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November 26, 2014

NeoCart is our lead product candidate and is currently being evaluated in a Phase 3 clinical trial, as described in our registration statement. Prior to our Phase 3 clinical trial, NeoCart was evaluated in Phase 1 and 2 clinical trials and the data from these clinical trials was published in the *Journal of Bone and Joint Surgery* (Phase 2) and the *American Journal of Sports Medicine* (Phase 1).

Our NeoCart Phase 2 clinical trial was initiated in 2007 to further evaluate the positive safety and early efficacy signals demonstrated in our Phase 1 clinical trial of NeoCart for articular cartilage damage in the knee. We also sought to identify clinically meaningful endpoints and identify appropriate patient populations to be studied in the design of future clinical studies. The trial was a five-year prospective, controlled, randomized, clinical study of 30 patients conducted at six U.S. centers and completed its enrollment in 2008. Twenty-one patients were randomized to receive a NeoCart implant and nine patients were randomized to undergo a microfracture procedure. In November 2013, the Phase 2 trial concluded its five-year observation period. During the course of the trial, no serious adverse events (expected or unexpected) were considered to be product- or implant-related. Two-year results of this trial were published in the article titled “NeoCart, an Autologous Cartilage Tissue Implant, Compared with Microfracture for Treatment of Distal Femoral Cartilage Lesions: An FDA Phase-II Prospective, Randomized Clinical Trial After Two Years” (*Journal of Bone and Joint Surgery*; Crawford, et al.; 2012;94:979-89) (the “Phase 2 Article”). The full text of the Phase 2 Article is reproduced below.

A Phase 1 clinical trial was conducted to demonstrate the safety of NeoCart for use when implanted into cartilage defects in the knee with the intention of repairing the articular cartilage defects. The two-year results of our Phase 1 clinical trial were published in the article titled “An Autologous Cartilage Tissue Implant NeoCart for Treatment of Grade III Chondral Injury to the Distal Femur: Prospective Clinical Safety Trial at 2 Years” (*American Journal of Sports Medicine*; Crawford, et al.; 2009;37:1334-43) (the “Phase 1 Article” and, together with the Phase 1 Article, the “Articles”). The full text of the Phase 1 Article is reproduced below.

We have filed a registration statement, including a preliminary prospectus and exhibits, with the SEC for the offering to which this communication relates. The registration statement has not yet become effective. Before you invest, you should read the registration statement, including the preliminary prospectus and exhibits, for more complete information about us and this offering. You may obtain these documents for free by visiting EDGAR on the SEC web site at www.sec.gov or by requesting copies from us at (781) 547-7900 or from Cowen and Company, on behalf of the underwriters, c/o Broadridge Financial Services, 1155 Long Island Avenue, Edgewood, NY, 11717, Attn: Prospectus Department, by telephone at (631) 274-2806 or by fax at (631) 254-7140.

You should not rely on the statements in the Articles for purposes of evaluating an investment in our securities, but rather should solely on the information in the registration statement, including the preliminary prospectus and exhibits. In particular, you should carefully read the risk factors described in the preliminary prospectus.

NeoCart, an Autologous Cartilage Tissue Implant, Compared with Microfracture for Treatment of Distal Femoral Cartilage Lesions

An FDA Phase-II Prospective, Randomized Clinical Trial After Two Years

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Background: Despite introduction of autologous chondrocyte therapy for repair of hyaline articular cartilage injury in 1994, microfracture remains a primary standard of care. NeoCart, an autologous cartilage tissue implant, was compared with microfracture in a multisite prospective, randomized trial of a tissue-engineered bioimplant for treating articular cartilage injuries in the knee.

Methods: Thirty patients were randomized at a ratio of two to one (two were treated with an autologous cartilage tissue implant [NeoCart] for each patient treated with microfracture) at the time of arthroscopic confirmation of an International Cartilage Repair Society (ICRS) grade-III lesion(s). Microfracture or cartilage biopsy was performed. NeoCart, produced by seeding a type-I collagen matrix scaffold with autogenous chondrocytes and bioreactor treatment, was implanted six weeks following arthroscopic cartilage biopsy. Standard evaluations were performed with validated clinical outcomes measures.

Results: Three, six, twelve, and twenty-four-month data are reported. The mean duration of follow-up (and standard deviation) was 26 ± 2 months. There were twenty-one patients in the NeoCart group and nine in the microfracture group. The mean age (40 ± 9 years), body mass index (BMI) (28 ± 4 kg/m^2), duration between the first symptoms and treatment (3 ± 5 years), and lesion size (287 ± 138 mm^2 in the NeoCart group and 252 ± 135 mm^2 in the microfracture group) were similar between the groups. Adverse event rates per procedure did not differ between the treatment arms. The scores on the Short Form-36 (SF-36), Knee Injury and Osteoarthritis Outcome Score (KOOS) activities of daily living (ADL) scale, and International Knee Documentation Committee (IKDC) form improved from baseline ($p < 0.05$) to two years postoperatively in both treatment groups. In the NeoCart group, improvement, compared with baseline, was significant ($p < 0.05$) for all measures at six, twelve, and twenty-four months. Improvement in the NeoCart group was significantly greater ($p < 0.05$) than that in the microfracture group for the KOOS pain score at six, twelve, and twenty-four months; the KOOS symptom score at six months; the IKDC, KOOS sports, and visual analog scale (VAS) pain scores at twelve and twenty-four months; and the KOOS quality of life (QOL) score at twenty-four months. Analysis of covariance (ANCOVA) at one year indicated that the change in the KOOS pain ($p = 0.016$) and IKDC ($p = 0.028$) scores from pretreatment levels favored the NeoCart group. Significantly more NeoCart-treated patients ($p = 0.0125$) had responded to therapy (were therapeutic responders) at six months (43% versus 25% in the microfracture group) and twelve months (76% versus 22% in the microfracture group). This trend continued, as the proportion of NeoCart-treated patients (fifteen of nineteen) who were therapeutic responders at twenty-four months was greater than the proportion of microfracture-treated participants (four of nine) who were therapeutic responders at that time.

continued

Disclosure: One or more of the authors received payments or services, either directly or indirectly (i.e., via his or her institution), from a third party in support of an aspect of this work. In addition, one or more of the authors, or his or her institution, has had a financial relationship, in the thirty-six months prior to submission of this work, with an entity in the biomedical arena that could be perceived to influence or have the potential to influence what is written in this work. No author has had any other relationships, or has engaged in any other activities, that could be perceived to influence or have the potential to influence what is written in this work. The complete **Disclosures of Potential Conflicts of Interest** submitted by authors are always provided with the online version of the article.

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Conclusions: This randomized study suggests that the safety of autologous cartilage tissue implantation, with use of the NeoCart technique, is similar to that of microfracture surgery and is associated with greater clinical efficacy at two years after treatment.

Level of Evidence: Therapeutic Level I. See Instructions for Authors for a complete description of levels of evidence.

Articular cartilage has a poor capacity for repair, and articular cartilage injuries often result in pain and compromised joint function. In addition, injury to hyaline cartilage may be considered a primary risk factor for the development and progression of osteoarthritis[1,2]. In 1994, Brittberg et al. reported using clonally expanded autogenous chondrocytes injected below a periosteal patch in sixteen patients[3]. Histological analysis of random biopsy specimens indicated the presence of type-II collagen within the healing tissue; this finding increased interest in this cell-based therapy and the potential for hyaline tissue repair. Subsequent clinical outcome trials have demonstrated the clinical benefit of this autologous chondrocyte implantation procedure; the clinical success of the technique appears to depend on anatomic location and associated co-morbidity[4-8]. However, temporal pain relief and functional improvements have been reported to be essentially equivalent to those after microfracture[9-12]. Autologous chondrocyte implantation is both technically demanding and associated with a high percentage of reoperations[13]. Modification of this cell therapy designed to reduce complications by replacing the periosteal patch with a biocompatible matrix or selecting cells of potentially improved chondrogenic potential have been described and considered third and fourth-generation techniques[14].

One strategy is the transition from implanting cells with putative chondrogenic potential to implantation of neocarti-laginous tissue. NeoCart (Histogenics, Waltham, Massachusetts) is an autologous cartilage tissue-engineered implant that uniquely combines a bovine type-I collagen matrix scaffold with autogenous chondrocytes and bioreactor treatment. Previously, this technique was reported to generate increased glycosaminoglycan (GAG) and type-II collagen production in comparison with implants cultured without bioreactor treatment[15,16]. The implant is then adhered with a novel collagen adhesive, creating a sutureless-fixation cartilage repair technique[17]. This cell-collagen construct is a matured tissue-like implant containing hyaline GAGs designed to provide a more rapid and potentially sustained therapeutic effect for articular cartilage repair. In an initial study, it safely repaired hyaline articular cartilage with sutureless application, integrated with native chondral tissue and to subchondral bone, and decreased pain and improved function for twenty-four months[18].

Microfracture is recommended as a primary treatment for focal chondral lesions in the knee[19-21]. Microfracture involves penetrating the subchondral bone after removing the damaged hyaline cartilage including the calcified layer[22]. This ‘‘marrow stimulation technique’’ provides cell elements to the area of injury, potentially forming a fibrin clot that may mature as repair tissue. The nature of this fibrocartilage repair includes a variable mixture of bone and disorganized matrix providing a mechanically inferior repair tissue as compared with native hyaline articular cartilage[12,23,24]. Functional improvement and pain relief after microfracture have variably appeared to be most effective in patients with a new injury (symptoms of less than twelve months), a small focal injury (<2.5 cm^2), a younger age (less than thirty years), and a lower body mass index (BMI) (<30 kg/m^2)[23,24].

In this study, we compared the efficacy of a tissue-engineered, cell-seeded matrix scaffold (NeoCart) with that of microfracture at a minimum of two years after treatment of isolated symptomatic articular cartilage lesions of the knee. This report describes the clinical results and measures as required for phase II of a U.S. Food and Drug Administration (FDA)-regulated exploratory clinical trial (ClinicalTrials.gov, NCT00548119).

Materials and Methods

Institutional review board approval was granted for this FDA-regulated phase-II prospective, randomized clinical trial designed to evaluate safety and clinical outcomes of the NeoCart implant compared with microfracture. Written consent was obtained from forty-nine subjects who were provisionally enrolled and screened for eligibility. The study was registered with ClinicalTrials.gov (#NCT00548119).

Eligible patients were between eighteen and fifty-five years of age, had a symptomatic International Cartilage Repair Society (ICRS) grade-III cartilage lesion of the femoral condyle, and satisfied the inclusion criteria (Table I). The final determination of eligibility was made at the time of knee arthroscopy, prior to randomization. Fourteen patients were excluded as a result of advanced degenerative osteoarthritis, nonqualifying lesion(s), withdrawal of consent, symptoms inconsistent with a femoral condyle cartilage lesion, or smoking status (Fig. 1). Five more patients were excluded at the time of arthroscopy (Fig. 1). A total of thirty patients were randomized at six investigational sites. Randomization was achieved by opening a sequentially numbered envelope in the operating room after arthroscopic determination of eligibility was made. A permuted block design, generated by a statistician independent of the study, was utilized to minimize the opportunity for guessing the next treatment assignment. These stringent precautions were taken to ensure that neither the surgeon nor the patient would know the treatment arm until the decision regarding eligibility was made at arthroscopy.

All potential study patients were evaluated preoperatively with use of cartilage-sequence magnetic resonance imaging to assess both the lesion and the surrounding articular cartilage. Only patients with one or two isolated articular cartilage lesions of the femoral condyle(s) were included. The patients underwent diagnostic arthroscopy. Following confirmation of the eligibility of the chondral lesion(s), the patient was randomized either to NeoCart treatment and, therefore, immediate biopsy of uninjured hyaline cartilage from the proximal trochlea or to microfracture treatment.

The patients randomized to the microfracture treatment arm underwent that procedure at the index arthroscopy. Microfracture included

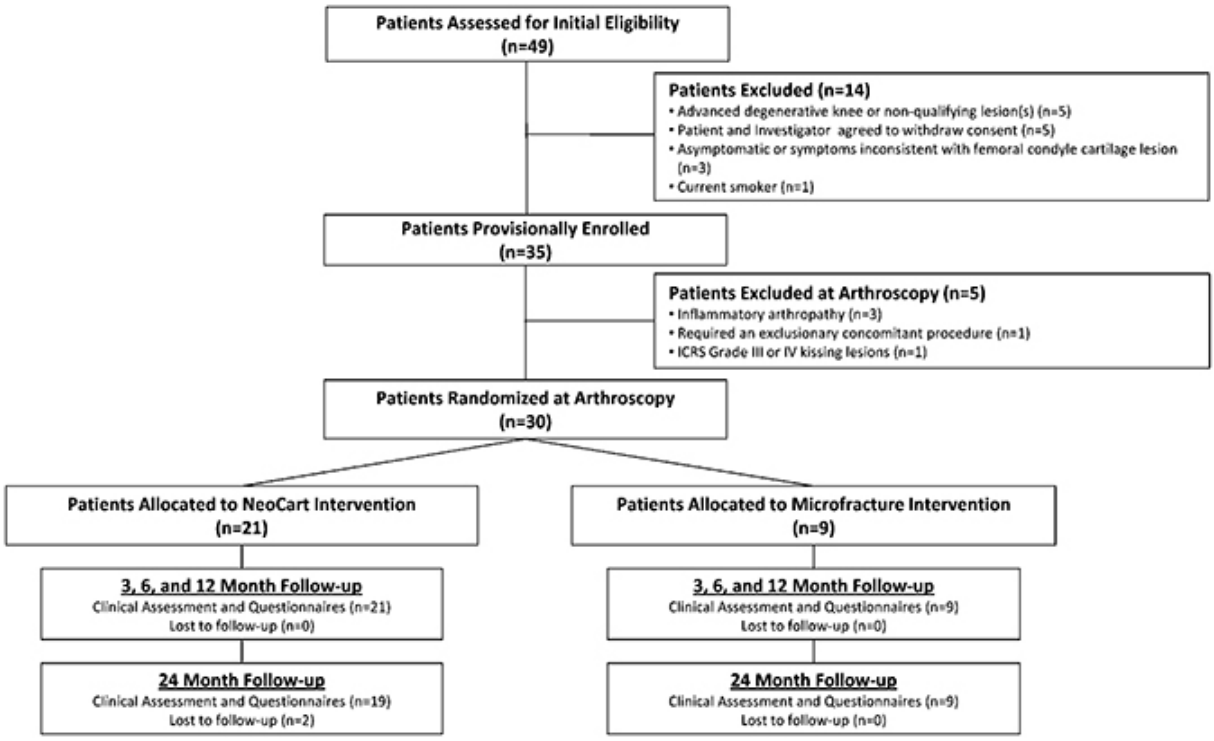

Fig. 1

Enrollment diagram, including patient randomization through clinical assessment.

lesion debridement to a stable cartilage margin, removal of the calcified cartilage layer, and the homogeneous creation of subchondral osseous penetrations within the base of the cartilage defect with use of 2 to 4-mm awls[21,24].

