



ORIGINAL ARTICLE

Comparison of intraarticular corticosteroid and autologous platelet rich plasma injection in patients with knee osteoarthritis.

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ABSTRACT... Objective: To compare IA corticosteroid and autologous PRP injection in OA knee, in terms of reducing stiffness, alleviating pain, and enhancing physical functionality. **Study Design:** Quasi-experimental study. **Setting:** AFIRM Rawalpindi. **Period:** Aug 2019 to April 2020. **Methods:** 70 patients with Knees OA, 40-70 years of age were included. Group A received IA 40 mg Triamcinolone Injection while Group B received PRP Injection in the affected knee. Pain evaluation was carried out using the Numerical Rating Scale (NRS) and the Western Ontario McMaster Osteoarthritis (WOMAC) Index at baseline, at 1 month, at 3rd month, and after 6 months and mean changes calculated. **Results:** Among 70 patients, 36 were male (52%) and 34 were female (48%). No notable differences were observed in age ($p=0.210$), sex ($p=0.811$), and body mass index (BMI) ($p=0.985$) between two groups. Pre-injection pain severity assessed on NRS ($p=0.217$) and OA severity assessed on WOMAC ($p=0.010$) differed not between the two groups. At 6 months postinjection, there was significant improvement (p -value < 0.05) in pain relief in Group B (IA PRP) compared to Group A (IA steroid injection). However, at 1 month post injection NRS and WOMAC has shown more improvement between Group A and Group B, a significant difference was observed ($p < 0.0001$). **Conclusion:** PRP Injection decreases joint pain and improve quality of life better than corticosteroid injection prolonged periods.

Key words: Corticosteroid, OA Knee, PRP.

INTRODUCTION

OA is predominant type of arthritis, significantly causing chronic discomfort and disability.¹ This complex, multifactorial disease has emerged as a significant issue in public health and a notable economic strain on the global economy.² Attributed to increase in life expectancy and obesity, OA is now affecting more than 50 percent of the adults older than 65 years.³

The aim of management in OA Knee is to provide clinical improvement, restore joint function and potential to modify the disease course.⁴ Various treatment options have been proposed, including IA injections, non-steroidal anti-inflammatory drugs and physical therapy, key components in the non-surgical approach to managing OA knee, however so far, no treatment has demonstrated all the features of a perfect therapeutic approach. As

there are currently no widely recognized medical interventions that modify the advancement of the condition and therapeutic approaches for OA Knee, predominantly focus on managing symptoms. Common approaches often involve administering IA injections containing corticosteroid, Hyaluronic Acid (HA) and PRP.

Corticosteroid have long been used to treat OA, and widely endorsed consensus statements suggest their use as an effective complementary therapy for managing OA Knee.⁵⁻⁷ Clinical studies and comprehensive reviews have consistently shown the efficacy of corticosteroid in management of OA Knee. IA corticosteroid injections are frequently administered with intention of providing symptomatic relief and potentially delaying the need for surgical intervention. Although corticosteroid injections

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seem to enhance pain scores for a short duration⁸, they are related with adverse effects⁹ and may not give relief beyond a period of 6 weeks.¹⁰

HA, a naturally occurring glycosaminoglycan, present in joints fluid, is a recently popularized treatment approach for its protective effects. It provides benefits like shock absorption, dispersion of traumatic energy, safeguarding the articular cartilage with a protective coating, and ensuring lubrication within the joint.¹¹ The initial biological basis for using HA in the OA Knee therapy stemmed from its capability to potentially increase viscosity of synovial fluid.¹² Numerous clinical studies have indicated prolonged positive outcomes in cases of OA Knee with the use of synthetic HA¹³, However, it's crucial to acknowledge that certain research has not shown benefits over a placebo.¹⁴

