Systematic Review

Effectiveness of platelet-rich plasma in partial-thickness rotator cuff tears: a systematic review

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ABSTRACT

Importance: Partial-thickness rotator cuff tears (PTRCTs) commonly affect overhead athletes, leading to a decline in sports performance. Platelet-rich plasma (PRP) is being explored as an alternative treatment modality for individuals with PTRCTs to reduce discomfort and enhance functional recovery. We conducted a systematic review study of randomized controlled clinical trials to determine the effectiveness of PRP in treating PTRCTs.

Aim: To determine the effectiveness of PRP in treating PTRCTs.

Evidence review: We conducted a comprehensive literature search for randomized controlled trials (RCTs) that compared the effectiveness of PRP with eccentric exercise and placebo injections as treatments for PTRCTs. We searched databases such as the Cochrane Library, Web of Science, PubMed, and EMBASE. The visual analog scale (VAS) score, American Shoulder and Elbow Surgeon (ASES) score, and Constant–Murley Score (CMS) was utilized as an outcome measure. Statistical analysis was performed using RevMan 5.3 software.

Findings: Our meta-analysis included 12 studies involving 762 patients. At six weeks post-treatment, the PRP group had significantly higher VAS scores compared to the control group, indicating improvement (standard mean difference (SMD): -2.04 [95% confidence interval (CI): -4.00 to -0.08], I² = 97%, P-value = 0.04).

Patients who received PRP showed statistically significant improvements in VAS scores at 3 months and 6 months follow-up (SMD, -1.78 [95% CI: -3.03 to -0.52], I² = 96%, P-value = 0.005) (SMD: -2.26 [95% CI: -3.77 to -0.76], I² = 97%, P-value = 0.003). A statistically significant difference was also observed in VAS scores at the long-term 1-year follow-up (SMD: -2.27 [95% CI: -4.07 to -0.47]; I² = 98%; P-value = 0.031). There were statistically significant differences in ASES scores and CMS scores in the short-term (SMD: 2.01 [95% CI: 0.14 to 3.88], I² = 97%, P-value = 0.04). However, in the long-term ASES and CMS scores did not show any statistical significance (SMD: 2.06 [95% CI: -0.54 to 4.65], I² = 99%, P-value = 0.12) (SMD: 4.36 [95% CI: -5.48 to 14.21], I² = 99%, P-value = 0.39).

Conclusions and relevance: Our findings suggest that PRP treatment is effective in reducing pain for individuals with PTRCTs, providing benefits in the short and long term. However, its impact on functional recovery appears somewhat constrained and doesn’t endure over time. Additionally, significant heterogeneity exists among studies, encompassing variations in PRP composition and control group treatments. Consequently, we conclude that compelling evidence for symptom improvement in PTRCT patients following PRP treatment remains elusive.

Level of evidence: Level I.
What is already known:

- Partial-thickness rotator cuff tears commonly affect overhead athletes, including handball players and pitchers, resulting in a decline in sports performance.
- Partial-thickness rotator cuff tears are a leading cause of shoulder pain and disability.
- Conservative treatments for Partial-thickness rotator cuff tears include sodium hyaluronate, corticosteroids, lidocaine, and physiotherapy.
- Surgical treatment carries risks, such as infections, delayed healing, shoulder stiffness, and tendon injuries.

What are the new findings:

- The findings of our study demonstrated that Platelet-rich Plasma treatment resulted in a statistically significant reduction in pain and improvement in function, as measured by the Constant–Murley score and American Shoulder and Elbow Surgeon score, compared to the control groups during the short-term follow-up period.

INTRODUCTION

Partial-thickness rotator cuff tears (PTRCTs) commonly affect overhead athletes [1], including handball players [2] and pitchers [3], resulting in a decline in performance. PTRCTs are a leading cause of shoulder pain and disability [4-6]. While PTRCTs may initially exhibit minimal symptoms [7,8], associated conditions such as scapula instability, dyskinesia, or tightness can contribute to pain and muscle weakness [9]. Surgical treatment carries risks, such as infections, delayed healing, shoulder stiffness, and tendon injuries [10]. Conservative treatments for PTRCTs include sodium hyaluronate, corticosteroids, lidocaine, and physiotherapy [11,12]. Given the limited healing capacity of the tendon, platelet-rich plasma (PRP) has gained interest as a potential treatment modality [13].

