

Original Article

Micro-needle knife combined with intra-articular and subchondral bone injection of platelet-rich plasma benefits moderate to advanced knee osteoarthritis

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Abstract: Objective: To evaluate the clinical efficacy and safety of micro-needle knife (MNK) therapy combined with intra-articular and subchondral platelet-rich plasma (PRP) injection compared to intra-articular PRP injection alone in patients with moderate to advanced knee osteoarthritis (KOA). Methods: This retrospective study enrolled 150 patients (≥ 45 years) diagnosed with moderate to advanced KOA treated at three centers (Tonglu County Traditional Chinese Medicine Hospital, Tonglu County Second People's Hospital, and Zhangshi Traditional Chinese Medicine Hospital) between April 1, 2021, and March 31, 2024. Patients were assigned to either the simple injection group (intra-articular PRP only; $n = 135$) or the combined treatment group (MNK plus intra-articular and subchondral PRP; $n = 106$). Clinical outcomes assessed at baseline and after one year included pain (visual analog scale [VAS]), function (Lysholm and WOMAC scores), inflammatory and oxidative stress markers (IL-1 β , TNF- α , SOD, MDA), overall clinical efficacy, incidence of adverse events, and structural changes assessed by radiography and Whole-Organ Magnetic Resonance Imaging Score (WORMS). Results: Baseline demographic and clinical characteristics were comparable between groups. One year after treatment, the combined treatment group exhibited significantly lower VAS and WOMAC scores and higher Lysholm scores than the simple injection group ($P < 0.05$ for all), indicating superior pain relief and functional improvement. Inflammatory and oxidative stress markers showed a more pronounced improvement in the combined group ($P < 0.05$). Overall efficacy and improvement rates were significantly higher in the combined group ($P < 0.05$), where cure rates and adverse event rates were similar. No significant differences were observed between groups in knee joint space width or WORMS at baseline or follow-up. Conclusion: MNK combined with intra-articular and subchondral PRP injection provides a greater clinical benefit in pain relief, functional recovery, and improvement of inflammatory and oxidative stress markers in moderate to advanced KOA, without increasing adverse events or altering structural outcome compared with intra-articular PRP alone.

Keywords: Knee osteoarthritis, platelet-rich plasma, micro-needle knife, pain management, joint function

Introduction

Knee osteoarthritis (KOA) is a prevalent, progressive, and disabling joint disorder that predominantly affects individuals over 45 years of age, imposing a substantial burden on patients and health care systems worldwide [1]. With the aging population, rising obesity rates, and sedentary lifestyles, both the incidence and severity of KOA are escalating, making it a leading causes of disability and reduced quality of life among older adults [2]. Characterized by degeneration of articular cartilage, subchon-

dral bone remodeling, synovial inflammation, and osteophyte formation, KOA results in chronic pain, stiffness, functional impairment, and eventual loss of mobility [3]. Its multifactorial pathogenesis, encompassing mechanical, biochemical, genetic, and inflammatory mechanisms, complicates both diagnosis and optimal management strategy [4].

Conventional treatment strategies for KOA include physical therapy, weight reduction, exercise, pharmacologic interventions with analgesic and nonsteroidal anti-inflammatory

drug (NSAID), intra-articular corticosteroid or hyaluronic acid injections, and, in advanced stages, surgical intervention [5]. Despite these options, many patients with moderate to advanced KOA continue to suffer from persistent pain, functional decline, and progressive structural deterioration [6]. Surgical interventions, while effective for end-stage disease, are associated with notable risk, high costs, and limited long-term durability, and are often unsuitable for patients with significant comorbidities or unwillingness to undergo surgery [7]. This substantial unmet need has driven growing interest in biological and regenerative therapies designed not only to alleviate symptoms but also to address the underlying pathophysiology of KOA and modify disease progression [8].

Among the emerging biological therapies, platelet-rich plasma (PRP) injections have gained attention for their ability to harness the body's intrinsic healing mechanisms [9]. PRP is an autologous concentration of platelets and growth factors prepared from the patient's own blood, which is injected into target tissues, including the intra-articular space and, more recently, the subchondral bone [9]. Upon activation, platelets release bioactive molecules - such as platelet-derived growth factor, transforming growth factor- β , insulin-like growth factor, and vascular endothelial growth factor - that exert immunomodulatory, anti-inflammatory, angiogenic, and regenerative effects on chondrocytes, synoviocytes, and subchondral bone cells [10]. Both preclinical and early clinical evidence suggests that PRP can alleviate symptoms, improve joint function, and modulate the microenvironment of OA joints, thereby slowing or modifying disease progression [11]. Increasing evidence further highlights the role of subchondral PRP delivery, because subchondral bone remodeling, bone marrow lesions, and biomechanical alterations are now recognized as central drivers of KOA initiation, progression, and pain [10, 11].

Another minimally invasive approach rooted in both traditional and contemporary medicine is micro-needle knife (MNK) therapy [12], also referred to as small needle-knife therapy or acupotomy. It is designed to mechanically release myofascial trigger points and periarticular adhesions, which contribute to local pain, contracture, and abnormal biomechanics in KOA [13]. The technique employs a fine needle

with a cutting edge to disrupt pathologic soft tissue, enhance microcirculation, and potentially restore normal biomechanics [13]. Evidence from randomized trials and meta-analyses have demonstrated that MNK can substantially reduce pain, improve knee function, and enhance walking capacity in KOA patients, with an acceptable safety profile [12, 13]. Additionally, experimental evidence indicates that MNK may exert its therapeutic effects by suppressing inflammatory cytokine expression, thereby reducing both local and systemic inflammation [14].

While both PRP and MNK have demonstrated efficacy as stand-alone treatments for KOA, evidence regarding their combined or synergistic effects remains limited - particularly if PRP is administered to both the intra-articular space and the subchondral bone [14]. The rationale for this combined approach reflects the current understanding of KOA as a "whole-organ disease", where cartilage, subchondral bone, synovium, and periarticular muscles comprise an integrated functional unit [15]. Subchondral bone pathology, including remodeling and bone marrow lesions, is increasingly recognized as an important contributor to the progression and pain of KOA, providing both biomechanical and biochemical signals that perpetuate cartilage degeneration and inflammation [15]. Emerging studies support the hypothesis that modifying the subchondral bone microenvironment - whether by biological or mechanical means - may disrupt the pathophysiologic feedback loop between bone and cartilage, resulting in more sustained symptom relief and functional improvement [15, 16].

