Clinical Efficacy of Intra-articular Mesenchymal Stromal Cells for the Treatment of Knee Osteoarthritis

A Double-Blinded Prospective Randomized Controlled Clinical Trial

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Background: Currently, there are limited nonoperative treatment options available for knee osteoarthritis (OA). Cell-based therapies have emerged as promising treatments for knee OA. Autologous stromal vascular fraction (SVF) has been identified as an efficient medium for intra-articular administration of progenitor cells and mesenchymal stem cells derived from adipose tissue.

Hypothesis: Patients receiving intra-articular SVF would show significantly greater improvement than patients receiving placebo injections, and this improvement would be dose dependent.

Study Design: Randomized controlled trial; Level of evidence, 1.

Methods: This was a multisite prospective double-blinded randomized placebo-controlled clinical trial. Adult patients with symptomatic knee OA were eligible. Thirty-nine patients were randomized to high-dose SVF, low-dose SVF, or placebo (1:1:1). SVF was obtained via liposuction, processed to create the cellular implant, and injected during the same clinical visit. Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) scores and magnetic resonance images were obtained preoperatively and at 6 and 12 months after injection. The Wilcoxon rank sum nonparametric test was utilized to assess statistical significance, and the Hodges-Lehmann location shift was used to assess superiority.

Results: The median percentage change in WOMAC score at 6 months after injection for the high-dose, low-dose, and placebo groups was 83.9%, 51.5%, and 25.0%, respectively. The high- and low-dose groups had statistically significant changes in WOMAC scores when compared with the placebo group (high dose, $P = .04$; low dose, $P = .02$). The improvements were dose dependent. The median percentage change in WOMAC score from baseline to 1 year after injection for the high-dose, low-dose, and placebo groups was 89.5%, 68.2%, and 0%, respectively. The high- and low-dose groups displayed a greater percentage change at 12 months when compared with the placebo group (high dose, $P = .006$; low dose, $P = .009$). Magnetic resonance image review revealed no changes in cartilage thickness after treatment. No serious adverse events were reported.

Conclusion: Intra-articular SVF injections can significantly decrease knee OA symptoms and pain for at least 12 months. The efficacy and safety demonstrated in this placebo-controlled trial support its implementation as a treatment option for symptomatic knee OA.

Registration: NCT02726945 (ClinicalTrials.gov identifier)

Keywords: progenitor cells; stem cells; osteoarthritis; cartilage; knee; stromal vascular fraction; adipose

Knee osteoarthritis (OA) is highly prevalent among older adults in developed countries and is a significant cause of chronic pain and disability.[24] The definitive surgical treatment for knee OA is total knee arthroplasty, a major surgical procedure. Given the complications and intense rehabilitation process associated with total knee arthroplasty, physicians

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attempt to manage knee OA symptoms with multiple nonoperative modalities before surgery. These modalities include the use of anti-inflammatory medications, physical therapy, corticosteroid injections, and viscosupplementation. Without the ability to prevent cartilage loss, these treatment modalities only delay symptomatic progression to total knee arthroplasty.

Recently, cell-based therapies have emerged as possible disease-modifying treatments. Mesenchymal stem cells (MSCs) and adipose-derived stem cells (ASCs) have demonstrated chondrogenic potential.[5] However, the isolation of MSCs and ASCs may require multiple weeks and special laboratories for cell expansion.[17] A more efficient method for collection and administration of ASCs is the use of autologous stromal vascular fraction (SVF) cells. SVF consists of a heterogeneous concentration of nucleated stromal and vascular cells that are normally present in the stromal and vascular structures of adipose tissue, including stromal and vascular progenitor cells, as well as endothelial cells.[19] SVF does not contain adipocytes; it has a very low concentration of leukocytes and a very low presence of extracellular matrix. Adipose tissue is easily acquired through the use of a small liposuction harvest (requiring only local anesthetic), which can then be processed to isolate the SVF cells. Furthermore, SVF processing does not require cell expansion or culture.[4] SVF can be processed at the bedside.

Multiple studies have supported the use of intra-articular SVF injections for knee OA symptom management.[2,15,16,26] These studies demonstrated improvement in knee OA symptoms ranging from 1 month to 2 years after SVF injection, without an increased risk of adverse effects.[16,26] Unfortunately, the clinical interpretations of these studies are limited by small sample sizes and the lack of control group comparisons or evaluation of SVF in conjunction with other treatment modalities, such as platelet-rich plasma or arthroscopic debridement.

Previous randomized controlled trials have demonstrated increased efficacy of intra-articular injections of autologous bone marrow MSCs or ASCs as compared with hyaluronic acid and normal saline.[6,12,17] The primary aim of this study was to investigate the efficacy and safety of intra-articular autologous SVF injections at 6 months as compared with placebo injection. The secondary aims of this study were to determine if SVF injections continue to reduce knee pain at 1 year after treatment and to assess any effects of SVF injections on articular cartilage with magnetic resonance imaging (MRI) evaluations 6 months and 1 year after injection. We hypothesized that patients receiving intra-articular SVF will show significantly greater improvement in symptoms than patients receiving placebo injections and that this improvement will be dose dependent.