IA PRP injections have demonstrated efficacy in reducing pain and enhancing functionality in statistical evaluations.¹⁵ PRP stimulates revascularization of soft tissue and increases the concentration of growth factors at injury location. PRP is effective in repair of tendons, muscles and ligaments.¹⁶ In one study it has been observed that patients treated with IA PRP injections experienced greater improvements in knee function and pain relief, compared to those treated with a single corticosteroid injection.¹⁷ PRP treatments have shown reported improvements that can last for up to one year.¹⁸ IA PRP treatments do not exhibit the adverse effects on cartilage that IA corticosteroid usage may entail. Despite its effectiveness, IA PRP has not become a standard practice. The reason for this is primarily because there is a shortage of comprehensive research that compares its efficacy with the traditional approach of using IA corticosteroid. As a result, while the biological plausibility suggests potential benefits of PRP injections, its incorporation as a standard therapeutic option for OA Knee hinges upon further research that directly compares PRP with the existing standard treatments.

The objective of the study is to compare IA corticosteroid and autologous PRP injection in OA Knee in terms of reducing stiffness, alleviating

pain, and enhancing physical functionality.

METHODS

A randomized controlled trial was conducted at AFIRM Rawalpindi from Aug 2019 to April 2020. In our study, A sample size of 70 patients was calculated using WHO sample size calculator (power of study 80%, and level of significance 5%). Nonprobability consecutive sampling technique was employed. Patients with Knees OA, 40-70 years of age were included. Patients with septic/ inflammatory arthritis, overlying soft tissue infection, uncontrolled diabetes mellitus, BMI > 35.5, prior knee joint injury, advanced grade iv OA with deformity and malalignment, OA confirmed on radiology, total knee replacement, coagulation disorders, patients using warfarin and heparin were excluded from the study. Patients were divided in two groups, Group A received IA 40 mg Triamcinolone Acetonide plus 1 ml 1% Lignocaine Injection in the affected knee while Group B received PRP Injection in the affected knee. The PRP injection was repeated every after 2 weeks with a total of 3 sessions done per patient, 2 weeks apart.

Patients were directed to apply cold compresses on the joint every six hours for three consecutive days and refrain from placing stress on knee for the initial 24 hours and to consider using acetaminophen for pain relief if necessary.

NRS and WOMAC questionnaires were used. NRS contains Pain Score 0-10 on Numerical Rating Scale. WOMAC contains questions on the three sections of Rate Your Pain, rate your Stiffness and Rate your Difficulty, was used in this study. The results of each section were calculated from 0-96.

The pain was assessed on the NRS and WOMAC Index at baseline that is pre-injection, at 1 month, at 3rd month, and after 6 months. The mean change in pain scores on NRS and WOMAC was calculated at baseline, 1 month, 3 months and 6 months post-intervention.

The approval from hospital's ethical committee (1/2019) was obtained. Patient was sampled

by consecutive convenience sampling. Written consent was taken from all the patients and the patients' privacy and confidentiality was maintained.

Patients were selected from the outpatient department of AFIRM, RWP. Diagnosis of OA Knee was confirmed by history and x-ray findings. Patients with OA Knees of Grade II and Grade III on Kellgren and Lawrence method of classification of OA knee were followed. Fundamental demographic information such as age, gender, ethnicity, and socioeconomic background was documented. WOMAC score and NRS at presentation were calculated. Patients were subsequently assigned to one of two groups through a random selection process, utilizing a lottery method. In group A, IA injection of corticosteroid (triamcinolone+ lidocaine) was given and in group B, a course of three autologous PRP was injected at two weeks' interval. Patients of both groups were called for post-treatment follow up after 1 month, 3 months and 6 months from the time of administering the injections. WOMAC and NRS score of each patient was calculated at each follow-up visit and findings was documented.