Given these considerations, the present study was designed to evaluate the clinical efficacy and mechanisms of MNK therapy in combination with intra-articular and subchondral PRP injections in patients with moderate to advanced KOA. By simultaneously targeting both myofascial pain generators and the osteochondral unit, this multidimensional strategy aims to address the complex spectrum of pathologic changes in KOA.

Materials and methods

Case selection

A retrospective analysis was conducted on 150 patients with moderate to advanced KOA

treated at multiple centers (Tonglu County Traditional Chinese Medicine Hospital, Tonglu County Second People's Hospital, and Zhangshi Traditional Chinese Medicine Hospital) between April 1, 2021, and March 31, 2024. Patients were categorized into two groups based on their treatment methods: the simple injection group received intra-articular PRP injections alone, and the combined treatment group received MNK treatment together with intra-articular and subchondral PRP injections at the medial femoral condyle and tibial plateau. All patients were followed up for one year.

This study was approved by the Institutional Review Board (IRB) and Ethics Committee of Tonglu County Traditional Chinese Medicine Hospital. Given that this study was based on de-identified retrospective data and posed no risk or adverse impact on participants, the requirement for written informed consent was waived in accordance with ethical and regulatory guidelines.

Inclusion Criteria: (1) Patients aged ≥ 45 years; (2) Diagnosis with KOA according to the American College of Rheumatology (ACR) criteria [17], with radiographic evidence of at least one osteophyte in the tibiofemoral joint of one or both knees (Kellgren Lawrence grade 3 or 4); (3) Pain score ≥ 3 on a 10-point visual analog scale (VAS) for most of the past month; (4) Hemoglobin level > 100 g/L and platelet count $> 150 \times 10^9/L$; (5) Complete medical records without missing data.

Exclusion Criteria: (1) Severe cardiovascular, cerebrovascular, hepatic, renal, or hematopoietic diseases; (2) Inflammatory arthritis (e.g., rheumatoid arthritis, gouty arthritis) or other conditions affecting the knee joint; (3) Psychiatric disorders interfering with cooperation; (4) History of adverse reactions to MNK; (5) Pregnancy or lactation.

Intervening method

Patients in the simple injection group received an intra-articular injection of 7 mL of PRP. Patients in the combined treatment group received MNK treatment (once every 3 days, twice per week, for a total of 3 weeks), an intra-articular injection of 3 mL PRP, and subchondral injections of 2 mL PRP each in the medial femoral condyle and tibial plateau. The initial

MNK session and PRP injection were performed simultaneously.

Preparation of PRP: Before treatment, 40 mL of venous blood was drawn from patients and transferred into a PRP preparation tube containing 4 mL of sodium citrate as anticoagulant. A balancing tube was used, and the samples were centrifuged for 10 minutes at 2500 r/min with a centrifuge radius of 15 cm. After centrifugation, the red blood cell layer was aspirated with a 20 mL syringe, retaining the plasma layer along with the platelet and white blood cell layers. After rebalancing, the sample underwent a second centrifugation at 3200 r/min for 12 minutes. The upper layer was identified as platelet-poor plasma (PPP), while the lower layer was leukocyte-poor platelet-rich plasma (LP-PRP), which was collected for injection.

MNK treatment: Three local myofascial trigger points were selected according to the principles outlined in *Micro-Knife Needle Therapy*. Taking the right knee as an example, the patella was conceptualized as a clock face, with the 12 o'clock position corresponding to the superior patellar ligament and the 6 o'clock position corresponding to the inferior patellar ligament. For patients experiencing pain during stair climbing, trigger points at the 11, 12, and 1 o'clock positions along the patellar margin were selected. For pain from descending stairs, the 5, 6, and 7 o'clock positions were chosen. For medial knee pain, the 2, 3, and 4 o'clock positions were targeted, and for lateral knee pain, the 8, 9, and 10 o'clock positions were chosen.

Patients were placed in a supine position with the knee and hip flexed to fully expose the knee joint. After routine disinfection of the peri-patellar region, the practitioner palpated along the patellar margin with the guiding thumb to identify painful myofascial trigger points, which were then pressed and fixed. A Laozongyi Brand Micro-Needle Knife (0.40 mm $\times$ 25.00 mm; Sichuan Laozongyi Medical Device Co., Ltd., China) was inserted parallel to the patellar bone margin to a depth of 3 mm to sever the myofascial trigger points. After withdrawal, a dry cotton ball was applied to the puncture site for 1 minute to prevent bleeding.

Intra-articular and subchondral PRP injection: Patients were placed in a supine position, and the knee joint area was routinely disinfected

and draped. With the knee flexed at 45°, intra-articular puncture and PRP injection were performed through a parapatellar approach. The knee was then extended, and C-arm fluoroscopy was used to guide localization at the medial joint line. Following local anesthesia at the puncture site, a femoral puncture needle was inserted from below the adductor tubercle of the medial femoral condyle at a 45° angle to the femoral shaft axis, advancing into the cancellous bone of the medial femoral condyle. For the tibial side, the puncture was made 2 cm below the joint line at the midpoint of the medial tibia surface, advancing beneath the medial tibial plateau, with the needle tip positioned approximately 1-1.5 cm from the articular surface. Pre-prepared PRP was injected into both sites. After injection, sterile dressings were applied, and pressure was maintained for 10 minutes. Patients were instructed to avoid weight-bearing for 24 hours.

Data collection

Patient data were collected from the medical record system, including demographic characteristics, disease characteristics, Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) scores, clinical laboratory parameters, radiological and magnetic resonance imaging (MRI) findings before and at one year after treatment.

Primary indicators

Clinical efficacy: Treatment efficacy was evaluated in both groups according to the following criteria: (1) Cure: Complete resolution of joint pain, swelling, and discomfort; restoration of functional activity; and a Lysholm score of 80-100. (2) Improvement: Reduction in joint pain and swelling with functional improvement; a Lysholm knee score of 60-79. (3) No Improvement: No obvious change in joint pain or swelling, with a Lysholm knee score < 60.