PRP's regenerative properties can be attributed to the growth factor it contains [14]. Platelet-derived growth factors stimulate cell proliferation and tissue regeneration, while transforming growth factor β-1 plays a crucial role in modulating the inflammatory response and promoting collagen synthesis [15,16]. Hepatocyte growth factor aids in tissue regeneration and angiogenesis insulin-like growth factor promotes cell growth and differentiation, and vascular endothelial growth factor stimulates the formation of new blood vessels, facilitating tissue repair [17]. Despite the potential benefits of PRP, its clinical effects can vary due to several factors. The composition of PRP can differ depending on individual patients and the specific preparation method employed. This can lead to variations in the concentration of growth factors and other bioactive substances, which may influence the treatment outcomes. Additionally, the inconsistent parameters used in PRP preparation, such as the choice of preparation kit can further contribute to the variability in clinical effects. Another factor that can impact the effectiveness of PRP is post-injection rehabilitation. The appropriate rehabilitation protocols following PRP injection play a crucial role in optimizing outcomes. The specific exercises, physical therapy modalities, and activity restrictions during the healing process can vary among patients and may affect the overall success of the treatment. The lack of standardized protocols for PRP preparation and administration hinders the ability to compare and replicate studies accurately. Moreover, the clinical evidence supporting the use of PRP is still evolving, and the optimal treatment protocols for specific musculoskeletal conditions are yet to be fully determined.

In recent years, several studies have focused on investigating the therapeutic effects of PRP injections for rotator cuff tears. A systematic review conducted on rotator cuff disease indicated that PRP injections may not provide significant benefits in the short-term follow-up period [18,19]. Furthermore, the latest guideline released by the American Academy of Orthopaedic Surgeons (AAOS) [20] does not recommend the routine use of PRP in the nonoperative management of PTRCTs due to limited evidence and its quality. However, it is worth noting that, despite the overall findings, several trials have reported positive effects associated with PRP injections for PTRCTs [21,22]. These studies have observed beneficial outcomes in terms of pain reduction, improved shoulder function, and enhanced healing. It is essential to consider the varying results and recommendations from different studies and guidelines when assessing the potential benefits of PRP injections for rotator cuff disease. Further research is necessary to establish a more comprehensive understanding of PRP's effectiveness, optimal administration protocols, and patient selection criteria in order to provide more definitive guidance on its usage in the management of rotator cuff tears. We have conducted a meta-analysis study of randomized controlled clinical trials aiming at the effectiveness of PRP in PTRCTs.

METHODS

Search strategy for the identification of studies

This study was performed in accordance with the 2020 Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines and the PRISMA of Individual Patient Data (PRISMA-IPD) statement [23]. Using PubMed, MEDLINE, and the Cochrane Library, a comprehensive search of the literature was carried out, with the last search in February 2023 for “PRP or platelet-rich plasma, partial rotator cuff tears and rotator cuff” filtered by “human” and “randomized controlled trial.”

Study selection and inclusion criteria

Full-length English-language articles that reported clinical outcomes were screened for inclusion.

The inclusion criteria were as follows: (1) patients diagnosed with PTRCs by MRI or ultrasound over 2 months; (2) patients aged 18 years or older; (3) patients in the intervention group received PRP; (4) quantifiable outcomes were reported; (5) RCT design. Exclusion criteria were as follows: (1) patients diagnosed with full-thickness rotator cuff tears, rheumatoid arthritis, adhesive capsulitis, and calcific rotator disease; (2) patients had a history of rotator cuff repair or injection; (3) case reports, letters, comments, trial protocols, editorials, reviews, or practice guidelines; (4) studies not written in English.

Data extraction and analysis

Outcome measures: The primary outcome was pain, assessed by visual analog scale (VAS) score; the secondary outcome was a function that consisted of the Constant–Murley score (CMS) and American Shoulder and Elbow Surgeons score (ASES). Two independent authors extracted data with standardized data forms. The following data were extracted: first author's name, publication year, type of study, level of evidence, type of intervention, mean age, number of patients, outcome measurements, details of PRP treatment, and duration of follow-up. The means and standard deviations (SD) of continuous outcomes were recorded. The VAS score (reported on 0–100 scales) was converted into 0–10 scales. The results of the VAS score, CMS, and ASES were extracted and categorized as follows: baseline, short-term (≤6 months) follow-up, and long-term (>1 year) follow-up. The data of the diagram was extracted by Engauge Digitizer (version 3.0) if no original data were available in the study. If SDs were missing for continuous data, other statistics (for example, 95% confidence interval, standard errors, T values, F values,
and P values) were used for the calculation of the standard deviation via the calculator tool from Review Manager, version 5.3 (Nordic Cochrane Centre, Cochrane Collaboration). Disagreements between the two reviewers were resolved by consensus and, if necessary, by consultation with a third reviewer. Data analysis was performed with Review Manager 5.3. A P-value < 0.05 and quantified with I^2 (significance level at I^2 > 50%) [23,24]. Random effects were used if the Q or I^2 value was statistically significant or a small number of studies were analyzed. The subgroup analysis was reported based on variables of interest, like the number of PRP injections.