The patients randomized to NeoCart treatment were allowed immediate unrestricted activity after the arthroscopic biopsy and returned for implantation approximately six weeks later. During the six-week period, the NeoCart was produced via collagenase extraction of chondrocytes, followed by seeding of these cells onto a bovine type-I collagen matrix. The seeded matrix construct was then incubated in a bioreactor with conditions simulating the knee environment—e.g., low oxygen tension and varying pressure. The implant was released after sterility and glycoprotein-composition quality-control confirmation. One specimen was discarded following a manufacturing process error, after which a repeat arthroscopic biopsy was performed.

Implantation was carried out during a second outpatient surgical procedure via mini-arthrotomy, debridement, and preparation of the defect bed in a manner analogous to microfracture, without subchondral penetration[3]. The NeoCart was secured without suture by using a proprietary collagen-based polymer (CT-3; Histogenics) to anneal the implant to the prepared condyle defect bed and adjacent tissue[18].

The rehabilitation protocol, standardized to that recommended after microfracture, was identical for both groups following the index procedure[21].The protocol included six weeks of toe-touch weight-bearing, six to eight hours of continuous passive motion daily beginning on post-operative day 1, and restriction of sports activity for six months. One patient began immediate unrestricted weight-bearing ten days after NeoCart implantation in concurrence with the rehabilitation protocol, which allowed accelerated weight-bearing by individuals with small, contained lesions.

Clinical measurements included the following self-reported patient surveys: the International Knee Documentation Committee (IKDC) form (subjective); Knee Injury and Osteoarthritis Outcome Score (KOOS) pain, symptoms, activities of daily living (ADL), quality of life (QOL), and sports scales; Short Form-36 (SF-36) questionnaire; and a visual analog scale (VAS) pain score. The IKDC is a combined outcomes assessment of objective clinician and subjective patient measures validated to assess short and long-term knee function[25,26].The KOOS contains five separately scored components designed and validated to assess short and long-term function in subjects with knee injury. These scores were obtained preoperatively and at three, six, twelve, and twenty-four months postoperatively. IKDC objective data were collected and reported to the study sponsor by one physician (not an author), blinded to the treatment, for nineteen of the thirty patients (patients of the first author) or by clinicians (not authors) blinded to all patient-reported subjective data scores. The study sponsor collected and managed all data and provided this information to the authors after all patients had completed two years of follow-up. Subsequent calculations and application of statistical methods are the work of the first author.

Statistical Analyses

All data were collected prospectively, with data entry, edit checks, and query tracking supported by a data management service (Synteract, Carlsbad, California). Descriptive statistics (mean and standard deviation) and the responder rate were calculated for each time point for the intent-to-treat population with the significance level set at $p < 0.05$ (two-sided). The intent-to-treat population included all patients who had been randomized, with the patients classified by the group to which they had been randomized, regardless of the treatment received and follow-up status. The mean change from baseline to the

TABLE I Study Inclusion/Exclusion Criteria

	Inclusion Criteria	Exclusion Criteria*
Initial	• Patient able and willing to give informed consent • Age between 18 and 55 yr • Patient presenting with symptomatic knee pain indicative of an articular cartilage injury • Patient medically able to undergo arthroscopic microfracture or biopsy and subsequent arthrotomy for NeoCart implantation	• Any previous surgical treatment of lesion other than debridement • BMI >35 kg/m^2 • Joint space narrowing of >1/3 compared with normal knee, or <3 mm of joint space measured on radiographs, osteophytes, sclerosis, or degenerative conditions in treatment knee noted on radiographs • Malalignment >3° outside mechanical axis of other knee, or need for surgery to correct malalignment • Other symptomatic pathology of contralateral knee • Surgery on contralateral knee within 8 wk prior to scheduled arthroscopy • Any form of inflammatory arthritis • Ankylosing spondylitis • Synovioma, hemangioma, pigmented villonodular synovitis, or neoplasms in knee • Patient on chemotherapy • Patient unable to undergo MRI • Patient who is pregnant or intends to become pregnant during yr following initial enrollment • Known history of allergy to bovine products or to collagen or more than a minimal reaction to an intradermal collagen injection challenge • History of autoimmune disease • Evidence of HIV or chronic hepatitis-B or C viral infection • Known allergy to gentamicin • Current drug or alcohol abuse • Patient deemed by investigator as unlikely to comply with protocol
At arthroscopy	• Patient with at least 1 treatable lesion located on either medial or lateral femoral condyle that would be a candidate for microfracture therapy • ICRS grade-III lesion • Lesions with a maximum linear dimension at least 1 cm and no more than 3 cm to healthy cartilage border • Lesions with total area less than area of NeoCart (7-8 cm^2)	• Subchondral bone loss • Patient requiring a concomitant procedure other than medial or lateral partial meniscectomy, removal of loose bodies, debridement of articular cartilage lesions other than that being treated and synovectomy • Untreated ACL and/or PCL deficiency or ligamentous instability in involved knee • Meniscus with rim <50% of normal thickness • ICRS grade-III or IV kissing lesion • More than slight anterior knee pain referable to patellofemoral joint and ICRS grade-III(B), III(C), or IV trochlear groove or patellar lesion

*MRI = magnetic resonance imaging, HIV = human immunodeficiency virus, ACL = anterior cruciate ligament, and PCL = posterior cruciate ligament.

three, six, twelve, and twenty-four-month follow-up points was calculated. The analysis included the IKDC (subjective) score, all five KOOS subscores (pain, symptoms, ADL, sports, and knee-related QOL), the VAS score for knee pain (maximum), and the SF-36 score. A paired t test was applied to each group to assess the score change from baseline. Changes in IKDC and KOOS scores from baseline to twelve months were evaluated with an analysis of covariance (ANCOVA), with the baseline scores used as the covariate.

TABLE II Patient Characteristics*

	NeoCart (N = 21)	Microfracture (N = 9)	Total (N = 30)
Age *(yr)*	41 ± 9	39 ± 10	40 ± 9
% male	90% (19 of 21)	67% (6 of 9)	83% (25 of 30)
BMI† *(kg/m²)*	29 ± 3	25 ± 4	28 ± 4
Time since first symptoms *(yr)*	3 ± 5	2 ± 4	3 ± 5
Baseline IKDC score *(points)*	44 ± 13	52 ± 12	47 ± 13
Baseline KOOS pain score *(points)*	65 ± 12	73 ± 16	67 ± 14
Post-debridement lesion size *(mm²)*	287 ± 138	252 ± 135	278 ± 135

*The values are given as the mean and standard deviation, except for % male. †There was a significant difference between arms ($p < 0.05$).

In the responder analysis, a patient was classified as a responder if he or she had at least a 12-point improvement in the pain score of the KOOS assessment and a 20-point improvement in the IKDC subjective score. This combination of two validated patient-reported outcome measures for cartilage repair (IKDC function and KOOS pain scores) to define a responder was derived conservatively from reports indicating the ability of each score to detect a minimal clinically important difference[27,28]. Roos and Lohmander suggested 10 points as a cutoff for clinical improvement or decline and described the minimal perceptible clinical improvement as 8 points for KOOS measures[29]. Similarly, Irrgang et al. and others showed that the IKDC was responsive with a change of 6 points and had reliable sensitivity and specificity for patients perceiving improvement with score changes ranging from 11 to 20 points[25,26].

Patients were dichotomized as either ‘‘responders’’ or ‘‘nonresponders’’ if they met the dual-threshold criteria[30]. The FDA panel indicated a preference for a single functional and separate pain measure for primary outcome determination. Thus, our definition of a therapeutic responder was both a change in the KOOS pain score of ³12 points and a change in the IKDC function score of ³20 points. Patients with a high baseline KOOS pain score (>80 points) were classified as responders if the improvement in their IKDC score met the 20-point criterion and their KOOS pain score had not deteriorated >8 points (the minimal perceptible clinical limit); this was done to account for potential confounding by ceiling effects or the phenomenon where measuring patient response to treatment is no longer influenced by their treatment.

Source of Funding

Institutional financial support was provided by Histogenics Corporation (Waltham, Massachusetts). The funding source provided data management and monitoring services as required by the FDA and had no authorship role.

Results

Study Group

Of the forty-nine subjects who provided consent, fourteen did not meet all inclusion criteria prior to arthroscopy. Of the thirty-five patients undergoing diagnostic arthroscopy, thirty were confirmed as eligible and randomized to treatment groups. Five patients were deemed ineligible at arthroscopy; three of them had confirmed inflammatory arthropathy, one required a concomitant procedure, and another had an ICRS grade-III or IV kissing lesion. Of the patients randomized to the study arm, one had two femoral condyle lesions that satisfied the eligibility criteria; therefore, both lesions were treated with the study implant. All randomized patients received treatment to which they had been assigned, and none were lost to follow-up prior to the twelve-month time point. Two patients were lost to clinical follow-up before the twenty-four-month time point for unknown reasons. Demographic characteristics of the study population and for each treatment arm are summarized in Table II.

Safety

There were three serious adverse events, as defined by the U.S. Department of Health and Human Services Office for Human Research Protections, across treatment groups. Two adverse events occurred in the NeoCart group, both in the same patient. The first event was a case of septic arthritis in the contralateral knee after meniscectomy, which subsequently required a total knee arthroplasty (the second event). In the microfracture group, there was one adverse event: cancer of gynecologic origin. None of these events were considered to be related to the treatment of the cartilage defect. No patients were dropped from the study because of an adverse event. One patient elected to undergo repeat arthroscopic biopsy when an autologous cartilage tissue implant was lost to contamination. A total of eighty-six adverse events were reported during the study period, with sixty-two in the twenty-one patients in the NeoCart group and twenty-four in the nine patients in the microfracture group. These events included a repeat arthroscopic biopsy; an arthroscopic microfracture of a lesion in the ipsilateral knee; an arthroscopic anterior cruciate ligament (ACL) reconstruction of the contralateral knee after the patient had returned to full activity; and postoperative pain, stiffness, swelling, back pain, arm pain, and peri-incisional numbness. Adverse events considered related to the study interventions were consistent with those associated with routine outpatient arthroscopy or mini-knee arthrotomy, with the exception of the repeat biopsy.

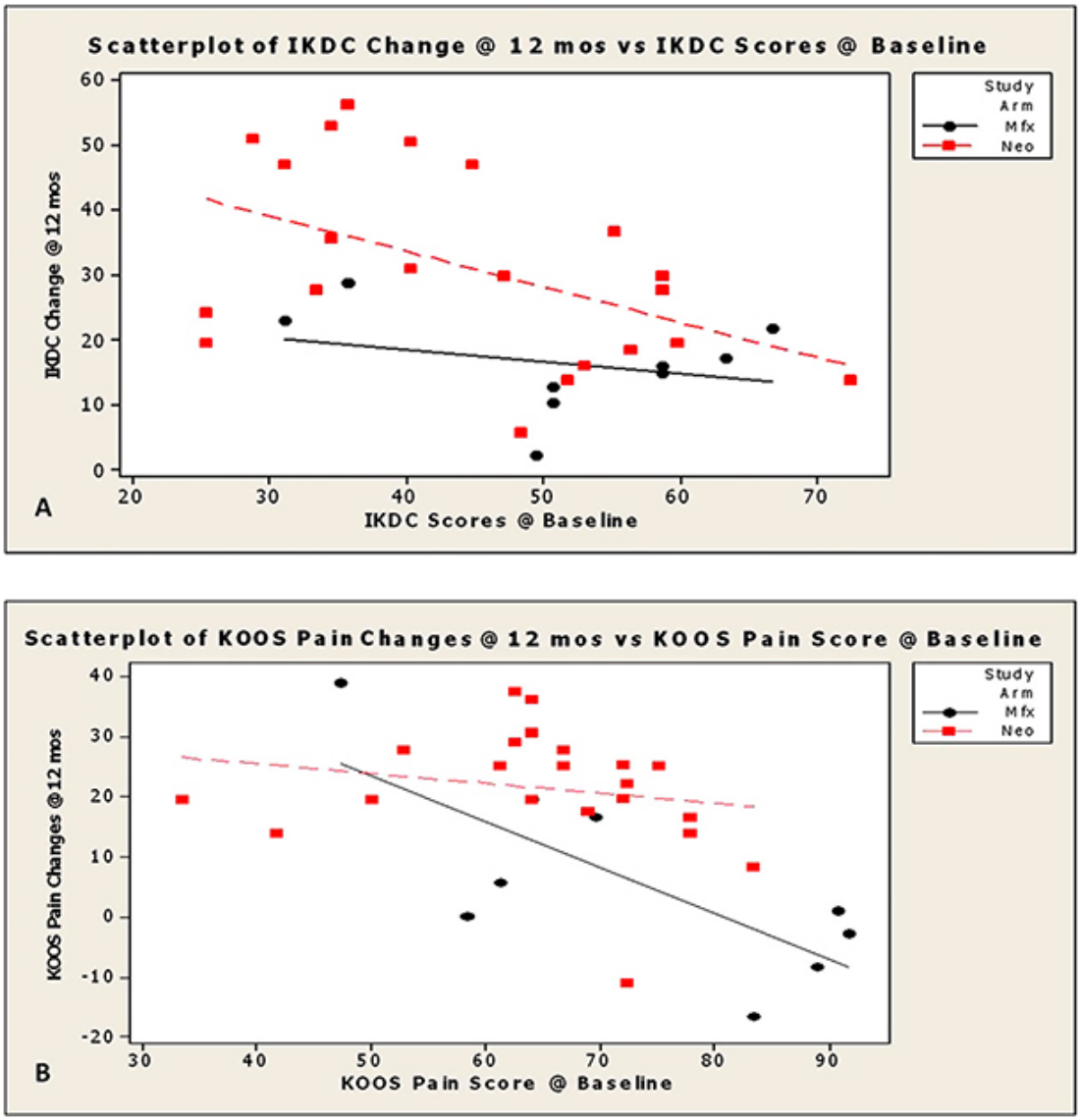

Fig. 2

ANCOVA analysis of change in IKDC **(Fig. 2-A)** and KOOS pain **(Fig. 2-B)** outcome scores at one year, as a function of baseline. Mfx = microfracture and Neo = NeoCart.

