Platelet Concentration Explains Variability in Outcomes of Platelet-Rich Plasma for Lateral Epicondylitis: A High Dose Is Critical for a Positive Response

A Systematic Review and Meta-analysis With Meta-regression

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Background: Randomized controlled trials (RCTs) evaluating the efficacy of platelet-rich plasma (PRP) for the management of lateral epicondylitis (LE) have been characterized by substantial variability in reported outcomes. The source of this heterogeneity is uncertain.

Purpose: To determine the effect of estimated platelet concentration on the efficacy of PRP for the management of LE.

Study Design: Systematic review and meta-analysis; Level of evidence, 2.

Methods: All RCTs evaluating the efficacy of PRP in managing LE were identified. RCTs were classified according to whether the study documented a platelet concentration factor of PRP representing a greater than 3-fold increase over whole blood or a supraphysiological platelet dose (high-dose vs low-dose PRP). The primary outcome was the mean difference (MD) in the visual analog scale (VAS) score at latest follow-up. Random-effects and mixed-effects meta-analyses were performed, and meta-regression was used to evaluate whether differences in outcomes after treatment with PRP could be explained by differences in the concentration of PRP used.

Results: Overall, 13 RCTs with a total of 791 patients were included in this analysis, with 5 that utilized low-dose PRP and 8 that used high-dose PRP. Meta-analysis of VAS scores reported by studies that used high-dose PRP resulted in an MD of −1.31 (95% CI, −1.87 to −0.75) in favor of PRP over all alternative treatment strategies ($P < .001$). Meta-analysis of VAS scores reported by studies that used low-dose PRP resulted in an MD of 0.08 (95% CI, −0.51 to 0.68), suggesting no difference in the effect between PRP and all alternative treatment strategies ($P = .79$). The platelet concentration factor of PRP used in each RCT was found to be strongly predictive of the VAS score at final follow-up in meta-regression ($P < .001$), with 58.5% of the heterogeneity in the outcomes of PRP between studies explained by the platelet concentration factor alone.

Conclusion: The platelet concentration of PRP may play a significant role in the outcomes of patients with LE. A direct linear relationship was observed between the platelet concentration factor of PRP used and the magnitude of patient-reported symptom relief after the management of LE with PRP. Clinicians should ensure a supraphysiological platelet concentration when preparing PRP for the management of LE.

Keywords: platelet-rich plasma; PRP; lateral epicondylitis; tennis elbow; dose; concentration

Lateral epicondylitis (LE), or tennis elbow, is a common cause of elbow pain in adults aged 35 to 55 years, affecting up to 3% of adults and causing characteristic pain and tenderness over the lateral epicondyle.[34] While several causes have been proposed, the underlying pathophysiology is thought to be related to cumulative microscopic tendon injuries, most commonly of the extensor carpi radialis brevis tendon. Although extensive infiltrates of classic inflammatory/immune cells are not typically seen in tissue biopsy specimens from patients with LE, it is well established that there is "molecular inflammation" in tendinopathy, with increased gene expression for a number of inflammatory cytokines.[5,30] Tissue damage promotes the release of

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nociceptive signaling molecules including substance P and calcitonin gene-related peptide, eliciting potentially severe pain.[21] Although roughly 80% of LE cases self-resolve within 10 to 18 months, a considerable number of patients experience persistent symptoms, ultimately resulting in muscle wasting and decreased grip strength.[3,36] Additionally, LE is a significant economic burden, accounting for 11.7% of work-related injury claims.[31] Thus, finding a consistent therapy for reducing LE-associated morbidity is an important unmet clinical need.