Secondary indicators

Visual analog scale (VAS): Pain intensity was assessed using the VAS before and at one year after treatment. The VAS a 10-cm horizontal line marked on a card, with endpoints labeled “no pain” (0) and “worst possible pain” (10). Scores range from 0 to 10, with higher values indicating greater pain intensity. The intra-class

correlation coefficient (ICC) of the VAS has been reported to range from 0.97 to 0.99 [18].

Lysholm knee joint score: Knee function was evaluated using the Lysholm Knee Scoring Scale before and one year after treatment. The scale includes limp (5 points), support (5 points), squatting (5 points), stair climbing (10 points), swelling (10 points), locking (15 points), stability (25 points), and pain (25 points), with a total possible score of 100 points. Higher scores indicate better knee function. The reported Cronbach’s α of the scale is 0.65 [19].

Western Ontario and McMaster Universities Arthritis Index (WOMAC): Knee function was assessed using the WOMAC before and one year after treatment. The WOMAC consists of three subscales: knee pain, knee stiffness, and knee function. Each subscale score was standardized to a range of 0-100, with higher scores indicating more severe symptoms. The Cronbach’s α for the subscales are 0.84 for pain, 0.86 for stiffness, and 0.96 for physical function [20].

Clinical laboratory parameter: Before and after treatment, inflammatory and oxidative stress markers were assessed. A 5 mL fasting venous blood sample was collected from each patient, centrifuged at 3000 rpm for 15 minutes at room temperature, to separate the serum. Levels of interleukin-1 β (IL-1 β) and tumor necrosis factor α (TNF- α) were determined using the Human Inflammation 20-Plex ProcartaPlex Assay Kit (EPX200-12185-901, Thermo Fisher Scientific, USA). Serum superoxide dismutase (SOD) activity was determined using the Superoxide Dismutase Colorimetric Activity Kit (EIASODC; Thermo Fisher Scientific, USA). Serum malondialdehyde (MDA) levels were assessed using the Human Malondialdehyde ELISA Kit (E-10376; Shanghai Yaji Biotechnology Co., Ltd., China).

Whole-Organ Magnetic Resonance Imaging Score (WORMS): At baseline and after one year of follow-up, WORMS was assessed to quantify KOA severity and progression. The scoring system divides the knee into anatomic regions, including the medial femorotibial joint, lateral femorotibial joint, and patellofemoral joint. For each anatomical region, WORMS scores cartilage, bone marrow lesions, menisci, ligaments, and joint effusion on MRI.

Table 1. Comparison of demographic characteristics between the two groups

Factor	Simple injection group (n = 84)	Combined treatment group (n = 66)	t/ χ^2	P
Age (years)	54.36 $\pm$ 5.62	55.42 $\pm$ 5.32	1.175	0.242
Male/Female [n (%)]	47 (55.95%)/37 (44.05%)	35 (53.03%)/31 (46.97%)	0.127	0.721
BMI (kg/m ²)	31.26 $\pm$ 11.92	30.65 $\pm$ 6.78	0.390	0.697
Ethnicity [n (%)]			0.018	0.893
Han	68 (80.95%)	54 (81.82%)		
Others	16 (19.05%)	12 (18.18%)		
Educational level [n (%)]			0.698	0.874
Illiteracy	7 (8.33%)	5 (7.58%)		
Grade school	9 (10.71%)	10 (15.15%)		
Junior middle school or Senior high school	51 (60.71%)	39 (59.09%)		
Bachelor or above	17 (20.24%)	12 (18.18%)		
Marital status [n (%)]			0.529	0.912
Single	11 (13.10%)	9 (13.64%)		
Married/cohabitant	61 (72.62%)	47 (71.21%)		
Widow/Widower	4 (4.76%)	2 (3.03%)		
Separated/divorced	8 (9.52%)	8 (12.12%)		
Smoking history [n (%)]			0.822	0.365
Yes	46 (54.76%)	41 (62.12%)		
No	38 (45.24%)	25 (37.88%)		
Alcohol consumption history [n (%)]			0.092	0.761
Yes	45 (53.57%)	37 (56.06%)		
No	39 (46.43%)	29 (43.94%)		
Diabetes history [n (%)]			0.156	0.693
Yes	28 (33.33%)	20 (30.30%)		
No	56 (66.67%)	46 (69.70%)		
Hypertension history [n (%)]			1.352	0.245
Yes	22 (26.19%)	12 (18.18%)		
No	62 (73.81%)	54 (81.82%)		

BMI: Body Mass Index.

Each feature is graded from 0 to a maximum value (3-6, depending on the specific feature being assessed). The scores from subscales were summed to yield a composite WOMBS score. Reported intra-class correlation coefficients (ICC) for WOMBS range from 0.74 to 1.00 [21].

Statistical analysis

Data analysis was performed using SPSS 29.0 (SPSS Inc., Chicago, IL, USA). Continuous variables were presented as means $\pm$ standard deviation (M $\pm$ SD) and compared using independent samples t-test or paired t test, as appropriate. Categorical variables were presented as frequencies and percentages [n (%)], and compared using chi-square tests (χ^2). All statistical tests were two-tailed, with P < 0.05 considered significant.

Results

Basic data

A total of 150 patients with moderate to advanced KOA were included in the study, with 84 in the simple injection group and 66 in the combined treatment group. Baseline demographic and disease characteristics did not differ significantly between the two groups (all P > 0.05), indicating comparability (**Tables 1, 2**).