RESULTS

There were 70 patients, 36 were male (52%) and 34 were female (48%). Mean age in group A was 58.54 ± 6.74 years and in group B was 56.40 ± 7.42 years. No notable disparities were observed in the ages between the groups ($p=0.210$), body mass index ($p=0.985$) and sex ($p=0.811$) distributions of the 2 groups (Table-I). Pain Severity assessed pre injection on NRS ($p=0.217$) and OA severity assessed on WOMAC pre injection ($p=0.010$) did not show any differences between the 2 groups (Table-II), while postinjection, the results have shown that there was significant improvement (p -value < 0.05) in mean pain relief in group B (IA PRP) compared to group A (IA steroid) at 6th month. However, NRS and WOMAC at 1 month has shown more improvement in Group A patients as compared to Group B patients with p value < 0.0001 .

Mean NRS Scale for pain of Group A at 1 month

was 1.20 ± 1.158 compared to 4.54 ± 2.99 of Group B (IA PRP) with p value < 0.0001 indicating marked improvement in Group A (IA steroid) as compared to Group B (IA PRP) at 1 month. Mean NRS Scale for pain of Group A (IA steroid) after 3 months was 6.14 ± 1.556 compared to 3.54 ± 2.318 of Group B (IA PRP) with p value < 0.0001 indicating improvement in Group B (IA PRP) as compared to Group A (IA steroid) at 3rd month. Mean NRS Scale for pain of Group A (IA steroid) after 6 months was 6.37 ± 1.832 compared to 3.63 ± 2.613 of Group B (IA PRP) with p value < 0.0001 indicating improvement in Group B (IA PRP) as compared to Group A (IA steroid) at 6th month. Mean WOMAC Score of Group A (IA steroid) at 1 month was 16.31 ± 8.210 compared to 28.00 ± 13.460 of Group B (IA PRP) with p value < 0.0001 indicating improvement in Group A (IA steroid) as compared to Group B (IA PRP) after 1 month. Mean WOMAC Score of Group A (IA steroid) after 3 months was 42.71 ± 13.028 compared to 24.51 ± 13.274 of Group B (IA PRP) with p value < 0.0001 indicating improvement in Group B (IA PRP) as compared to Group A (IA steroid) after 3 months. Mean WOMAC Score of Group A (IA steroid) after 6 months was 49.06 ± 15.076 compared to 23.03 ± 15.050 of Group B with p value < 0.0001 indicating improvement in Group B (IA PRP) as compared to Group A (IA steroid) after 3 months.

DISCUSSION

In our study with 70 OA Knee patients, both IA PRP and IA steroid injection groups had similar demographics, age, sex distribution, and body mass index. No significant differences in pain and OA severity were observed initially. Post-injection, the steroid group showed more pain relief at 1 month, while the PRP group exhibited significantly better outcomes in pain relief and OA severity at 3 and 6 months. Numeric Rating Scale (NRS) and WOMAC scores supported these findings, indicating sustained improvement with PRP over 3 to 6 months. Group B (IA PRP) displayed greater pain relief than Group A (IA steroid) at the 6-month mark. While Group A showed early improvement at 1 month, Group B demonstrated superior improvement at 3 and 6 months.

Descriptive Statistics					
	N	Minimum	Maximum	Mean	Std. Deviation
Age	70	45	69	57.47	7.122
BMI	70	24	34	30.70	2.261
PNRS	70	2	10	6.81	1.828
1NRS	70	0	16	2.87	2.813
3NRS	70	0	9	4.84	2.357
6NRS	70	0	9	5.00	2.632
PWOMAC	70	18	79	48.43	12.661
1WOMAC	70	4	52	22.16	12.535
3WOMAC	70	2	62	33.61	15.952
6WOMAC	70	3	75	36.04	19.885