METHODS

Study Design and Participants

Before voluntary patient enrollment, this clinical trial was approved by the Institutional Review Board at each research site, as well as the US Food and Drug Administration (IDE 16347). This trial was listed on ClinicalTrials.gov (NCT02726945). The complete protocol will not be available for access. A synopsis is available on ClinicalTrials.gov. This study was sponsored and funded by the GID Group. This was a prospective double-blinded randomized placebo-controlled interventional safety and efficacy study conducted at multiple centers (3 sites). The dose-escalated study used a parallel-group design with 3 arms: high-dose treatment group (3.0×10^7 SVF cells), low-dose treatment group (1.5×10^7 SVF cells), and placebo control group (zero SVF cells).

English-speaking men and nonpregnant women between the ages of 40 and 75 years were screened. Patient eligibility was determined per the degree of clinical and radiographic disease. Eligibility criteria included (1) a Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) pain (A1) subscore >6 and ≤ 16 on a 20-point scale in 1 knee and a WOMAC pain score ≤ 6 for the contralateral knee; (2) grade 2 or 3 Kellgren-Lawrence OA on radiograph with no full-thickness lesion >1 cm in any dimension by MRI assessment; and (3) failure of a minimum of 2 nonoperative therapies (oral pain medications, physical therapy, corticosteroid injection, or viscosupplementation injection).

Exclusion criteria included the following: a body mass index ≥ 35, American Society of Anesthesiologists score ≥ 3, history of symptomatic OA (hips, spine, or ankle), rheumatologic disease, avascular necrosis, severe bone deformity, previous infection of the knee joint, pes anserine

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bursitis, pain attributed to diffuse edema, pain attributed to displaced meniscal tear or osteochondritis dissecans, neurogenic or vascular claudication, bleeding disorders, chemotherapy, radiation therapy to treatment leg or adipose harvest site, and tobacco use. Patients were also excluded if their target knee had an injection within 3 months before screening, surgery within 6 months before screening, or major injury within 12 months before enrollment. Those who could not discontinue use of the following drugs 7 days before injection have also excluded prescription pain medication, chronic oral steroids, anticoagulants, thrombolytics, or antiplatelet medication.

Patient screening and evaluations took place in private physician examination rooms. All procedures related to adipose collection, SVF processing, and SVF injection took place in private physician examination rooms. Patient-reported outcomes were collected during clinical visits.

Randomization, Blinding, and Dose Escalation

Patients were randomized 1:1:1. Randomized opaque folders containing the treatment dose assignment were sent to each site. On the day of intervention, a folder was randomly selected for the patient and delivered to the site technician. Only the site technician had access to the randomization information. Investigators and participants were blinded to treatment group assignment. Once the appropriate SVF or placebo dose was created by the technician, the dose syringe and tip were completely wrapped in sterile white labels to mask the contents. After the 6-month follow-up evaluation, patients and physicians were unblinded. Patients were unblinded at this time because the primary efficacy endpoint was the 6-month follow-up.

Under the dose escalation protocol, the first 15 consecutive participants were randomized to either the low-dose or the placebo group and followed for 6 weeks with a safety and adverse events analysis. The remaining 24 patients were assigned to the high-dose, low-dose, or placebo group. The criterion for trial continuation was $\leq$2 adverse events of grade $<$4 on the Common Terminology Criteria for Adverse Events scale. No adverse events were observed, and the study proceeded. Adverse events were monitored continuously during the study period.

Intervention

The complete adipose harvesting, processing, and injection procedure is described in the Appendix Methods (available in the online version of this article). Adipose was harvested from the abdomen with patients under local anesthetic. A mean 75 mL of adipose was aspirated directly into a sterile GID SVF-2 tissue-processing device (GID Group). The GID SVF-2 device is designed to produce a standardized and fully characterized dose of stromal vascular cells. The filled device was handed to a technician for tissue processing and cell characterization. Complete cell characterization and results are detailed in the Appendix Methods.

All tissue processing was done under sterile conditions within the single-use GID SVF-2 device. The appropriate dose (per treatment group) was created in a blinded 5-mL syringe, and the total volume was brought to 3 to 4 mL with lactated Ringer solution. The dose was then injected into the knee joint via a superior-lateral approach under sterile technique. Verification of joint space location of the needle was verified with ultrasound imaging or by aspiration of visible synovial fluid into the syringe. All participants were advised to maintain minimal weightbearing for 2 days. Full range of motion (nonweightbearing) was encouraged. Participants were advised to maintain only light activity and to avoid previously painful activities for the first 3 weeks after injection.

The cell dose was evaluated for viability, endotoxin level, and gram-negative contamination before release. A sample from each subject was sent to a central laboratory for evaluation of residual collagenase, cultured sterility, colony-forming unit analysis, phenotype analysis (flow cytometry), and cytokine/growth factor assessment, see Appendix Methods (available online).

Outcome Measures

The prespecified primary efficacy outcome was the percentage change from baseline per the short-form WOMAC scale, a patient-reported OA symptom questionnaire. The WOMAC instrument consists of 3 subscores used to evaluate pain, stiffness, and functionality. Total scores range from 0 to 56 points. A decreasing score is indicative of decreased pain and stiffness and increased functionality. The total score was normalized to 100 points. The WOMAC was completed by the patient before intervention and at 6 weeks and 3, 6, and 12 months after injection.

MRI of the treatment knee was obtained before treatment and at 6 months and 1 year after treatment. MRI scans were taken according to the following parameters: sagittal plane only, 2.5-mm proton density fat saturation sequence, and 3.0 or 1.5 T with knee coil magnet (8-16 channels). The MRI studies were reviewed for anatomic changes and for cartilage changes in the anteroposterior dimension for medial and lateral tibiofemoral lesions via the sagittal view. Cartilage degeneration was rated with the modified Outerbridge classification. Two fellowship-trained radiologists reviewed all images independently and then reached consensus agreement. Reviewers were blinded to the treatment arm. The resolution of the MRI measurement was 1 mm.

