SYSTEMATIC REVIEW

Effectiveness of Platelet-Rich Plasma for Fracture Non-Union Management: A Systematic Review and Meta-Analysis

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ABSTRACT

Introduction: Fracture non-union is the most common and problematic complication post fracture with the incidence of 5-10%. Despite adequate standard treatment with autologous bone graft, failure of healing still occurs. The availability of platelet-rich plasma (PRP) to enhance bone tissue regeneration presents as potential solution for persistent non-union cases. The aim of this study was to conduct a systematic review and meta-analysis of literatures evaluating the effectiveness of PRP for fracture non-union management. **Methods:** Literature searching was conducted in PubMed, Cochrane Library, and ScienceDirect to obtain current evidence for the clinical application of PRP for fracture non-union. Relevant studies were critically appraised by Oxford Central for Evidence-Based Medicine critical appraisal tool. Data were pooled and tested for its heterogeneity with I^2. Study weight was analyzed using Mantel-Haenszel method and inverse variance when appropriate. **Results:** We selected seven relevant studies consist of two randomized controlled trials, three placebo-controlled clinical trials, and two intervention-only clinical trials. All studies showed high rate of union with PRP application. Four controlled studies showed significantly higher union rate and faster union time in PRP applied surgery compared to control. Pooled study analysis showed overall Z score of 3,15 (p=0,002) for union rate and 3,07 (p=0,002) for union time with the mean difference of 5 weeks. Secondary outcomes, such as functional score and visual analogue scale, favored PRP applied surgery compared to control. **Conclusion:** The application of PRP is effective to increase fracture union rate and shorten fracture union time in the management of fracture non-union.

Keywords: Fracture non-union, Platelet-rich plasma, Fracture union rate, Fracture union time

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INTRODUCTION

Fracture non-union is the most common and problematic complication post fracture. As high as 100,000 cases of fracture non-union is estimated per year in the USA. Reports on its incidence are generally varied among literatures ranging from 5-10%.(1) A study in Australian population showed 8.1% readmission rate within two years post-fracture, mostly due to fracture non-union.(2) Meanwhile, consensus on which age groups and sites of fracture contribute most to fracture non-union are still conflicting across studies.(1,2) Pain, psychological, and functional disability originated from fracture non-union are usually devastating and cause significant distress for patients.

Fracture non-union management focuses on creating proper biological environment and adequate mechanical strength properties on fracture site. Autologous bone graft (ABG) is often chosen as standard treatment for its capability to reenact proper environment for optimum bone healing. However, failure of healing still occurs and often results to additional treatment to achieve desirable fracture healing.(3,4)

Platelet-rich plasma (PRP) is among the most promising solution for persistent non-union that is readily obtained from autologous blood with minimal to zero morbidity. PRP is defined as volume of plasma with higher concentration of platelet compared to physiological levels. Platelets are essential especially for complex inflammatory stage of healing. It promotes angiogenesis, formation of mesenchymal cells, as well as growth factors. PRP, being able to provide platelets in increased amount, is believed to potentially enhance effectiveness in bone healing process, especially in persistent non-union cases.(5) Therefore, this study aims to conduct

a systematic review and meta-analysis of literatures evaluating the effectiveness of PRP in the management of fracture non-union.

MATERIALS AND METHODS

We conducted a systematic review and meta-analysis to obtain current evidence for the clinical application of PRP for fracture non-union. This systematic review and meta-analysis was conducted according to preferred reporting items for systematic reviews and meta-analyses (PRISMA) guideline as shown in Fig 1.