Efficacy Analysis

In the NeoCart arm, improvement compared with baseline was significant for all measures at six, twelve, and twenty-four months. In contrast, improvement over baseline measures in the microfracture arm was limited to the IKDC and SF-36 scores at six months and the IKDC, KOOS ADL, and SF-36 scores at twelve and twenty-four months. The NeoCart group had more improvement in the KOOS pain and symptom scores at six months than the microfracture group. At twelve months, the patients treated with the NeoCart showed more improvement in the IKDC, KOOS pain, KOOS sports, and VAS pain scores than the microfracture cohort. At twenty-four months, the improvements in the IKDC, KOOS pain, KOOS sports, KOOS QOL, and VAS pain scores in the NeoCart group were significantly greater than those in the microfracture group ($p < 0.05$). The mean baseline data in the two groups, although not identical, were not significantly different. ANCOVA was performed to adjust for any variation in baseline scores. Changes in both the IKDC function (Fig. 2-A) and KOOS pain (Fig. 2-B) scores were evaluated with use of the baseline score as the covariant. The IKDC score changes from baseline differed significantly between the microfracture and NeoCart study arms (p =

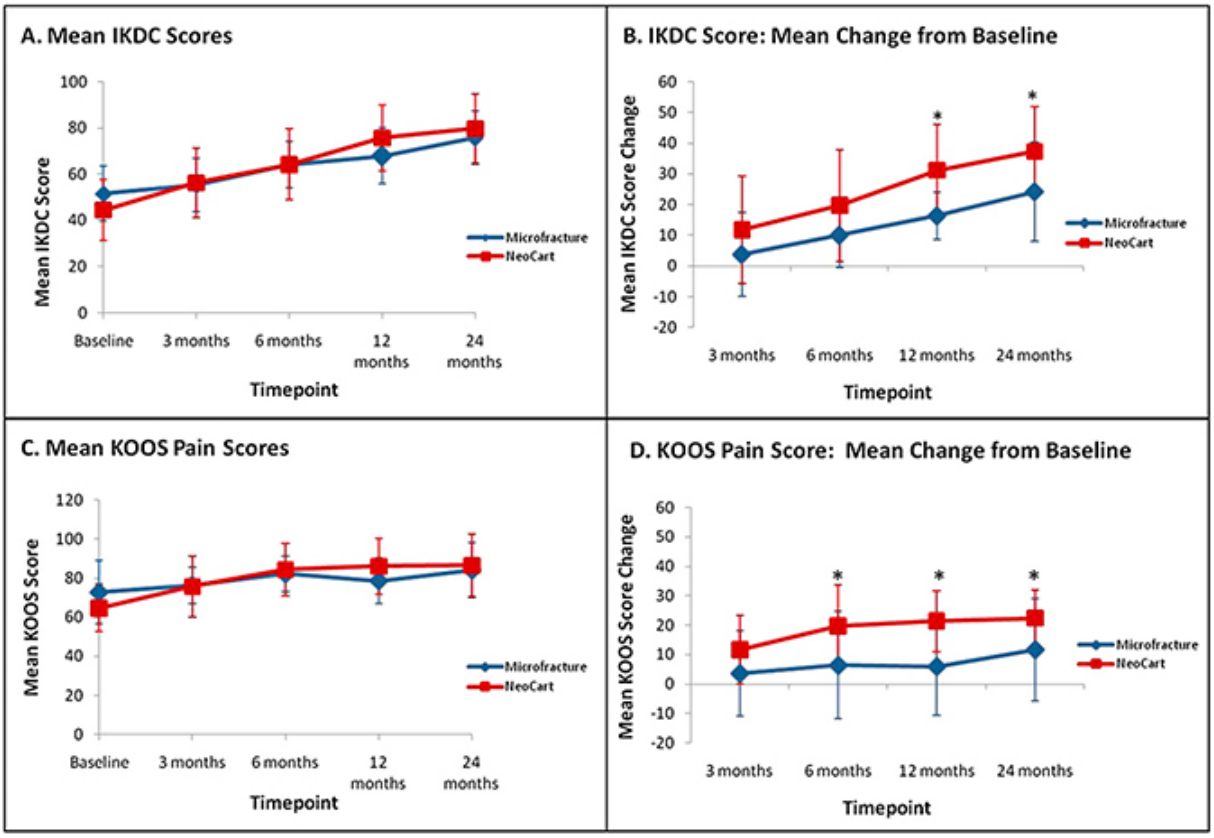

Fig. 3

Temporal representation of IKDC and KOOS pain score means **(Figs. 3-A and 3-C)** and mean changes from baseline **(Figs. 3-B and 3-D)**. All thirty patients (twenty-one in the NeoCart group and nine in the microfracture group) are represented at three, six, and twelve months; twenty-eight patients (nineteen in the NeoCart group and nine in the microfracture group) are represented at twenty-four months. Standard deviation bars are shown. Asterisks represent significant differences, as shown by the unpaired t test, between the NeoCart and microfracture arms ($p < 0.05$).

0.028), when the scores were adjusted. The difference in the adjusted means (NeoCart minus microfracture) was 11.59 with a 95% confidence interval (CI)of 1.353, 21.82. The KOOS pain score changes from baseline also differed significantly between the microfracture and NeoCart study arms ($p = 0.016$). The difference in the adjusted KOOS pain score means (NeoCart minus microfracture) was 12.06 with a 95% CI of 2.388, 21.74.

Temporal improvement, reflected as the changes in the KOOS pain and IKDC function scores, is represented in Figure 3. Direct comparison by applying an unpaired t test indicated significantly greater improvement from baseline in the NeoCart arm compared with the microfracture arm for the KOOS pain score at six months and both the KOOS pain and the IKDC function score at twelve and twenty-four months.

Responder analysis was applied to assess efficacy with use of the dual-criteria threshold of improvement that included both the KOOS pain and the IKDC score (Fig. 4). The proportion of responders in the NeoCart group was 43% (nine of twenty-one) at six months, 76% (sixteen of twenty-one) at twelve months, and 79% (fifteen of nineteen) at twenty-four months. In contrast, a minority of patients treated with mi-crofracture were considered responders: 25% (two of eight) at six months, 22% (two of nine) at twelve months, and 44% (four of nine) at twenty-four months. The proportion of responders in NeoCart group was found to be significantly greater than that in the microfracture group ($p = 0.0125$) when a Fisher exact test was applied to the data at both six months and twelve months after treatment. At twenty-four months, 79% of NeoCart-treated and 44% of the microfracture-treated patients were considered responders ($p = 0.097$), with two microfracture-treated patients considered responders when the correction for a possible ceiling effect was applied. Similarly, 81% (seventeen) of the twenty-one patients in the NeoCart group responded to treatment compared with 44% (four) of the nine in the microfracture cohort at the time of final follow-up (Fig. 4-B).

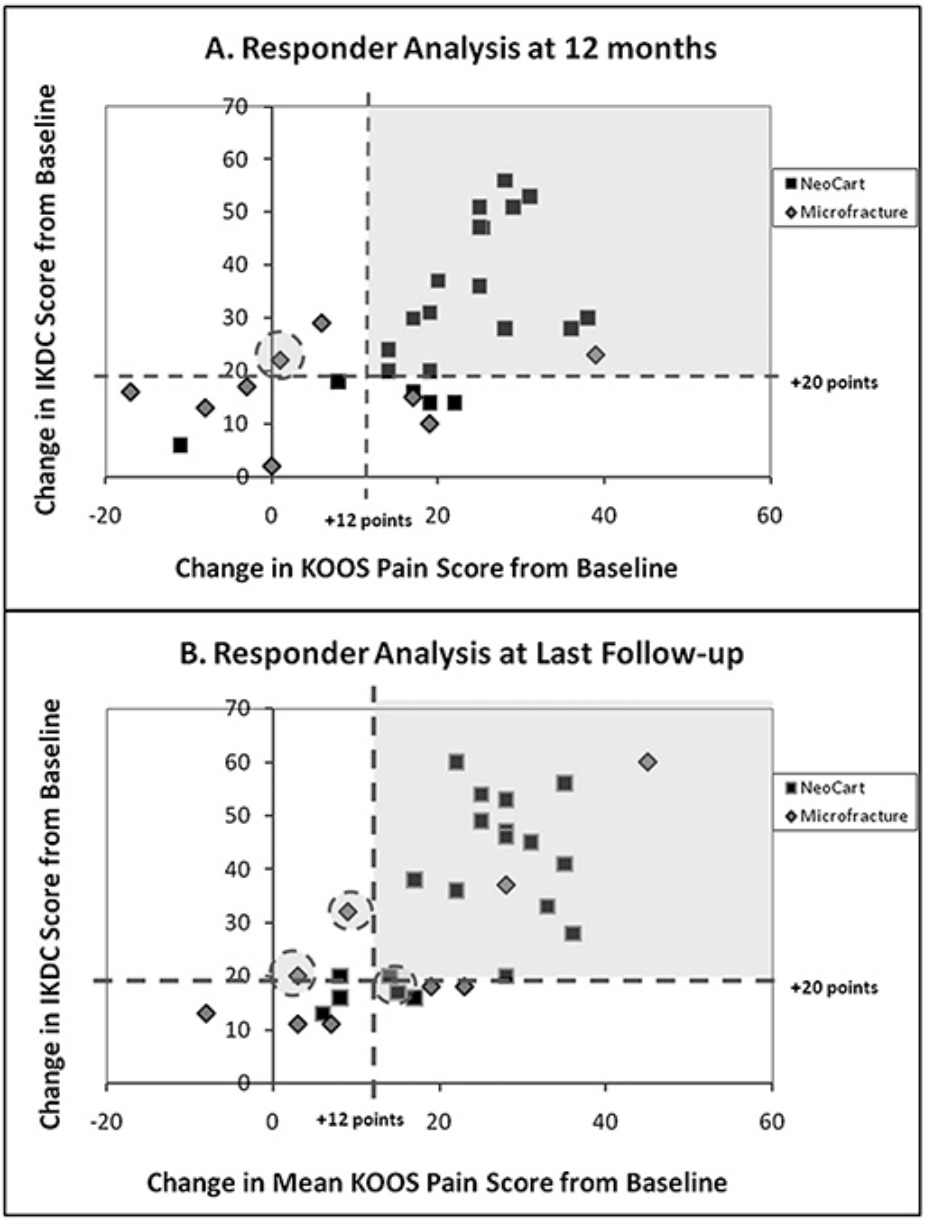

Fig. 4

Responder analysis at twelve months postoperatively **(Fig. 4-A)** and the time of the last follow-up **(Fig. 4-B)**, at an average of 25.6 months postoperatively. Responders (shown in the shaded areas) are defined by improvement in the IKDC score of ³20 points and the KOOS pain score of ³12 points. The proportion of responders in the NeoCart group is greater than that in the microfracture group at twelve months (sixteen of twenty-one and two of nine, respectively) and at the time of the last follow-up (seventeen of twenty-one and four of nine, respectively). The circled symbols indicate subjects considered therapeutic responders on the basis of a single criterion due to a potential KOOS pain ceiling effect (two in the microfracture group and one in the NeoCart group).

Discussion

Currently, microfracture is the standard surgical treatment for isolated cartilage lesions in the U.S.[19-21]. Microfracture results include inconsistent fibrocartilage formation within the lesion and a highly variable therapeutic response[12,21,23,24,27].We describe an exploratory multisite randomized trial that evaluated a tissue-engineered bioimplant for treatment of hyaline cartilage knee injury in comparison with microfracture.