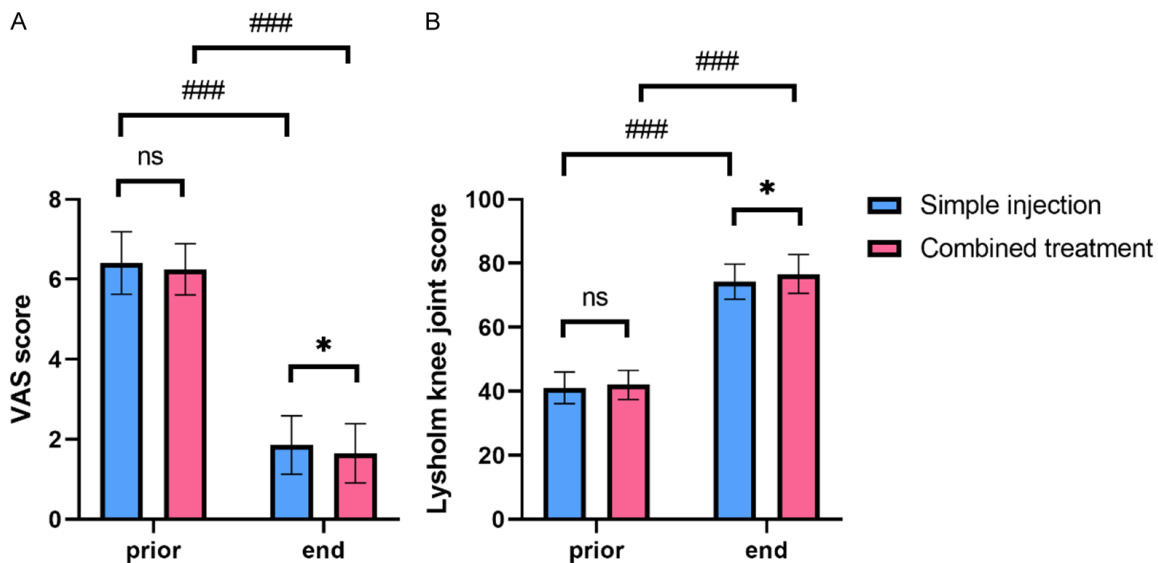
VAS and Lysholm knee joint score

At baseline, there were no significant differences between the two groups in VAS score (P = 0.167) or Lysholm knee joint score (P = 0.239) (**Figure 1**). At 1-year follow-up, the combined treatment group demonstrated a significantly lower VAS score (P = 0.041) and a significantly

Table 2. Comparison of disease characteristics between the two groups

Factor	Simple injection group (n = 84)	Combined treatment group (n = 66)	t/ χ^2	P
Cause of disease [n (%)]			1.454	0.693
Overuse or damage	42 (50.00%)	29 (43.94%)		
Muscle weakness	22 (26.19%)	16 (24.24%)		
Genetic factors	8 (9.52%)	10 (15.15%)		
Others	12 (14.29%)	11 (16.67%)		
Pathogenic site [n (%)]			2.139	0.343
Right knee	23 (27.38%)	18 (27.27%)		
Left knee	22 (26.19%)	24 (36.36%)		
Both knees	39 (46.43%)	24 (36.36%)		
K-L sore [n (%)]			0.253	0.615
3	48 (57.14%)	35 (53.03%)		
4	36 (42.86%)	31 (46.97%)		
Duration of pain (years)	6.54 $\pm$ 1.58	6.67 $\pm$ 1.35	0.500	0.618
Knee circumference (cm)	40.21 $\pm$ 3.31	40.52 $\pm$ 3.27	0.570	0.570
Previous treatment (past half year) [n (%)]			1.744	0.418
Pharmaceutical intervention	43 (51.19%)	27 (40.91%)		
Physiotherapy	16 (19.05%)	17 (25.76%)		
Assistive devices	25 (29.76%)	22 (33.33%)		

K-L: Kellgren Lawrence.


Figure 1. Comparison of the VAS score and the Lysholm knee joint score between the two groups. A. VAS score; B. Lysholm knee joint score. VAS: Visual Analog Scale; ns: no significant difference; *: P < 0.05; ###: P < 0.001.

higher Lysholm score (P = 0.012) compared to the simple injection group, indicating superior pain relief and functional improvement in patients receiving MNF combined with intra-articular and subchondral PRP injection. Within-group analysis demonstrated significant improvements in both VAS and Lysholm scores

after treatment compared to baseline (all P < 0.001).

WOMAC

Before treatment, WOMAC subscale scores (pain, stiffness, function) and total scores did

