International Orthopaedics (2021) 45:365–373
https://doi.org/10.1007/s00264-020-04571-4

ORIGINAL PAPER

Human bone marrow mesenchymal stem cell injection in subchondral lesions of knee osteoarthritis: a prospective randomized study versus contralateral arthroplasty at a mean fifteen year follow-up

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Received: 10 January 2020 / Accepted: 6 April 2020 / Published online: 23 April 2020

Abstract

Purpose Recently, mesenchymal stem cells (MSCs) have been proposed as potential treatment modalities for knee osteoarthritis. However, indications and long-term results have not been frequently reported. The purpose of this study was to determine whether bone marrow lesion on MRI are predictive of risk progression to total knee arthroplasty during the first ten years after subchondral cell therapy.

Methods This study included 140 adults aged 65 to 90 years. These 140 patients (mean age 75.4 ± 14.2 years) planned to undergo staged-bilateral total knee arthroplasty (TKA) for medial osteoarthritis, had "comparable" pain in both knees, and accepted randomization of the knees for surgery. They received TKA on one side and a subchondral injection of MSCs (from iliac bone marrow concentrate) on the contralateral knee during the same anaesthetic. The bone marrow graft of 20 cm^3 volume (10 cc in the tibia and 10 cc in the femur) contained average 7800 MSCs/mL (range 3120 to 11,560). The baseline volume of bone marrow lesions (BMLs) on the tibia and on the femoral condyle determined on MRI was average 3.4 cm^3 (range 0.4 to 6.4 cm^3). The risk of subsequent knee arthroplasty due to absence of bone marrow lesions regression as well as osteoarthritis (OA) grade was evaluated with Cox proportional-hazards ratio after control of baseline variables (number of cells injected, age, knee alignment).

Results After treatment with MSCs injection in bone marrow lesions of the subchondral bone, medial femorotibial compartment BML volume experienced regression over 24 months (mean regression 1.5 cm^3, range 0.8 to 3.2 cm^3). At the most recent follow up (average of 15 years, range 10 to 20 years), a total of 25 (18%) of the 140 patients underwent total knee arthroplasty performed at a mean of ten years (range, 5 to 15 years) after the date of the cell therapy. The overall incidence of knee arthroplasty after cell therapy was 1.19% per person-year which was equivalent to the risk of a revision for a primary TKA in the contralateral knees of the same patient population (21 revisions, corresponding to 1.00% revision per person-year; $p = 0.34$). After adjusting for confounders, persistent BMLs larger than 3 cm^3 after cell therapy was a strong independent risk factor for total knee arthroplasty (hazard ratio HR = 4.42 [95% CI = 2.34 to 7.21]; $p < 0.001$), regardless of OA grade, with higher risks demonstrated for larger BMLs. Incidence rates of arthroplasty were also higher for young patients and for knees presenting severe malalignment.

Conclusions This study showed that subchondral bone marrow concentrate (as compared with TKA) had a sufficient effect on pain to postpone or avoid the TKA in the contra lateral joint of patients with bilateral osteoarthritis. Bone marrow lesions were predictive factors for future knee arthroplasty in the knee with subchondral cell therapy at ten years follow-up.

Keywords Mesenchymal stem cells · Knee osteoarthritis · Bone marrow · Subchondral bone injection

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Many patients with bilateral osteoarthritis (OA) do not want simultaneous total knee arthroplasty (TKA); however, when they plan to undergo staged-bilateral TKA, they frequently elect to differ or to forego the second arthroplasty [1]; therefore, we proposed to them to receive, during the same anaesthetic, a subchondral injection of bone marrow mesenchymal

stem cells on the knee contralateral to the arthroplasty as a first line of treatment before an eventual second arthroplasty [2]. Our rational for subchondral bone injection of bone marrow concentrate (BMC) was that physiopathology [3] of osteoarthritis has revealed that functional fibrocartilage tissue is synthesized only when the subchondral bone is involved through bone marrow stimulating procedures such as drilling and microfractures [4]. Our hypothesis was that stimulation of subchondral bone marrow by direct injection of MSCs without drilling the cartilage would succeed in the production of fibrocartilage in the knee. Furthermore, MSCs stick quickly to bone [5] and this may avoid the phenomenon of washing by synovial fluid when MSCs are injected intra-articular. Pain related to OA is multifactorial; as cartilage has no innervation, the source of pain may have origin in the highly innervated subchondral bone. MSCs have a neuroprotection effect [6] by secretion of cytokines; therefore, a subchondral injection could be directly efficient on pain whether production of cartilage or not. The effects could be long-lasting if the transplanted cells become durable residents in the bone or induce chemotaxis and migration of other deficient autogenous cells in the subchondral bone as observed in other pathologies [7].

Although non-arthroplasty therapies are considered the first line of treatment in knee osteoarthritis, we have limited ability to determine which patients will not respond well to a conservative treatment as stem cell therapy [8] and are at high risk for an eventual subsequent total knee arthroplasty. There is some evidence that magnetic resonance imaging (MRI)-based assessment of OA [9] may help to determine the risk of failure of some conservative treatment. However, no study specifically assessed the risk of TKA after cell therapy in the long term.