Statistical Analysis

The sample size determination was based on a difference to detect at least 17 points (representing a 33% change relative to baseline for a median baseline score of 50 points on the WOMAC scale [100 points, full scale]), a 1-sided superiority test, a standard deviation of 14 points, an α value of .05, and a power of 80%, resulting in 11 participants per group. To account for possible losses to follow-up, a loss rate of 20% was assumed, and an additional 2 participants per treatment group were added for a total

sample size of 13 per group and a total enrolled sample size of 39.

The hypothesis in the prespecified analysis was as follows: primary efficacy will be achieved if either dose group is shown to be superior to the placebo group at 6 months after treatment by using the percentage change from baseline as the primary variable and the following null and alternative hypotheses:

H_0: There is no difference between the dose group and the placebo group.

$$H_0: M_L = M_C \text{ and } M_H = M_C.$$

H_a: Either or both dose groups are superior to the placebo group.

$$H_a: M_L > M_C \text{ and/or } M_H > M_C.$$

C is control (placebo); L is low dose; H is high dose; and M is median.

Percentage change in total WOMAC score for each group was calculated and compared by between-group comparisons of each treatment group with the placebo per the Wilcoxon rank sum test (nonparametric). Hodges-Lehmann estimation (nonparametric) was used to construct 1-sided 95% CIs of the location shift (median of pairwise differences) to assess superiority. Concordance statistics (nonparametric) were used to calculate the area under the operating characteristic curve to evaluate the effect size. A Bonferroni-corrected α value of .025 (2.5%) was used for the primary efficacy evaluation to account for the multiple comparisons (2) of the null hypothesis.

The data set was analyzed with an intent-to-treat principle and used the last observation carried forward method for missing data. To assess clinical meaningfulness of treatment, a threshold of 33% change from baseline in total WOMAC score was set as the minimal clinically important difference (MCID). This indicated that a 33% improvement in the WOMAC score was needed for patients to experience a clinically meaningful change in knee OA symptoms. The MCID for the percentage change in WOMAC score was based on an analysis of current treatments for OA of the knee. Ten peer-reviewed randomized and concurrent-controlled studies involving >2500 participants for treatment of OA of the knee with corticosteroids, hyaluronic acid, total knee replacement, and controls (saline)[¶] were analyzed (Table 1).

The MCID of 33% was selected as the largest of the 3 injection approaches. The primary endpoint was prespecified at 6 months with a safety follow-up at 1 year. Data at 6 weeks and 3 months were used descriptively but not prespecified for statistical comparison. Between-group differences in cartilage thickness and within-group differences in Outerbridge classifications were analyzed per the Mann-Whitney *U* and Wilcoxon signed rank tests, respectively.

[¶]References 1, 3, 7, 10, 13, 14, 18, 20, 23, 25.

TABLE 1
Previously Published WOMAC Percentage Change for MCID Calculation[a]

Treatment	Percentage Change From Baseline at 6 mo	Participants, n
Total knee arthroplasty[7,10,13,14]	54	1451
Corticosteroids[3,18,20,23]	26	362
Hyaluronic acid[1,3,18,23]	33	433
Normal saline[1,20,25]	22	250

[a]MCID, minimal clinically important difference; WOMAC, Western Ontario and McMaster Universities Osteoarthritis Index.

RESULTS

Of the 329 patients screened, 67 consented for further MRI evaluation of their OA. After review, 1 patient unenrolled owing to relocation, and 27 were excluded for full-thickness lesions >1.5 cm in any dimension or displaced meniscal tears (Figure 1). A total of 39 patients (22 women and 17 men) were enrolled with 13 in each treatment group. Patients were enrolled between July 2016 and September 2017. The last patient completed 1-year follow-up in September 2018. Patient characteristics are summarized in Table 2.

Six-Month WOMAC Evaluation

Of the 39 patients enrolled, 37 completed the 6-month WOMAC evaluation. Missing data were completed with the aforementioned last observation carried forward method. Two missing 6-month values were imputed, resulting in an imputation rate of 2.6% (2 of 78). Patient attrition and data carried forward are described in Figure 1. One patient in the placebo group was disqualified after receiving the knee injection because of a protocol error in the initial MRI evaluation, which was identified immediately after treatment. The distribution of studentized residuals of the primary variable (percentage change) was evaluated for normality with the Shapiro-Wilk test for normality, showing a strongly nonnormal distribution ($W = 0.907$, $P = .004$). The nonnormal distribution was partially caused by the floor effect of the WOMAC and partially by the nature of the WOMAC, which is a Likert-type ordinal scale. Parametric analysis was not tenable, and nonparametric methods (distributed free about medians) were used to evaluate the hypotheses.