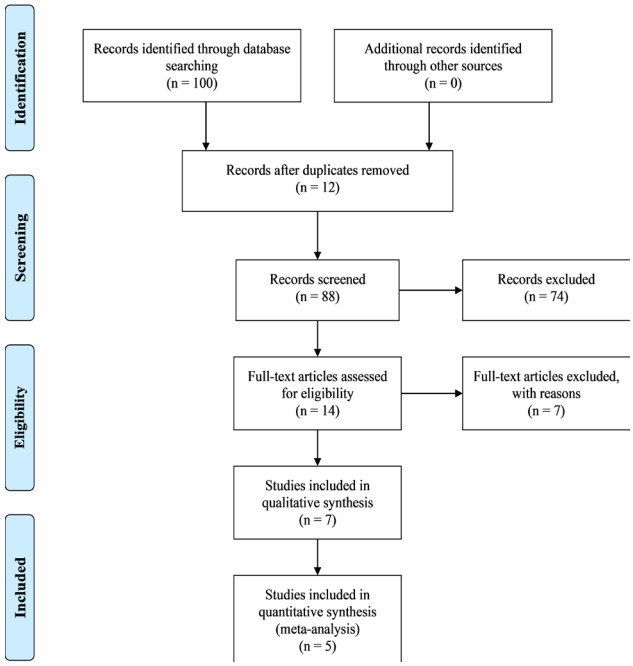

Fig. 1 PRISMA flow diagram

Eligibility criteria

Literatures were selected in this systematic review and meta-analysis following these selection criteria:

Study design

We included only randomized controlled trial (RCTs), controlled trial, and clinical trials. Case reports, case series, and commentary were excluded from the study.

Participants

Participants with aseptic fracture non-union were considered in this study. Non-union is defined as a failure of fracture union within 6 months after injury without further progression.

Interventions

Human clinical studies with any application of PRP as an adjuvant therapy with or without autologous bone graft and surgical intervention were included in this study. There was no restriction for intervention comparison in this study.

Outcomes

Primary outcomes for this study were subsequent fracture union rate and fracture union time. Fracture union time was measured by weeks. Patient related outcomes measured with visual analogue scale (VAS) and functional score were considered as secondary outcomes.

- Primary outcomes:
  - Fracture union rate
  - Fracture union time
- Secondary outcome:
  - VAS
  - Functional score

Search strategy

Literature searching was conducted through MEDLINE (PubMed), Cochrane Library, and ScienceDirect online electronic databases. Literature searching was conducted on December 26th, 2020. We used combination of following keywords to optimize the literature searching sensitivity: "platelet-rich plasma", "non-union", "fusion", "callus formation", and "healing" (Table I). Reference lists of included studies and reviews were further reviewed to ensure literature saturation. Literature searching was not limited by language, publication date, nor publication status.

Table I. Specific keywords on literature searching

Online database	Keywords
Pubmed	(("platelet rich plasma"[All Fields]) AND (("non-union"[All Fields]) OR ("non union"[All Fields]))) AND ((("fusion"[All Fields]) OR ("callus formation"[All Fields])) OR ("healing"[All Fields]))
Cochrane	(platelet rich plasma):ti,ab,kw AND (nonunion OR non union):ti,ab,kw AND (fusion OR callus formation OR healing):ti,ab,kw
Science-Direct	platelet rich plasma AND (nonunion OR non union) AND (fusion OR callus formation OR healing)

Study selection

Study titles and abstracts were screened and selected according to eligibility criteria by two contributors independently. Any disagreement in the selection process was resolved by discussion. Duplication of published literature was sought and excluded after the initial screening. Full article for the selected studies were obtained and reviewed independently by the contributors.

Data collection process

Data extraction was conducted by a data extraction sheet that was constructed and customized according to the selected study results. One of the authors extracted the data from the selected study, while another author checked the extracted data to ensure the data's accuracy. Any disagreement in the data extraction and collection was resolved by discussion. No additional data were obtained from authors of selected studies. Information

extracted from the studies includes characteristic of the study (year, sample size, method); characteristics of the participants (age, fracture location, non-union duration, mechanism of trauma); types of intervention and comparison; and type of outcome measures (fracture union rate, fracture union time, VAS, functional outcome). The outcome measured for fracture union rate was odds ratio and 95% confidence interval. The outcome measured for fracture union time was mean and standard deviation (SD).

Risk of bias

Evaluation for the risks of bias was conducted in this study to ascertain the validity of selected studies. Oxford Central for Evidence-Based Medicine (CEBM) critical appraisal tool for the randomized controlled trial was used to evaluate the risk of bias of each selected individual study. Both authors evaluated selected studies independently with further discussion over any disagreement.