While up to 25% of patients with LE may be surgical candidates, only 4% to 11% of patients seeking medical help will undergo surgery,[3] with the majority of LE cases treated with nonoperative therapies. In addition to rest, physical therapy, bracing, and nonsteroidal anti-inflammatory drugs, injection therapies including botulinum toxin, corticosteroids, autologous whole blood, and platelet-rich plasma (PRP) have also been utilized.[1] PRP in particular has become an attractive option for treating the tendinopathic processes of LE, as early randomized controlled trials (RCTs) demonstrated improved efficacy over alternative therapies.[2,12,13,15,18,28,29,33] However, more recent RCTs have been unable to replicate these initial benefits,[14,20] leading to a high degree of heterogeneity in previous meta-analyses[8,11,25] and the inability to draw meaningful conclusions as to whether PRP is a reliable and effective management option for LE.

Platelet concentrations in PRP can be highly variable, and very few studies in the current literature have measured and reported platelet concentrations.[4] While it has been theorized that higher platelet counts are associated with improved outcomes clinically, this has not been systematically studied or determined, resulting in a wide range of dosages utilized in RCTs evaluating PRP for LE. It is possible that the variability in platelet concentrations used by RCTs may contribute to the heterogeneity in outcomes associated with the application of PRP for LE. Thus, the purpose of this study was to determine the effect of the platelet concentration on the efficacy of PRP for the management of LE. We hypothesized that there would be substantial heterogeneity in the platelet concentrations of PRP used in RCTs evaluating the effect of PRP on LE and that greater clinical efficacy would be associated with higher platelet concentration factors.

METHODS

Search Strategy

This study was conducted in accordance with the PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) guidelines. In June 2023, the PubMed, MEDLINE/Ovid, and Cochrane databases were systematically searched to identify all studies evaluating PRP for the management of LE. The search contained the following parameters: (elbow tendon injury OR elbow tendinopathy OR elbow tendonitis OR tennis elbow OR lateral epicondylitis OR lateral epicondylosis OR epicondylitis) AND (platelet-rich plasma OR prp). Inclusion criteria included RCTs that consisted of a full report of outcomes, the use of statistical methods (including mean and standard deviation values), a comparison of PRP with a control (ie, hyaluronic acid, corticosteroid, placebo), patient-reported outcome measures, and reporting on the platelet concentration of PRP utilized. Only titles and abstracts published in English were reviewed. Exclusion criteria included abstracts, technique articles, cadaveric or animal studies, review articles, letters to the editor, and studies that did not focus on PRP for the treatment of LE.

Article Review and Data Abstraction

There were 2 independent reviewers (J.F.O., C.J.M.) who screened titles and abstracts and manually reviewed reference lists for additional studies not identified in the primary search. A total of 86 full-text articles were reviewed, with 13 RCTs meeting inclusion and exclusion criteria (Figure 1).

From each study, data were extracted by 2 authors (J.F.O., C.J.M.) using a standardized form that included the sample size, mean values of primary outcomes, number of patients lost to follow-up, PRP formulation (leukocyte-poor PRP, leukocyte-rich PRP, or not specified), and platelet concentration. Given that so few studies reported data on the actual platelet concentration, the platelet concentration factor was defined as the volume of PRP obtained divided by the initial volume of blood collected. The study quality and risk of bias were assessed via the Cochrane risk-of-bias tool.[16]

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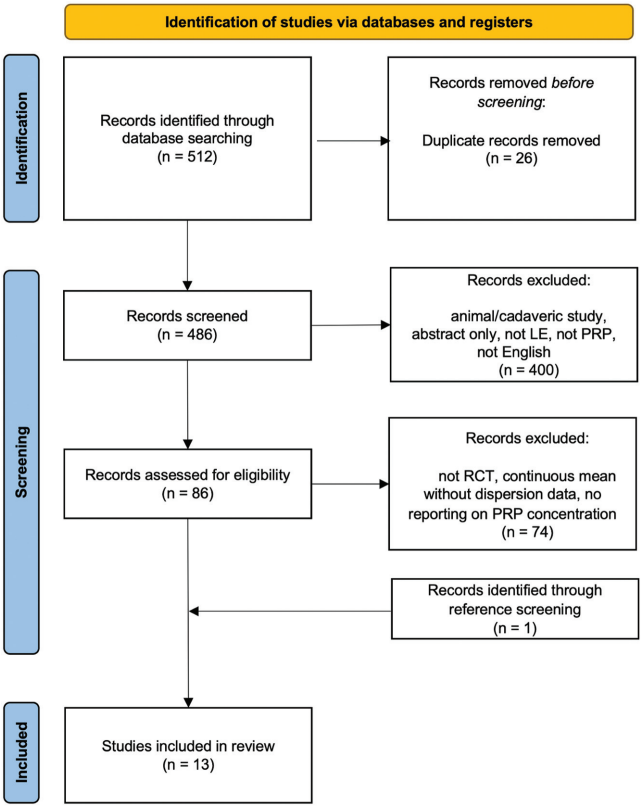

