Autologous Protein Solution Injections for the Treatment of Knee Osteoarthritis

3-Year Results

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Background: Blood derivative injections have been recently proposed to address osteoarthritis (OA) with overall positive results, although long-term data on their efficacy are lacking. A novel blood derivative has been developed to concentrate growth factors and antagonists of inflammatory cytokines and shown promising early findings.

Purpose: To investigate if the positive effects of a single intra-articular injection of autologous protein solution (APS) in patients affected by knee OA—previously documented at 1 year in a multicenter double-blind randomized saline-controlled trial—last up to 3 years.

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

Methods: A total of 46 patients with Kellgren-Lawrence 2 or 3 knee OA were randomized into 2 groups: 1 ultrasound-guided APS injection (n = 31) or 1 saline injection (n = 15). At 1 year, the saline group was allowed to cross over. Patients were re-evaluated at 24 and 36 months through the visual analog scale for pain (VAS), Western Ontario and McMaster Universities Osteoarthritis Index Likert 3.1 (WOMAC LK 3.1), Knee injury and Osteoarthritis Outcome Score (KOOS), 36-Item Short Form Health Survey (SF-36), and Outcome Measures in Rheumatology–Osteoarthritis Research Society International (OMERACT-OARSI) responder rate. Magnetic resonance imaging evaluation was performed with the MRI Osteoarthritis Knee Score (MOAKS) before and at 24 months after treatment, and radiographs were assessed per Kellgren-Lawrence before and annually after treatment.

Results: In the APS cohort, WOMAC pain improved from 11.5 ± 2.4 (mean ± SD) to 4.3 ± 4.0 at 1 year and to 5.7 ± 5.0 at 3 years ($P < .0001$ vs baseline). The APS cohort also showed a statistically significant improvement in its KOOS pain score from 39.4 ± 13.1 to 70.6 ± 21.5 at 1 year and to 64.1 ± 24.6 at 3 years ($P < .0001$ vs baseline) and VAS pain scores from 5.5 ± 2.2 to 2.6 ± 2.5 at 1 year and to 3.4 ± 2.9 at 3 years ($P = .0184$ vs baseline). VAS pain score significantly worsened from 12 to 36 months ($P = .0411$). All patients in the saline group decided to cross over to APS, and their final scores were better than baseline, although not significantly better than at the crossover point. Overall, 7 of 26 (26.9%) APS cases and 4 of 14 (28.6%) crossover cases were considered failures as patients underwent further injective treatments or surgical procedures between the 12- and 36-month follow-up. MOAKS findings showed no statistically significant differences. Patients with better cartilage had greater WOMAC pain improvement when their baseline scores were worse, whereas the trend was reversed for patients with cartilage loss at baseline.

Conclusion: Intra-articular use of APS for mild to moderate knee OA was safe, and significant pain improvement was documented 3 years after a single injection. Patients with better cartilage status seem to respond better than patients with more cartilage loss, with more clinical improvement even when starting from more painful conditions.

Registration: NCT02138890 (ClinicalTrials.gov identifier).

Keywords: blood derivative; platelets; knee osteoarthritis; growth factors; cytokines; injection

Autologous therapies such as platelet-rich plasma (PRP), bone marrow concentrate, and autologous anti-inflammatories such as autologous conditioned serum are currently being investigated for the treatment of mild to moderate knee osteoarthritis (OA) to determine if they can improve symptoms longer or

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better than traditional intra-articular injections such as hyaluronic acid and steroids.[11] In particular, blood derivatives are gaining increasing attention for the safety, limited costs, and promising results as a minimally invasive approach to provide symptom relief and function improvement.[6,7,14]