The pathogenesis of osteoarthritis is poorly understood. One putative etiology for osteoarthritis involves focal damage

to the articular cartilage in a localized, nonuniform manner with altered joint biomechanics, tissue loss, and subsequent remodeling of subchondral bone[1,2]. The chondral injury, when painful and focal, is potentially managed with a variety of surgical techniques. Microfracture is widely recommended as a primary treatment, but it requires compromise of the normal subchondral bone by multiple 3-mm awl penetrations in an effort to supply mesenchymal cells to the chondral injury. Clinically, outcome measures are reported to plateau between twelve and twenty-four months after microfracture treatment, and the procedure appears less efficacious for larger, more chronic lesions in older patients and those with a higher BMI[11,23,24].

Surgical therapies designed to improve the clinical efficacy of cartilage injury treatments by improving the potential for a hyaline healing response have followed the original report by Brittberg et al.[3,7,31,32]. However, among well-tested alternatives to microfracture, none have provided a significant clinical improvement when directly compared with microfracture up to two years after treatment[11,12]. One recent report indicated a significant improvement, after thirty-six months, in one measure, the group mean of a combined KOOS score (excluding the KOOS sports score), for patients treated with an autologous chondrocyte implant variant, ChondroCelect (TiGenix, Brussels, Belgium) as compared with those treated with microfracture[33]. These investigators applied a heterogeneous compound symmetry structure mode analysis to a partially combined KOOS score and found a statistical difference ($p < 0.05$). Prior reports on this study population demonstrated no clinical differences at either one or two years, despite showing improved histological quality of the repair tissue. Other investigators have found minimal distinction in clinical outcomes between autologous chondrocyte implantation and microfracture treatment after either two or five years[10,11,34].

NeoCart is a bioengineered tissue patch containing an autologous chondrocyte population matured in a biodegradable collagen matrix with use of bioreactor technology. The bioreactor culture technique (hydrostatic pressure with modified flow rates and low oxygen) has been previously shown to stimulate extracellular matrix accumulation and suppress long-term degradation of matrix accumulated by isolated chondrocytes in vitro studies[15,16,35]. Using an in vivo porcine model, Kusanagi et al. showed improved temporal and absolute integration and matrix production with bioreactor-treated human chondrocyte constructs (chondrocytes in a type-I-collagen honeycomb matrix), in comparison with untreated constructs, when applied to femoral chondral defects[36,37]. This is a unique approach for providing potentially improved replacement tissue to the damaged articular surface in comparison with that produced by currently available clinical therapy in the U.S. The initial clinical report on the use of NeoCart for treatment of cartilage injury indicated an ability to integrate with adjacent normal tissue and an associated progressive collagen organization within the treatment area[18]. Perhaps these biologic distinctions are responsible for the findings reported in our small prospective randomized study. Trends through the first two years indicate not only improved pain and function scores, but also a more rapid onset of improvement in the NeoCart group. Similarly, a larger percentage of patients appear to respond to the NeoCart treatment at six, twelve, and twenty-four months. This difference appears to be a result of the response to NeoCart, rather than some failure of the microfracture technique, as the outcomes in this study are comparable with those reported in other studies for microfracture treatment[11,23,34,38].

Our purpose, as with most FDA phase-II trials, is to continue to assess safety and, preliminarily, efficacy. Although the current study is limited by size and not powered to detect differences in the group mean, a secondary goal is to assess the range of treatment-effect differences in the NeoCart group, to allow sample size calculation for the definitive phase-III study. Similarly, use of the 2:1 randomization format, common for clinical trials with novel therapeutics, allowed this smaller-scale pilot study to accumulate an adequate number of patients for such calculations while providing the necessary control cohort of microfracture-treated patients[27,39].

The four adverse events requiring surgery, unrelated to the study treatment, and the minor complications that we observed were typical of outpatient surgical procedures and did not differ between the two small cohorts. Despite randomization, the study groups were similar, although not identical. The observed responders in the NeoCart group had lower mean baseline IKDC and KOOS pain scores, indicating NeoCart-treated patients had more room to improve than those treated with microfracture. In fact, three patients (two in the microfracture group and one in the NeoCart group) were considered responders at the time of final follow-up as they met the IKDC threshold and had improvement in the KOOS pain score but may have been limited by a ceiling effect. Application of ANCOVA, however, indicated that differences in the change in the reported outcomes from baseline between the NeoCart and microfracture groups were not disproportionately influenced by the different baseline scores. Notably, the lesions were slightly smaller; BMI, lower; symptom duration, shorter; and age at the time of injury, lower in the microfracture population. These circumstances are known to be associated with more favorable outcomes of microfracture treatment and are associated with higher Patient Reported Outcome scores[23,24]. The larger, properly statistically powered phase-III study should control and account for these potentially confounding issues and identify the superior treatment.

In randomized clinical trials, assessment of clinical effectiveness is frequently determined by quantifying the average change within a population and comparing group means with control treatment. Alternatively, responder analysis has been applied as a method of allowing a more discrete assessment of clinically relevant treatment efficacy, failure, and applicability to individual patient care[40,41]. In this analysis, we reported positive responders exclusively and required an improvement above the reported perceptible clinical improvement for both

metrics for a patient to be considered a therapeutic responder. We recognize a potential limitation to dichotomizing continuous or ordinal variables into a binary variable[30]. However, recent FDA guidance on patient-reported outcomes specifically endorses responder analysis as a preferred alternative to assessing clinical relevance[42]. Also, it is recognized that applying responder analysis assessments to randomized clinical trials is suitable when comparing new techniques with standard-of-care therapy[43-46].

Our preliminary findings strongly suggest that using the NeoCart autologous cartilage tissue implant significantly decreases knee pain and improves function within six months after treatment and provides significantly greater improvements than microfracture. Collectively, this report on NeoCart may represent a potentially important evolution in the treatment of cartilage injury and joint-specific chondral pathology. Concomitant application of responder analysis to identify individuals‘ or subpopulations most likely to benefit may further define therapeutic application. A larger-scale, confirmatory study comparing NeoCart safety and efficacy with those of the microfracture technique has been sanctioned by the FDA under special protocol assessment. n

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References

1. Brandt KD, Dieppe P, Radin E. Etiopathogenesis of osteoarthritis. Med Clin North Am. 2009;93:1-24,xv.

2. Lane NE. Clinical practice. Osteoarthritis of the hip. N Engl J Med. 2007;357: 1413-21.

3. Brittberg M, Lindahl A, Nilsson A, Ohlsson C, Isaksson O, Peterson L. Treatment of deep cartilage defects in the knee with autologous chondrocyte transplantation. N Engl J Med. 1994;331:889-95.

4. Krishnan SP, Skinner JA, Carrington RW, Flanagan AM, Briggs TW, Bentley G. Collagen-covered autologous chondrocyte implantation for osteochondritis dissecans of the knee: two- to seven-year results. J Bone Joint Surg Br. 2006;88:203-5.

5. Bartlett W, Skinner JA, Gooding CR, Carrington RW, Flanagan AM, Briggs TW, Bentley G. Autologous chondrocyte implantation versus matrix-induced autologous chondrocyte implantation for osteochondral defects of the knee: a prospective, randomised study. J Bone Joint Surg Br. 2005;87:640-5.

6. Peterson L, Minas T, Brittberg M, Nilsson A, Sjögren-Jansson E, Lindahl A. Two- to 9-year outcome after autologous chondrocyte transplantation of the knee. Clin Orthop Relat Res. 2000;374:212-34.

7. Peterson L, Vasiliadis HS, Brittberg M, Lindahl A. Autologous chondrocyte implantation: a long-term follow-up. Am J Sports Med. 2010;38:1117-24.

8. Minas T. Autologous chondrocyte implantation for focal chondral defects of the knee. Clin Orthop Relat Res. 2001;391Suppl:S349-61.

9. Vasiliadis HS, Wasiak J. Autologous chondrocyte implantation for full thickness articular cartilage defects of the knee. Cochrane Database Syst Rev. 2010;10:CD003323.

10. Knutsen G, Drogset JO, Engebretsen L, Grøntvedt T, Isaksen V, Ludvigsen TC, Roberts S, Solheim E, Strand T, Johansen O. A randomized trial comparing autolo-gous chondrocyte implantation with microfracture. Findings at five years. J Bone Joint Surg Am. 2007;89:2105-12.

11. Knutsen G, Engebretsen L, Ludvigsen TC, Drogset JO, Grøntvedt T, Solheim E, Strand T, Roberts S, Isaksen V, Johansen O. Autologous chondrocyte implantation compared with microfracture in the knee. A randomized trial. J Bone Joint Surg Am. 2004;86:455-64.

12. Saris DB, Vanlauwe J, Victor J, Haspl M, Bohnsack M, Fortems Y, Vandekerckhove B, Almqvist KF, Claes T, Handelberg F, Lagae K, van der Bauwhede J, Vandenneucker H, Yang KG, Jelic M, Verdonk R, Veulemans N, Bellemans J, Luyten FP. Characterized chondrocyte implantation results in better structural repair when treating symptomatic cartilage defects of the knee in a randomized controlled trial versus microfracture. Am J Sports Med. 2008;36:235-46.

13. Wood JJ, Malek MA, Frassica FJ, Polder JA, Mohan AK, Bloom ET, Braun MM, Coté TR. Autologous cultured chondrocytes: adverse events reported to the United States Food and Drug Administration. J Bone Joint Surg Am. 2006;88:503-7.

articular chondrocyte in collagen sponges [abstract]. In: Transactions of the 49th Annual Orthopaedic Research Society; 2003 Feb 2-5; New Orleans, LA. Rosemont, IL: Orthopaedic Research Society; 2003. p 0634.

16. Mascarenhas AC, Kusanagi A, Blahut EB, Johnson J, Berlowitz-Tarrant LJ, Mizuno S. Hydrostatic pressure stimulates extracellular matrix accumulation by swine articular chondrocytes in 3D collagen sponges [abstract]. In: Transactions of the 49th Annual Orthopaedic Research Society; 2003 Feb 2-5; New Orleans, LA. Rosemont, IL: Orthopaedic Research Society; 2003. p 0127.

17. Hettrich CM, Crawford D, Rodeo SA. Cartilage repair: third-generation cell-based technologies—basic science, surgical techniques, clinical outcomes. Sports Med Arthrosc. 2008;16:230-5.

18. Crawford DC, Heveran CM, Cannon WD Jr, Foo LF, Potter HG. An autologous cartilage tissue implant NeoCart for treatment of grade III chondral injury to the distal femur: prospective clinical safety trial at 2 years. Am J Sports Med. 2009;37:1334- 43.

19. Alford JW, Cole BJ. Cartilage restoration, part 2: techniques, outcomes, and future directions. Am J Sports Med. 2005;33:443-60.

20. Cole BJ, Pascual-Garrido C, Grumet RC. Surgical management of articular cartilage defects in the knee. J Bone Joint Surg Am. 2009;91:1778-90.

21. Steadman JR, Rodkey WG, Singleton SB, Briggs KK. Microfracture technique for full-thickness chondral defects: technique and clinical results. Oper Tech Orthop. 1997;7:300-4.

22. Mithoefer K, Williams RJ 3rd, Warren RF, Potter HG, Spock CR, Jones EC, Wickiewicz TL, Marx RG. Chondral resurfacing of articular cartilage defects in the knee with the microfracture technique. Surgical technique. J Bone Joint Surg Am. 2006;88 Suppl. 1 Pt 2:294-304.

23. Williams RJ 3rd, Harnly HW. Microfracture: indications, technique, and results. Instr Course Lect. 2007;56:419-28.

24. Mithoefer K, Williams RJ 3rd, Warren RF, Potter HG, Spock CR, Jones EC, Wickiewicz TL, Marx RG. The microfracture technique for the treatment of articular cartilage lesions in the knee. A prospective cohort study. J Bone Joint Surg Am. 2005;87:1911-20.

25. Irrgang JJ, Anderson AF, Boland AL, Harner CD, Kurosaka M, Neyret P, Richmond JC, Shelborne KD. Development and validation of the international knee documentation committee subjective knee form. Am J Sports Med. 2001;29:600-13.

26. Greco NJ, Anderson AF, Mann BJ, Cole BJ, Farr J, Nissen CW, Irrgang JJ. Responsiveness of the International Knee Documentation Committee Subjective Knee Form in comparison to the Western Ontario and McMaster Universities Osteoarthritis Index, modified Cincinnati Knee Rating System, and Short Form 36 in patients with focal articular cartilage defects. Am J Sports Med. 2010;38: 891-902.

27. Roos EM, Engelhart L, Ranstam J, Anderson AF, Irrgang JJ, Marx

14. Tuan RS. A second-generation autologous chondrocyte implantation approach to the treatment of focal articular cartilage defects. Arthritis Res Ther. 2007;9:109.
15. Blahut EB, Kusanagi A, Mascarenhas AC, Johnson J, Berlowitz-Tarrant LJ, Mizuno S. Effects of medium flow rate on extracellular matrix accumulation by swine
RG, Tegner Y, Davis AM. ICRS recommendation document: patient-reported outcome instruments for use in patients with articular cartilage defects. Cartilage. 2011;2: 122-36.

28. Bekkers JE, de Windt TS, Raijmakers NJ, Dhert WJ, Saris DB. Validation of the Knee Injury and Osteoarthritis Outcome Score (KOOS) for the treatment of focal cartilage lesions. Osteoarthritis Cartilage. 2009;17:1434-9.

29. Roos EM, Lohmander LS. The Knee injury and Osteoarthritis Outcome Score (KOOS): from joint injury to osteoarthritis. Health Qual Life Outcomes. 2003;1:64.

30. Snapinn SM, Jiang Q. Responder analyses and the assessment of a clinically relevant treatment effect. Trials. 2007;8:31.