Figure 1. PRISMA flowchart of study inclusion. LE, lateral epicondylitis; PRP, platelet-rich plasma; RCT, randomized controlled trial.

Definitions of High-Dose PRP and Low-Dose PRP

Currently, PRP is defined as a plasma volume with a platelet concentration above that of baseline whole blood, which is 150,000/μL to 450,000/μL.[9,10,35] Thus, high-dose PRP (ie, "true" PRP) was defined as a platelet concentration factor representing a greater than 3-fold increase over whole blood (such that even patients with platelet concentrations at the lowest end of the normal range achieved a supraphysiological concentration) or that with a documented supraphysiological platelet dose. PRP that did not meet this criterion was considered low-dose PRP.

Statistical Analysis

While all reported outcome scores were collected, the only outcome reported by a sufficient number of studies to conduct meta-analyses was the visual analog scale (VAS) score at final follow-up. The summary measure was the mean difference (MD) between PRP and the control. Inverse variance random-effects meta-analysis using the DerSimonian and Laird method[7] to estimate between-study variances and to determine MDs with 95% confidence intervals (CIs) was conducted to compare high-dose PRP (ie, "true" PRP) and low-dose PRP. In addition to a single meta-analysis including all studies, regardless of the PRP composition, an independent meta-analysis was conducted for each type of PRP (high-dose and low-dose). Next, mixed-effects models with meta-regression were generated with the reported platelet concentration factor, a binary PRP characterization variable (high-dose vs low-dose), and the control to which PRP was compared, with all explored as potential covariates that could explain the heterogeneity in, and may be associated with, outcomes reported after the treatment of LE with PRP. Assessments of heterogeneity were performed independently using the Higgins and Thompson I^2 statistic,[17] the DerSimonian and Laird τ^2 estimator,[7] and the Cochran Q test of heterogeneity. All analyses were performed in R (Version 4.2.2) and Excel (Microsoft).

RESULTS

Study Characteristics

Overall, 13 RCTs with a total of 791 patients (mean age, 42.4 years) were ultimately included in this analysis, with 5 that utilized low-dose PRP and 8 that used high-dose PRP (Table 1).

Results of Meta-analysis

The forest plot for each random-effects meta-analysis is provided in Figure 2. An MD in the VAS score of −0.72 (95% CI, −1.26 to −0.18) in favor of PRP over all alternative treatment strategies was determined when evaluating the pooled treatment effect of all RCTs, irrespective of the PRP composition (P = .009) (Figure 2A). Meta-analysis of VAS scores reported by studies that used high-dose PRP resulted in an MD of −1.31 (95% CI, −1.87 to −0.75) in favor of PRP over all alternative treatment strategies (P < .001) (Figure 2B). Meta-analysis of VAS scores reported by studies that used low-dose PRP resulted in an MD of 0.08 (95% CI, −0.51 to 0.68), suggesting no difference in the effect between PRP and all alternative treatment strategies (P = .79) (Figure 2C).