PRP is prepared from a small sample of autologous peripheral blood, and after a centrifugation process, it can provide a high concentration of platelets and bioactive molecules to be administered back into the patient via an intra-articular injection.[2] In a systematic review and meta-analysis of 10 level 1 randomized controlled trials (RCTs), platelet concentrates were found to provide more pain relief and better functional outcomes, without an increase in the risk of adverse events, than hyaluronic acid and saline in patients with knee OA at 1 year after injection.[8] However, the literature lacks data on the long-term effects of these blood derivatives. In this landscape, autologous protein solution (APS) has been developed to provide a milieu of bioactive factors starting from a PRP and then undergoing a further process involving polyacrylamide beads to secure the production of desired molecules. This process leads to a high level of anti-inflammatory cytokines (derived from white blood cells) while ensuring low levels of proinflammatory molecules, thus combining the anabolic effects of PRP technology with autologous anti-inflammatory homeostatic properties. The results of a multicentric double-blind randomized placebo-controlled study were recently published,[19] showing that a single APS injection led to better results than saline.

The aim of this follow-up study is to document if the positive outcome previously documented in that RCT is maintained over time, by investigating clinical and imaging findings up to 3 years in patients who underwent a single APS injection for the treatment of knee OA.

METHODS

Patient Selection and Study Design

The double-blind randomized saline-controlled trial was approved by the hospital ethics committees (ClinicalTrials.gov NCT02138890), as published in the 12-month study.[19] This multicenter trial was conducted over a 2-year span (2014-2016) in 3 highly specialized referral centers for sports medicine and orthopaedics in Europe (Belgium, Italy, and Norway). Major inclusion criteria included women or men aged 40 to 75 years with knee OA Kellgren-Lawrence grade 2 or 3, body mass index $\leq$40, Western Ontario and McMaster Universities Osteoarthritis Index Likert 3.1 (WOMAC LK 3.1) questionnaire mean total score $>$1.75 and $<$4, and at least 1 failed nonoperative OA therapy (eg, physiotherapy, simple analgesics, intra-articular injection). Major exclusion criteria included symptomatic OA in the nonstudy knee, rheumatoid arthritis or arthritis secondary to other inflammatory diseases or of metabolic origin, diagnosis of isolated patellofemoral OA, intra-articular steroid injection into any joint within 3 months before screening, and hyaluronic acid injection into any joint within 6 months before screening. Demographics and pretreatment score levels were similar between groups; details were described previously, as well as a full list of enrollment criteria.[19]

A total of 46 patients underwent a 2:1 randomization process (chosen for ethical reasons to reduce the number of participants receiving placebo) to a single injection of APS (31 patients) or saline (15 patients). All patients had a blood draw to ensure double blinding, and a "blinding sleeve" was used to mask the syringe content. APS was prepared for patients randomized to the APS group with the nSTRIDE APS Kit with Anticoagulant Citrate Dextrose Solution–Formula A, a single-use device designed to concentrate growth factors and anti-inflammatory cytokines from whole blood through 2 steps. The nSTRIDE Cell Separator separated the cellular components from plasma and red blood cells in whole blood, and the resulting suspension was loaded into the nSTRIDE Concentrator, where filtration through polyacrylamide beads concentrated the cytokines in the injectable output. After joint fluid was aspirated, approximately 2.5 mL of APS or saline was injected into the joint via ultrasound guidance. After the injection, patients were sent home with instructions to limit leg use for at least 24 hours and to use cold therapy/ice on the affected area to relieve pain. After treatment, OA medication was allowed during the study and standardized as oral acetaminophen/paracetamol (maximum, 3 g/d). A gradual resumption of normal sports or recreational activities was allowed as tolerated.