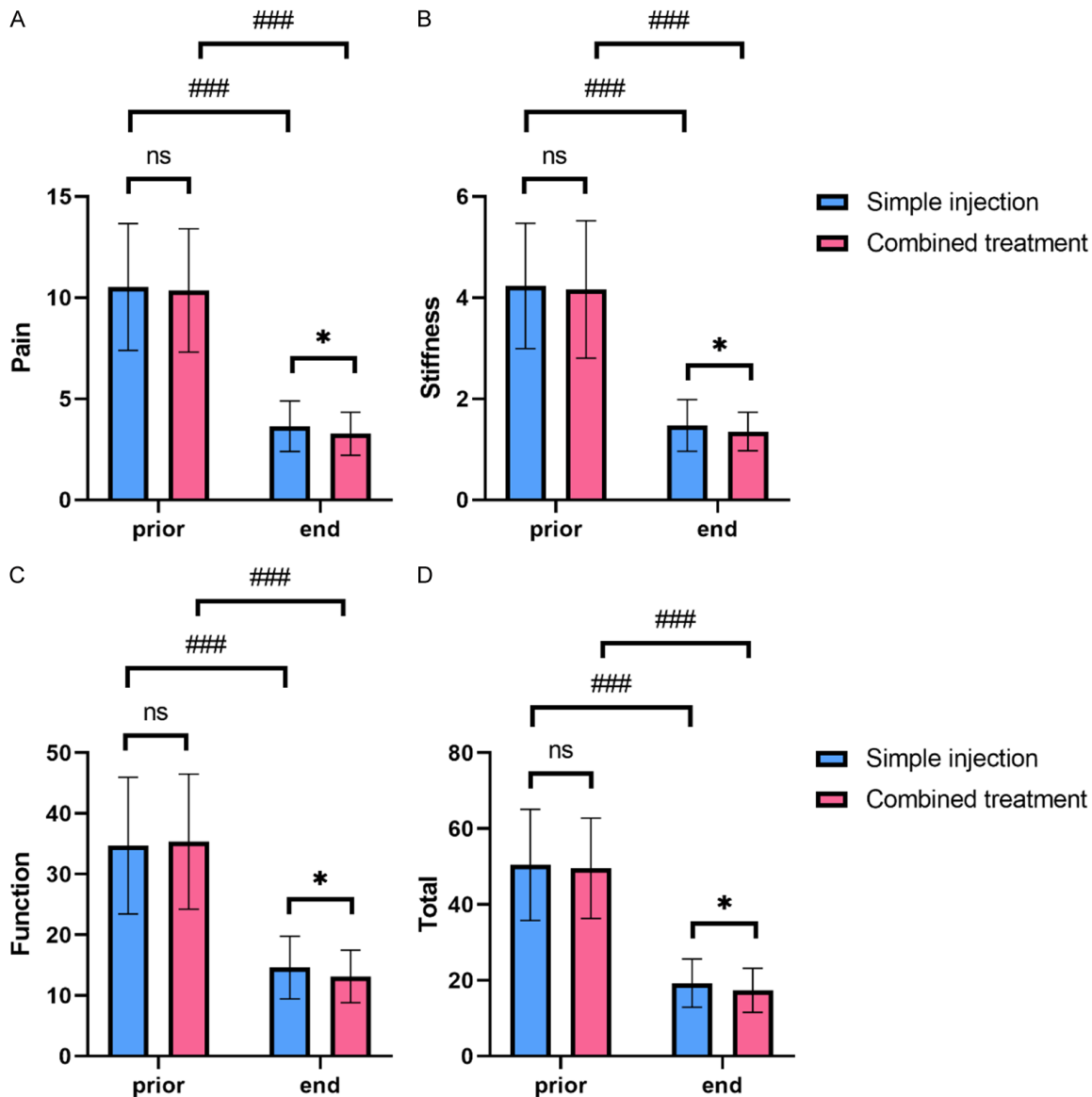


Figure 2. Comparison of the WOMAC score between the two groups. A. Pain; B. Stiffness; C. Function; D. Total. WOMAC: Western Ontario and McMaster Universities Arthritis Index; ns: no significant difference; *: $P < 0.05$; ###: $P < 0.001$.

not differ significantly between groups (all $P > 0.05$) (Figure 2). At 1-year follow-up, however, the combined treatment group demonstrated significantly lower WOMAC pain ($P = 0.013$), stiffness ($P = 0.018$), function ($P = 0.050$), and total scores ($P = 0.048$) compared to the simple injection group. Within-group comparisons revealed significant reductions in WOMAC scores from baseline in both groups ($P < 0.001$).

Clinical laboratory values

At baseline, there were no significant differences between the two groups in IL-1 β , TNF- α ,

SOD, or MDA levels (all $P > 0.05$) (Table 3). At 1-year follow-up, the combined treatment group exhibited significantly lower IL-1 β ($P = 0.010$) and TNF- α ($P = 0.015$) levels, higher SOD content ($P = 0.023$), and lower MDA levels ($P = 0.017$) compared to the simple injection group. This indicated that MNF combined with intra-articular and subchondral PRP injection more effectively ameliorated inflammatory responses and oxidative stress in patients with moderate to advanced KOA. Within-group comparisons showed significant improvements in all four values after treatment relative to baseline ($P < 0.001$).

Table 3. Comparison of the inflammatory response and oxidative stress reaction indexes between the two groups

Factor	Simple injection group (n = 84)	Combined treatment group (n = 66)	t	P
IL-1 β (ng/L) (baseline level)	38.67 $\pm$ 3.45	39.26 $\pm$ 3.48	1.020	0.309
IL-1 β (ng/L) (at 1-year follow-up)	19.63 $\pm$ 2.64	18.68 $\pm$ 1.87 ^{###}	2.595	0.010
TNF- α (ng/L) (prior treatment)	87.64 $\pm$ 9.52	88.63 $\pm$ 10.62	0.604	0.547
TNF- α (ng/L) (at 1-year follow-up)	52.41 $\pm$ 5.06	50.36 $\pm$ 5.07 ^{###}	2.458	0.015
SOD (NU/mL) (baseline level)	6.09 $\pm$ 1.35	6.14 $\pm$ 1.24	0.220	0.826
SOD (NU/mL) (at 1-year follow-up)	12.12 $\pm$ 1.41	12.63 $\pm$ 1.24 ^{###}	2.295	0.023
MDA (nmol/L) (baseline level)	8.59 $\pm$ 1.62	8.52 $\pm$ 1.53	0.276	0.783
MDA (nmol/L) (at 1-year follow-up)	5.84 $\pm$ 1.23	5.37 $\pm$ 1.18 ^{###}	2.405	0.017

IL-1 β : Interleukin-1 β ; TNF- α : Tumor Necrosis Factor α ; SOD: Serum Superoxide Dismutase; MDA: Malondialdehyde; ^{###}: P < 0.001, compared to baseline level.

Table 4. Comparison of clinical efficacy between the two groups

Factor	Simple injection group (n = 84)	Combined treatment group (n = 66)	χ^2	P
Cure [n (%)]	23 (27.38%)	15 (22.73%)	0.423	0.515
Improve [n (%)]	43 (51.19%)	45 (68.18%)	4.400	0.036
Ineffective [n (%)]	18 (21.43%)	6 (9.09%)	4.186	0.041
Effective [n (%)]	66 (78.57%)	60 (90.91%)	4.186	0.041

Table 5. Comparison of adverse events between the two groups

Factor	Simple injection group (n = 84)	Combined treatment group (n = 66)	χ^2	P
Local pain [n (%)]	19 (22.62%)	9 (13.64%)	1.964	0.161
Redness [n (%)]	9 (10.71%)	6 (9.09%)	0.108	0.742
Swelling [n (%)]	12 (14.29%)	15 (22.73%)	1.784	0.182

Clinical efficacy

There was no significant difference in cure rate between the two groups (P = 0.515) (**Table 4**). However, the combined treatment group demonstrated a significantly higher improvement rate (P = 0.036) and a lower proportion of ineffective outcomes (P = 0.041) at one-year follow-up, indicating superior clinical effectiveness of MNF combined with intra-articular and subchondral PRP injection for the management of moderate to advanced KOA.

Adverse events

There were no significant differences in the incidence of adverse events between the two groups (**Table 5**). Local pain, redness, and swelling were comparable between groups, indicating that the addition of MNF to intra-articular and subchondral PRP injection did not increase the risk of adverse events in patients with moderate to advanced KOA.

Radiological and MRI findings

There were no significant differences between the two groups in knee joint space width or WOMS at baseline or 12 months after treatment (**Table 6** and **Figure 3**). Within-group analysis, however, demonstrated significant changes from baseline to post-treatment (P < 0.001).