31. Mithoefer K, McAdams T, Williams RJ, Kreuz PC, Mandelbaum BR. Clinical efficacy of the microfracture technique for articular cartilage repair in the knee: an evidence-based systematic analysis. Am J Sports Med. 2009;37:2053-63.

32. Kon E, Gobbi A, Filardo G, Delcogliano M, Zaffagnini S, Marcacci M. Arthroscopic second-generation autologous chondrocyte implantation compared with microfracture for chondral lesions of the knee: prospective nonrandomized study at 5 years. Am J Sports Med. 2009;37:33-41.

33. Van Assche D, Van Caspel D, Vanlauwe J, Bellemans J, Saris DB, Luyten FP, Staes F. Physical activity levels after characterized chondrocyte implantation versus microfracture in the knee and the relationship to objective functional outcome with 2-year follow-up. Am J Sports Med. 2009;37 Suppl. 1:42S-49S.

34. Saris DB, Vanlauwe J, Victor J, Almqvist KF, Verdonk R, Bellemans J, Luyten FP; TIG/ACT/01/2000&EXT Study Group. Treatment of symptomatic cartilage defects of the knee: characterized chondrocyte implantation results in better clinical outcome at 36 months in a randomized trial compared to microfracture. Am J Sports Med. 2009;37 Suppl. 1:10S-19S.

35. Kusanagi A, Johnson J, Blahut EB, Mascarenhas AC, Mizuno S. Hydrostatic pressure suppresses long term degradation of matrix accumulated by porcine articular chondrocytes in 3D collagen gel/sponges [abstract]. In: Transactions of the 50th Annual Orthopaedic Research Society; 2004 Mar 7-10; San Francisco, CA. Rosemont, IL: Orthopaedic Research Society; 2004. p 0726.

36. Kusanagi A, Blahut EB, Johnson JM, Mascarehas AC, Murata T, Tsuchiya A, Mochizuki M, Mizuno S. Cartilage repair with engineered chondrocyte constructs: a 6 month study in a swine model [abstract]. In: Transactions of the 51st Annual Orthopaedic Research Society; 2005 Feb 20-3; Washington, DC. Rosemont, IL: Orthopaedic Research Society; 2005. p 0302.

37. Kusanagi A, Mascarenhas AC, Blahut EB, Johnson JM, Murata T, Mizuno S. Hydrostatic pressure with low oxygen stimulates extracellular matrix accumulation by human articular chondrocytes in a 3-D collagen gel/sponge [abstract]. In: Transactions of the 51st Annual Orthopaedic Research Society; 2005 Feb 20-3; Washington, DC. Rosemont, IL: Orthopaedic Research Society; 2005. p 0384.

38. Kreuz PC, Steinwachs MR, Erggelet C, Krause SJ, Knorad G, Uhl M, Südkamp N. Results after microfracture of full-thickness chondral defects in different compartments of the knee. Osteoarthritis Cartilage. 2006;14:1119-25.

39. Dumville JC, Hahn S, Miles JN, Torgerson DJ. The use of unequal randomisation ratios in clinical trials: a review. Contemp Clin Trials. 2006;27:1-12.

40. Farrar JT, Dworkin RH, Max MB. Use of the cumulative proportion of responders analysis graph to present pain data over a range of cut-off points: making clinical trial data more understandable. J Pain Symptom Manage. 2006;31:369-77.

41. Burkhoff D, Parides M, Borggrefe M, Abraham WT, Kadish A. ''Responder analysis'' for assessing effectiveness of heart failure therapies based on measures of exercise tolerance. J Card Fail. 2009;15:108-15.

42. U.S. Department of Health and Human Services FDA Center for Drug Evaluation and Research; U.S. Department of Health and Human Services FDA Center for Biologics Evaluation and Research; U.S. Department of Health and Human Services FDA Center for Devices and Radiological Health. Guidance for industry: patient-reported outcome measures: use in medical product development to support labeling claims: draft guidance. Health Qual Life Outcomes. 2006;4:79.

43. Eron J Jr, Yeni P, Gathe J Jr, Estrada V, DeJesus E, Staszewski S, Lackey P, Katlama C, Young B, Yau L, Sutherland-Phillips D, Wannamaker P, Vavro C, Patel L, Yeo J, Shaefer M; KLEAN study team. The KLEAN study of fosamprenavir-ritonavir versus lopinavir-ritonavir, each in combination with abacavir-lamivudine, for initial treatment of HIV infection over 48 weeks: a randomised non-inferiority trial. Lancet. 2006;368:476-82.

44. Tyring S, Gottlieb A, Papp K, Gordon K, Leonardi C, Wang A, Lalla D, Woolley M, Jahreis A, Zitnik R, Cella D, Krishnan R. Etanercept and clinical outcomes, fatigue, and depression in psoriasis: double-blind placebo-controlled randomised phase III trial. Lancet. 2006;367:29-35.

45. Edwards JC, Szczepanski L, Szechinski J, Filipowicz-Sosnowska A, Emery P, Close DR, Stevens RM, Shaw T. Efficacy of B-cell-targeted therapy with rituximab in patients with rheumatoid arthritis. N Engl J Med. 2004;350:2572-81.

46. McMillan-Price J, Petocz P, Atkinson F, O'neill K, Samman S, Steinbeck K, Caterson I, Brand-Miller J. Comparison of 4 diets of varying glycemic load on weight loss and cardiovascular risk reduction in overweight and obese young adults: a randomized controlled trial. Arch Intern Med. 2006;166:1466-75.

An Autologous Cartilage Tissue Implant NeoCart for Treatment of Grade III Chondral Injury to the Distal Femur

Prospective Clinical Safety Trial at 2 Years

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Background: The healing potential of damaged articular cartilage is limited. The NeoCart is a tissue-engineered collagen matrix seeded with autogenous chondrocytes designed for the repair of hyaline articular cartilage.

Hypothesis: The NeoCart implant is well tolerated in the human knee.

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

Methods: Eight patients (treatment group) with full-thickness cartilage injury were treated with the NeoCart and evaluated prospectively. Autogenous chondrocytes provided by arthroscopic biopsy were seeded into a 3-dimensional type I collagen scaffold. The seeded scaffold was subjected to a tissue-engineering protocol including treatment with a bioreactor. Implantation of the prepared cartilage tissue patch was performed via miniarthrotomy and secured with a collagen bioadhesive. Evaluations through 24 months postoperatively included the subjective International Knee Documentation Committee questionnaire, visual analog scale, range of motion, and cartilage-sensitive magnetic resonance imaging (MRI), including quantitative T2 mapping.

Results: Pain scores after NeoCart implantation were significantly lower than baseline at 12 and 24 months after the procedure ($P < .05$). Improved function and motion were also noted at 24 months. Six patients had 67% to 100% defect fill at 24 months with MRI evaluation. One patient had moderate (33%-66%) defect fill, and another patient had poor (less than 33%) defect fill. Partial stratification of T2 values was observed for 2 patients at 12 months and 4 patients at 24 months. No patients experienced arthrofibrosis or implant hypertrophy.

Conclusion: Pain was significantly reduced 12 and 24 months after NeoCart treatment. Trends toward improved function and motion were observed 24 months after implantation. The MRI indicated implant stability and peripheral integration, defect fill without overgrowth, progressive maturation, and more organized cartilage formation.

Keywords: cartilage repair; chondrocyte; chondral repair; articular cartilage injury

Full-thickness cartilage injury is common in the knee.[12] Although mechanical pain may be addressed through removal of damaged tissue, articular cartilage has little capacity to undergo spontaneous repair, and defects may progress to degenerative joint conditions. Marrow stimulation techniques such as microfracture provide largely fibrocartilage repair,[19] which is known to have inferior ability to withstand shear and indentation forces.[1,11] While fibrocartilage repair may be an effective short-term treatment for young patients with small defects,[19,20,30] the essential differences in tissue quality may contribute to

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the clinical deterioration noted as early as 18 months after microfracture.[20]

Other approaches use autologous chondrocytes in an effort to create durable hyaline repair. In autologous chondrocyte implantation (ACI), a suspension of cultured chondrocytes is injected under a periosteal cover.[5] This technique does not consistently achieve hyaline cartilage repair.[16,19,21] Furthermore, complications such as arthrofibrosis and hypertrophy of the implanted periosteum are reported.[7,8,19,26,27,33]

Seeding a chondrocyte population into a scaffold eschews many procedural difficulties and potential postsurgical complications that are attributed to periosteum harvest and implantation.[2,10,14,37] In addition, the improved structural support provided to chondrocytes by a 3-dimensional environment may promote cartilage maturation.[10] The NeoCart implant (Histogenics Corporation, Waltham, Massachusetts), a novel treatment for articular cartilage defects, partners a 3-dimensional type I collagen scaffold seeded with autologous chondrocytes with a tissue-engineering protocol that includes treatment with a bioreactor.[17,31,35] The resulting product is a proteoglycan- and glycosaminoglycan-rich, viable, and dynamic tissue-like implant. Preclinical trials, including a porcine model of full-thickness femoral cartilage injury, demonstrated that NeoCart implantation leads to hyaline-like repair cartilage, as compared with empty defects or matrix alone (Kusanagi, personal communication, 2003). Preclinical safety evaluations suggested that the NeoCart would be well tolerated in the human knee. The purpose of this report is to describe the initial experience with NeoCart therapy in a clinical population for the treatment of full-thickness cartilage injury. All data are derived from an ongoing prospective Food and Drug Administration (FDA) phase I clinical trial.

MATERIALS AND METHODS

Study Population

This FDA- and Institutional Review Board (IRB)–approved phase I prospective clinical trial was designed to evaluate safety as well as early clinical and radiographic outcomes of the NeoCart implant. Ten patients were enrolled at 2 investigational sites. Written consent was obtained from all patients. Eligible patients were between 18 and 55 years of age, had a symptomatic grade III (full-thickness) cartilage lesion of the femoral condyle, and otherwise satisfied inclusion and exclusion criteria (see Appendix, available in the online version of this article at http://ajs.sagepub.com/supplemental/).

Study Design

NeoCart development is summarized in Figure 1. After screening and consent, all patients underwent arthroscopic evaluation of the cartilage defect and surrounding structures. Articular cartilage (200-400 mg) was taken at the time of arthroscopy from a nonweightbearing portion of

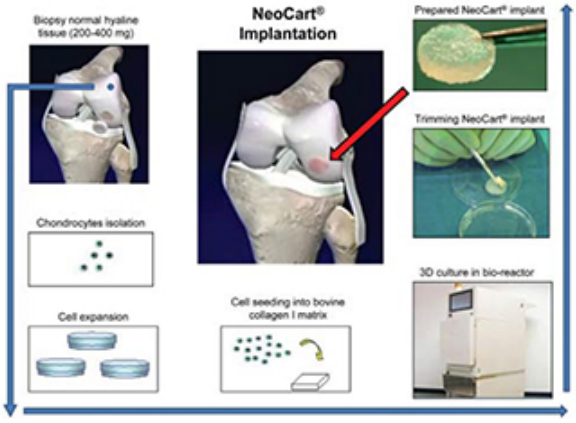
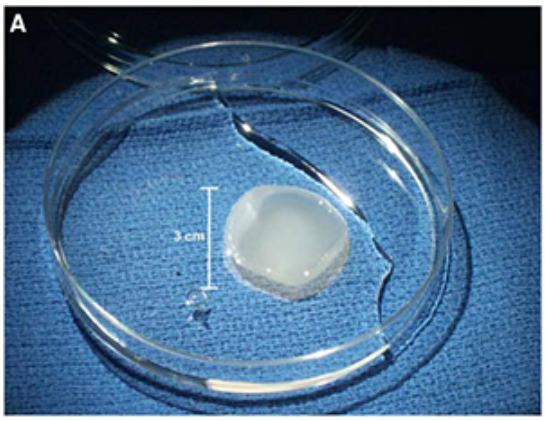


Figure 1. NeoCart production and implantation. Chondrocytes are harvested via arthroscopic biopsy and seeded into a bovine type I collagen matrix. Ex vivo maturation in a bioreactor provides the implant, which is surgically fixed to the damaged area with CT3 bioadhesive.

the femoral condyle or from the femoral notch of the ipsilateral knee. The subchondral bone was not penetrated. One patient had ACL reconstruction with hamstring tendon autograft at the time of arthroscopy. Five patients underwent arthroscopic debridement, and one patient had removal of a loose body. The biopsy specimen was packaged for sterile transport and shipped to Histogenics Corporation for processing.

The biopsy specimen was processed to yield chondrocytes for NeoCart. Chondrocytes were expanded and seeded into a bovine type I collagen 3-dimensional honeycomb matrix. The seeded scaffold was processed in a bioreactor in which culture conditions, including hydrostatic pressure, induced chondrocytes to synthesize cartilage glycoproteins. Subsequent static culture further encouraged chondrocyte expression. The total implant development averaged 67 ± 18 days.

The NeoCart implantation procedure was performed successfully in 8 of 10 patients. The implant bed was prepared by debridement of the damaged chondral tissue, including removal of the calcified cartilage. Surgical goals included avoiding both penetration of the subchondral bone and osseous bleeding. The NeoCart was then cut to size and secured within the defect bed (Figure 2). The first 2 implantation procedures were deemed unsuccessful as a result of damage to the NeoCart from suturing and intraoperative motion testing. In subsequent procedures (the 8 reported patients), the NeoCart was secured without sutures, using only a thin layer of CT3 (Histogenics), a proprietary tissue adhesive polymer composed of collagen and polyethylene glycol components, spread below and atop the NeoCart implant. Intraoperative manipulation of the surgical knee was subsequently limited to bringing the knee into full extension for incision closure. A knee immobilizer was used for 10 ± 2 days after implantation at all times.