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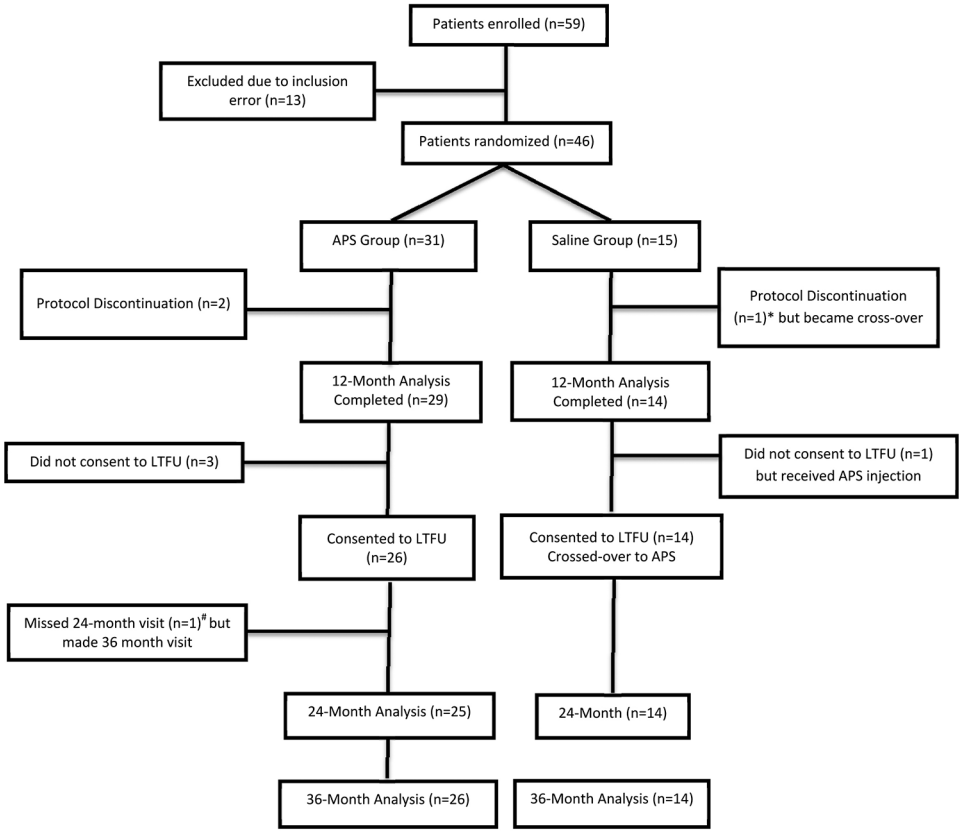

Figure 1. CONSORT (Consolidated Standards of Reporting Trials) flow diagram: efficacy analysis (number of patients). Patients who received a saline injection crossed over to APS. One patient in the APS# group and 1 in the saline/crossover* group missed their 24- and 12-month visits, respectively, but continued in LTFU. APS, autologous protein solution; LTFU, long-term follow-up.

Patient Clinical and Imaging Evaluation

One-year results were previously published on the efficacy analysis population (29 patients for APS and 14 patients for saline).[19] Forty participants (26 APS, 14 crossover) consented to the long-term follow-up. After the 12-month follow-up, participants in the saline group were offered APS treatment, and all received the crossover injection. Data for the original APS cohort (n = 31) are presented from their enrollment baseline scores. Data for the saline group are presented as their performance scores from baseline to 12 months, but then they are followed as the crossover group for the follow-up visits (ie, 24 and 36 months after enrollment, which corresponds to 12 and 24 months after the APS injection).

Long-term clinical efficacy endpoints were improvements in pain and function over time per the WOMAC, Knee injury and Osteoarthritis Outcome Score (KOOS), and visual analog score for pain (VAS) scores, measured as a change from baseline to each time point. The 36-Item Short Form Health Survey (SF-36) (8 subscales) was used to measure change from baseline in general quality-of-life outcomes. The Outcome Measures in Rheumatology–Osteoarthritis Research Society International (OMERACT-OARSI) set of responder criteria was used to calculate the number of responders/nonresponders to APS treatment over time. The percentage of patients taking analgesics for their knee OA was measured and reported as well. Morphological and structural changes in the knee joint over time after APS treatment were measured with magnetic resonance imaging (MRI) at 0, 3, 12, and 24 months and annual radiographs. In particular, images were sent to an imaging core lab and were evaluated by 2 independent musculoskeletal radiologists blinded to each other's assessments. Disagreements between the primary reviewers were resolved by a third independent reviewer. The change from screening for each MRI Osteoarthritis Knee Score (MOAKS) parameter was evaluated. In addition, joint space narrowing/Kellgren-Lawrence on radiographs was evaluated for differences between treatment groups. Patients were followed up until 36 months. For failed cases (ie, patients undergoing further treatments), the basal score was carried forward at follow-up to include them in the analysis up to 36 months. The CONSORT (Consolidated Standards of Reporting Trials) flow diagram showing patient inclusion and follow-up is reported in Figure 1.