Results of Meta-regression

In meta-regression, the platelet concentration factor was found to be significantly associated with the VAS score at final follow-up (−0.21 [95% CI, −0.90 to −0.32] relative improvement on the VAS per point increase in the platelet concentration factor; P < .001), with 58.5% of the heterogeneity in outcomes between RCTs explained by the platelet concentration factor alone (Figure 3). These results became even more pronounced in multivariable meta-regression when also adjusting for the comparative treatment (−0.26 [95% CI, −0.38 to −0.14] relative improvement on the VAS per point increase in the platelet concentration factor; P < .001), with 70.4% of the heterogeneity explained. Similarly, when adjusting for a binary variable corresponding to whether each study utilized high-dose PRP (ie, "true" PRP) or low-dose PRP, the use of high-

TABLE 1
Study Characteristics[a]

First Author (Year)	No. of Patients	Mean Age, y	PRP Type	Level of Evidence	Risk of Bias	Power Analysis	Platelet Concentration Factor
PRP vs corticosteroid							
Kamble[18] (2023)	64	40	NR	1	Low	NR	10.0
Gungor[14] (2022)	48	43.6	LP-PRP	2	Unclear	A priori	2.0
Gupta[15] (2020)	80	40.8	NR	1	High	A priori	5.0
Gautam[12] (2015)	30	NR	NR	2	Unclear	NR	10.0
Omar[26] (2012)	30	39	NR	2	Unclear	NR	2.0
Gosens[13] (2011)	100	47	LR-PRP	1	Low	A priori	9.0
PRP vs autologous whole blood							
Linnanmaki[20] (2020)	80 (40 vs 40, respectively)	47	NR	2	High	A priori	3.0
Raeissadat[27] (2014)	61	45.3	LR-PRP	1	High	A priori	10.0
Raeissadat[28] (2014)	40	46.3	LR-PRP	1	High	NR	10.0
Thanasas[33] (2011)	27	36.3	LR-PRP	1	High	A priori	9.0
PRP vs saline							
Linnanmaki[20] (2020)	79 (40 vs 39, respectively)	47	NR	2	High	A priori	3.0
Yerlikaya[37] (2018)	90	38.6	LP-PRP, LR-PRP	2	Low	NR	5.3
Montalvan[23] (2016)	50	46.7	NR	1	Low	A priori	1.6
PRP vs low-level laser therapy							
Tetschke[32] (2015)	52	52.3	NR	2	Unclear	NR	2.5

[a]LP, leukocyte poor; LR, leukocyte rich; NR, not reported; PRP, platelet-rich plasma.

dose versus low-dose PRP was found to be associated with significantly better VAS scores at final follow-up (−1.38 [95% CI, −2.20 to −0.55] in favor of PRP compared with alternative treatment; $P < .001$), accounting for 54.1% of the heterogeneity in outcomes (Figure 4). Results again held in multivariable meta-regression when additionally adjusting for the comparative treatment (−1.52 [95% CI, −2.44 to −0.60] in favor of PRP compared with alternative treatment; $P < .001$), with 55.0% of the heterogeneity explained.

DISCUSSION

The main findings of this study are as follows: (1) there was substantial variability in the outcomes of RCTs evaluating PRP for LE as well as substantial variability in the composition of PRP blood products used by these RCTs; (2) the platelet concentration factor of PRP used by each RCT was found to be strongly predictive of outcomes after the intervention; and (3) PRP with an estimated concentration of platelets greater than 3 times that of whole blood resulted in significantly improved outcomes compared with all alternative treatment options, while PRP with an estimated platelet concentration representing less than a 3-fold increase over whole blood resulted in no difference. These results contribute to the ongoing discourse in the field of orthopaedics by addressing a critical knowledge gap regarding the role of platelet concentration in PRP therapy for LE, reveal a potential breakthrough in our understanding of PRP, and provide critical evidence that should inform PRP treatment moving forward.

