outcome scores over that same time period. It is possible that the onset or benefits of repair tissue may have been expedited compared with other techniques, given that juvenile cartilage is hypercellular compared with adult cartilage and metabolically more active, although further studies will be required.[1] The decrease in observed T2 relaxation times was essentially linear (R^2 = 0.99), demonstrating that reorganization and maturity of the tissue proceeded over time for at least 2 years. The T2 scores also revealed a gradual maturing of the nascent tissue (Figure 5), and by 24 months, only 7.4% of new tissue showed greater than 3 areas of increased T2 relaxation time.

The AE profile was very similar to those previously reported and suggests that the procedure poses few or no unexpected complications compared with other available options.[3,5] The observed rate of full delamination (4%) was the same as the overall reported delamination rate for ACI.[9]

On elective surgeries for biopsy, all grafts were visibly intact with the exception of 2, one of which was found to be partly delaminated (approximately 10% of the treated area) and the second of which was found to be fully delaminated. In both cases, the patients had not presented to the investigator with excessive pain or other clinical symptoms, and it is likely that these would not have been identified if it were not for the second-look procedure.

Biopsy specimens were obtained from about one third of the patients at 24 months after surgery and generally displayed a mix of hyaline and fibrocartilage, with a preponderance of hyaline cartilage in 3 of 8 samples. It is acknowledged that biopsy samples must be interpreted with caution, as a single biopsy site may not be representative of the entire repair tissue. With that limitation in mind, the integration of areas of hyaline cartilage with areas of fibrocartilage was extremely good, and gross arthroscopic findings during elective biopsies suggested seamless integration with the native tissue. Two revision

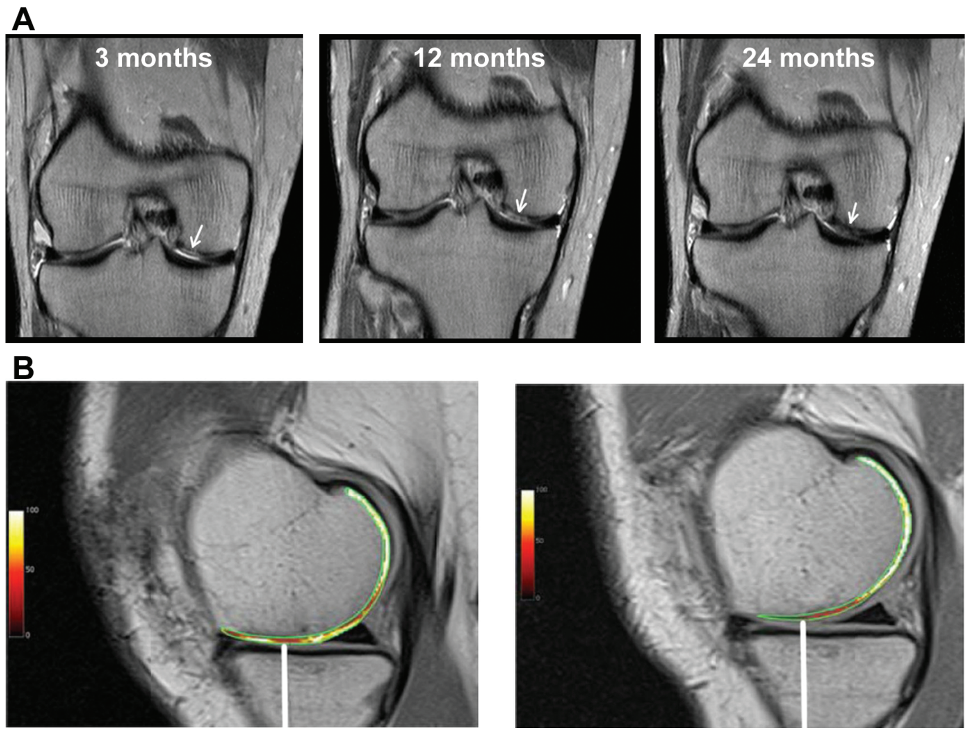

Figure 5. (A) A DeNovo NT implant in the medial femoral condyle (MFC) of a 52-year-old patient. The 3-month image shows the implant with incomplete fill and a high T2 signal compared with the adjacent cartilage. The 12- and 24-month images show increasing fill and a decreased T2 signal, consistent with graft maturation. (B) Representative T2 map from a 49-year-old female patient with a 3.2-cm^2 lesion in the MFC. The T2 map values are shown in the color scale on the left from 0 to 100 milliseconds. The area just to the right of the line going down to the bottom of the image shows decreased T2 relaxation times in the area of the implant between 6 and 12 months postoperatively.

surgeries were performed during the course of the elective biopsy: 1 because of partial delamination and 1 because of complete delamination noted while evaluating the repair site. These patients' KOOS values did not differ significantly from the mean at the 24-month follow-up visit (77.3 and 72.1, respectively). However, 1 patient (having partial delamination) had a VAS pain score of 23 mm at this visit. It is possible that these lesions would not have led to reoperations under traditional nonstudy clinical circumstances.

Limitations

Inherently, there are several limitations from a small study without an appropriate surgical control. For example, the sample size is inadequately powered for anything other than an analysis of safety, only 3 surgeons participated, and the use of a single but experienced radiologist and pathologist prevents intrarater reliability measurements.

Noting the above limitations, a few qualified options for the use of PJAC may be suggested:

1. The current lesion size and postoperative course would suggest similar lesion treatment indications as for ACI.
2. As no randomized comparative data are available, no recommendations can be advanced as to using PJAC over another cartilage restoration treatment.
3. Compared with other cell-based procedures or osteochondral allograft transplantation, PJAC remains a cost-effective alternative that essentially can be used as a point-of-care solution in a single-stage procedure.
4. Arguably, using healthy young donor tissue compared with the patient's own tissue (older, possibly genetically predisposed to failure) or compared with an adult osteochondral allograft offers an appealing alternative, given the inherent biological challenges.

In summary, PJAC is an option for the treatment of chondral defects, with short-term results similar to other treatments. Specific advantages of this procedure include the lack of donor site morbidity and the ability to treat the defect with a single operation with lower resource intensity. Further studies on this novel approach will be required, owing to the small number of lesions and relatively short follow-up time in this study.

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