Statistical Analysis

Data were calculated from the modified intention-to-treat population. This population included all participants who had baseline and $\geq$1 follow-up WOMAC pain scores and who had no major entry violations likely to affect outcomes, as determined by a blind review of the data before

TABLE 1
Study Exit Accountability[a]

Randomization: Patient	Exit	Reason
APS		
1002	Early termination before 12-mo visit	Exited study owing to bladder cancer
1008	12-24 mo	Did not consent to LTFU
1017	12-24 mo	Did not consent to LTFU
2006	12-24 mo	Received TKA
2008	12-24 mo	Arthrolysis
2010	24-36 mo	Received TKA
4007	12-24 mo	HA and steroid injection
4008	12-24 mo	Did not consent to LTFU
4013	24-36 mo	HA injection
4014	24-36 mo	Steroid injection
4024	24-36 mo	Steroid injection
Crossover		
1011	12-24 mo after crossover	Received TKA
1012	First 12 mo after crossover	Did not consent to LTFU
2009	12-24 mo after crossover	Received TKA
2016	First 12 mo after crossover	HA injection
4003	12-24 mo after crossover	HA injection

[a]APS, autologous protein solution; HA, hyaluronic acid; LTFU, long-term follow-up; TKA, total knee arthroplasty.

analysis. Missing data for patients exiting the study for knee OA (7 APS and 4 crossover) were imputed with their baseline scores (ie, zero change). Continuous variables were tabulated via mean, standard deviation, and number of observations. If the data were normally distributed, then a t test or analysis of variance was used for significance testing; otherwise, the Wilcoxon test was used. Categorical variables were tabulated per the number of observations and percentages, and 2×2 tables were analyzed with the Fisher exact test. Tables with >2 rows or columns were analyzed with the likelihood ratio chi-square test. Differences among and between means and proportions were declared statistically significant if $P < .05$, with no adjustment done for multiple comparisons.

RESULTS

No adverse events of interest—excessive injection site pain, burning, swelling and/or effusion, injection site infection, damage to blood vessels, hematoma, temporary or permanent nerve damage near the injection site resulting in pain or numbness, early or late postoperative infection, or other events possibly associated with APS or the injection procedure—occurred in the treated knees between the 12- and 36-month follow-up visits in the APS cohort, nor were any adverse events reported with crossover injections. Six patients took rescue medication in the 12- to 24-month time frame, and 5 of these exited the study for alternative treatments. Two patients took rescue medication in the 24- to 36-month time frame but continued in the study.

Overall, 7 of 26 (26.9%) cases in the APS cohort and 4 of 14 (28.6%) in the crossover group were considered to have treatment failure, as patients underwent further injective treatments or surgical procedures between the 12- and 36-month follow-ups. In the APS group, 3 patients had failure within the 24-month follow-up and 4 within 36 months; in the crossover group, 1 patient had failure within 12 months after crossover and 3 within the 24 months (Table 1).