Discussion

A key rationale for integrating MNK therapy with intra-articular and subchondral PRP injections lies in their complementary mechanisms of action. MNK therapy, rooted in principles of traditional Chinese medicine and modern pain science, mechanically disrupts pathologic myofascial trigger points and periarticular tissue adhesions that arise in KOA from chronic overload, maladaptive gait patterns, and persistent inflammation [22]. The controlled micro-injuries produced by needle manipulation may activate endogenous repair processes, including the

Table 6. Comparison of imaging findings of the knee joint space width (mm) and WOMS between the two groups

Factor	Simple injection group (n = 84)	Combined treatment group (n = 66)	t	P
Knee joint space width (baseline)	2.01 ± 0.67	2.08 ± 0.52	0.759	0.449
Knee joint space width (one year after treatment)	1.57 ± 0.43	1.59 ± 0.46###	0.223	0.824
WORMS (baseline)	74.68 ± 20.96	73.64 ± 19.25	0.312	0.755
WORMS (one year after treatment)	78.41 ± 21.85	81.86 ± 20.37###	0.986	0.326

WORMS: Whole-Organ Magnetic Resonance Imaging Score; ###: P < 0.001, compared to baseline value.

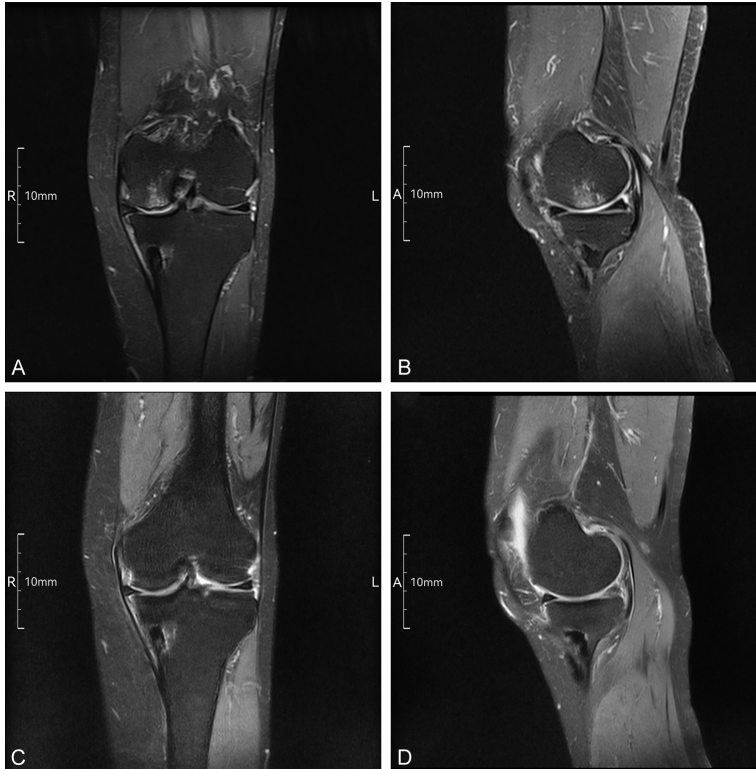


Figure 3. Comparison of MRI images between the two groups. A. MRI of the simple injection group (baseline); B. MRI of the combined treatment group (baseline); C. MRI of the simple injection group (12 months after treatment); D. MRI of the combined treatment group (12 months after treatment). MRI: Magnetic Resonance Imaging.

recruitment of reparative cells and the release of growth factors, thereby priming the local microenvironment for the reparative effects of subsequently administered PRP [23].

PRP exerts therapeutic effects through its concentrated autologous growth factors, cytokines, and chemokines [24]. Once activated, PRP releases a spectrum of bioactive mediators, including platelet-derived growth factor (PDGF), transforming growth factor- β (TGF- β), vascular endothelial growth factor (VEGF),

and insulin-like growth factor-1 (IGF-1) [25]. In articular cartilage and subchondral bone, these factors exert anti-inflammatory effects (e.g., downregulation of IL-1 β and TNF- α), promote cellular proliferation and matrix synthesis, enhance chondrocyte viability, and modulate the catabolic microenvironment typical in KOA. Notably, PRP also facilitates mesenchymal stem cell recruitment, stimulates angiogenesis, and fosters subchondral bone remodeling, all of which contribute to joint repair and symptom alleviation in chronic osteoarthritis [25]. Similarly, Qiao et al. [26] reported that PRP demonstrated the best WOMAC scores at 3, 6, and 12 months compared to other treatments, corroborating the sustained functional benefits of PRP. Taken together, these findings suggest that MNK may enhance the biological responsiveness of periarticular tissues to PRP, thereby

amplifying its clinical efficacy in moderate to advanced KOA.

The improved pain relief and functional gains observed in the combined therapy group can be explained within a mechanistic framework. Mechanical release of myofascial trigger points by MNK may disrupt chronic pain circuits and restore joint kinematics, thereby enhancing both distribution and biological efficacy of intra-articular and subchondral PRP [27]. Furthermore, subchondral PRP may directly tar-

get bone marrow lesions-recognized contributors to pain generation and KOA progression-while attenuating the inflammatory signaling sustained by subchondral bone and synovium [27]. The suppression of pro-inflammatory cytokines such as IL-1 β and TNF- α observed in this study supports the current view of KOA as an inflammation-driven disease, influenced by dys-regulated innate immunity and oxidative stress. Thus, the reduction in these markers is consistent with both symptomatic and histopathologic improvements.

Another key finding was the modulation of oxidative stress indices, including increased SOD and decreased MDA [28]. Oxidative stress is a well-established driver of cartilage matrix degradation, chondrocyte apoptosis, and progression of OA [28]. Enhanced SOD activity and decreased MDA reflect reduced oxidative damage and improved antioxidant defenses, likely mediated by the composite anti-inflammatory and cellular effects of PRP, as well as improved local vascularization and tissue metabolism following MNK [29]. By mitigating oxidative stress, combined therapy may help slow matrix breakdown and joint senescence, offering a more durable chondroprotective effect than symptom-relief alone [30].

In terms of clinical effectiveness, the superior improvement and overall efficacy rates in the combined treatment group may reflect not only additive but also synergistic effects of the two modalities. By releasing pathologic myofascial adhesions, MNK optimizes the periarticular microenvironment, alleviates pain, and may enhance compliance with rehabilitation. This, in turn, facilitates the distribution and action of intra-articular and subchondral PRP, enabling growth factors to exert reparative effects more broadly and deeply within joint and subchondral compartments. The inclusion of subchondral infiltration is consistent with the growing evidence that bone-cartilage unit pathology is integral to pain and structural outcomes in KOA [31]. This approach could explain the greater magnitude of clinical improvement and patient-reported outcomes compared to intra-articular injection alone, which does not adequately address subchondral pathology.