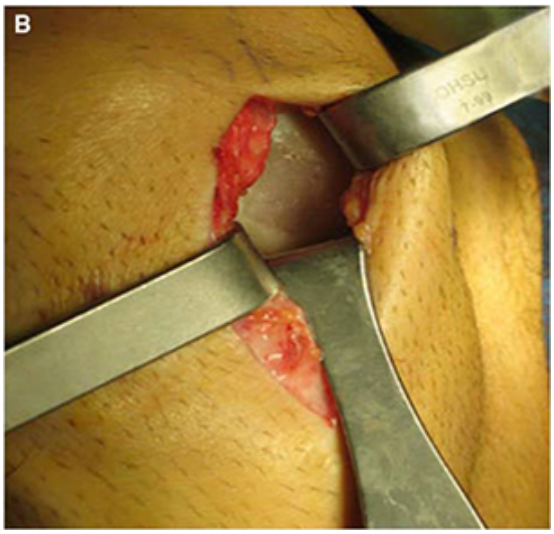


Figure 2. A, The NeoCart implant. B, The implant in situ, viewed via miniarthrotomy.

Rehabilitation Program

Unrestricted active or passive range of motion was encouraged after the knee immobilizer was discontinued. Patients used a continuous passive motion machine for a minimum of 6 hours a day during the 6 weeks after immobilizer use. For the first 6 weeks after implant surgery, patients were nonweightbearing. Unrestricted weightbearing was allowed afterward, with patients restricted to rehabilitation analogous to femoral condylar microfracture and ACI type protocols.[3,15,38]

Assessment of Clinical and MRI Outcomes

Clinical evaluations were conducted preoperatively, at 6 and 12 weeks, and at 6, 12, and 24 months after implantation. Each evaluation included routine reporting of the visual analog scale (VAS) pain score at rest, completion of the International Knee Documentation Committee (IKDC) subjective questionnaire, and measurement of knee range of motion. An MRI was performed at 12 weeks and 12 and 24 months after implantation. Two patients had an additional MRI at 2 and 4 weeks, and 3 patients had MRI between 7 and 9 months after implantation.

All MRI images were obtained on a clinical 1.5-T MRI unit (Signa HD Excite or HDx, General Electric Healthcare, Milwaukee, Wisconsin) using a standard receive-only 8-channel phased array or quadrature knee coil (in vivo extremity coil, InVivo Coporation, Orlando, Florida). Images were obtained using a previously validated fast spin echo pulse sequence for assessing articular cartilage.[34] Coronal, axial, and sagittal fast spin echo images were performed using a repetition time (TR) of 3500 to 6000 milliseconds, echo time (TE) of 34 milliseconds (effective), field of view (FOV) of 14 (axial) to 16 (sagittal) cm, matrix of 512 × 256 (axial) to 384 (sagittal), slice thickness of 3 mm (coronal) to 4 mm (sagittal) with no gap, receiver bandwidth (over the entire frequency range) of 31.25 kHz, at 2 excitations. An additional fat-suppressed fast spin echo sequence (Chemsat, General Electric Healthcare) was obtained in the sagittal plane using an effective TE of 45 milliseconds, matrix of 256 × 224, and slice thickness of 4 mm with no gap. Echo train length varied between 6 and 12. Cartilage morphological characteristics were evaluated using a previously reported series of imaging parameters including signal intensity of the repair cartilage relative to the surrounding cartilage (hypointense, isointense, or hyperintense, measured using a standardized region of interest [ROI] in the center of the repair with a standard of deviation on an MRI workstation), gross appearance (depressed, flush, or proud), the presence or absence of hypertrophy or displacement, subchondral edema (mild [<1 cm^2], moderate [1-2 cm^2], or severe [>2 cm^2]), bony overgrowth (absence or presence), interface with adjacent cartilage (absence, presence, size of fissure [<2 mm or >2 mm]), percentage of fill based on both coronal and sagittal images (0%-33%, 34%-66%, or 67%-100%), integrity of adjacent cartilage (modified International Cartilage Repair Society [ICRS] classification),[6] integrity of opposing cartilage (modified ICRS classification), fat pad scarring (mild, moderate, or severe), and synovial reaction (none, mild, moderate, severe).[7]

T2 mapping was performed using a multislice, multi-echo, modified Carr Purcell Meiboom Gill (CPMG) pulse sequence, which uses interleaved slices and tailored refocusing pulses to minimize contribution from stimulated echoes.[24] Standard T2-mapping pulse sequence parameters used were a TR of 800 milliseconds, 8 echoes sampled using sequential multiples of the first TE (9-10 milliseconds), FOV of 16 cm^2, matrix of 256 to 384 × 256, providing a minimum in-plane resolution of 254 μ in the frequency direction by 312 μ in the phase direction, by slice resolution of 2.0 to 3.0 mm with no gap, and a receiver bandwidth of 62.5 kHz. After image acquisition, data sets were analyzed on a pixel-by-pixel basis with a 2-parameter weighted least-squares fit algorithm, assuming a monoexponential fit (Functool 3.1, General

TABLE 1
Patient and Lesion Demographics[a]

Case No.	Sex	Age at Implant, y	Body Mass Index	Location of Injury (distal femur)	Size of Injury, cm	Time Since Injury, y	Previous Surgeries of the Ipsilateral Knee[b]
1010	M	25	28	Medial condyle	1.9 × 1.5	>2	ACL reconstruction; Partial meniscectomy and debridement
3001	M	43	27	Medial condyle	1.0 × 2.0	>2	ACL reconstruction
3002	F	43	24	Lateral condyle	1.0 × 1.2	>20	Removal of loose body; Debridement and removal of loose body
3003	M	43	28	Medial condyle	1.4 × 2.0	>25	Partial meniscectomy (2); Meniscus repair (2)
3004	M	34	38	Medial condyle	1.0 × 2.0	0.5	None
3006	M	46	29	Medial condyle	1.5 × 2.0	>6	Arthroscopic debridement (2)
3007	F	46	20	Medial condyle	1.1 × 1.6	0.75	Partial meniscectomy and debridement
3008	F	26	25	Lateral condyle	1.3 × 1.3	>3	None
Mean (range)		38 (25-46)	27 (20-38)		1.3 × 1.7	>7	1.5 (0-4)

[a] M, male; F, female; ACL, anterior cruciate ligament.
[b] Not including arthroscopic biopsy harvest.

Electric Healthcare). Quantitative T2 values were calculated by taking the natural logarithm of the signal decay curve in a selected ROI. The ROIs were obtained in a standardized fashion from the articular cartilage over the center of the cartilage repair, both within the deep and superficial 50% of the cartilage repair, as well as of the adjacent and opposite articular cartilage surfaces. Care was taken not to sample close to the tidemark/subchondral plate to avoid partial volume effects of sampling any misregistration due to residual chemical shift. All MRI images were evaluated by 2 experienced musculoskeletal MRI radiologists.

Safety

Safety was assessed by physical examinations, clinical laboratory tests, and monitoring of adverse events (AEs) throughout the study period. All AEs were monitored until resolved and reported as required by each IRB and the FDA.

RESULTS

Patient Enrollment

Ten patients were enrolled as the intention to treat group. The 8 patients who had the NeoCart surgically secured with CT3 alone are reported as the treatment group. For 2 other patients, the implant was damaged as a result of suturing and intraoperative motion testing. Data for these 2 patients are not reported here.

The treatment group included 3 women and 5 men, with a mean age of 38 years (range, 25-46) and a mean body mass index (BMI) of 27 (range, 20-38) (Table 1). All patients had an isolated grade III chondral injury to the weightbearing region of either the medial or lateral femoral condyle. Injuries were typically chronic; 6 patients reported symptoms greater than 2 years before treatment. Defects were associated with trauma (n = 3), focal osteoarthritis (n = 3), prior ACL injury (n = 2), and osteonecrosis (same knee, opposite condyle) (n = 1). Defects averaged 1.3 × 1.7 cm, or 2.2 cm^2 (range, 1.2-3.0 cm^2). No patient had prior treatment of the study defect other than arthroscopic debridement. All patients were evaluated up to or exceeding 24 months.

Safety

No serious AEs occurred, and no patients were discontinued from the study because of AEs. Twelve AEs were considered at least possibly related to the implant. They included normal postsurgical sequelae such as pain, swelling, and numbness at the site of incision. A subchondral cyst of dimensions 1.9 × 1.8 × 1.3 cm was noted at 12-month MRI for one patient and appeared larger at 24 months. This patient had preimplant evidence of osteonecrosis (without collapse) of the ipsilateral knee lateral condyle (medial condyle treated with the NeoCart), as well as prior ACL reconstruction (Figure 3). There were 2 severe AEs possibly related to the study implant. One involved a patellar fracture that occurred after a fall in the index knee 6 months after NeoCart treatment. Another patient suffered a meniscal tear in the index compartment within 3 months of implantation (Figure 4). The implant remained stable in both cases. No infections or study-related interventions occurred in the treatment group. Incomplete attachment was described for the initial MRIs (3 and 9 months) for 2 patients. At subsequent MRIs, these implants remained stable, with good defect fill (67%-100%) seen at both 12 and 24 months and partial stratification of T2 values observed at 24 months.

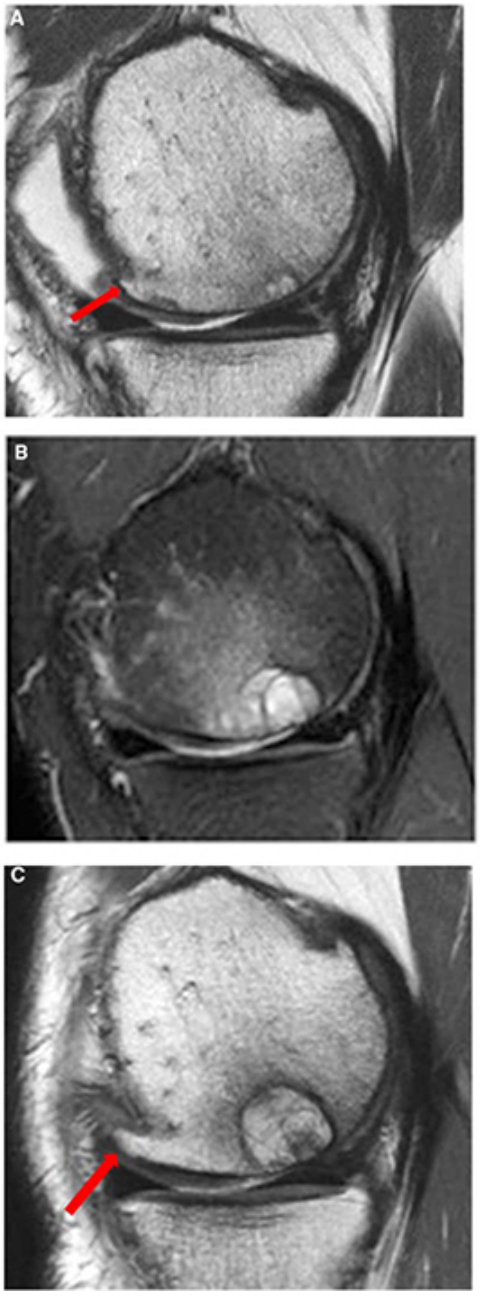


Figure 3. Sagittal fast inversion recovery and fast spin echo magnetic resonance images over the site of cartilage repair in a patient with osteonecrosis of the opposite femoral condyle. Persistent poor fill of the defect demonstrated by hyperintense repair cartilage is accompanied by a progressive subchondral cystic change and bone marrow edema. A, Three months after NeoCart implantation with adjacent osteophyte (arrow). B, Thirteen months after NeoCart implantation. C, Twenty-five months after NeoCart implantation. Osteophyte noted with arrow.

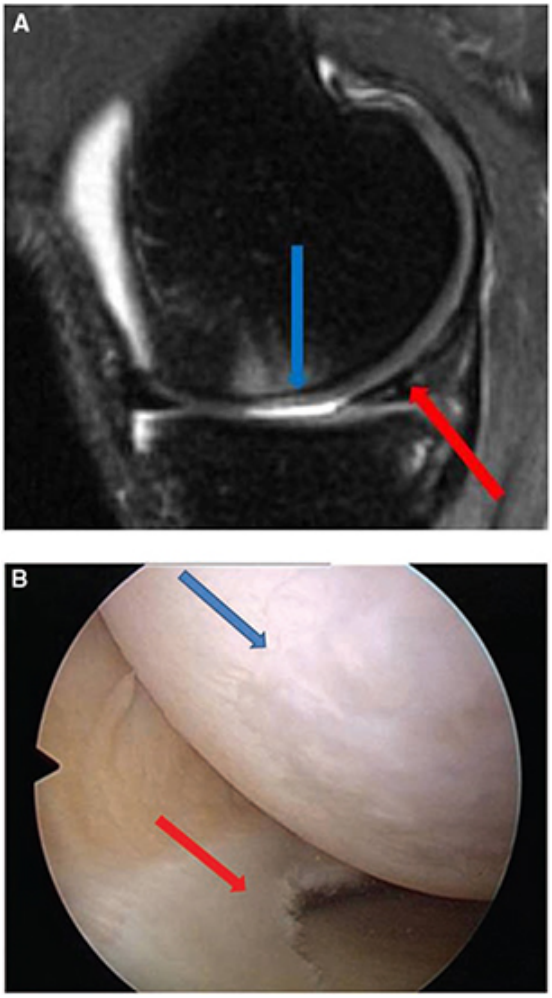


Figure 4. A, Sagittal magnetic resonance image of the NeoCart implant (gray arrow) at 12 weeks with adjacent posterior horn meniscal tear (white arrow). B, Arthroscopic image of the NeoCart implant in situ after arthroscopic meniscus debridement. The NeoCart (blue arrow) appears flush and integrated to the area of medial meniscectomy (red arrow).