Long-term Follow-up

APS Cohort. At 36 months, the mean WOMAC pain improvement was 50.4% ± 42.1% (Figure 2) in the APS cohort, which was a significant improvement as compared with the baseline score ($P < .0001$). The APS cohort also showed a statistically significant improvement versus baseline scores in its WOMAC stiffness and function scores, KOOS findings (5 subscales), and VAS pain scores (Table 2). Although significantly higher than the basal value, the level of improvement at 36 months was significantly lower than that at the 12-month evaluation for the VAS pain score ($P = .0411$). The number of OMERACT-OARSI responders at 12 months was 19 of the 29 patients available to follow-up, 15 of the 25 available at 24 months, and 13 of the 26 available at 36 months. At 36 months, the SF-36 physical and mental components improved 8.0 ± 10.5 points ($P = .0006$) and 4.4 ± 8.9 points ($P = .0184$), respectively, as compared with the baseline evaluation (Table 3).

Crossover Cohort. For the patients who received saline and then crossed over to APS after the 12-month follow-up, no statistical improvements in any efficacy measurements were found when their 12-month time point was used as their new baseline measurement. However, when compared with their initial baseline score (time zero), the crossover group after 36 months had 44.0% ± 37.6% mean WOMAC pain improvement ($P = .0008$) (Figure 2), 25.0% ± 33.4% mean WOMAC stiffness improvement ($P = .015$), and 36.4% ± 34.3% mean WOMAC function improvement ($P = .0016$) (Table 2). As compared with the original baseline scores, the crossover cohort also had significant improvements in VAS pain and in KOOS Pain,

TABLE 2
WOMAC, KOOS, and VAS Scores for the APS and Crossover Cohorts[a]

	WOMAC: Pain			WOMAC: Stiffness			WOMAC: Function			VAS		
	Mean	SD	*P* Value	Mean	SD	*P* Value	Mean	SD	*P* Value	Mean	SD	*P* Value
APS												
Baseline (n = 29)	11.5	2.4		4.8	1.7		34.9	12.4		5.5	2.2	
Month 12 (n = 29)	4.3	4.0		2.7	1.7		15.6	13.8		2.6	2.5	
Month 24 (n = 25)	4.5	4.8		2.4	1.9		14.4	14.5		2.8	2.7	
Month 36 (n = 26)	5.7	5.0		2.8	2.1		18.0	14.6		3.4	2.9	
Improvement, %	50.4	42.1	<.0001	38.4	42.8	.0002	45.3	46.8	<.0001	31.2	63.1	.0184
Crossover												
Baseline (n = 14)	11.8	1.9		5.0	1.2		38.1	9.3		6.5	1.8	
Month 12 (n = 14)	6.3	3.9		3.0	2.0		20.4	12.3		4.8	2.2	
Month 24 (n = 14)	5.1	4.3		3.1	1.9		19.5	15.7		3.8	2.5	
Month 36 (n = 14)	6.8	4.6		3.9	1.9		24.9	15.6		4.1	2.6	
Improvement, %	44.0	37.6	.0008	25.0	33.4	.015	36.4	34.3	.0016	33.7	43.4	.0123

	KOOS: Pain			KOOS: Symptoms			KOOS: ADL			KOOS: Sports/Rec			KOOS: QOL		
APS															
Baseline (n = 29)	39.4	13.1		47.8	19.3		48.6	18.3		23.1	25.0		26.5	13.8	
Month 12 (n = 29)	70.6	21.5		68.4	21.4		77.0	20.3		47.1	29.4		51.3	25.3	
Month 24 (n = 25)	71.0	25.0		69.9	21.2		78.9	21.3		42.0	31.3		53.5	26.5	
Month 36 (n = 26)	64.1	24.6		65.8	22.4		73.5	21.5		38.3	32.5		48.3	27.5	
Improvement, %	24.4	24.0	<.0001	18.1	23.4	.0006	25.3	25.9	<.0001	15.2	37.1	.047	21.9	30.8	.0013
Crossover															
Baseline (n = 14)	37.9	10.1		46.4	12.1		44.0	13.7		14.3	9.4		22.3	8.7	
Month 12 (n = 14)	61.1	18.5		58.2	17.4		70.0	18.1		37.1	27.9		38.4	19.3	
Month 24 (n = 14)	65.7	21.4		60.7	22.8		71.1	23.4		32.9	24.0		41.1	19.9	
Month 36 (n = 14)	59.9	23.5		55.4	21.4		63.5	22.9		25.7	28.2		35.7	19.7	
Improvement, %	23.0	21.3	.0014	12.2	15.0	.0092	20.9	18.6	.001	12.1	23.4	.0745	14.3	15.0	.0035