Method of analysis

Randomized controlled trials and non-randomized controlled trials in this study were pooled and tested for heterogeneity test to identify any inconsistency between studies related to measured outcomes. The outcome of the inconsistency analysis I^2 was used to describe the percentage of total variation among selected studies. The study weight was analyzed using Mantel-Haenszel method for dichotomous outcomes, while inverse variance was used for continuous outcomes. Results of the analysis were reported using Forrest plot. Data analysis was calculated using the Review Manager (REVMAN) version 5.4.

RESULTS

Study selection

We identified 100 articles through online database searching. We did not find any additional records from other sources of publication. After we eliminated all duplicated studies, 88 articles were further evaluated for its study design, population, intervention, and outcomes through title and abstract screening. 74 articles were excluded, and 14 remaining full-text articles were assessed for articles eligibility. Finally, 7 studies were included in qualitative analysis, from which 5 studies were selected for quantitative analysis.(5–11)

We included two RCT, three n-RCT, and two clinical trial with total of 376 patients enrolled in this systematic review and meta-analysis(see Table II for study characteristics and data collection). Out of all included studies, five studies compared PRP to conventional surgical intervention. All studies applied PRP by local injection to non-union site. Six studies used freshly activated PRP and one study applied frozen PRP gel. Surgical interventions that were conducted on most subjects varied from intramedullary (IM) nailing, ORIF, to external fixation. However, a few selected patients underwent conservative treatment with closed reduction and cast immobilization. The outcome of union rate and union time was observed in six studies and five studies respectively. The pain improvement measured by VAS was evaluated in three studies, while functional outcomes were only observed in one study.

Table II. Study characteristics

Author (year)	Study design	Treatment / control (number)	Treatment /control (age)	Fracture location	PRP			Intervention		Outcome	Outcome Measurement	Results
					Preparation	Activator	Administration	Treatment	Control			
Galasso (2008)	Clinical trial	22 / 0	39 / -	Tibia, femur, humerus	6mL PRP extracted from 55mL centrifuged autologous blood	Batroxobin and CaCl	Direct gel application	IM nail exchange + PRP	-	Union rate	Clinical and radiological (every 45 days until 13 months)	91% union rate
Maricionda (2008)	n-RCT	20 / 20	34 / 30	Tibia, humerus, forearm	14mL PRP extracted from 75 mL centrifuged autologous blood	Autologous thrombin and Ca Gluconate	Percutaneous gel injection	External fixation + PRP	External fixation	Union rate, union time	Clinical (every 2 weeks); radiological (every months)	No significant difference between groups
Malhotra (2015)	Clinical trial	94 / 0	-	Femur, tibia, humerus, forearm	15-20mL PRP (platelet count of minimum 2,000,000/µl and leuko-reduction) centrifugated from autologous blood	No activator	Percutaneous gel injection	ORIF / closed reduction + PRP	-	Union rate	Clinical and radiological (every months until 3 months)	82% union rate

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Table II. Study characteristics

Author (year)	Study design	Treatment / control (number)	Treatment / control (age)	Fracture location	PRP		Intervention			Outcome	Outcome Measurement	Results
					Preparation	Activator	Administration	Treatment	Control			
Ghaffarpasand (2016)	RCT	37 / 38	26,5 / 26,3	Femur, tibia, humerus, ulna	5mL PRP extracted from 54mL venous autologous blood	No activator	Direct PRP application	IM nail/ ORIF+ bone graft + PRP	IM nail/ ORIF + bone graft + 5mL saline	Union rate, union time, VAS	Clinical and radiological (every 45 days until 9 months)	Favors PRP significantly in union rate, time, and VAS
Zhao (2017)	RCT	46 / 46	-	Femoral shaft fracture	5mL PRP extracted from 30mL venous blood	Autologous thrombin and tranexamic acid	Injection to the injury site	Conventional surgery + PRP	Conventional surgery	Union rate, union time, VAS	Clinical and radiological (at the 9th month postoperative)	Favors PRP significantly in union rate and time
Duramaz (2018)	n-RCT	15 / 14	35 / 41	Femur and tibia	10mL PRP extracted from 55mL venous autologous blood	Bovine thrombin and 10% calcium chloride	Percutaneous injection	IM nail exchange with reaming + PRP	IM nail exchange with reaming	Union rate, union time, VAS	Clinical (every 2 weeks) and radiological (every month)	Favors PRP significantly in union time and VAS
Basdelioglu (2020)	n-RCT	14 / 10	50 / 48	Femur, tibia, humerus, ulna, clavicle	10mL PRP extracted from 75mL venous autologous blood	No activator	Direct PRP application	ORIF/ External fixator + graft + PRP	ORIF/ External fixator + graft	Union time, functional outcome	Clinical and radiological (every months until 3 months, every 3 months after that until union occur)	Favors PRP significantly in union time