MDs in the VAS score ranged from 0.60 in favor of corticosteroids over PRP[14] to −2.43 in favor of PRP over corticosteroids.[18] Interestingly, the worst outcome of PRP relative to all other treatment options (MD of 0.60 in favor of corticosteroids over PRP) corresponded to the RCT with the second lowest platelet concentration factor (2×),[14] while the best outcome of PRP relative to all other treatment options (MD of −2.43 in favor of PRP over corticosteroids) corresponded to the RCT with the highest platelet concentration factor (10×).[18] With such a wide range of outcomes reported by RCTs evaluating PRP for LE, it is not surprising that substantial uncertainty remains around the efficacy of PRP as a nonoperative treatment option. Indeed, in a recent systematic review and meta-analysis of RCTs evaluating ultrasound-guided PRP injections for tendinopathies, the authors found that PRP did not result in significantly lower pain and function scores in the short, medium, or long term compared with control treatment methods, which included ultrasound-guided injections of steroids, saline, and anesthetics.[22] Even in previous meta-analyses that have suggested a beneficial effect of PRP for LE, there has been a substantial degree of heterogeneity and warnings about interpreting the results, given such variability.[8,11,25]

The high degree of heterogeneity in the current PRP literature creates difficulty for the practicing clinician who is faced with the decision of whether to offer PRP as a nonoperative treatment option to patients with LE. Clinical decision–making is further complicated by the out-of-pocket cost for patients. In these contexts, the surgeon may often rely on his or her own experience and decide to continue or discontinue the use of PRP based on the