The purpose of the current study was therefore to determine whether bone marrow lesions (BMLs) are predictive of future total knee arthroplasty in patients who received cell therapy for knee osteoarthritis. This study has a control group for evaluating the results of cell therapy since each patient has a contralateral TKA at the same surgery; this allowed to consider that the eligibility criteria for TKA were correct when the knee treated with stem cells required revision with total knee arthroplasty (the patient was aware of the result obtained by a TKA on the other knee).

Materials and methods

Patient selection and study design

After institutional review board approval, this study included 140 adults (demographics on Table 1) aged 65 to 90 years from 2000 to 2010. These 140 patients (mean age 75.4 ± 14.2 years) planned to undergo staged-bilateral TKA for medial osteoarthritis, had "comparable" pain in both knees (Table 1) according to the visual analog scale (VAS) pain score, and accepted randomization of the knees for surgery. We proposed to them to receive a subchondral injection of MSCs (from iliac bone marrow concentrate) on one of the knees during the same anaesthetic. Our hypothesis was that the subchondral injection of BMC should give at midterm a clinical improvement and could postpone or avoid TKA in some patients. The randomization method was conducted by a staff person who was blinded to the patients' data; patients were asked to choose two identical envelopes indicating for each side the MSC or TKA group. Subchondral bone MSC injection and the TKA were performed during the same anaesthetic on the same day.

Evaluation was scheduled at three months, six months, and one year and yearly thereafter. At the most recent follow up (end of 2019), 50 of 140 patients had died after a mean follow up of six years (range 3 to 15 years); death was not related to knee surgery; three of these 50 patients had another surgery on one knee. These three revisions were included in the analysis of data. At the most recent follow up, ten patients were unable or unwilling to return for full clinical evaluation and in these patients, local radiographic evaluation was arranged and obtained, and clinical examination performed locally by their medical doctor. The follow up after surgery for the 90 living patients (mean age 88.4; range 80 to 99 years) was an average of 15 years (range 10 to 20 years).

Surgical technique

Surgery was performed under general anaesthesia in three time-periods.

Mesenchymal stem cell aspiration from bone marrow was first performed: after installation, the patient for knee arthroplasty and before TKA incision, bone marrow was harvested on the contralateral anterior iliac crest of the TKA knee and prepared as previously reported [2].

Knee arthroplasty was performed secondly: For TKA, the same cemented implants were utilized in all patients and all were posterior-stabilized prostheses including patellar component. Protected weight-bearing with crutches was instituted during the two post-operative weeks for these patients who had surgery on both knees at the same time.

MSCs were injected in the subchondral bone of the contralateral knee at the end of the anaesthetic:

A tourniquet was not typically needed. Treatment was performed by percutaneous injection of 20 mL BMC in the subchondral medial femorotibial compartment of each knee, i.e., 10 mL in the medial tibial plateau and 10 mL in the medial condyle. We used the pre-operative radiographs and the MRI to determine the precise location of the subchondral lesion and to select the optimal trajectory of the trocar. For the femoral side, we used a retrograde

Table 1 Characteristics of the 140 patients and 280 knees

Characteristic	140 patients		
Gender (male/female)	53/87		
Age at inclusion (mean, range)	75.4 years (65–90)		
Weight (mean, St deviation)	81.0 ± 17.1 kg		
Body mass index, kg/m^2	28.1 (19.5–32.4)		
Follow-up (mean, range)	15 years (10–20)		
Age at follow-up	88.4 years (80–99)		
Knees	Knees		
Pre-operative scores	With cell therapy	With TKA	*P* value
(VAS) pain score	3.5 (2 to 5)	3.4 (2 to 6)	0.48
Knee society scores	57 ± 12	54 ± 8	0.24
Range of motion	115° ± 18	120° ± 21	0.26
H-K-A angle	174° (178°-165°)	175°(179°-167°)	0.41
Kellgren and Lawrence grading			
Grade 2 on both sides	32	32	
Grade 2 and Grade 3	4	4	
Grade 3 and Grade 2	6	6	0.53
Grade 3 on both sides	62	62	
Grade 4 on both sides	36	36	

"outside-in" femoral condyle bone marrow implantation. When advancing the trocar, an effort was made to avoid penetration of the cartilage. Sometimes computerized navigation was used as proposed for condyle osteonecrosis [10]. For the tibia, the trocar was introduced percutaneously parallel to the cartilage of the tibial plateau under image intensification fluoroscopy and directed to the center of the lesion in the subchondral bone, avoiding perforation of the cartilage by remaining 0.5 cm below the tidemark. The number of MSCs in the bone marrow graft of 20 cm^3 volume (10 cc in the tibia and 10 cc in the femur) was determined as previously reported [11] and contained average 7800 MSCs/mL (range 3120 to 11,560).