The rate of 6-month WOMAC follow-up for the high-dose, low-dose, and placebo groups was 92.3% (12 of 13), 100% (13 of 13), and 92.3% (12 of 13), respectively. Six months after SVF injection, all groups displayed a reduction in total WOMAC score from baseline. The median percentage change in WOMAC score for the high-dose, low-dose, and placebo groups was 83.9%, 51.5%, and 25.0%, respectively (Table 3, Figures 2 and 3). The median percentage change in the WOMAC score for the high- and low-dose groups was greater than the MCID, and that for

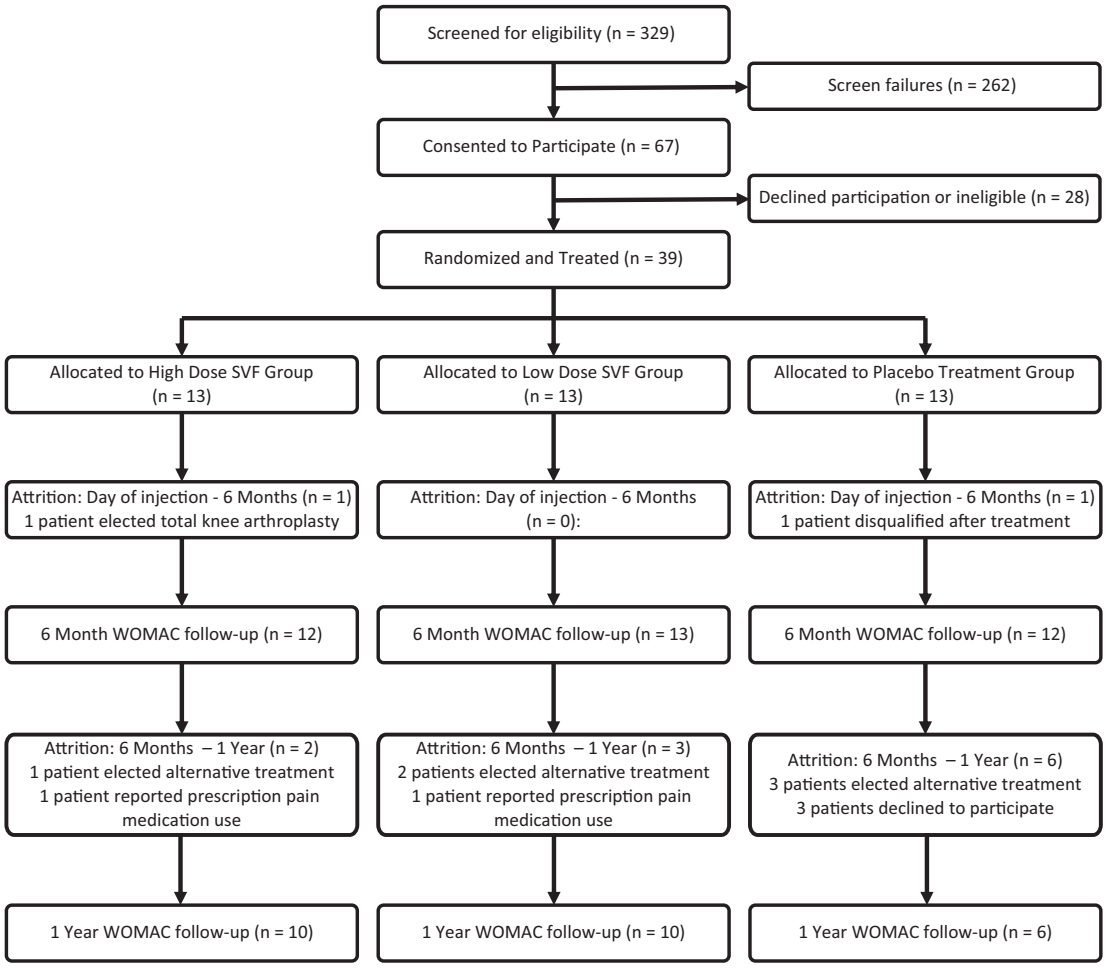

Figure 1. Study flow diagram depicts patient follow-up and attrition. SVF, stromal vascular fraction; WOMAC, Western Ontario and McMaster Universities Osteoarthritis Index.

TABLE 2
Patient Characteristics[a]

	Treatment Group			
Parameter	Placebo	Low	High	Total
Age, y	57.1 ± 9.1 (41-74)	60.5 ± 7.9 (48-71)	59.5 ± 11.7 (41-74)	59.0 ± 9.9 (41-74)
BMI	27.1 ± 2.7 (22.3-32.8)	27.6 ± 4.1 (19.6-34.9)	28.8 ± 4.3 (21.7-34.9)	27.8 ± 3.9 (19.6-34.9)
Race/ethnicity				
White	69.2 (9)	92.3 (12)	84.6 (11)	82.0 (32)
Black	7.7 (1)	0 (0)	0 (0)	2.6 (1)
Hispanic	23.1 (3)	7.7 (1)	15.4 (2)	15.4 (6)
Sex				
Female	53.8 (7)	69.2 (9)	46.2 (6)	56.4 (22)
Male	46.2 (6)	30.8 (4)	53.8 (7)	43.6 (17)
Kellgren-Lawrence grade				
2	30.8 (4)	30.8 (4)	30.8 (4)	30.8 (12)
3	69.2 (9)	69.2 (9)	69.2 (9)	69.2 (27)
Knee laterality				
Right	30.8 (4)	76.9 (10)	69.2 (9)	59.0 (23)
Left	69.2 (9)	23.1 (3)	30.8 (4)	41.0 (16)
ASA score				
I	61.5 (8)	69.2 (9)	46.2 (6)	59.0 (23)
II	38.5 (5)	30.8 (4)	53.8 (7)	41.0 (16)

[a]Values are presented as mean ± SD (range) or % (n). ASA, American Society of Anesthesiologists; BMI, body mass index.