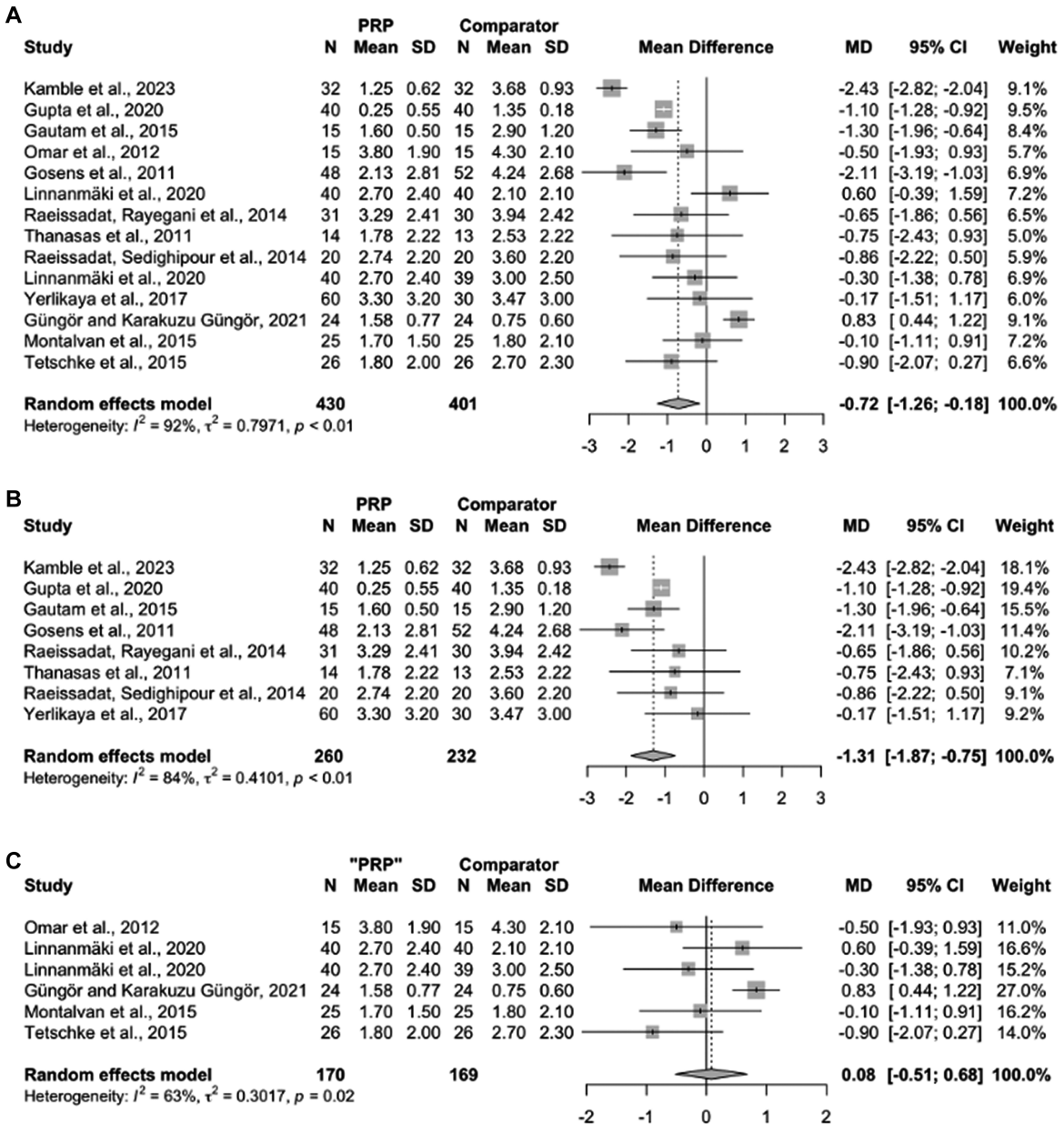

Figure 2. Random-effects meta-analysis results for (A) all randomized controlled trials (RCTs), regardless of the platelet-rich plasma (PRP) composition; (B) RCTs that utilized high-dose PRP (ie, ''true'' PRP); and (C) RCTs that utilized low-dose PRP. All pairwise treatment comparisons are represented in a forest plot, with treatment effects reported as mean differences with 95% confidence intervals. Intervals that do not cross the zero reference line represent statistically significant differences.

anecdotal results of previous patients. In light of the findings of the current study, demonstrating that 58.5% of the heterogeneity in outcomes reported by RCTs on PRP for LE can be accounted for by the platelet concentration factor of the PRP blood product alone, this empirical approach makes sense. Specifically, given the wide range of platelet concentrations used by RCTs in this meta-analysis, it is likely that similar or even greater variability exists in the platelet concentrations of PRP used by practicing orthopaedic surgeons. In fact, in a recent review of 33 PRP collection systems and protocols, the authors found that only 11 had >1 million platelets/μL or a 4- to 5-fold increase over the platelet concentrations of whole blood.[9] There were 3 systems that even produced "PRP" products with platelet counts less than those of whole blood.[9] Given the relationship that we observed between outcomes after PRP therapy and the platelet concentration factor, it is possible that patients presenting with LE could experience substantially different outcomes based on their clinician's individual PRP preparation protocol, leading to many advocating for PRP and equally many who suggest no benefit of PRP. Thus, there is a substantial need for clinicians