IM nail (intramedullary nail), n-RCT (non-randomized controlled trial), ORIF (open reduction internal fixation), PRP (platelet rich plasma), RCT (randomized controlled trial), VAS (visual analogue scale)

Risk of bias assessment

The validity of the studies showed adequate randomization in two studies (Ghaffarpasand et al, Zhao et al).(9,10) The baseline condition comparison showed similarities in three studies (Mariconda et al, Ghaffarpasand et al, Basedioglu et al).(5,7,9) All comparation studies accounted all subjects in their final analysis and all subjects were analyzed according to their assigned groups. Only 1 study showed consistent blinding of treatment of their subjects and clinician (Ghaffarpasand et al).(9) The importance appraisal showed significant result of union rate in favor of PRP in two studies and significant result of union time in favor of PRP in three studies (Ghaffarpasand et al, Zhao et al).(9,10) The applicability assessment showed that all studies were applicable to their included study populations. Complete critical appraisal assessment are shown in Table III.

Table III. Critical Appraisal

Validation				
Author (year)	Was the assignment of patients randomized?	Were the group similar at the start of the study?	Were all patients who entered the trial accounted for? And were they analyzed in the groups to which they were randomized?	Were measures objective, or were the patients and clinician kept "blind" to which treatment was being received?
Galasso et al. (2008)	No	No	No	No
Mariconda et al. (2008)	No	Yes	Yes	No
Malhotra et al. (2015)	No	No	No	No
Ghaffarpasand et al. (2016)	Yes	Yes	Yes	Yes

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Table III. Critical Appraisal (CONT.)

Validation				
Author (year)	**Was the assignment of patients randomized?**	**Were the group similar at the start of the study?**	**Were all patients who entered the trial accounted for? And were they analyzed in the groups to which they were randomized?**	**Were measures objective, or were the patients and clinician kept "blind" to which treatment was being received?**
Zhao et al. (2017)	Yes	No information	Yes	No
Duramaz et al. (2018)	No	No	Yes	No
Basdelioglu et al. (2020)	No	Yes	Yes	No

Importance				
Author (year)	**Union rate**		**Union time**	
	Relative risk	**P value**	**Mean difference**	**P value**
Galasso et al. (2008)	-	-	-	-
Mariconda et al. (2008)	1,59 (0,2-10,7)	0,633	-0,85 (-6,7 / 5,0)	0,784
Malhotra et al. (2015)	-	-	-	-
Ghaffarpasand et al. (2016)	3,47 (1,2 / 9,8)	0,025*	-1,6 (-3,3 / 0,1)	0,003
Zhao et al. (2017)	3,98 (1,0 / 15,5)	0,036*	-3,37 (-3,8 / -2,9)	0,009
Duramaz et al. (2018)	3,25 (0,3 / 35,6)	-	-2,36 (-4,5 / -0,1)	0,654
Basedioglu et al. (2020)	-	-	-23,6 (-29,9 / -17,3)	0,000

Applicability			
Author (year)	**Is my patient so different from those in the study that the results cannot apply?**	**Is the treatment feasible in my setting?**	**Will the potential benefits of treatment outweigh the potential harms of treatment for my patient?**
Galasso et al. (2008)	No	Yes	Yes
Mariconda et al. (2008)	No	Yes	No
Malhotra et al. (2015)	No	Yes	Yes
Ghaffarpasand et al. (2016)	No	Yes	Yes
Zhao et al. (2017)	No	Yes	Yes
Duramaz et al. (2018)	No	Yes	No
Basedioglu et al. (2020)	No	Yes	Yes

Intervention effects

The summary findings of all primary and secondary outcomes were shown in Fig. 2,3 and 4. Four studies compared the rate of fracture union in surgical management with PRP application to surgical management alone.(7,9–11) Pooled data of studies (336 patients) showed significantly higher union rate in PRP applied subjects (OR=3,20, 95%CI=1,55/6,61) with the overall Z value of 3,15 (p=0,002). Heterogeneity analysis showed low percentage of heterogeneity between studies (I2= 0%).