Assessment of risk of bias

To assess bias with the Cochrane collaboration’s risk of bias tool [12, 25], two reviewers independently assessed each of the following domains: selection bias, performance bias, detection bias, attrition bias, reporting bias, and other sources of bias. Each component was recorded as low, unclear, or high risk of bias. Studies were considered to have a low risk of bias only when a “+” was recorded for all items; a high risk of bias was considered if studies scored a “−”. Others were scored as having a moderate risk of bias [17,26]. Any disagreements were resolved through discussion or by referral to a third author.

RESULTS

Selection of study

Our initial search yielded a total of 241 articles. Following the screening and application of inclusion and exclusion criteria, we included 12 articles with 762 patients with PTRCTs in our meta-analysis (Fig. 1). Table 1 details the general information included in the study.

PRP treatment characteristics varied widely among the included studies, though all injections were under ultrasound guidance. For example, PRP parameters included leukocyte poor in four studies [27–30], single injection in five studies [28,30–33] and multiple injections in seven studies [27,29,34–38]. Included study characteristics are provided (Table 1).

Risk of bias in included studies

The results of the risk of bias assessment are presented (Fig. 2). Among the 12 studies, six studies were deemed to be at high risk of bias [27,32,34,35,37,38], and six studies were deemed to be at low risk of bias [28–31,33,36]. Selection bias was the most common source of bias across the included studies.
<table>
<thead>
<tr>
<th>Sr. no</th>
<th>First author</th>
<th>Year</th>
<th>Study/level of evidence</th>
<th>Overview</th>
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<td>PTRCTs</td>
<td>PRP vs injection</td>
<td>Lidocaine</td>
<td>VAS, ASES, WORC</td>
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<td>Dry Needling</td>
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<td>Ozlem et al. [35]</td>
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<td>Cai et al. [27]</td>
<td>2019</td>
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<td>Schweitzgabel et al. [29]</td>
<td>2019</td>
<td>Randomized control trial/Level 1</td>
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<td>PRP vs physiotherapy</td>
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<td>Thepsoparn M et al. [30]</td>
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<td>PTRCTs</td>
<td>PRP vs injection</td>
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**Abbreviations:** PRP = platelet-rich plasma; PTRCTs = partial thickness rotator cuff tears; NR = not reported; LP = leukocyte poor; LR = leukocyte rich; VAS = visual analog scale; ASES = American Shoulder and Elbow Surgeons score; CMS = Constant-Murley score; DASH = disabilities of the arm, shoulder and hand; SPADI = shoulder pain and disability index; SST = simple shoulder test; WORC = Western Ontario rotator cuff index; SDQ = shoulder disability questionnaire; SANE = single assessment numerical evaluation; ROM = range of motion; OSS = Oxford shoulder score.
The results of the meta-analysis of 12 studies are reported as follows.

Main outcome indicators, visual analog scale score

The VAS score was assessed in our meta-analysis at 6 weeks, 3 months, 6 months, and 1 year. Four studies used VAS scores at 6 weeks, six studies used VAS scores at 3 months, seven studies used VAS scores at 6 months, and five studies used VAS scores at 1 year following treatment. The results were sufficiently dissimilar that a combined analysis was done using a random-effect model. The results were shown on a forest plot diagram in terms of standard mean difference (SMD), 95 % confidence interval (CI), Z score and $I^2$ (Figs. 3–6). At six weeks post-treatment, the PRP group had significantly higher VAS scores compared to the control group, suggesting an improvement in pain relief associated with PRP treatment (SMD, $-2.04$ [95 % CI: $-4.00$ to $-0.08$], $I^2 = 97\%$, $P$-value = 0.04). However, the high $I^2$ value (97%) indicates a substantial degree of variability across studies, and the interpretation of the findings should consider this heterogeneity. Patients who received PRP showed statistically significant improvements in VAS scores at 3 months and 6 months follow-up (SMD, $-1.78$ [95 % CI: $-3.03$ to $-0.52$], $I^2 = 96\%$, $P$-value = 0.005) (SMD, $-2.26$ [95 % CI: $-3.77$ to $-0.76$], $I^2 = 97\%$, $P$-value = 0.003). A statistically significant difference was also observed in VAS scores at the long-term 1-year follow-up (SMD, $-2.27$ [95 % CI: $-4.07$ to $-0.47$]; $I^2 = 98\%$, $P$-value = 0.031).