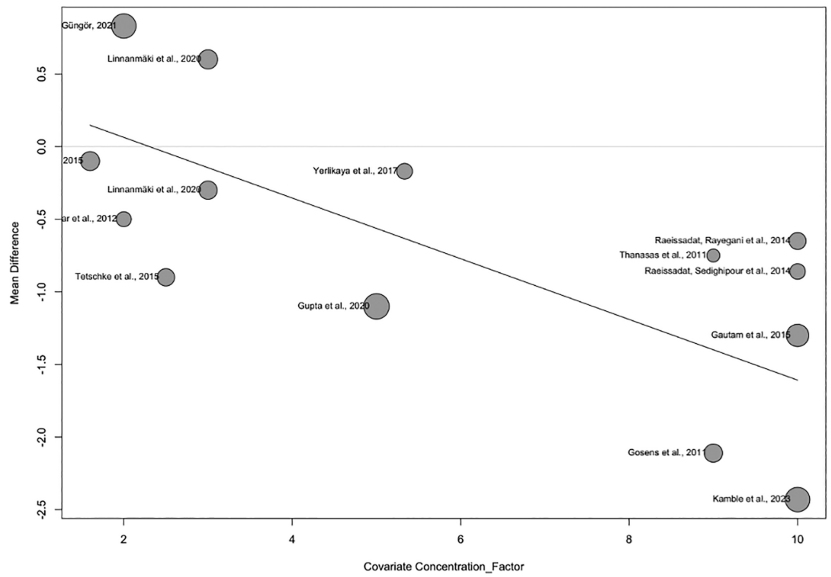

Figure 3. Meta-regression results with platelet concentration factor as the covariate of interest. The treatment effect for each study is plotted on the y-axis versus the platelet concentration factor on the x-axis. The size of each circle is inversely proportional to the variance of the estimated treatment effect. Platelet concentration factor was found to be significantly associated with the visual analog scale (VAS) score at final follow-up ($P < .001$), with 58.5% of the heterogeneity in outcomes accounted for by the platelet concentration factor alone.

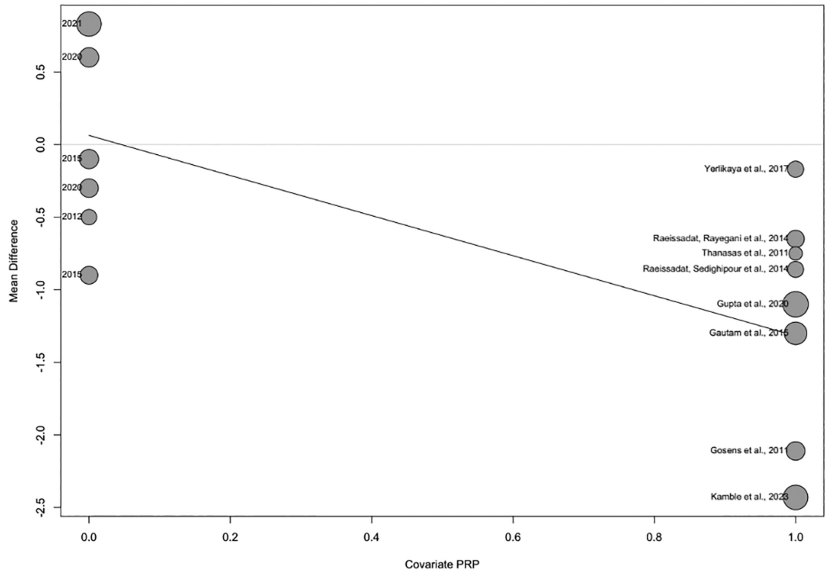

Figure 4. Meta-regression results with a binary platelet-rich plasma (PRP) composition variable corresponding to low-dose versus high-dose PRP as the covariate of interest. The treatment effect for each study is plotted on the y-axis versus the type of PRP (0, low-dose PRP; 1, high-dose PRP) on the x-axis. The size of each circle is inversely proportional to the variance of the estimated treatment effect. This binary variable corresponding to whether each study utilized high-dose PRP (ie, ''true'' PRP) or low-dose PRP was found to be significantly associated with the visual analog scale (VAS) score at final follow-up ($P < .001$), with 54.1% of the heterogeneity in outcomes accounted for by this variable alone.

and researchers to adopt a standardized approach to PRP preparation and methods to evaluate and ensure the quality of individual PRP compositions for patients.

Our meta-regression found a significant linear relationship between the platelet concentration of PRP used by each RCT and the corresponding MD in improvement of