Clinical outcome

Patients were discharged with instructions for partial weight-bearing using crutches for the first post-operative week, and then total weight-bearing without crutches. Physical therapy was not necessary for the knee with cell therapy. All inflammatory or analgesic drugs were stopped at the entry to the study, three to four weeks before the injection of MSCs. Glucosamine was allowed, when the patient was using it before the study. During surgery, no joint fluid aspiration was performed and no steroid was injected. Analgesics were given in relation with TKA; anti-inflammatory drugs were not given after the procedure. Patients were asked to avoid any intra-articular injection in both knees.

At the most recent follow up (average of 15 years, range 10 to 20 years), the clinical results were graded according to the Knee Society knee score and the Knee Society function score [12]. During follow up, patients were evaluated with radiographs as well as functional outcomes. In radiographic evaluation, standing anteroposterior and lateral views of the knee as well as skyline and long-standing AP views were obtained. Details of any revision surgery were also recorded from hospital charts and clinic records. Patients were asked to compare their knees.

Imaging assessments

Radiographs

Baseline bilateral knee radiographs were standard fixed-flexion posteroanterior (PA) views and lateral views were obtained. For the purpose of this study, severe radiographic OA [13] was defined as "Kellgren and Lawrence" (KL) grade 4; moderate OA as KL grade 3; mild OA, as KL grade 2 (Table 1). Inclusion needed that the two knees of the same patient had no more difference than 1 grade with Kellgren-Lawrence grading (Table 1) and absence of prior surgery in the two knees. Data on coronal knee alignment were obtained from full-limb radiographs made according to a standard protocol and measured with the hip-knee-ankle (HKA) angle. The average HKA angle in knees treated with stem cells was

average before surgery 174 degrees (range 178 degrees to 165 degrees).

MRI bone marrow lesions

Patient-specific guides (PSGs) were developed [14] to improve implant positioning in TKA. Manufacture PSGs requires a pre-operative MRI. Since these patients had planned bilateral TKA, they had bilateral knee MRI for this reason. Each subject had an MRI performed at baseline before cell therapy and two years later. Knees were imaged in the sagittal plane on the same 2.5-T whole-body magnetic resonance unit (Siemens) by using a commercial extremity coil.

Osteoarthritis BMLs were defined [15] as ill-defined areas of decreased signal intensity (Fig. 1) on T2 MRI sequences. To eliminate false-positive, the distance of a lesion to the articular surface should be less than 10 mm and a BML should be present on two contiguous MR images. Bone marrow lesion (BML) volume were measured with a semi-automated segmentation method that detects, extracts, and quantifies the structure of BMLs based on the coronal and sagittal MRI sequences, using a graphical user interface (Photoshop) to manually identify the crude boundaries of the tibia and femur in each slice of the MR imaging dataset by marking multiple points along the articular surface. The reproducibility for measurement of the BMLs was assessed by using 30 randomly selected MRIs (Kappa value 0.86; $p = 0.001$).

The cartilage volume was measured by using a computer program. The change in the volume of the knee's cartilage was calculated by subtracting the volume at follow-up from the baseline volume. Bone marrow lesions and knee cartilage volume was determined by means of image processing on an independent work station by using the software program Osiris.

Statistical analysis

Analyses were performed with a standard software statistical package. Differences in variables by surgery status (arthroplasty compared with stem cell therapy) were assessed for categorical variables by chi-square tests and for continuous variables by Student *t* tests or by Wilcoxon rank-sum tests. Differences in baseline symptom severity by cartilage defect status and OA grade were assessed by Wilcoxon rank-sum tests. Kaplan-Meier survival curves were generated to assess the yearly incidence of total knee arthroplasty after cell therapy. The crude (unadjusted) associations between baseline variables and the risk of total knee arthroplasty were determined by univariate Cox proportional-hazards models. Finally, to determine the independent (adjusted) association of the presence or absence of bone marrow lesions with the risk of total knee arthroplasty, a multivariate Cox proportional-hazards model was created. All risks factors as demographic, clinical, and radiographic factors presented in this study were included and particularly age, sex, weight, knee alignment, and osteoarthritis severity.

Results

Subchondral bone marrow MSC injection postponed TKA

At the most recent follow-up (mean 15 years; range 10 to 20 years) with inclusion of revisions among patients who had died, 21 (among 140) TKA knees had needed subsequent revision (12 loosening, two mobilizations with general anesthesia, two periprosthetic fractures, 5 patella fractures) versus 25 knees among the 140 knees with cell therapy (25 conversions to TKA).