Function

Patients who received PRP showed statistically significant differences in ASES and CMS in the short-term (SMD, 1.21 [95 % CI: 0.19 to 2.24]; $I^2 = 96\%$, $P$-value = 0.02) (SMD, 2.01 [95 % CI: 0.14 to 3.88]; $I^2 = 97\%$, $P$-value = 0.04). However, in the long-term, ASES and CMS were not statistically significantly different (SMD, 2.06 [95 % CI: $-0.54$ to 4.65]; $I^2 = 99\%$, $P$-value = 0.12) (SMD, 4.36 [95 % CI: $-5.48$ to 14.21]; $I^2 = 99\%$, $P$-value = 0.39) (Figs. 7–10). According to the forest plot, the control group had a better outcome.

**Fig. 2.** Cochrane risk of bias assessment.
Fig. 3. Forest plot for VAS score between PRP and placebo at 6 weeks after treatment. VAS = visual analog scale; PRP = platelet-rich plasma.

Fig. 4. Forest plot for VAS score between PRP and placebo at 3 months after treatment. VAS = visual analog scale; PRP = platelet-rich plasma.

Fig. 5. Forest plot for VAS score between PRP and placebo at 6 months after treatment. VAS = visual analog scale; PRP = platelet-rich plasma.

Fig. 6. Forest plot for VAS score between PRP and placebo at 1 year after treatment. VAS = visual analog scale; PRP = platelet-rich plasma.

Fig. 7. Forest plot for ASES score between PRP and placebo at short term follow-up (<6 months) after treatment. ASES = American Shoulder and Elbow Surgeons score; PRP = platelet-rich plasma.

Fig. 8. Forest plot for ASES score between PRP and placebo at long term follow-up (1 year) after treatment. ASES = American Shoulder and Elbow Surgeons score; PRP = platelet-rich plasma.
Subgroup analysis

The factor selected for subgroup analysis among the VAS scores and CMS was the number of PRP injections. The forest plots of subgroup analysis are shown in Figs. 11–15. The results of long-term CMS with single injections as well as multiple injections were not pooled due to insufficient data. For the number of PRP injections subgroup, patients who received the single injection had a statistically significant difference in the short-term VAS score (SMD, −0.48 [95% CI: −1.01 to 0.05], I² = 66%, P-value = 0.08), which was similar to patients who received multiple injections (SMD, −2.36 [95% CI: −4.45 to −0.27], I² = 98%, P-value = 0.03). There was a statistically significant difference in the multiple PRP injections subgroup for long-term VAS scores (SMD, −2.91 [95% CI: −5.54 to −0.27], I² = 98%, P-value = 0.03). There was no statistically significant difference in CMS in patients who received PRP with single injection in the short-term (P-value = 0.41) as well as multiple injection subgroup in the short-term (P-value = 0.15).

DISCUSSION

Our meta-analysis aimed to assess the effectiveness of PRP injections in the treatment of PTRCTs. The findings of our study demonstrated that PRP treatment resulted in a statistically significant reduction in pain compared to the control groups during the short-term follow-up period. However, no statistically significant long-term improvements were observed in terms of function. Our results differ from previous systematic reviews [15,17] that suggested PRP injections may not provide short-term benefits as a nonoperative treatment for rotator cuff tears. It is...
important to note that the evidence supporting these previous reviews was limited. In contrast, our study's findings align with a study conducted by Chen et al. [39], which reported statistically significant improvements in functional outcomes among patients with rotator cuff tears who received PRP treatment compared to control groups. It is worth mentioning that most of the controls in the aforementioned study underwent rotator cuff repair, indicating that PRP therapy was not inferior to surgical intervention.