the VAS score between PRP and the control treatment. In other words, the estimated platelet concentration of PRP used in an RCT strongly predicted the outcomes that patients in that RCT would experience. As the estimated platelet concentration increased, the relative improvement in the VAS score became more pronounced in favor of PRP over the control. Tendinopathies such as LE involve chronic microscopic tears occurring in hypovascular tendon tissue, with the associated expression of inflammatory mediators at the molecular level, which initiate healing via scar formation rather than regeneration of the microstructure and composition of normal tendon tissue.[6] Individual anti-inflammatory mediators and growth factors contained in platelets including transforming growth factor–beta 1, platelet-derived growth factor, vascular endothelial growth factor, and epidermal growth factor have been shown to increase type I collagen synthesis and tenocyte proliferation.[6] The present study builds upon in vitro studies by providing evidence of a direct relationship between the concentration of platelets (and thus platelet-derived growth factors) and outcomes after treatment with PRP. These results suggest that achieving adequate levels of platelets and platelet-derived growth factors is essential for optimal outcomes, with inadequate concentrations of platelets potentially leading to the minimal release of anti-inflammatory mediators and growth factors. Further, we found that this lower limit of platelet concentration may be higher than previously thought, as many have considered blood products with platelet concentrations as low as 2 times greater than whole blood to be "PRP."[6] Importantly, we did not observe an upper bound on the platelet concentration level to achieve symptom relief, although more data are needed to determine whether a maximum concentration level exists beyond which no additional benefit is achievable.

Currently, PRP is commonly defined as a plasma volume with a platelet concentration above that of baseline whole blood, which is 150,000/μL to 450,000/μL.[9,10,35] Thus, we defined any PRP with a platelet concentration factor representing a greater than 3-fold increase over whole blood or that with a documented supraphysiological platelet dose as high-dose or "true" PRP (ie, such that a patient with the minimum physiological platelet level in his or her blood would still receive a supraphysiological dose on average). PRP that did not meet this criterion was considered low-dose PRP. The range of platelet concentration factors used by RCTs in the high-dose group was 5× to 10×, while the range of platelet concentration factors used by RCTs in the low-dose group was 1.6× to 3×. One of the most important findings of this study was that meta-analysis of VAS scores reported by studies using high-dose PRP resulted in an MD of −1.31, corresponding to a significant difference in the effect between PRP and control treatment ($P < .001$), while meta-analysis of VAS scores reported by studies using low-dose PRP resulted in an MD of 0.08, suggesting no difference in the effect between PRP and control treatment ($P = .79$). This discrepancy provides perhaps the clearest evidence that clinicians should prioritize attaining a platelet concentration representing a greater than 3-fold increase over that of whole blood. Given the potential effect of this factor and the wide range of platelet concentrations reported by various commercial systems, it may be worthwhile for clinicians to quantify these values for themselves (eg, through platelet/thrombocyte counters or by sending samples to a nearby hematology laboratory).

Limitations

This study has several limitations. First, this systematic review and meta-analysis is subject to the limitations of the individual RCTs, including bias, study design, patient characteristics, PRP formulations, and heterogeneity in outcome measures. Next, it is possible that the platelet concentration does not perfectly correlate with the platelet concentration factor. Unfortunately, so few studies have reported on platelet concentration that a direct analysis was not possible. The current results suggest that future studies of PRP should be required to report on the platelet concentration, and it is possible that the variability in outcomes could be explained to an even greater degree with an exact measurement. Similarly, inadequate reporting precluded an analysis of the exact platelet dose (ie, the total number of platelets injected). To this end, Murray and coauthors[24] have previously reported on the minimum information that should be reported in studies evaluating the effect of PRP in musculoskeletal tissue, and the results of this study further underscore the importance of this reporting. Relatedly, fewer than half of the included studies reported on the leukocyte profile of PRP, precluding an analysis of this factor; notably, however, multiple previous reviews found no difference in the effect of leukocyte-rich and leukocyte-poor PRP on outcomes for LE,[19,25] suggesting that this may be unlikely to meaningfully affect results. Finally, many studies defined a number of dichotomous outcomes such as achievement of greater than a 25% decrease in the VAS score from baseline, which, because of the heterogeneity in the way that these binary outcomes were defined by studies, prevented their analysis.

CONCLUSION

The platelet concentration of PRP may play a significant role in the outcomes of patients with LE. A direct linear relationship was observed between the platelet concentration factor of PRP used and the magnitude of patient-reported symptom relief after the management of LE with PRP. Clinicians should ensure a supraphysiological platelet concentration when preparing PRP for the management of LE.

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