TABLE 3
WOMAC Total Scores for Groups: 100-Point Full Scale[a]

Group: Time Point	Mean	Median (IQR)	Median Percentage Change	Minimum	Maximum
High dose					
Baseline	47.1	49.8 (35.6-55.2)	0	19.6	69.4
6 wk	25.7	27.0 (14.2-36.0)	37	0.0	55.2
3 mo	26.5	27.0 (10.7-34.7)	56	3.6	60.5
6 mo	20.0	8.9 (3.6-32.0)	84	0.0	53.4
1 y	13.2	3.6 (0.0-26.7)	89	0.0	53.4
Low dose					
Baseline	56.2	51.6 (46.3-62.3)	0	39.2	99.7
6 wk	24.8	20.0 (10.7-37.4)	50	0.0	64.1
3 mo	19.7	14.0 (5.3-35.6)	75	0.0	64.1
6 mo	23.7	26.7 (8.9–32.0)	52	0.0	60.5
1 y	21.8	12.5 (7.1-35.6)	68	0.0	60.5
Placebo					
Baseline	49.3	49.8 (37.4-57.0)	0	28.5	80.1
6 wk	26.0	23.0(14.2-37.4)	46	6.2	55.2
3 mo	22.9	20.0 (16.0-32.0)	62	0.0	55.2
6 mo	37.2	30.2 (21.4-55.2)	25	16.0	81.9
1 y	41.9	41.0 (19.5-55.2)	0	5.3	81.9
Treatment[b]					
Baseline	51.7	51.0 (41.4-58.7)	0	19.6	99.7
6 wk	25.2	24.0 (14.2-37.0)	45	0.0	64.1
3 mo	23.1	20.0 (7.1-35.4)	61	0.0	64.1
6 mo	21.8	22.0 (4.0-32.0)	62	0.0	60.5
1 y	17.5	8.0 (0.9-29.6)	85	0.0	60.5

[a]IQR, interquartile range; WOMAC, Western Ontario and McMaster Universities Osteoarthritis Index.
[b]Treatment group comprises both the high- and low-dose groups.

the placebo group was below the MCID. Sixty-two percent of patients in the treatment groups (high and low doses) had a response greater than the MCID, in contrast to only 38% participants in the placebo group. Three patients in the high-dose treatment group and 3 in the low-dose group experienced a 100% reduction in WOMAC score, while no patients in the placebo group did.

In the comparative analysis, the high- and low-dose groups displayed statistical significance as compared with the placebo group (high dose, $P = .043$; low dose, $P = .023$; below Bonferroni-corrected value for multiple comparisons). The lower-bound 95% 1-sided confidence intervals (CIs) of the location shift showed that the high dose and the low dose were superior to placebo (location shift >0): high dose, 0.339 (95% CI, 0.012-0.662); low dose, 0.314 (95% CI, 0.042-0.606). The effect sizes for the high and low doses were 0.701 and 0.734, respectively, indicating large effect sizes for both doses relative to placebo. Both dose groups showed statistical significance relative to placebo (with superiority based on CIs relative to placebo), had similar large effect sizes, and were combined in a treatment group (see Table 3).

One-Year WOMAC Evaluation

Of the initial 39 patients, 37 were available for follow-up 1 year after SVF injection; however, only 26 were able to complete the WOMAC. The 1-year WOMAC follow-up rate in the high-dose, low-dose, and placebo groups was 76.9% (10 of 13), 76.9% (10 of 13), and 46.2% (6 of 13), respectively. Missing data were completed with the last observation carried forward method. Reasons for patients' inability to complete the WOMAC at 1 year are displayed in Figure 1.

All groups continued to demonstrate lower total WOMAC scores 1 year after injection as compared with baseline scores. The percentage change from baseline for the high-dose, low-dose, and placebo groups was 89.5%, 68.2%, and 0%, respectively (Table 3). The high- and low-dose groups continued to display significantly greater percentage improvement in WOMAC scores as compared with the placebo group (high dose, $P = .006$; low dose, $P = .009$).

The lower-bound 95% 1-sided CIs of the location shift showed that the high dose and the low dose were superior to placebo (location shift >0): high dose, 0.524 (95% CI, 0.252-0.917); low dose, 0.435 (95% CI, 0.122-0.810). The effect sizes for the high and low doses were 0.793 and 0.775, respectively, indicating large effect sizes for both doses relative to placebo.

The analysis at 1 year showed continued improvement from 6 months to 1 year for both high- and low-dose groups and a return toward baseline for the placebo group (Figure 2). Both treatment groups maintained statistical significance and superiority relative to the placebo group and large effect sizes at 1 year.

MRI Review

Of the initial 39 patients, 37 completed MRI evaluation 6 months after SVF injection. There were no signs of new cyst