[a]Percentage improvement and *P* values calculated between baseline and 36 months. Crossover patients received APS injection after the 12-month visit. ADL, Activities of Daily Living; APS, autologous protein solution; KOOS, Knee injury and Osteoarthritis Outcome Score; QOL, Quality of Life; Sports/Rec, Sports and Recreation; VAS, visual analog scale for pain; WOMAC, Western Ontario and McMaster Universities Osteoarthritis Index.

TABLE 3
SF-36 Cumulative Scores for the Physical and Mental Components for the APS and Crossover Cohorts[a]

	SF-36 Physical Component			SF-36 Mental Component		
	Mean	SD	*P* Value	Mean	SD	*P* Value
APS						
Month 12	8.11	8.47		3.25	9.12	
Month 24	8.77	9.67		3.12	9.38	
Month 36	8.03	10.45	.0006	4.39	8.88	.0184
Crossover						
Month 12	5.53	7.12		0.96	11.60	
Month 24	6.46	10.11		6.51	9.37	
Month 36	4.18	6.29	.0337	3.11	8.09	.1908

[a]Crossover patients received APS injection after the 12-month visit. APS, autologous protein solution; SF-36, 36-Item Short Form Health Survey.

Symptoms, Activities of Daily Living, and Quality of Life (Table 2). The number of OMERACT-OARSI responders before crossover at 12 months was 7 of the 14 patients available to follow-up; after crossover, the number increased to 9 at 24 months and 36 months. At 36 months, the SF-36 physical component improved 4.2 ± 6.3 points (P = .0337) from baseline, and the mental component improved 3.1 ± 8.1 points (P = .1908) (Table 3).

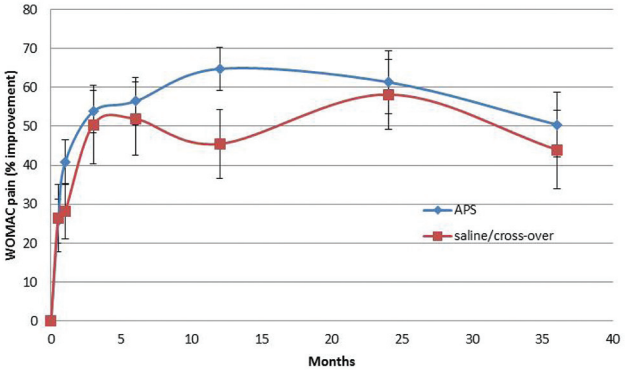

Figure 2. Percentage change in WOMAC pain score for the APS cohort. Data are presented as mean ± SE. All data points are statistically different from baseline ($P \leq .0001$). APS, autologous protein solution; WOMAC, Western Ontario and McMaster Universities Osteoarthritis Index.

Image Evaluation

MOAKS scoring of the APS cohort comparing baseline and 24 months showed no statistically significant differences (improved or worsened). There were also no statistically significant associations between the MOAKS scoring and the clinical outcome measures in the APS cohort between screening and 24 months. However, there was a statistically significant association between the change in WOMAC pain from baseline to 24 months and the percentage of full-thickness cartilage loss present at baseline ($P = .0061$); this association was related to the WOMAC baseline pain score. Participants without any region of full cartilage loss at baseline had a greater change in WOMAC pain score when their baseline WOMAC pain scores were worse (above the median/more pain). The trend was reversed for patients with a region of full cartilage loss at baseline: the mean change in WOMAC pain was greater for patients with better baseline WOMAC pain scores (below the median/less pain).