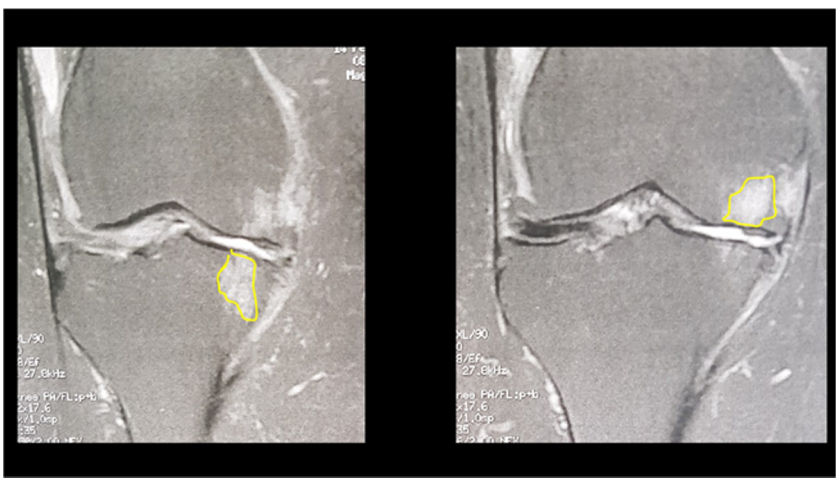

Fig. 1 Bone marrow lesion (BML) in the medial tibia and in the medial condyle. The yellow lines identify areas of high signal intensity and surround them. The following sequence and parameters were used: a T1-weighted fat-suppressed 3D gradient recall acquisition in the steady state for bone marrow lesion and T2-weighted

Clinical pre-operative knee scores of cell therapy knees before treatment (57 points ± 12) were similar ($p = 0.08$) to knee's scores of the TKA group before treatment (knee: 54 points ± 8; function: 55 points ± 13). The mean overall changes in knee scores at three months follow up were similar ($p = 0.24$) for the cell therapy group (81.3 points ± 12) when compared with the TKA group (79 points ± 21). At the time of the most recent follow-up, the Knee Score remained similar in both groups (respectively 80.3 points ± 11 versus 78.3 ± 23). More than half of the population (74 patients) has preference for the knee treated with MSCs despite no improvement was observed for range of motion. This preference was related to a better pain improvement (Fig. 5).

Radiological and imaging outcome after cell therapy

Cartilage volume changes over time

Cartilage volume changes over time were measured on knees with cell therapy; the percentage cartilage volume measured with MRI (excluding osteophytes) increased compared to baseline (2.3 ± 1.1% at 2 years). This cartilage production was however fibrocartilage as demonstrated during revision in knees with conversion to TKA.

Subchondral bone marrow lesion (BML) changes: The average baseline BML volume of the medial femorotibial compartments (both femur and tibia) was 3.4 cm^3 (range 0.4 to 6.9 cm^3). Larger total baseline BML volumes were associated with greater knee pain ($p = 0.01\%$). There was no correlation between bone marrow lesions and KL grade ($p = 0.21$). After treatment with MSCs injection in the subchondral bone, medial femorotibial compartment BMLs volume experienced regression over 24 months (mean 2.1 cm^3, range 1.2 to 5.7 cm^3) in all the knees (Fig. 2). Therefore, the residual volume of the BMLs was average 1.3 cm^3, range 0 to 5 cm^3. We analyzed the effect of the number of progenitor cells on the decrease of bone marrow lesions and found higher decrease of BMLs associated with higher number of MSCs ($p = 0.04$). Smaller lesion had better regression as compared to larger lesions (for the same number of progenitor cells injected in the subchondral bone).

According to the KL classification, the pre-operative (Table 1) grading was in the cell therapy group: 36 "Grade-2" knees, 68 "Grade-3", and 36 "Grade-4" knees. At the most recent follow-up, radiographic changes had progressed to 28 Grade-2 knees, 66 Grade-3, and 46 Grade-4 knees.

BML as predictors for another surgery (TKA) after cell therapy

We analyzed the effect of the baseline bone marrow lesions (BMLs) on the risk of a subsequent TKA after cell therapy. There was a trend for TKA for larger BMLs at baseline, but this trend was not significant ($p = 0.07$). Since regression was observed, we extended our observation by examining on the two year MRI the effect of residual persistent BMLs on risk for another surgery (TKA) over a mean of ten years (range, 5 to 15 years). Increasing severity of residual subchondral bone abnormality two years after cell therapy was staged as follows: stage 1, no bone marrow lesions; stage 2, BMLs less than 2 cm^3; stage 3, BMLs between 2 and 4 cm^3; stage 4, BMLs larger than 4 cm.

Descriptive statistics and incidence of total knee arthroplasty after cell therapy

A total of 25 (18%) of the 140 patients underwent total knee arthroplasty at a mean of ten years (range, 5 to 15 years) after