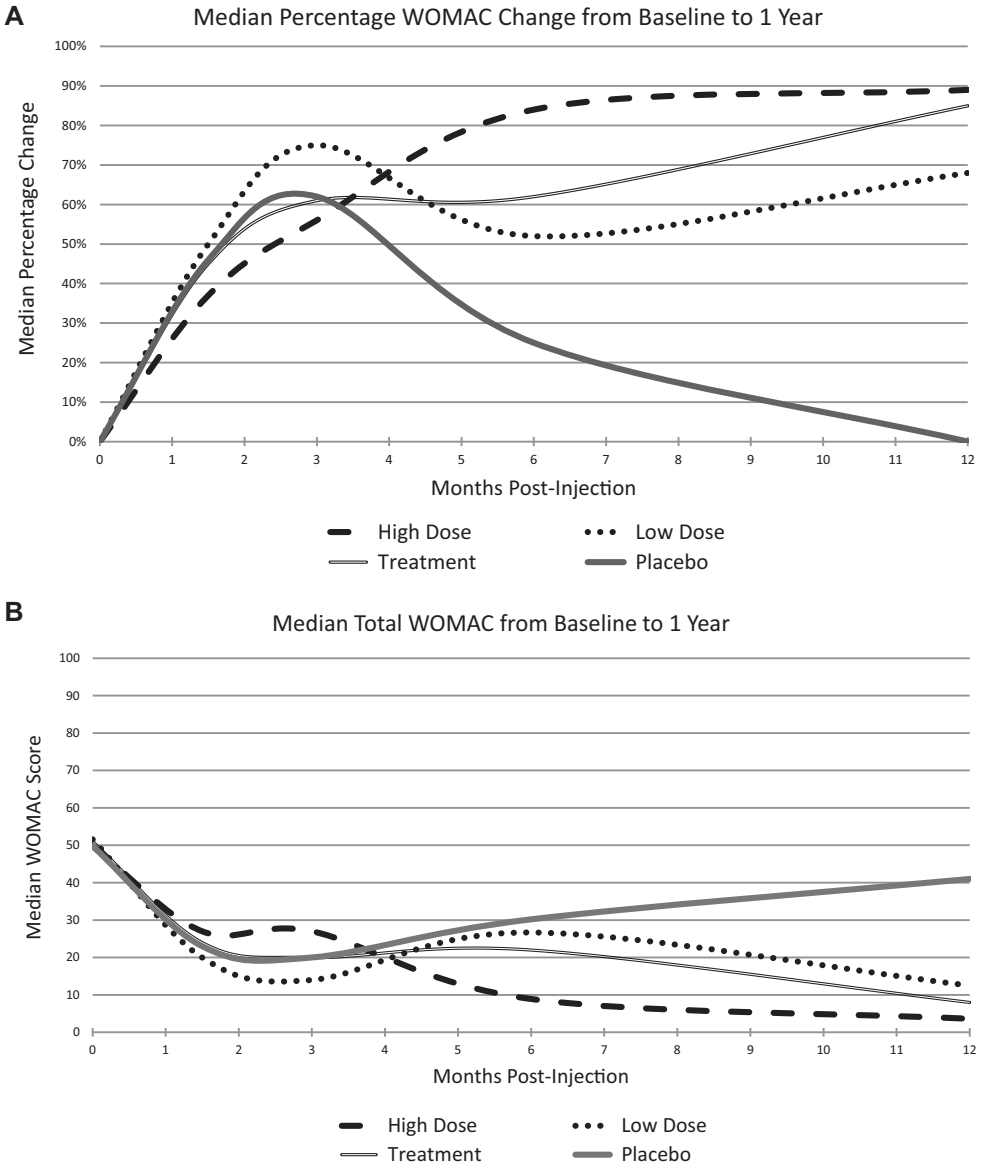

Figure 2. (A) Median overall percentage improvement in WOMAC scores over time. (B) Median total WOMAC scores over time. The high- and low-dose groups demonstrated an improvement in WOMAC scores 6 months and again at 1 year after injection. Treatment group represents the low- and high-dose groups combined. WOMAC, Western Ontario and McMaster Universities Osteoarthritis Index.

formation, heterotopic ossification, or neoplasms (benign or malignant) of the bone, cartilage, synovium, or vasculature. Sixty lesions in the 39 patients were evaluated for changes in cartilage thickness and changes in Outerbridge classification. Outerbridge classification ranged from 1 to 4, with patients having 1 to 4 lesions of various grades. At 6-month follow-up, the mean change in cartilage thickness for all participants was 0 mm (Table 4). The mean changes in cartilage thickness for the treatment group and the placebo group were –0.2 mm and 0.5 mm, respectively, with no statistical difference between groups ($U = 316$, $P = .89$). The median change in Outerbridge classification at 6 months was 0 for the treatment group and the placebo group, with no statistically significant difference between baseline and 6 months per within-group evaluation ($V = 30$, $P = .46$; $V = 0$, $P \geq .99$ [respectively]).

Of the initial 39 patients, 23 completed MRI evaluation at 1 year. Patient attrition is detailed in Table 5. There were no visibly quantifiable changes in knee cartilage thickness (Table 6). One MRI scan (high-dose group) was notable for showing new subchondral cystic changes, and another (placebo group) was notable for showing a new parameniscal cyst. All other MRI scans revealed no changes from baseline or any evidence of disease progression.