The findings regarding pain reduction are consistent with previous studies that have suggested the potential benefits of PRP in musculoskeletal conditions, including tendinopathy and joint degeneration [40, 41]. The high concentration of growth factors in PRP promotes tissue repair and healing, which could explain the observed pain reduction in PTRCTs. However, it is worth noting that the meta-analysis showed a significant heterogeneity among the included studies, which may be attributed to the variation in PRP treatment characteristics, such as leukocyte content and the number of injections.

In terms of functional outcomes, the meta-analysis revealed no significant improvements in ASES and CMS scores. This finding suggests that PRP injections may have a limited effect on functional recovery in PTRCTs. However, it is important to consider the limitations of the included studies, such as small sample sizes and the variation in follow-up durations. Further research with larger sample sizes and a longer follow-up period is needed to better understand the long-term effects of PRP on functional outcomes.

The assessment of the risk of bias in the included studies revealed that most studies were at a high risk of bias, particularly regarding selection bias. This highlights the need for well-designed RCTs with rigorous methodology to minimize bias and improve the overall quality of evidence.

Despite the positive findings regarding pain reduction and short-term functional improvements, it is important to interpret the results of this meta-analysis with caution. The clinical significance of the observed outcomes should be considered, along with the potential limitations of PRP treatment, such as its high cost and the lack of standardized protocols for PRP preparation and administration. Additionally, it is crucial to consider individual patient factors, such as the severity of the PTRCT, comorbidities, and patient preferences, when making treatment decisions.

We had well-defined inclusion and exclusion criteria for the study, and hence, only randomized controlled trials (RCTs) with clinical evidence of effectiveness were included in this meta-analysis.

This study has several limitations that should be acknowledged. The primary limitation is the presence of heterogeneity among the original studies included in our analysis. Specifically, there was inconsistency in the preparation of PRP across the RCTs included in our study. Moreover, important details regarding the specific kits used, platelet concentration, and leukocyte concentration were often not adequately reported, which hindered the possibility of conducting further subgroup analyses based on these parameters. Hence, it was difficult to classify PRP composition according to the universal coding system by Kon et al. [42]. Another significant limitation is the potential confounding effect of subsequent rehabilitation. The postinjection rehabilitation protocols varied among the included studies, and this variability could have introduced a cointervention effect that may have influenced the results of the PRP intervention. This cointervention effect has the potential to overestimate the observed effects of PRP treatment.

There is a need for future RCTs, to reduce heterogeneity in the existing research literature. Future studies should focus on addressing the heterogeneity in PRP treatment protocols, optimizing patient selection criteria, and conducting long-term follow-up assessments to evaluate the sustained effects of PRP treatment. Also, future studies should be carried out following the universal coding system by Kon et al. [42] for PRP composition.

In most studies we included, there is a lack of detailed information regarding the specific characteristics of rotator cuff tears, such as lesion sites (articular-sided, bursal-sided, or intra-tendinous tears) or classification. This missing information may have contributed to the observed heterogeneity across the included studies. Moreover, there was considerable variability in the shoulder functional assessments used across the studies, including measures such as the disabilities of the arm, shoulder and hand (DASH), CMS, and Western Ontario rotator cuff index, Single Assessment Numeric Evaluation, Shoulder Pain and Disability Index (SPADI), ASES, and quick DASH. This variation in assessment tools posed challenges in pooling the data for this meta-analysis, as it limited the ability to directly compare and combine the results across studies. Also, there was inconsistency in the interventions used in the control groups across the included studies. The control interventions encompassed a
range of approaches, including exercise, physical therapy, dry needling, and other types of injections. This variability in the control group interventions add complexity and may introduce additional sources of heterogeneity.

**CONCLUSION**

Our findings suggest that PRP treatment is effective in reducing pain for individuals with PTRCTs, providing benefits in the short term, and long term. However, the effects on functional recovery may be more limited and not sustained over the long term. Additionally, significant heterogeneity exists among studies, encompassing variations in PRP composition and control group treatments. Consequently, we conclude that compelling evidence for symptom improvement in PTRCT patients following PRP treatment remains elusive.

**Ethical approval**

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**Author’s contribution**

All authors have contributed equally to the preparation of the manuscript.

**Informed consent**

Not Applicable.

**Availability of data and materials**

This published article contains all the data generated or analyzed during this study.

**Declaration of competing interest**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

**References**