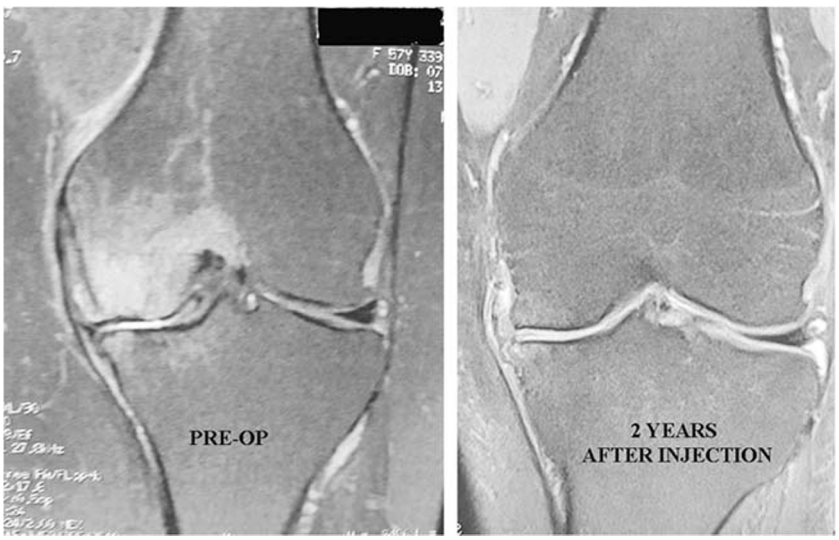

Fig. 2 Bone marrow regression of 5.7 cm^3 from a 6.9- to a 1.2-cm^3 lesion

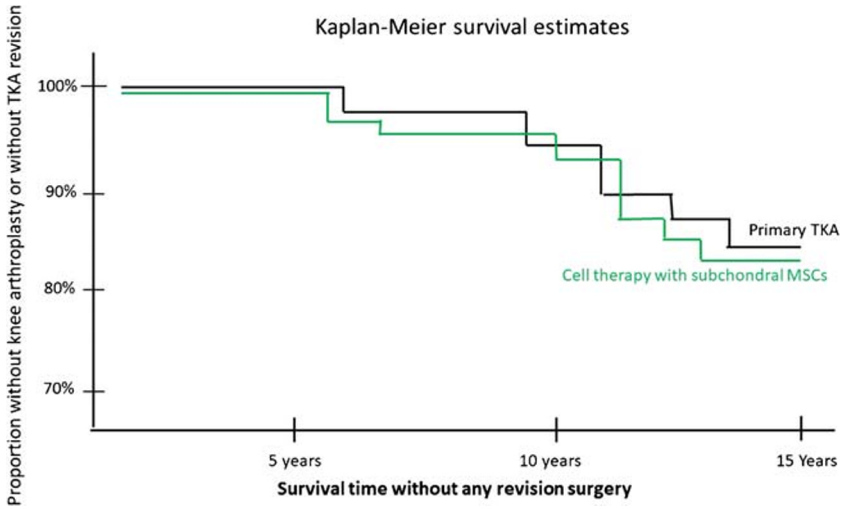

Fig. 3 Arthroplasty-free survival knees (green line) with cell therapy (arthroplasty incidence of 1.19% per person-year) compared with revision TKA survival knees (dark line) in the contralateral knee (revision incidence of 1.00% per person-year) among the same patients

the date of the cell therapy. The overall incidence of knee arthroplasty was 1.19% per person-year which is equivalent to the risk of a revision of primary TKA (Fig. 3) in the same patients (21 revisions, corresponding to 1.00% revision per person-year; $p = 0.34$). Among the 63 knees with persistent large BMLs (stage 3 or 4) after cell therapy, a total of 21 knees underwent TKA, and the yearly arthroplasty incidence was 2.22% for these patients; for the 77 patients without persistent large BMLs (stage 1 or 2), the yearly arthroplasty incidence was only 0.34% ($p = 0.01$). The results of Kaplan-Meier survival analyses are shown in Figs. 4 and 5.

Incidence rates in the group with persistent larger BMLs were higher ($p = 0.04$) among the 87 patients younger than 80 years (20 subsequent TKA; 18%) at the time of surgery as compared with the 53 older than 80 years (5 subsequent TKA; 9%). Incidence rates were higher (8 among 35; 21%) for the 35 knees with severe malalignment (HKA angle < 170 degrees).

Independent risk factors for future total knee arthroplasty

After adjusting for confounders, the presence of persistent BMLs larger than 3 cm^3 after cell therapy was a strong independent risk factor for total knee arthroplasty (hazard ratio HR = 4.42 [95% CI = 2.34 to 7.21]; $p < 0.001$), regardless of OA grade, with higher risks demonstrated for larger BMLs. The risk of total knee arthroplasty increased with BML size (HR = 2.38 [95% CI = 1.67 to 5.42]; $p < 0.001$ for lesion size between 3 cm^3 and 4 cm^3; and HR = 6.34[95% CI = 3.84 to 9.82]; $p < 0.001$ for lesion size > 4 cm^3). The BML location did not significantly add to the predictive ability of the adjusted model (likelihood ratio chi-square test, $p = 0.14$).