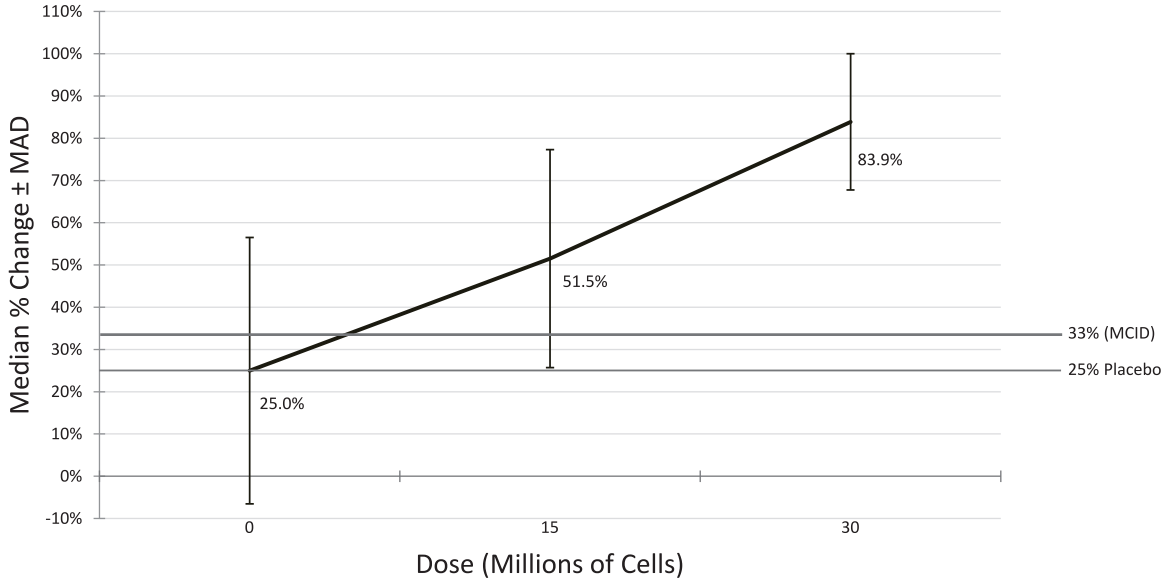

Figure 3. Dose-response curve at 6 months. Error bars represent median absolute deviation (MAD), a measure of variation around the median, representing the median of the values on each side of the group median. MCID, minimal clinically important difference.

TABLE 4
Changes in Cartilage from Baseline to 6 Months using MRI[a]

		Cartilage Loss		Outerbridge Classification	
Group	Lesions, n	Baseline Mean, mm	Mean Change at 6 mo, mm	Baseline Median (Range)	Median Change at 6 mo
All	60	12.6	0	3 (1-4)	0
Treatment group	46	11.5	−0.2	3 (1-4)	0
Placebo group	14	16.3	0.5	4 (1-4)	0
Responders, >MCID	38	13.2	0.2	3 (1-4)	0
Nonresponders, <MCID	22	11.6	−0.4	3 (1-4)	0

[a]MCID, minimal clinically important difference.

Adverse Events

During the initial 6 months, no serious adverse events were reported, and 3 adverse events were reported with none greater than grade 1 on the common terminology criteria for adverse events rating scale.[21] One patient from the high-dose group reported knee swelling, and aspirated fluid was sent for culture, with no growth. Two SVF sample cultures at the central laboratory for the study indicated possible bacteria growth, having only 1 colony in the culture plate. Those patients were evaluated, with no infection identified. None of these events were associated with infections. No adverse events of any type were reported during the 6-month to 1-year follow-up period.

DISCUSSION

Nonoperative management is the primary treatment for knee OA symptoms. While current nonoperative modalities can offer symptomatic relief, these treatment modalities often fail, ultimately leading to knee arthroplasty. There is a need for more effective nonoperative knee OA treatment modalities, especially ones that may arrest or even reverse disease progression. The results from our study demonstrate a clinically meaningful improvement in knee OA symptoms and pain 6 months and 1 year after intra-articular injection of a high dose (3.0×10^7 cells) or low dose (1.5×10^7 cells) of SVF cells. The percentage improvement in WOMAC scores for both SVF dose treatment groups was >33%, the predetermined MCID for this study, at 6 months and 1 year. The MCID of 33% represents the magnitude of improvement needed for patients to experience a clinically meaningful improvement in their symptoms; therefore, the superiority of their improvement was clinically meaningful. The improvements in WOMAC scores in the treatment groups were significantly greater than the improvement experienced by patients in the placebo treatment group at 6 months and 1 year. Furthermore, WOMAC scores continued to improve for the high- and low-dose SVF groups from 6 months to 1 year after treatment. In contrast, the WOMAC scores for the placebo group declined after 3

TABLE 5
Six-Month and 1-Year Magnetic Resonance Imaging Attrition

			Attrition Cause		
Treatment Group	Initially Enrolled	Completed Assessment	Patient Exited Study	Alternative Treatment[a]	Declined to Participate
6 mo					
High dose	13	12	1	0	0
Low dose	13	13	0	0	0
Placebo	13	12	0	0	1
1 y					
High dose	13	9	1	1	2
Low dose	13	10	0	2	1
Placebo	13	4	1	3	5

[a]Total knee replacement or intra-articular injection (corticosteroids, hyaluronic acid, or platelet-rich plasma).

TABLE 6
Changes in Cartilage from Baseline to 6 Months using MRI[a]

Group	Lesions, n	Mean at Baseline, mm	Mean Change, mm
All	38	10.4	0.0
Treatment group	33	9.9	−0.1
Placebo group	5	14.2	0.8
Responders, >MCID	27	10.6	0.1
Nonresponders, <MCID	11	10.2	−0.2

[a]MCID, minimal clinically important difference.

months and continued to decline toward baseline during the 6-month to 1-year period. This demonstrated the potential for SVF to provide symptomatic relief for a greater time frame than other OA treatments.

Our results are similar to previous studies assessing the efficacy of SVF injections. Fodor and Paulseth[8] and Garza et al[11] identified similar results in pilot studies assessing the safety and feasibility of intra-articular SVF injections in 6 patients (8 knees) and 6 patients (10 knees) with knee OA via the same method of SVF preparation, respectively. Similarly, Yokota et al[26] identified a significant 32% improvement in WOMAC scores and 40% improvement in pain visual analog scale scores 6 months after SVF injection in 13 patients.