Implant failures associated with the first 2 implantation procedures were considered to be AEs. To date, additional AEs for these patients include only superficial venous thrombosis of the left forearm for one patient, a complication of intravenous placement on the day of surgery.

Clinical Outcomes

All 10 patients completed all benchmarks through 24 months. Clinical outcomes represent the treatment group (8 patients). The VAS significantly decreased to an average of 0.9 ± 1.5 at 12 months, down from 3.3 ± 2.8 at baseline ($P < .05$). Pain scores at 24 months remained significantly

TABLE 2
Clinical Outcomes After NeoCart Implantation[a]

Assessments		Case No. 1010	3001	3002	3003	3004	3006	3007	3008
Visual analog scale	Baseline	5.92	3.35	1.48	1.33	0.47	6.78	0.47	6.67
	6 weeks	2.31	0.95	0.68	0.79	0.40	4.16	1.14	2.23
	3 months	0.41	1.66	0.46	0.34	0.20	6.47	5.27	2.03
	6 months	0.61	1.14	0.34	0.47	0.74	5.80	2.15	0.81
	1 year	0.00	1.08	0.54	0.07	0.34	4.59	0.27	0.13
	2 years	0.00	1.02	0.95	0.00	0.82	2.30	0.20	0.30
Range of motion, deg	Baseline	144	126	137	115	117	130	130	125
	6 weeks	140	115	125	125	105	120	125	135
	3 months	130	125	130	130	115	125	130	135
	6 months	136	140	125	135	115	125	135	135
	1 year	140	135	130	135	120	125	135	135
	2 years	145	135	125	135	130	135	140	145
International Knee Documentation Committee score	Baseline	13.79	68.97	68.97	75.86	68.60	25.29	80.46	52.87
	6 weeks	22.78	48.28	29.89	43.68	58.62	6.90	42.53	37.93
	3 months	45.78	64.37	65.52	68.97	54.02	13.79	49.43	50.57
	6 months	56.32	68.97	73.56	73.56	64.37	33.33	59.77	65.52
	1 year	72.00	89.66	87.36	100.00	62.07	34.94	62.07	81.61
	2 years	73.56	85.06	77.01	100.00	72.41	49.43	59.77	93.10

[a] NeoCart from Histogenics Corporation.

lower than at baseline ($P < .05$). Average range of motion improved from 128° ± 10° at baseline to 136° ± 7° at 24 months. Range of motion improved in 7 of 8 patients during the study period. No patients developed arthrofibrosis. Knee function, assessed with the IKDC, improved in 7 of 8 patients from 57 ± 25 at baseline to 76 ± 17 at 24 months (Table 2).

MRI Outcomes

The MRI at 24 months showed 6 of 8 patients with good to complete (67%-100%) defect fill. One patient had moderate (33%-66%) defect fill, and another patient had poor (<33%) defect fill. This last patient developed a subchondral cyst adjacent to the implant. No soft tissue hypertrophy was noted throughout the study period. Two patients demonstrated partial stratification of T2 values similar to hyaline cartilage at 12 months. At 24 months, 4 of 8 patients had stratification of T2 values; however, all repairs showed prolongation of quantitative T2 values in both the superficial and deep components of the repair tissue (Figures 5 and 6).

Assessment of peripheral integration at 12 weeks found 2 patients with fissures less than 2 mm and 6 patients with fissures greater than 2 mm. Improved integration was seen in 4 of 8 patients at 12 months. At this time point, 1 implant was completely integrated with surrounding tissue, 4 had fissures less than 2 mm, and 3 had fissures greater than 2 mm. Integration continued to improve at 24 months. Two implants were completely integrated, including one that at 12 months had fissures less than 2 mm. Three implants had fissures less than 2 mm, and 3 had fissures greater than 2 mm.

DISCUSSION

Suboptimal repair of articular cartilage defects with current methods inspires investigation into the potential use of tissue-engineered cartilage. The autologous cartilage tissue implant (ACTI) NeoCart is an implantable cell/matrix-based implant with characteristics of hyaline cartilage. This product is developed from a 3-dimensional collagen scaffold seeded with autologous cells and incubated using physiological stimuli.

This study provides the first evaluation of the NeoCart in the human knee. Data collected through 24 months for all patients suggest implant safety. No serious AEs were associated with the NeoCart implant, procedures, or rehabilitation. The implant remained stable for all patients when secured with the CT3 bioadhesive alone. It was determined from the first 2 (unsuccessful) implantation procedures that suture fixation and/or immediate intraoperative motion testing may damage the NeoCart implant and cause detachment. No serious AEs were noted in the 2 patients who did not receive the implant. One underwent uneventful microfracture treatment.

Two-year preliminary data for this small number of phase I patients suggest beneficial clinical outcomes. A significant reduction in subjective pain was observed as early as 12 months after NeoCart treatment and sustained through 24 months ($P < .05$). Knee function as measured

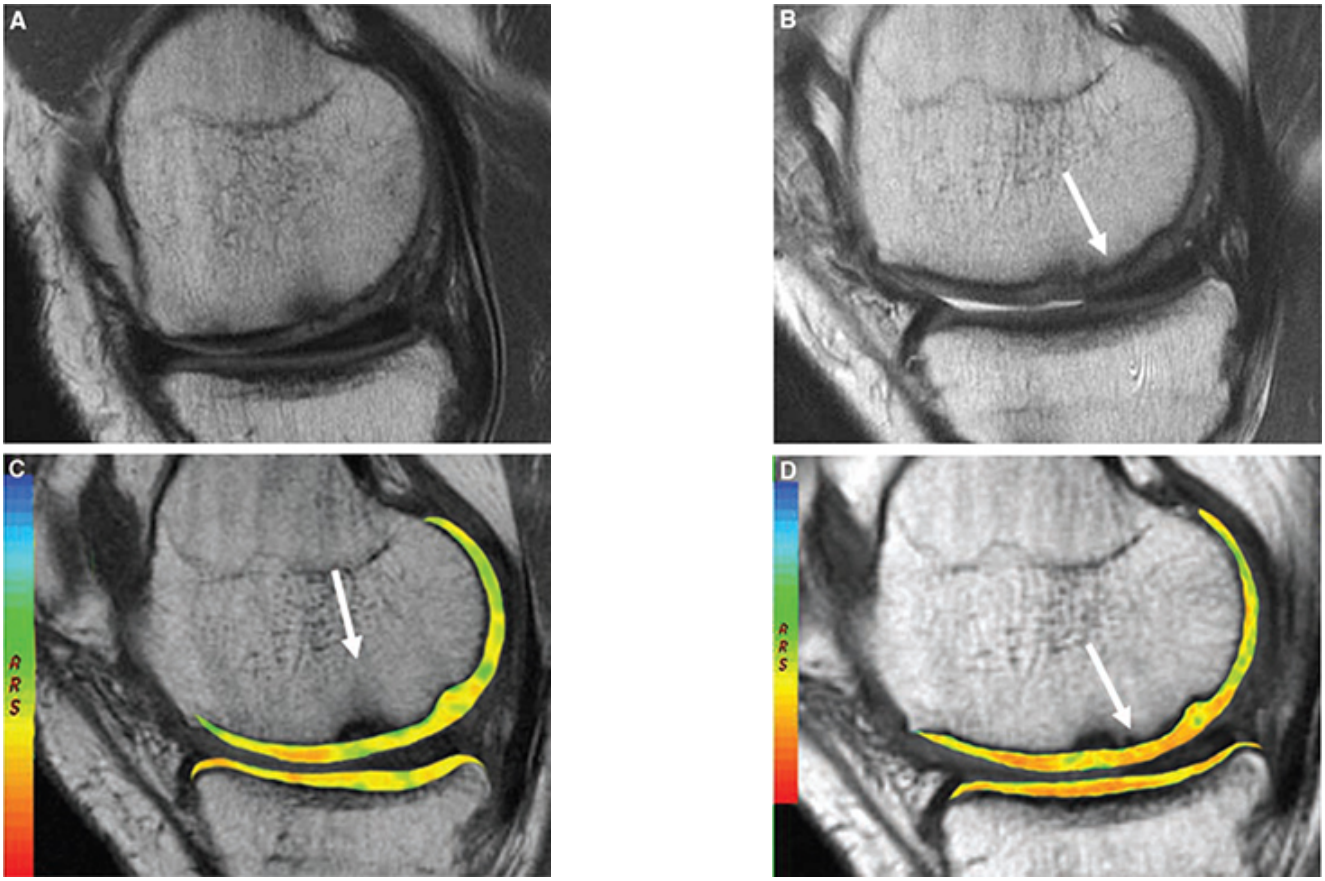


Figure 5. A, Sagittal magnetic resonance images after implantation of a NeoCart patch over the medial femoral condyle. Cartilage-sensitive magnetic resonance image at 1 year demonstrates fill of the defect by repair tissue that is hyperintense relative to native cartilage. B, Good fill was maintained at 2 years, with progressive decrease in signal intensity of the repair cartilage (arrow) undergoing "maturation." C, Corresponding T2 mapping at 1 year demonstrates prolongation of T2 values at the site of cartilage repair (arrow). D, Corresponding T2 mapping at 2 years demonstrates partial stratification of T2 values (arrow).

by the IKDC score also improved for 7 of 8 patients. The exception suffered a new meniscal tear of the index knee. All patients gained or preserved range of motion arc versus presurgery measures. This was not statistically significant but may be an important finding based on contrast with the association of cartilage restoration procedures such as ACI with potential arthrofibrosis. In a study of 169 patients treated with ACI, Minas reported a 5% incidence of arthrofibrosis.[27] Outcomes review of the Carticel outcomes registry, with 891 patients at the time of the authors' review, found the incidence of arthrofibrosis after ACI to be 3.1%.[25] Of particular interest, our 8 patients all demonstrated improvement in at least 2 of 3 key efficacy measurements: VAS pain, IKDC score, or range of motion at 12 and 24 months after NeoCart treatment. At 12 months, 3 patients improved in all 3 efficacy categories compared with baseline. By 24 months, 5 of 8 demonstrated improvement from baseline evaluation for all 3 measurements.

Good to complete defect fill (67%-100%) was observed in MRI studies for 6 of 8 patients at 24 months. While clinical outcomes and fill grade are positively correlated,[29,30] volume of fill has varied considerably for the microfracture procedure.[9,13,29] The MRI of 19 patients at an average of 3 years after microfracture evidenced that only 42% of patients had 67% to 100% defect fill, while 31% to 66% and 0% to 30% defect fill were noted in 21% and 37% of patients, respectively.[13] No patients had overgrowth of the NeoCart patch. By contrast, graft and periosteum hypertrophy are known complications of the ACI technique that often necessitate removal or debridement.[7,8,19,33] In one randomized comparison of ACI and microfracture, 25% of patients in the ACI group, as opposed to 10% of patients in the microfracture group, required arthroscopic debridement before second-look arthroscopy at 2 years.[19] Another comparison demonstrated an incidence of hypertrophy at 1 year after study surgery of 25% for patients receiving characterized

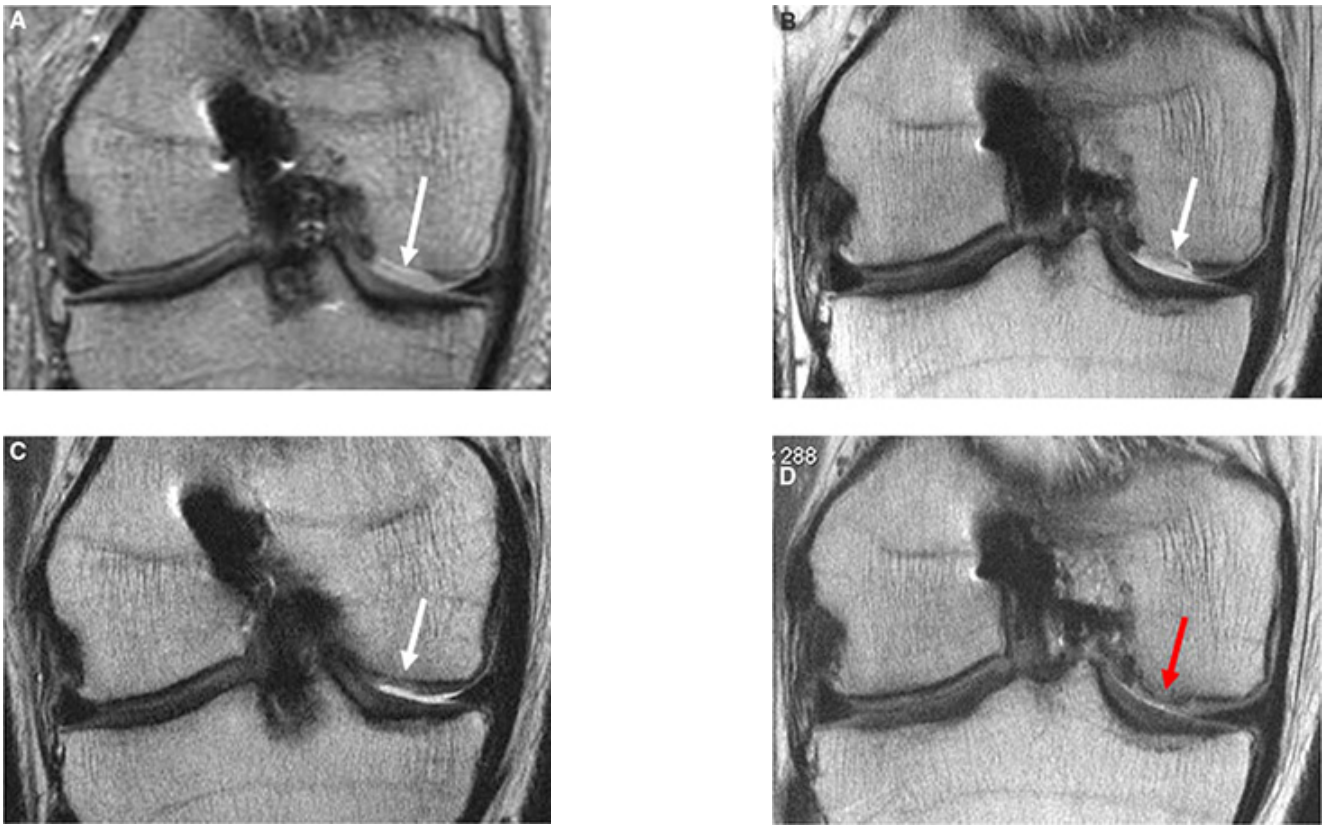


Figure 6. Coronal magnetic resonance images after implantation of the NeoCart patch over the medial femoral condyle demonstrate good fill of the cartilage defect as well as progressive decreased signal intensity of the repair tissue, approaching that of adjacent native cartilage (white arrows). A, Ten days after NeoCart implantation. B, One month after NeoCart implantation. C, Three months after NeoCart implantation. D, Eleven months after NeoCart implantation. Development of focal underlying subchondral bony remodeling is noted (arrow).