Increased age over 80 years at the time of cell therapy was independent factor to decrease the risk of future total knee arthroplasty (per year, HR = 0.85 [95% CI = 0;78 to 1.05]; $p = 0.001$) during the first ten years follow-up; male participants were less likely to undergo total knee arthroplasty, but

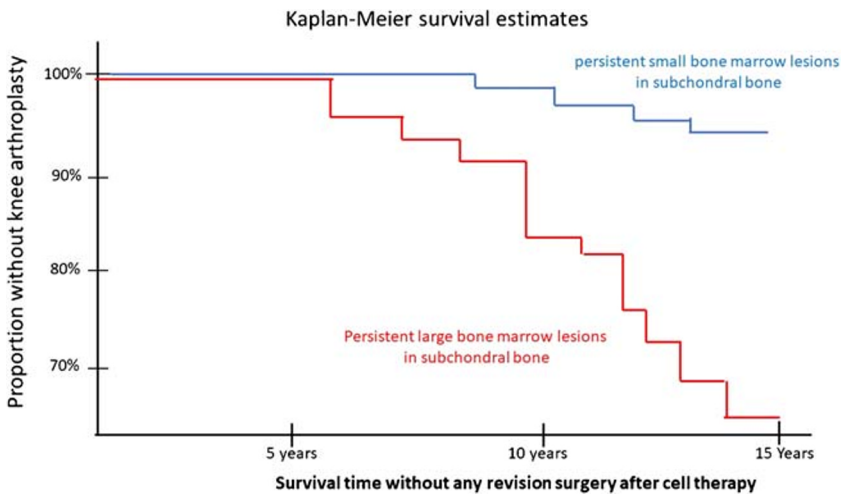

Fig. 4 Arthroplasty-free survival knees (red line) with cell therapy among patients with persistent stage 3 or 4 large BMLs (arthroplasty incidence of 2.22% per person-year) after cell therapy, compared with arthroplasty-free survival knees (blue line) with cell therapy among patients with persistent small stage 1 or 2 BMLs (arthroplasty incidence of 0.34% per person-year)

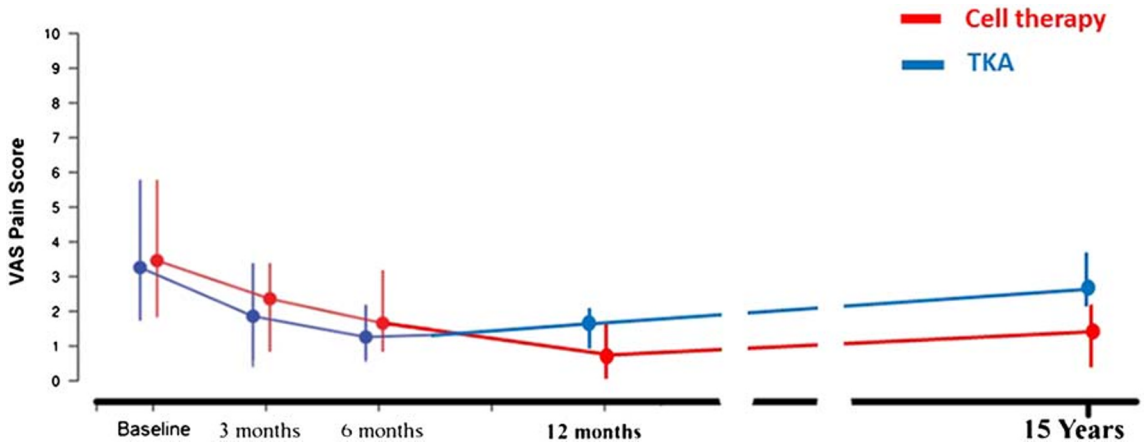

Fig. 5 Visual analog scale (VAS) pain score for cell therapy knee and TKA knee groups at each time point

this factor is not significant ($p = 0.24$). Weight also was not significant ($p = 0.36$).

OA grade did not increase the risk of total knee arthroplasty when severe grades were compared with moderate grade (HR = 0.67 [95% CI = 0.32 to 1.48]; $p = 0.48$). The effect of KL grade-3 OA was significantly dependent on BMLs status: among the patients without large BMLs, KL grade-3 OA did not increase the risk of total knee arthroplasty compared with grade-1 to 2 OA (HR = 0.61[95% CI = 0.22 to 1.37]; $p = 0.46$); among patients with large BMLs, KL grade-3 OA increased total knee arthroplasty risk (HR = 5.4[95% CI = 2.16 to 8.36]; $p = 0.01$).

The effect of malalignment also was dependent of the BMLs status (likelihood ratio chi-square test, $p = 0.04$ for interaction): among the patients without large BMLs, malalignment (HKA < 170 degrees) did not increase the risk of total knee arthroplasty ($p = 0.25$); among patients with large BMLs, HKA < 170 degrees baseline malalignment increased total knee arthroplasty risk ($p = 0.03$).

Discussion

Knee pain, frequently due to OA, is subjective symptom but a major source of disability in older adults. Socioeconomic factors and patient expectations also play an important role in the demand for treatment whatever this treatment: total joint arthroplasty or cell therapy. To our knowledge, this is the first comparison between cell therapy and TKA in knee osteoarthritis. When proposed to patients, the idea was only to postpone the second arthroplasty and not to avoid it. Indeed, most of the patients elected to forego the second arthroplasty in the knee that had received bone marrow injection. However, if none of the patients were dissatisfied during the first five years follow-up, some pain recurrence was observed in 25 patients after five years with TKA as consequence.