Kellgren-Lawrence assessments were completed yearly, and results represent data for patients included in the long-term follow-up. Radiograph assessment of patients with baseline and final follow-up showed 12 stable, 1 improved, and 5 worsened in the APS group at 36 months and 4 stable, 3 improved, and 3 worsened at 24 months in the crossover group. There was no correlation between the pretreatment Kellgren-Lawrence scale and study exit.

DISCUSSION

The main finding of this study is that intra-articular injections of APS for mild to moderate knee OA are safe and significant pain improvement was documented 3 years after a single injection.

Blood derivatives have been proposed as a safe, easy, cost-effective, and minimally invasive strategy to influence the joint environment, favoring the restoration of homeostatic balance and possibly the regeneration of degenerating tissues. Previous studies demonstrated in vitro the reduction of MMP-13 production from IL-1- and TNFα-activated chondrocytes and GAG release from IL-1- and TNFα-stimulated cartilage explants; cartilage protection was also demonstrated histologically in a meniscal tear animal model.[17,21,31] However, while most of the existing RCTs provide evidence of superiority in comparison with placebo—overall, they show short-term benefit versus viscosupplementation[2,19,28,29]—the literature on the duration of the beneficial effect is scarce. In fact, clinical evaluations of RCTs published so far have largely been limited to 6 or 12 months after treatment, precluding assessment of whether the biological approach might lead to a longer-lasting beneficial effect. Beyond the mere increase in clinical scores, which have been shown to increase in the short-term evaluation in all published studies, the stability of the results is equally relevant for physicians and patients. A long-term observation is required to understand results over time, which is key when choosing one treatment approach over another in clinical practice. Injective treatments are common and can be repeated over time, but they carry the risk of infective sequelae, which could be devastating[33]; therefore, products that provide long-lasting results should be favored. Currently, only 2 studies have investigated this aspect: an RCT showing scarce improvement maintained over time[9] and a case series suggesting that the median duration of clinical improvement is limited at 9 months.[10] These preliminary findings are in contrast with the present study, which could shed new light on the potential of blood derivatives over time.

The present study showed that significant improvement was maintained at 3 years. This is an unexpected finding given that available preclinical literature suggests homeostatic improvement of the joint environment rather than a cartilage-regenerative effect.[13] However, inflammation is a key OA feature associated with joint symptoms and disease progression. When the balance between pro- and anti-inflammatory cytokines leads to the perpetuation of inflammation via continued activation of innate inflammatory pathways, OA progression is inevitable in the affected joint, with chronic inflammation leading to slow progression of structural change and chronic disability.[20] In this light, anti-inflammatory approaches could counteract this key mechanism of disease progression.

Preclinical studies support the potential to modulate the disease process in OA joints.[31] Kanwat et al[15] investigated the pathway for disease-modifying effects in knee OA with a study in Dunkin-Hartley guinea pigs. Synovitis and synovial vascularity were significantly lower in PRP-treated knees at 3 and 6 months. Additionally, mean articular cartilage degeneration was significantly lower as well, which supports the possibility of having longer-lasting effects based on structural and homeostatic modifications. Khatab et al[16] showed in a mouse OA model that PRP-injected knees had a thinner synovial membrane with more anti-inflammatory cells (CD206+ and CD163+) and a trend toward less cartilage damage. These findings demonstrated that, besides pain reduction, PRP injections reduced synovial thickness, possibly through the modulation of macrophage subtypes. This might be of clinical

relevance, as it suggests the possibility of reducing not only pain but also synovial inflammation with disease-modifying and short-term chondroprotective effects, which could explain the durable results observed up to 3 years in patients undergoing APS injection.