Nevertheless, no significant differences in radiographic or MRI-based structural outcomes (e.g., joint space width, WOMS) were observed between the two groups over one-year follow-

up. This aligns with recent trials reporting that biological and minimally invasive therapies provide marked symptomatic relief without inducing robust imaging-detectable structural changes over relatively short observation periods. Structural progression in KOA is slow, heterogeneous, and often discordant with clinical symptoms. MRI-based composite scores such as WOMS are sensitive to early alterations, these changes may be insufficiently pronounced over a one-year period, especially in moderate to advanced disease stages. Furthermore, PRP and MNK likely exert their principal benefits by modulating microstructural, cellular, and biochemical processes rather than eliciting macroscopic cartilage or bone repair detectable by conventional imaging.

Another consideration is the comparable safety profile between the two groups. The addition of MNK and subchondral PRP did not increase adverse events, suggesting that this combined modality is well tolerated. This finding is clinically meaningful given the risks associated with surgical interventions in this population. Moreover, the minimally invasive nature of MNK and autologous origin of PRP not only minimize immunogenic and infectious risks but also make repeated interventions feasible when necessary.

The present study has several limitations. First, the retrospective design is subject to inherent selection bias and unmeasured confounders, although it provides valuable real-world insight. Second, the one-year follow-up was sufficient to capture symptomatic and biochemical changes but may have been inadequate to assess long-term structural modification. Third, although inter-group comparisons were emphasized, detailed intra-group analyses before and after treatment were limited, and this may have provided a more comprehensive understanding of therapeutic effects. Future prospective, randomized studies with extended follow-up and additional mechanistic biomarkers are warranted to confirm and expand upon our findings.

Conclusion

The combined use of MNK and intra-articular and subchondral PRP injections provides a multi-faceted approach for the management of moderate to advanced KOA. By addressing pain, inflammation, oxidative stress, and micro-structural changes, this strategy appears to

provide synergistic clinical benefits beyond PRP alone. While this approach shows potential in the non-surgical management of moderate to advanced KOA, further high-quality studies are needed to determine its long-term effects on joint preservation and disease modification.

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Disclosure of conflict of interest

None.

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References

- [1] Wang YC, Lee CL, Chen YJ, Tien YC, Lin SY, Chen CH, Chou PP and Huang HT. Comparing the efficacy of intra-articular single platelet-rich plasma (PRP) versus novel crosslinked hyaluronic acid for early-stage knee osteoarthritis: a prospective, double-blind, randomized controlled trial. *Medicina (Kaunas)* 2022; 58: 1028.
- [2] Siddiq MAB, Clegg D, Jansen TL and Rasker JJ. Emerging and new treatment options for knee osteoarthritis. *Curr Rheumatol Rev* 2022; 18: 20-32.
- [3] Raeissadat SA, Rahimi M, Rayegani SM and Moradi N. Cost-utility analysis and net monetary benefit of Platelet Rich Plasma (PRP), intra-articular injections in compared to Plasma Rich in Growth Factors (PRGF), Hyaluronic Acid (HA) and ozone in knee osteoarthritis in Iran. *BMC Musculoskelet Disord* 2023; 24: 22.
- [4] Pesare E, Vicenti G, Kon E, Berruto M, Caporali R, Moretti B and Randelli PS. Italian Orthopaedic and Traumatology Society (SIOT) position statement on the non-surgical management of knee osteoarthritis. *J Orthop Traumatol* 2023; 24: 47.
- [5] Du D and Liang Y. A meta-analysis and systematic review of the clinical efficacy and safety of platelet-rich plasma combined with hyaluronic acid (PRP + HA) versus PRP monotherapy for knee osteoarthritis (KOA). *J Orthop Surg Res* 2025; 20: 57.
- [6] Thompson K, Shankar DS, Huang S, Kirsch T, Campbell KA, Gonzalez-Lomas G, Alaia MJ, Strauss EJ and Jazrawi LM. The effectiveness of Alpha-2-macroglobulin injections for osteoarthritis of the knee. *Bull Hosp Jt Dis* (2013) 2024; 82: 245-256.
- [7] Dubin J, Leucht P, Murray M and Pezold R; Staff of the American Academy of Orthopaedic Surgeons on Behalf of the Platelet-Rich Plasma (PRP) for Knee Osteoarthritis Technology Overview Workgroup and Contributors. American academy of orthopaedic surgeons technology overview summary: Platelet-Rich Plasma (PRP) for knee osteoarthritis. *J Am Acad Orthop Surg* 2024; 32: 296-301.
- [8] Tschopp M, Pfirrmann CWA, Fucentese SF, Brunner F, Catanzaro S, Kühne N, Zwyssig I, Sutter R, Götschi T, Tanadini M and Roskopf AB. A randomized trial of intra-articular injection therapy for knee osteoarthritis. *Invest Radiol* 2023; 58: 355-362.
- [9] Khalid S, Ali A, Deepak F, Zulfiqar MS, Malik LU, Fouzan Z, Nasr RA, Qamar M and Bhattarai P. Comparative effectiveness of intra-articular therapies in knee osteoarthritis: a meta-analysis comparing platelet-rich plasma (PRP) with other treatment modalities. *Ann Med Surg (Lond)* 2024; 86: 361-372.
- [10] Tanguilig G, Dhillon J and Kraeutler MJ. Platelet-rich plasma for knee and hip osteoarthritis pain: a scoping review. *Curr Rev Musculoskelet Med* 2024; 17: 415-421.
- [11] Li Z and Weng X. Platelet-rich plasma use in meniscus repair treatment: a systematic review and meta-analysis of clinical studies. *J Orthop Surg Res* 2022; 17: 446.
- [12] Di Martino A, Boffa A, Andriolo L, Romandini I, Altamura SA, Cenacchi A, Roverini V, Zaffagnini S and Filardo G. Leukocyte-rich versus leukocyte-poor platelet-rich plasma for the treatment of knee osteoarthritis: a double-blind randomized trial. *Am J Sports Med* 2022; 50: 609-617.
- [13] Lewis E, Merghani K, Robertson I, Mulford J, Prentice B, Mathew R, Van Winden P and Ogden K. The effectiveness of leucocyte-poor platelet-rich plasma injections on symptomatic early osteoarthritis of the knee: the PEAK randomized controlled trial. *Bone Joint J* 2022; 104-B: 663-671.
- [14] Bensa A, Previtali D, Sangiorgio A, Boffa A, Salerno M and Filardo G. PRP injections for the treatment of knee osteoarthritis: the improvement is clinically significant and influenced by platelet concentration: a meta-analysis of randomized controlled trials. *Am J Sports Med* 2025; 53: 745-754.