We have a limited understanding of the biologic factors that predict risk of failure of a cell therapy with as consequence a future total knee arthroplasty. The current study highlights the greater importance of bone marrow lesion loss over the radiographic OA grade as a determinant of OA severity, specifically regarding the risk of future knee arthroplasty after cell therapy. In adults with joint-space narrowing on weight-bearing radiographs, it is well known that radiographic severity is not a reliable predictor of symptom severity in patients with tibiofemoral osteoarthritis [16]; the MRI-based findings demonstrated that radiographic joint-space narrowing provides an incomplete picture of OA severity, as patients with persistent important subchondral bone marrow lesions despite cell therapy had greater likelihood of progressing to total knee arthroplasty than were patients with the same KL grade of OA with limited subchondral bone marrow lesions after cell therapy treatment. As the reduction of bone marrow lesions with cell therapy appears predictable in volume (but limited; the maximum volume of regression was 3.7 cm^3), this may help to the discussion with the patient for the choice of treatment when the volume of BMLs can be evaluated pre-operatively. The clinical indications for obtaining MRI for middle-aged patients with knee OA symptoms are narrow, but we recommend obtaining MRI as part of the initial evaluation for knee OA when the treatment proposed is cell therapy. In the patient with moderate radiographic OA who does not respond to cell therapy, demonstration of absence of regression of BMLs on MRI may allow earlier consideration for another treatment (arthroplasty, or osteotomy). This approach could avoid a long period of poor symptom control until there is sufficient radiographic OA progression to meet current appropriate use criteria for surgery.

Other clinical and demographic factors were evaluated as determinants of future total knee arthroplasty after subchondral cell therapy. Older (> 80 years) patients were less likely to undergo total knee arthroplasty than were younger patients. Important malalignment has been shown to be

associated with increased knee symptoms in patients with medial compartment tibiofemoral OA; we found an independent association between malalignment and future arthroplasty risk in the multivariate analysis, particularly when the HKA angle was < 170 degrees. Persistence of large bone marrow lesions two years after cell therapy were consistently associated with higher total knee arthroplasty risk, even after adjusting for malalignment. We propose two possible mechanisms for the increased total knee arthroplasty risk associated with severe bone marrow lesions after cell therapy: (1) patients with persistent large bone marrow lesions have responded poorly to cell therapy for the degenerative knee due to an insufficient number of cells or due to an insufficient function of cells, (2) and having a large subchondral BML is associated in some knees with more severe structural changes less reversible as compared with knees having small BMLs. As older patients despite decreased number and function of MSCs had lower risks of TKA, we suspect that large BMLs in younger patients correspond to more severe structural (less reversible) changes as compared to the same lesion size in older patients as probably also when severe malalignment.

Several limitations to our study exist as the moderate number of patients in the current study; contrary to many other studies, we had no control with a placebo (saline injection in the contralateral subchondral bone); nevertheless, placebo treatment is not a recommended treatment of knee OA and was not accepted in old patients as comparison by our Institutional Review Board. Therefore, we compared the results of our subchondral cell therapy to a recommended and accepted treatment (arthroplasty) for knee OA; however, the improvement of walking obtained by only one arthroplasty on one side might have decrease pain in the contralateral as previously reported in some other reports [17, 18]; it could be therefore postulated that the MSC treatment did not really improve pain, but that decrease of pain was related to easier manoeuvrability of the "stem cell knee" obtained after TKA in the contralateral; however, more than half of the population had preference for the cell therapy knee, and decrease of BMLs was observed which is difficult to relate to the contralateral arthroplasty.

Whatever the reason of improvement of pain, our findings suggest that clinicians do not need to routinely monitor the contralateral knee aggressively with simultaneous TKA [1]. Results of this study showed that simple subchondral MSCs injections had favourable clinical outcome in terms of pain and function in knee osteoarthritis with some limited production of fibrocartilage. Most of the publications about intra-articular MSCs [19] have demonstrated evidence in pain relief and functional improvement in patients with knee osteoarthritis, but these publications had a short follow-up. This study with a long-term follow-up confirms the benefit of MSCs treatment with subchondral injection. While intra-articular MSCs may result in different clinical outcomes (with or without recommended higher concentration), this study with concentrate bone marrow MSCs is rather a low concentration treatment. The good effects observed with subchondral injection is probably related to the fact that transplanted cells become durable residents [5] in bone. Optimal cell concentration is difficult to demonstrate due to heterogeneity of cell concentrations in this study. In conclusion, this study showed that subchondral bone marrow concentrate (as compared with TKA) had a sufficient effect on pain to postpone or avoid the TKA in the contra lateral joint of patients with bilateral osteoarthritis. Severe bone marrow lesions were predictive factors for future knee arthroplasty in the knee with subchondral cell therapy.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval All procedures were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed consent Informed consent was obtained from all individual participants included in the study.