Table-I. Descriptive statistics of patients

	Grp	Mean	Std. Deviation	P-Value
Age	Grp A	58.54	6.740	0.210
	Grp B	56.40	7.425	
BMI	Grp A	30.71	2.066	0.985
	Grp B	30.69	2.471	
PNRS	Grp A	7.09	1.358	0.217
	Grp B	6.54	2.187	
1NRS	Grp A	1.20	1.158	<0.0001
	Grp B	4.54	2.994	
3NRS	Grp A	6.14	1.556	<0.0001
	Grp B	3.54	2.318	
6NRS	Grp A	6.37	1.832	<0.0001
	Grp B	3.63	2.613	
PWOMAC	Grp A	52.26	12.809	0.010
	Grp B	44.60	11.449	
1WOMAC	Grp A	16.31	8.210	<0.0001
	Grp B	28.00	13.460	
3WOMAC	Grp A	42.71	13.028	<0.0001
	Grp B	24.51	13.274	
6WOMAC	Grp A	49.06	15.076	<0.0001
	Grp B	23.03	15.050	

Table-II. Mean, Std deviation, p values for Age, BMI, Pre-injection NRS and Post injection 1st, 3rd and 6th month NRS, Pre-injection WOMAC and Post Injection 1st, 3rd and 6th month WOMAC
*Independent sample T-test (P-value <0.05 is significant)

NRS and WOMAC assessments six months after injections indicated considerable PRP superiority over corticosteroid, despite corticosteroid showing more pain relief at 1 month. Corticosteroid effects diminished at 3 months compared to PRP.

There are several studies that compare the effects of IA corticosteroid with HA, and PRP with HA for OA treatment. In Cochrane reviews comparing IA corticosteroid with HA injections for OA Knee, no significant differences were observed at 4 weeks post-injection. However,

HA demonstrated greater effectiveness from 5 to 13 weeks post-injection in OA Knee patients.¹⁹ Similar outcomes were noted for other measures like reduced stiffness and improved function following IA HA administration.²⁰ Various studies have demonstrated the efficacy of visco-supplementation in providing pain relief, improving functional abilities, and receiving positive assessments from individuals suffering from OA Knee. Corticosteroid are mainly linked to pain relief in OA Knee but there's limited evidence showing they significantly improve function or

overall patient assessment.²⁴ The Cochrane review indicated that initially, for up to four weeks, there was no notable difference in the efficacy of IA corticosteroid and visco-supplements. Yet, in the period of 5 to 13 weeks following the injection, visco-supplements proved to be more beneficial than IA corticosteroid.²²

In a study by Kon et al.²³, the comparison revealed that PRP effectively improved symptoms in OA Knee treatment compared to both low and high-molecular-weight HAs. The results further demonstrated that PRP led to greater pain reduction and had longer-lasting effects compared to HA treatments.²³ In another study, comparing corticosteroid injections with PRP treatments, corticosteroid demonstrated swift symptomatic improvement with the most significant effect around 6 to 8 weeks, followed by symptom recurrence.²⁴ Conversely, PRP exhibited a slower but continuous improvement extending up to 24 to 104 weeks across three studies. One study highlighted that corticosteroid displayed faster symptom relief compared to PRP up to the three-month study endpoint. Additionally, at the six-week mark, one study found comparable therapeutic effects between corticosteroid and PRP.²⁴

Cerza et al. found that PRP provided better clinical outcomes than HA (HA) for up to 6 months, effectively reducing pain and symptoms while improving quality of life.²⁵ Cole et al.'s study revealed that PRP treatment led to better pain scores at 24 and 52 weeks, particularly in patients with mild OA and lower BMI.²⁶

Andrejs et al. found both PRP and corticosteroid (CS) treatments effective for short-term pain relief and knee function enhancement, observed up to 5 weeks with no notable score differences. However, at 15 weeks post-treatment, the PRP group showed significant improvements compared to the corticosteroid group. PRP-treated patients demonstrated better outcomes in longer-term follow-up, extending up to one year, compared to corticosteroid-treated individuals.²⁷ In a study involving 120 patients split into three groups, all experienced notable score improvements

(WOMAC, VAS) post-treatment compared to their pre-treatment values. The group that received IA-PRP showed significantly lower scores at 6-, 9-, and 12-months following treatment ($P < 0.05$).²⁸ The study by Gvendi et al. showed that PRP is a safe and effective treatment for controlling OA symptoms for up to six months after application. They found that the response to treatment with corticosteroid injections lasts for a shorter duration compared to PRP treatment.²⁹