In this investigation, the treatment (both doses) and placebo groups obtained the majority of improvement during the first 3 months; however, knee function in the treatment group continued to improve between 3 and 6 months and thereafter to 1 year. In contrast, knee function in the placebo group began to decline after the 3-month point, with continued decline toward baseline at 6 months and 1 year. At 1 year after injection, the treatment group showed a median improvement of 85%, the placebo group showed a median improvement of 0%. Similarly, previous studies have also identified a sustained improvement in knee pain and function 1 year after SVF injections.[8,12] In contrast, the efficacy of corticosteroid or hyaluronic acid injections 1 year after treatment has not been established.

Although patients receiving SVF injections had significantly better knee function, MRI review revealed no changes in modified Outerbridge classifications over time and no differences in the changes in chondral thickness between groups. However, it should be noted that the mean change in cartilage thickness (anteroposterior dimension) for all groups was less than the resolution of the MRI measurement. Our results contrast with those from a study performed by Hong et al,[12] which evaluated the efficacy of SVF injections for knee OA as compared with hyaluronic acid injections. MRI performed at 1-year follow-up demonstrated significantly better defect filling and cartilage repair in knees that received SVF as compared with those that received hyaluronic acid. However, these patients underwent arthroscopic debridement before treatment injection, and MRI scans were evaluated with WORMS (whole-organ magnetic resonance imaging score) and the MOCART score (magnetic resonance observation of cartilage repair tissue) for MRI analysis. This may account for the differences in results. Bansal et al[2] also performed MRI analysis 1 year after SVF injections. They observed an increase in cartilage thickness of at least 0.2 mm in 6 patients, no change in 2 patients, and a decrease in cartilage thickness of 0.2 mm in 2 patients. However, the mean change in cartilage thickness was not reported, and platelet-rich plasma injections were administered concomitantly with the SVF injections; therefore, direct comparisons are not possible.

The large effect sizes observed in this clinical trial are noteworthy, with a large area under the curve (>0.70) in both dose groups. This indicates that the statistical superiority of SVF as compared with placebo was large and likely had a clinically meaningful effect on patients' symptoms. The effect size observed in this study can also be compared with previous studies investigating the efficacy of bone marrow–derived MSCs. Emadedin et al[6] reported the effect size of bone marrow–derived MSCs as compared with saline placebo injections as medium to large at 6 months after injection, with a Hedges *g* of 0.7 for function measured on the WOMAC. Of note, the effect size of SVF injections identified in this study and that of bone marrow–derived MSC injections observed by Emadedin et al

are similar; however, the MSCs used by Emadedin et al were isolated and cultured in a separate laboratory, while SVF was obtained and injected at the same clinical visit.

The dose-response curve provides meaningful guidance with regard to a dose-response relationship. Both dose groups were shown to be safe with respect to adverse events and to have similar statistical comparisons relative to placebo. The dose-response curve and the superiority and effect size assessments show that the high dose provided additional therapeutic relief of OA pain over the low dose.

While multiple studies have reported outcomes after SVF injections, differences in methodology make our trial unique. To our knowledge, this is the first randomized blinded multisite trial to assess the efficacy of SVF injection as compared with intra-articular placebo injections. Given the known positive response to placebo injections within populations with knee OA, the inclusion of a placebo arm in this trial helped strengthen the conclusions.[22] Our study similarly showed symptomatic improvement with placebo injections, although significantly less than our treatment groups, beyond 3 months. Although only 39 patients were included in this trial, it represents one of the largest to assess the utility of SVF injections. Moreover, the SVF suspension was not combined with any other treatment modalities, allowing for specific evaluation of SVF therapy. We collected, processed, and injected SVF cells during 1 patient visit, simulating the ideal treatment scenario. The multisite design increased the generalizability of our results. Finally, the comparison of percentage reduction in WOMAC scores with the calculated MCID allowed for clear clinical interpretation of our results.

While informative, this trial does have limitations. The high percentage of Caucasians in this study may limit its generalizability. Furthermore, patients with a body mass index $\geq$35 and other comorbidities were excluded, thus limiting generalizability. The primary purpose of the MRI scans was to assess safety and not for statistical analysis of efficacy (pain/function) among groups. Patients were also unblinded after 6 months, potentially biasing the 1-year results. Finally, there was considerable attrition in the control group at 1 year, which may have biased the results; however, its 6-month WOMAC scores were imputed for the 1-year results. Four of the 6 patients lost to follow-up sought additional therapy for their knee pain in the 6-month to 1-year period and thus were lost to follow-up. The use of the 6-month imputed scores for the 12-month missing values is considered conservative given the additionally sought therapies. Further research is needed to assess the efficacy of SVF treatment in patients with other comorbidities. Long-term outcomes and delay or elimination of progression to total knee arthroplasty after SVF treatment should also be investigated. Finally, the cost and risks of any treatment should be weighed against the benefit. While this trial demonstrated that SVF injections are safe and efficacious, the cost cannot be accurately estimated at this time. If SVF injections become commercially available for the treatment of knee OA, a cost analysis should be performed for comparison with other available treatment options.

CONCLUSION

In conclusion, intra-articular SVF injections can significantly decrease knee OA symptoms and pain at 6 months and 1 year. Both low- and high-dose treatments had a large effect size, with the greatest change in the high-dose group. The efficacy and safety of SVF support its use as a treatment option for symptomatic OA of the knee. Longer-term results are needed to determine if there is any effect of SVF on disease progression.

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