References

1. Meehan JP, Danielsen B, Tancredi DJ, Kim S, Jamali AA, White RH (2011) A population-based comparison of the incidence of adverse outcomes after simultaneous-bilateral and staged-bilateral total knee arthroplasty. J Bone Joint Surg Am 93(23):2203–2213
2. Hernigou P, Auregan JC, Dubory A, Flouzat-Lachaniette CH, Chevallier N, Rouard H (2018) Subchondral stem cell therapy versus contralateral total knee arthroplasty for osteoarthritis following secondary osteonecrosis of the knee. Int Orthop 42(11):2563–2571
3. Shapiro F, Koide S, Glimcher MJ (1993) Cell origin and differentiation in the repair of full-thickness defects of articular cartilage. J Bone Joint Surg Am 75(4):532–553
4. Orth P, Goebel L, Wolfram U, Ong MF, Gräber S, Kohn D, Cucchiarini M, Ignatius A, Pape D, Madry H (2012) Effect of subchondral drilling on the microarchitecture of subchondral bone: analysis in a large animal model at 6 months. Am J Sports Med 40(4):828–836
5. Mebarki M, Coquelin L, Layrolle P, Battaglia S, Tossou M, Hernigou P, Rouard H, Chevallier N (2017) Enhanced human bone marrow mesenchymal stromal cell adhesion on scaffolds promotes cell survival and bone formation. Acta Biomater 59:94–107
6. Lyden J, Grant S, Ma T (2019) Altered metabolism for neuroprotection provided by mesenchymal stem cells. Brain Circ 5(3):140–144
7. Hernigou P, Dubory A, Homma Y, Guissou I, Flouzat Lachaniette CH, Chevallier N, Rouard H (2018) Cell therapy versus simultaneous contralateral decompression in symptomatic corticosteroid osteonecrosis: a thirty year follow-up prospective randomized study of one hundred and twenty five adult patients. Int Orthop 42(7):1639–1649

8. Di Matteo B, Vandenbulcke F, Vitale ND, Iacono F, Ashmore K, Marcacci M, Kon E (2019) Minimally manipulated Mesenchymal stem cells for the treatment of knee osteoarthritis: a systematic review of clinical evidence. Stem Cells Int 2019:1735242
9. Driban JB, Price L, Lo GH, Pang J, Hunter DJ, Miller E, Ward RJ, Eaton CB, Lynch JA, McAlindon TE (2013) Evaluation of bone marrow lesion volume as a knee osteoarthritis biomarker longitudinal relationship with pain and structural changes: data from the osteoarthritis initiative. Arthritis Res Ther 15(5):R112
10. Hernigou P, Gerber D, Auregan JC (2020) Knee osteonecrosis: cell therapy with computer-assisted navigation. Surg Technol Int 36: sti36/1151
11. Hernigou P, Homma Y, Flouzat Lachaniette CH, Poignard A, Allain J, Chevallier N, Rouard H (2013) Benefits of small volume and small syringe for bone marrow aspirations of mesenchymal stem cells. Int Orthop 37(11):2279–2287
12. Scuderi GR, Sikorskii A, Bourne RB, Lonner JH, Benjamin JB, Noble PC (2016) The knee society short form reduces respondent burden in the assessment of patient-reported outcomes. Clin Orthop Relat Res 474(1):134–142. https://doi.org/10.1007/s11999-015-4370-2
13. Kellgren J, Lawrence J (1957) Radiological assessment of osteo-arthrosis. Ann Rheum Dis 16(4):494
14. Schotanus MG, Sollie R, van Haaren EH, Hendrickx RP, Jansen EJ, Kort NP (2016) A radiological analysis of the difference between MRI- and CT-based patient-specific matched guides for total knee arthroplasty from the same manufacturer: a randomised controlled trial. Bone Joint J 98-B(6):786–792. https://doi.org/10.1302/0301-620X.98B6.36633
15. Kon E, Ronga M, Filardo G, Farr J, Madry H, Milano G, Andriolo L, Shabshin N (2016) Bone marrow lesions and subchondral bone pathology of the knee. Knee Surg Sports Traumatol Arthrosc 24(6): 1797–1814. https://doi.org/10.1007/s00167-016-4113-2
16. Bedson J, Croft PR (2008) The discordance between clinical and radiographic knee osteoarthritis: a systematic search and summary of the literature. BMC Musculoskelet Disord 9:116
17. McMahon M, Block JA (2003) The risk of contralateral total knee arthroplasty after knee replacement for osteoarthritis. J Rheumatol 30(8):1822–1824
18. Smith HK, Wylde V, Lingard EA, Blom A, Metcalfe C, Ben-Shlomo Y (2013) The effect of pain after total knee arthroplasty on the contralateral, nonreplaced knee. J Bone Joint Surg Am 95(4): 315–322
19. Kim SH, Ha CW, Park YB, Nam E, Lee JE, Lee HJ (2019) Intraarticular injection of mesenchymal stem cells for clinical outcomes and cartilage repair in osteoarthritis of the knee: a meta-analysis of randomized controlled trials. Arch Orthop Trauma Surg 139(7):971–980