APS is a blood derivative that provides a milieu of bioactive factors with a high level of anti-inflammatory cytokines while ensuring low levels of proinflammatory molecules[26]; it also shows in vitro the inhibition of inflammatory cytokines and destructive proteases, with the stimulation of cell proliferation in cartilage tissue.[22,24,26,32] The high concentrations of anti-inflammatory molecules and growth factors can be obtained by all patients regardless of the degree of articular cartilage degeneration and age, thus prompting the use of APS in patients with OA.[25] In a prospective randomized clinical trial, horses with naturally occurring OA demonstrated decreased lameness, which was maintained up to 52 weeks,[3] supporting the rationale to address OA by providing platelets, plasma, and white blood cells. The role of leukocytes is currently a debated aspect; in vitro experiments showed release of catabolic and proinflammatory molecules,[4,30,31] which could be detrimental to the joint. Nonetheless, a recent in vivo study demonstrated that 1 week after the injection of leukocyte-rich PRP, no increase occurred in the concentration of inflammatory molecules in the synovial fluid.[21] While it is possible that different blood derivatives exert different effects,[1,5] it is also possible that in vivo final effects might be different than those suggested by in vitro tests, and the role of cellular components must still be investigated and clarified in regard to clinical outcome. In fact, the only available comparative trial revealed similar results between leukocyte-rich and leukocyte-poor formulations,[12] leaving unanswered the question of the in vivo role of leukocytes. The mere measurement of inflammatory cytokines provides an incomplete picture of the processes that balance inflammation, especially for this blood derivative, where the production passage in polyacrylamide beads secures the production of cytokines with an overall favorable anti-inflammatory profile.[23,25] The pleiotropic effects of these bioactive molecules and the synergistic interaction of the different cell components were demonstrated by a clinical study of APS, which correlated the presence of white blood cells with favorable cell release and ultimately positive clinical findings that lasted over time, effectively extending the half-life of the molecules.[18,22]

The clinical benefit of this injective approach to address knee OA was confirmed in a double-blind RCT against saline,[19] with symptom improvement persisting up to this 3-year evaluation. Interestingly, as previously reported and as largely documented for knee injection studies,[19,27] there are substantial placebo effects, and it took some time for the potential changes in homeostasis in the joint induced by APS to overcome the waning placebo effect. The placebo effect is even greater in biologic trials where patients perceive that they are getting "regenerative medicine" therapy; accordingly, and also for this study, a large placebo effect was documented over time, with treatment effects becoming significant after only 6 months. Whether this clinical benefit is coupled with significant structural and disease-modifying effects remains to be determined, as the radiograph and MRI evaluations were not statistically significant regarding improvements or worsening. As patients who underwent saline injections all opted for crossover APS treatment, it is not possible to have a comparison of the long-term group differences and therefore to understand the effects of APS on joint structures over time. Other limitations include the limited number of patients—particularly those undergoing saline treatment—which limits the statistical significance of the evaluation of this small patient group and which could explain, with the higher basal scores at crossover, the lack of significance in the further improvement provided by APS over time. However, this is a follow-up study of a previous study where 2:1 randomization was chosen to limit the number of patients receiving saline and thus the less effective option. Nonetheless, this study allowed us to confirm safety and benefit, as well as to demonstrate the duration of the improvement over time. Interestingly, a different response pattern was observed, with patients affected by severe cartilage lesions having less improvement when starting from a more painful condition, which suggests lower potential in overly compromised cases. However, when cartilage status was less compromised, patients could benefit significantly even when affected by more painful conditions. Although these data should be interpreted with caution owing to the low number of MRI scans, this suggests the importance of exploring the potential of different patient subpopulations, and future studies could help optimize the indications and therefore the improvement and duration of the clinical benefit of patients undergoing APS injections for OA treatment.

CONCLUSION

Intra-articular use of APS for mild to moderate knee OA was safe, and significant pain improvement was documented 3 years after a single injection. Patients with better cartilage status (without any region of full cartilage loss at baseline MRI) seemed to respond better than patients with more cartilage loss, with higher clinical improvement even when starting from more painful conditions. While future studies are warranted to determine the benefit of this new autologous treatment with respect to other available injective therapies, this study confirms improvement and duration of the clinical effects, supporting the conclusion that APS injections can be considered among the treatment options for patients affected by knee OA.

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