- [15] Laver L, Filardo G, Sanchez M, Magalon J, Tischer T, Abat F, Bastos R, Cugat R, Iosifidis M, Kocaoglu B, Kon E, Marinescu R, Ostojic M, Beaufils P and de Girolamo L; ESSKA - ORBIT Group. The use of injectable orthobiologics for knee osteoarthritis: a European ESSKA-ORBIT consensus. Part 1-Blood-derived products (platelet-rich plasma). *Knee Surg Sports Traumatol Arthrosc* 2024; 32: 783-797.
- [16] Berrigan WA, Bailowitz Z, Park A, Reddy A, Liu R and Lansdown D. A greater platelet dose may yield better clinical outcomes for platelet-rich plasma in the treatment of knee osteoarthritis: a systematic review. *Arthroscopy* 2025; 41: 809-817, e802.
- [17] Recommendations for the medical management of osteoarthritis of the hip and knee: 2000 update. American College of Rheumatology Subcommittee on Osteoarthritis Guidelines. *Arthritis Rheum* 2000; 43: 1905-1915.
- [18] Williamson A and Hoggart B. Pain: a review of three commonly used pain rating scales. *J Clin Nurs* 2005; 14: 798-804.
- [19] Kocher MS, Steadman JR, Briggs KK, Sterett WI and Hawkins RJ. Reliability, validity, and responsiveness of the Lysholm knee scale for various chondral disorders of the knee. *J Bone Joint Surg Am* 2004; 86: 1139-1145.
- [20] Symonds T, Hughes B, Liao S, Ang Q and Bellamy N. Validation of the chinese western ontario and mcmaster universities osteoarthritis index in patients from mainland china with osteoarthritis of the knee. *Arthritis Care Res (Hoboken)* 2015; 67: 1553-1560.
- [21] Peterfy CG, Guermazi A, Zaim S, Tirman PF, Mi- aux Y, White D, Kothari M, Lu Y, Fye K, Zhao S and Genant HK. Whole-Organ Magnetic Resonance Imaging Score (WORMS) of the knee in osteoarthritis. *Osteoarthritis Cartilage* 2004; 12: 177-190.
- [22] Filbay SR, Dowsett M, Chaker Jomaa M, Rooney J, Sabharwal R, Lucas P, Van Den Heever A, Kazaglis J, Merlino J, Moran M, All- wright M, Kuah DEK, Durie R, Roger G, Cross M and Cross T. Healing of acute anterior cruciate ligament rupture on MRI and outcomes following non-surgical management with the cross bracing protocol. *Br J Sports Med* 2023; 57: 1490-1497.
- [23] Jawanda H, Khan ZA, Warriar AA, Acuña AJ, Al- lahhabadi S, Kaplan DJ, Ritz E, Jackson GR, Mameri ES, Batra A, Dornan G, Westrick J, Verma NN and Chahla J. Platelet-rich plasma, bone marrow aspirate concentrate, and hyal- uronic acid injections outperform corticoste- roids in pain and function scores at a minimum of 6 months as intra-articular injections for knee osteoarthritis: a systematic review and network meta-analysis. *Arthroscopy* 2024; 40: 1623-1636, e1621.
- [24] Belk JW, Lim JJ, Keeter C, McCulloch PC, Houck DA, McCarty EC, Frank RM and Kraeutler MJ. Patients with knee osteoarthritis who receive platelet-rich plasma or bone marrow aspirate concentrate injections have better outcomes than patients who receive hyaluronic acid: sys- tematic review and meta-analysis. *Arthroscopy* 2023; 39: 1714-1734.
- [25] Paget LDA, Reurink G, de Vos RJ, Weir A, Moen MH, Bierma-Zeinstra SMA, Stufkens SAS, Goe- degebuure S, Krips R, Maas M, Meuffels DE, Nolte PA, Runhaar J, Kerkhoffs GMMJ and Tol JL. Platelet-rich plasma injections for the treat- ment of ankle osteoarthritis. *Am J Sports Med* 2023; 51: 2625-2634.
- [26] Qiao X, Yan L, Feng Y, Li X, Zhang K, Lv Z, Xu C, Zhao S, Liu F, Yang X and Tian Z. Efficacy and safety of corticosteroids, hyaluronic acid, and PRP and combination therapy for knee osteo- arthritis: a systematic review and network me- ta-analysis. *BMC Musculoskelet Disord* 2023; 24: 926.
- [27] Yurtbay A, Say F, Çinka H and Ersoy A. Multiple platelet-rich plasma injections are superior to single PRP injections or saline in osteoarthritis of the knee: the 2-year results of a random- ized, double-blind, placebo-controlled clinical trial. *Arch Orthop Trauma Surg* 2022; 142: 2755-2768.
- [28] Singh H, Knapik DM, Polce EM, Eikani CK, Bjornstad AH, Gursoy S, Perry AK, Westrick JC, Yanke AB, Verma NN, Cole BJ and Chahla JA. Relative efficacy of intra-articular injections in the treatment of knee osteoarthritis: a system- atic review and network meta-analysis. *Am J Sports Med* 2022; 50: 3140-3148.
- [29] Oeding JF, Varady NH, Fearington FW, Pareek A, Strickland SM, Nwachukwu BU, Camp CL and Krych AJ. Platelet-rich plasma versus alter- native injections for osteoarthritis of the knee: a systematic review and statistical fragility in- dex-based meta-analysis of randomized con- trolled trials. *Am J Sports Med* 2024; 52: 3147- 3160.
- [30] Budhiparama NC, Putramega D and Lumban- Gaol I. Orthobiologics in knee osteoarthritis, dream or reality? *Arch Orthop Trauma Surg* 2024; 144: 3937-3946.
- [31] Journal Of Healthcare Engineering. Retracted: intra-articular injection of PRP in the treatment of knee osteoarthritis using big data. *J Healthc Eng* 2023; 2023: 9828406.