chondrocyte implantation and 8% for patients receiving microfracture.[36] A large study of 349 ACI procedures reported that the incidence of symptomatic hypertrophy was 15.8% for patients receiving periosteum-covered ACI (ACI-P) and only 1.9% and 4.9% for patients receiving membrane-covered ACI (ACI-C) and matrix-associated ACI (MACI), respectively.[32] These findings suggest that hypertrophy may be avoided by the use of collagen matrix in place of periosteum as in the NeoCart procedure.

Quantitative T2 mapping was used to assess the repair tissue. Stratification of T2 values, with shorter relaxation times in the basilar components and longer values in the superficial components, has been correlated with the organization of collagen fibrils similar to that of normal articular cartilage.[7] At 12 months after surgery, 25% of patients demonstrated partial T2 stratification. While the numbers are small, this figure increased to 50% at 24 months. All patients with partially stratified cartilage at 12 months maintained this pattern of stratification at 24 months. These results are consistent with continued maturation of the NeoCart implant into hyaline-like repair cartilage. T2 stratification was not observed in 4 patients. Of these 4, one patient sustained a new meniscal tear in the index knee on return to activity, and another patient had 4 previous debridement surgeries and preimplantation radiographs that suggested early osteoarthritis. Multiple knee surgeries before cartilage restoration surgery are known to correlate with inferior clinical scores[21] and delayed return to sport.[30]

Clinical and MRI outcomes are particularly encouraging in relation to the chronicity of symptoms, age range, and BMI. Of the 8 patients receiving NeoCart treatment, 6 reported symptoms greater than 2 years previous to study surgery. In other studies, patients with chronic injuries were more likely to demonstrate less functional improvement[2,13,21,29] and inferior fill grade[29] compared with patients with acute injury. Five patients were above age 40 years at the time of index surgery. Of the 4 patients

with T2 stratification, 2 were at least 40 years of age. Younger patients, often defined as under 30 years of age, have the most success with microfracture[19,20,29] and ACI.[2,19,21,22] Patients with T2 stratification had a BMI encompassing the entire study range of 20 to 38. The only patient to not report improved knee function, as based on the IKDC score, had the lowest BMI. By contrast, a BMI greater than 30 has been shown to correlate with inferior clinical outcomes for the microfracture procedure.[29]

Early clinical and MRI outcomes suggest that the NeoCart therapy may be a safe and promising alternative to current restorative techniques for partial to full-thickness cartilage defects. This initial safety trial suggests that concerns such as arthrofibrosis and graft hypertrophy associated with ACI-type techniques are potentially avoided with the NeoCart technique. Comparison of NeoCart clinical outcomes with other cartilage restoration therapies such as microfracture, ACI-P,[5] and scaffold techniques such as ACI-C,[5,14] MACI,[10] and other scaffold techniques, for example, Hyalograft C[39] and Cartipatch,[37] is currently premature given the small number of patients in this trial. Previous comparisons of the effectiveness of these techniques for the treatment of articular cartilage injury have produced mixed results. A recent review of cartilage restoration techniques including microfracture, ACI-P, ACI-C, and MACI found no one superior cartilage restoration technique.[23] The authors of the review suggest that microfracture should be considered a first-line treatment, based upon findings that microfracture is not inferior to ACI techniques, may not preclude secondary treatments, and may require less planning and equipment.[23] Unfortunately, microfracture has been associated with a failure rate of 23% at 5 years[18] and a deterioration of clinical outcomes as early as 18 months after surgery.[20] In addition, inferior results for ACI after failed microfracture have been reported, raising concern for microfracture as a primary therapy.[28] Taken together, this suggests a need for an alternative first-line treatment that restores the hyaline matrix and does not violate the subchondral bone. A prospective, randomized controlled investigation comparing clinical and MRI outcomes in the NeoCart versus microfracture is currently underway.

REFERENCES

1. Akeson WH, Bugbee W, Chu C, Giurea A. Differences in mesenchymal tissue repair. *Clin Orthop Relat Res*. 2001;391:S124-141.
2. Bartlett W, Skinner JA, Gooding CR, et al. Autologous chondrocyte implantation versus matrix-induced autologous chondrocyte implantation for osteochondral defects of the knee. *J Bone Joint Surg Br*. 2005;87:640-645.
3. Bartz RL, Steadman JR, Rodkey WG. The technique of microfracture of full-thickness chondral lesions and postoperative rehabilitation. *Tech Knee Surg*. 2004;3:198-203.
4. Briggs TWR, Mahroof S, David LA, Flannelly J, Pringle J, Bayliss M. Histological evaluation of chondral defects after autologous chondrocyte implantation of the knee. *J Bone Joint Surg Br*. 2003;85: 1077-1083.
5. Brittberg M, Lindahl A, Nilsson A, Ohlsson C, Isaksson O, Peterson L. Treatment of deep cartilage defects in the knee with autologous chondrocyte transplantation. *N Engl J* Med. 1994;331:889-895.
6. Brittberg M, Winalski CS. Evaluation of cartilage injuries and repair. *J Bone Joint Surg Am*. 2003;85(Suppl 2):58-69.
7. Brown WE, Potter HG, Marx RG, Wickiewicz TL, Warren RF. Magnetic resonance imaging appearance of cartilage repair in the knee. *Clin Orthop Relat Res.* 2004;422:214-223.
8. Browne JE, Anderson AE, Arciero R, et al. Clinical outcomes of autologous chondrocyte implantation at 5 years in US patients. *Clin Orthop Relat Res.* 2005;436:237-245.
9. Buckwalter JA, Mow VC, Ratcliffe A. Restoration of injured or degenerated articular cartilage. *J Am Acad Orthop Surg.* 1994;2:192-201.
10. Cherubino P, Grassi FA, Bulgheroni P, Ronga M. Autologous chondrocyte implantation using a bilayer collagen membrane: a preliminary report. *J Orthop Surg.* 2003;11:10-15.
11. Coletti JM, Akeson WH, Woo SL. A comparison of the physical behavior of normal articular cartilage and the arthroplasty surface. *J Bone Joint Surg Am.* 1972;54:147-160.
12. Curl WW, Krome J, Gordon ES, Rushing J, Smith BP, Poehling GG. Cartilage injuries: a review of 31516 knee arthroscopies. *Arthroscopy.* 1997;13:456-460.
13. Gill TJ, MacGillivray JD. The technique of microfracture for the treatment of articular cartilage defects in the knee. *Oper Tech Orthop.* 2001;11:105-107.
14. Haddo O, Mahroof S, Higgs D, et al. The use of chondrogide membrane in autologous chondrocyte implantation. *Knee.* 2004;1:51-55.
15. Hambly K, Bobic V, Wondrasch B, Van Assche D, Marlovits S. Autologous chondrocyte implantation postoperative care and rehabilitation. *Am J Sports Med.* 2006;34:1020-1038.
16. Horas U, Pelinkovic D, Herr G, Aigner T, Schnettler R. Autologous chondrocyte implantation and osteochondral cylinder transplantation in cartilage repair of the knee joint. *J Bone Joint Surg Am.* 2003; 85:185-192.
17. Kim HT, Zaffagnini S, Mizuno S, Abelow S, Safran MR. A peek into the possible future of management of articular cartilage injuries: gene therapy and scaffolds for cartilage repair. *J Orthop Sports Phys Ther.* 2006;36:765-773.
18. Knutsen G, Drogset JO, Engebretsen L, et al. A randomized trial comparing autologous chondrocyte implantation with microfracture: findings at five years. *J Bone Joint Surg Am.* 2007;89:2105-2112.
19. Knutsen G, Engebretsen L, Ludvigsen TC, et al. Autologous chondrocyte implantation compared with microfracture in the knee. *J Bone Joint Surg Am.* 2004;86:455-464.
20. Kreuz PC, Steinwachs MR, Erggelet C, et al. Results after microfracture of full-thickness chondral defects in different compartments of the knee. *Osteoarthritis Cartilage.* 2006;14:1119-1125.
21. Krishnan SP, Skinner JA, Bartlett W, et al. Who is the ideal candidate for autologous chondrocyte implantation? *J Bone Joint Surg Br*. 2006;88:61-64.
22. Krishnan SP, Skinner JA, Carrington RWJ, Flanagan AM, Briggs TWR, Bentley G. Collagen-covered autologous chondrocyte implantation for osteochondritis dissecans of the knee. *J Bone Joint Surg Br.* 2006;88:203-205.
23. Magnussen RA, Dunn WR, Carey JL, Spindler KP. Treatment of focal articular cartilage defects in the knee: a systematic review. *Clin Orthop Relat Res.* 2008;466:952-962.
24. Maier CF, Tan SG, Hariharan H, Potter HG. T2 quantitation of articular cartilage at 1.5 T. *J Magn Reson Imaging.* 2003;17:358-364.
25. Mandelbaum BR, Browne JE, Fu F, et al. Articular cartilage lesions of the knee. *Am J Sports Med.* 1998;26:853-861.
26. Micheli LJ, Browne JE, Erggelet C, et al. Autologous chondrocyte implantation of the knee: multicenter experience and minimum 3-year follow-up. *Clin J Sport Med.* 2001;11:223-228.
27. Minas T. Autologous chondrocyte implantation of focal chondral defects of the knee. *Clin Orthop Relat Res.* 2001;391:S349-361.
28. Minas T, Gomoll AH, Rosenberger R, Royce RO, Bryant T. Increased failure rate of autologous chondrocyte implantation after previous treatment with marrow stimulation techniques. *Am J Sports Med.* 2009 Mar 4 [Epub ahead of print].
29. Mithoefer K, Williams RJ, Warren RF, et al. The microfracture technique for the treatment of articular cartilage lesions in the knee: a prospective cohort study. *J Bone Joint Surg Am.* 2005;87:1911-1920.
30. Mithoefer K, Williams RJ, Warren RF, Wickiewicz TL, Marx RG. High-impact athletics after knee articular cartilage repair: a prospective

evaluation of the microfracture technique. *Am J Sports Med.* 2006; 34:1413-1418.
31. Mizuno S, Johnson K. Cell and tissue culture technology for cartilage repair. *Orthopaedic Journal at Harvard Medical School.* 2005;7:71-73.
32. Niemeyer P, Pestka JM, Kreuz PC, et al. Characteristic complications after autologous chondrocyte implantation for cartilage defects of the knee joint. *Am J Sports Med.* 2008;36:2091-2099.
33. Peterson L, Minas T, Brittberg M, Nilsson A, Sjogren-Jansson E, Lindahl A. Two- to 9-year outcome after autologous chondrocyte transplantation of the knee. *Clin Orthop Relat Res.* 2000;374: 212-234.
34. Potter HG, Linklater LM, Allen AA, Hannafin JA, Haas SB. Magnetic resonance imaging of articular cartilage in the knee: an evaluation with use of fast-spin-echo imaging. *J Bone Joint Surg Am.* 1998; 80:1276-1284.
35. Safran MR, Kim H, Zaffagnini S. The use of scaffolds in the management of articular cartilage injury. *J Am Acad Orthop Surg.* 2008;16:306-311.
36. Saris DBF, Vanlauwe J, Victor J, et al. Characterized chondrocyte implantation results in better structural repair when treating symptomatic cartilage defects of the knee in a randomized controlled trial versus microfracture. *Am J Sports Med.* 2008;36:235-246.
37. Selmi TAS, Verdonk P, Chambat P, et al. Autologous chondrocyte implantation in a novel alginate-agarose hydrogel: outcome at two years. *J Bone Joint Surg Br.* 2008;90:597-604.
38. Steadman JR, Rodkey WG, Singleton SB, Briggs KK. Microfracture technique for full-thickness chondral defects: technique and clinical results. *Oper Tech Orthop.* 1997;7:300-304.
39. Tognana E, Borrione A, De Luca C, Pavesio A. Hyalograft C: hyaluronan-based scaffolds in tissue-engineered cartilage. *Cells Tissues Organs.* 2007;186:97-103.