CONCLUSION

PRP injections provided longer-lasting benefits compared to corticosteroid injections in alleviating joint pain, improving symptoms, and enhancing daily activities and quality of life over a six-month period. While corticosteroid injections showed promising results for the first month, their effects diminished thereafter. Overall, PRP demonstrated more sustained and lasting effects compared to corticosteroid.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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REFERENCES

1. Hunter DJ, Guermazi A, Roemer F, Zhang Y, Neogi T. **Structural correlates of pain in joints with OA.** *OA Cartilage.* 2013; 21:1170-78.
2. Patel S, Dhillon MS, Aggarwal S, Marwaha N, Jain A. **Treatment with PRP is more effective than placebo for OA Knee: A prospective, double-blind, randomized trial.** *Am J Sports Med.* 2013; 41:356-64.
3. Egemen A, Hayrettin K, Isik A. **IA injections (corticosteroid, HA, PRP) for the OA Knee.** *World J Orthop.* 2014 Jul 18; 5(3):351-61.
4. Neogi T. **The epidemiology and impact of pain in OA.** *OA Cartilage.* 2013; 21:1145-53.
5. **Recommendations for the medical management of OA of the hip and knee: 2000 update.** American College of Rheumatology Subcommittee on OA Guidelines. *Arthritis Rheum.* 2000; 43:1905-15.

6. Zhang W, Moskowitz RW, Nuki G, Abramson S, Altman RD, Arden N, et al. **OARSI recommendations for the management of hip and OA Knee, part II: OARSI evidence-based, expert consensus guidelines.** *OA Cartilage.* 2008; 16:137-62.
7. Jordan KM, Arden NK, Doherty M, Bannwarth B, Bijlsma JW, Dieppe P, et al. **EULAR recommendations 2003: an evidence-based approach to the management of OA Knee.** Report of a Task Force of the Standing Committee for International Clinical Studies Including Therapeutic Trials (ESCISIT). *Ann Rheum Dis.* 2003; 62:1145-55.
8. Bellamy N, Campbell J, Robinson V, Gee T, Bourne R, Wells G. **IA corticosteroid for treatment of OA of the knee.** *Cochrane Database Syst Rev.* 2006; 2:CD005328.
9. McAlindon TE, LaValley MP, Harvey WF, Price LL, Driban JB, Zhang M, et al. **Effect of IA triamcinolone vs saline on knee cartilage volume and pain in patients with OA Knee: A randomized clinical trial.** *JAMA.* 2017; 317(19):1967-75.
10. Juni P, Hari R, Rutjes AW, Fischer R, Silletta MG, Reichenbach S, et al. **IA corticosteroid for OA Knee.** *Cochrane Database Syst Rev.* 2015; 10(10):1-80.
11. Balazs EA, Denlinger JL. **Viscosupplementation: A new concept in the treatment of OA.** *J Rheumatol Suppl.* 1993; 39:3-9.
12. Moreland LW. **IA hyaluronan (HA) and hylans for the treatment of OA: mechanisms of action.** *Arthritis Res Ther.* 2003; 5:54-67.
13. Wobig M, Dickhut A, Maier R, Vetter G. **Viscosupplementation with hylan G-F 20: A 26-week controlled trial of efficacy and safety in the osteoarthritic knee.** *Clin Ther.* 1998; 20:410-23.
14. Henderson EB, Smith EC, Pegley F, Blake DR. **IA injections of 750 kD hyaluronan in the treatment of OA: a randomised single centre double-blind placebocontrolled trial of 91 patients demonstrating lack of efficacy.** *Ann Rheum Dis.* 1994; 53:529-34.
15. Dai WL, Zhou AG, Zhang H, Zhang J. **Efficacy of PRP in the treatment of OA Knee: A meta-analysis of randomized controlled trials.** *Arthroscopy.* 2017; 33(3):659-70 e1.
16. Gautam VK, Verma S, Batra S, Bhatnagar N, Arora S. **PRP versus corticosteroid injection for recalcitrant lateral epicondylitis: Clinical and ultrasonographic evaluation.** *J Orthop Surg.* 2015; 23(1):1-5.
17. Forogh B, Mianehsaz E, Shoaee S, Ahadi T, Raissi GR, Sajjadi S. **Effect of Single Injection of PRP in comparison with corticosteroid on OA Knee: A double blind randomized clinical trial.** *J Sports Med Phys Fitness.* 2016 Jul-Aug; 56 (7-8):901-8.
18. Dai WL, Zhou AG, Zhang H, Zhang J. **Efficacy of PRP in the treatment of OA knee: A meta-analysis of randomized controlled trials.** *Arthroscopy.* 2017; 33(3):659-70 e1.
19. Sundman EA, Cole BJ, Karas V, Della Valle C, Tetreault MW, Mohammed HO, et al. **The anti-inflammatory and matrix restorative mechanisms of plateletrich plasma in OA.** *Am J Sports Med.* 2014; 42(1):35-41.
20. Bannuru RR, Natov NS, Obadan IE, Price LL, Schmid CH, McAlindon TE. **Therapeutic trajectory of HA versus corticosteroid in the treatment of OA Knee: A systematic review and meta-analysis.** *Arthritis Rheum.* 2009; 61:1704-11.
21. Housman L, Arden N, Schnitzer TJ, Birbara C, Conrozier T, Skrepnik N, et al. **IA hylastan versus steroid for OA Knee.** *Knee Surg Sports Traumatol Arthrosc.* 2014 Jul; 22(7):1684-92.
22. Bellamy N, Campbell J, Robinson V, Gee T, Bourne R, Wells G. **IA corticosteroid for treatment of OA of the knee.** *Cochrane Database Syst Rev.* 2006; (2):CD005328.
23. Kon E, Filardo G, Martino AD, Maracacci M. **PRP to treat sports injuries: Evidence to support its use.** *Knee Surg Sports Traumatol Arthrosc.* 2011; 19:516-527.
24. Ben-Nafa W, Munro W. **The effect of corticosteroid versus PRP injection therapies for the management of lateral epicondylitis: A systematic review.** *SICOT J.* 2018; 4:11.
25. Cerza F, Carnì S, Carcangiu A, Di Vavo I, Schiavilla V, Pecora A, et al. **Comparison between HA and PRP, IA infiltration in the treatment of gonarthrosis.** *Am J Sports Med.* 2012 Dec; 40(12):2822-7.
26. Cole BJ, Karas V, Hussey K, Pilz K, Fortier LA. **HA versus PRP: A prospective, double-blind randomized controlled trial comparing clinical outcomes and effects on IA biology for the treatment of OA knee.** *Am J Sports Med.* 2017 Feb; 45(2):339-46.
27. Elksniņš-Finogejevs A, Vidal L, Peredistijs A. **IA PRP vs corticosteroid in the treatment of moderate OA Knee: A single-center prospective randomized controlled study with a 1-year follow up.** *J Orthop Surg Res.* 2020 Jul 10; 15(1):257.

28. Huang Y, Liu X, Xu X, Liu J. **IA injections of PRP, HA or corticosteroid for OA Knee: A prospective randomized controlled study.** Orthopade. 2019 Mar; 48(3):239-47. English.
29. Uslu Gündendi E, Aşkin A, Gündendi G, Koçyiğit H. **Comparison of efficiency between corticosteroid and PRP injection therapies in patients with OA knee.** Arch Rheumatol. 2017 Nov 2; 33(3):273-81.

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2	Sahibzada Nasir Mansoor	Conception, Study design, Drafting the manuscript and approval of the final version to be published.	
3	Shaista Kanwal	Data acquisition, data analysis and interpretation, critica review, approval of the final version to be published.	
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