Fracture union time was observed in five comparison studies contrasting surgical management with PRP application and surgical management alone.(5,7,9–11) Pooled data of studies (257 patients) showed significantly lower union time in PRP applied subjects (MD= -5,0, 95% CI= -29,9/-17,3) with the overall Z value of 3,07 (p=0,002). Heterogeneity analysis showed a high percentage of heterogeneity across studies (91%). Three comparison studies observed post-operative VAS between intervention and control group.(9–11) Pooled data (196 patients) showed no significant difference of postoperative VAS between PRP applied subjects and surgical only subjects (MD= -0,30, 95% CI= -0,63/0,38) with the overall Z value of 1,75 (p=0,08). There was moderate heterogeneity across studies (I2= 15%).

Functional outcome measured with Lower Extremity Functional Scale (LEFS) and Upper Extremity Functional Index (UEFI) was only observed in one study.(5) Thus, the statistical analysis could not be done for this outcome.

DISCUSSION

Summary of evidences

In this study, we found significant improvement of union rate and union time in PRP applied patients compared to

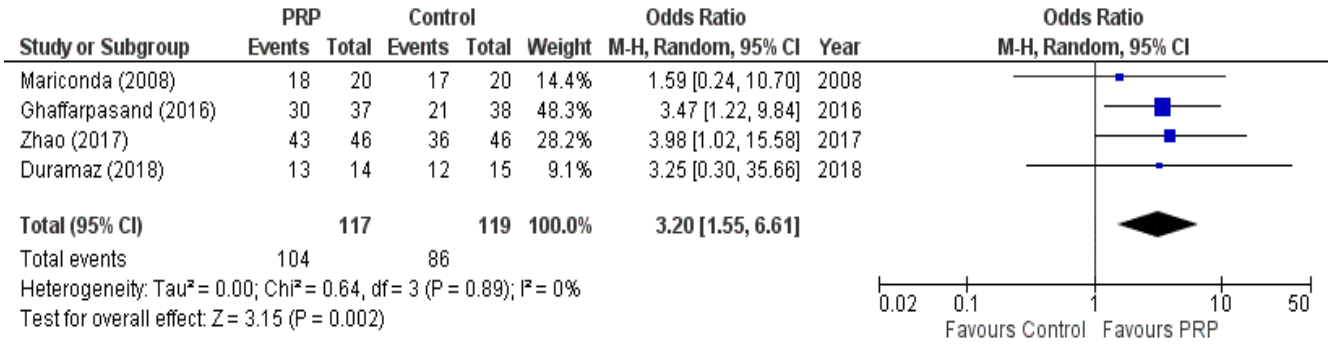

Fig. 2 Forrest plot of fracture union rate

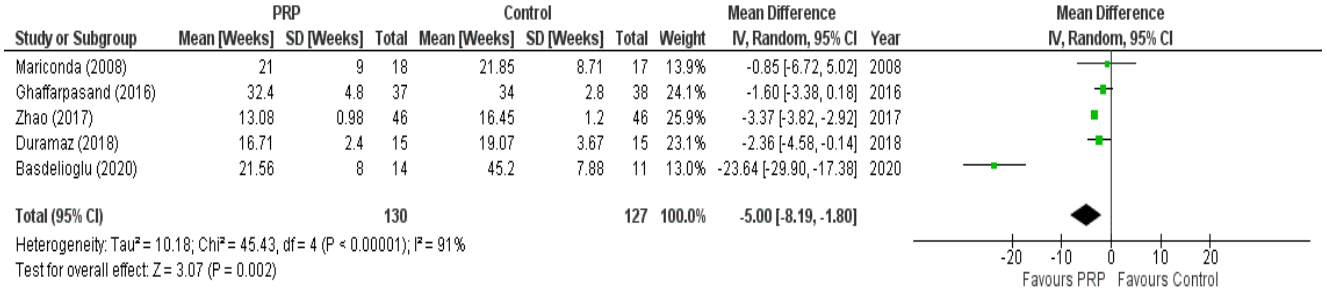

Fig. 3 Forrest plot of fracture union time

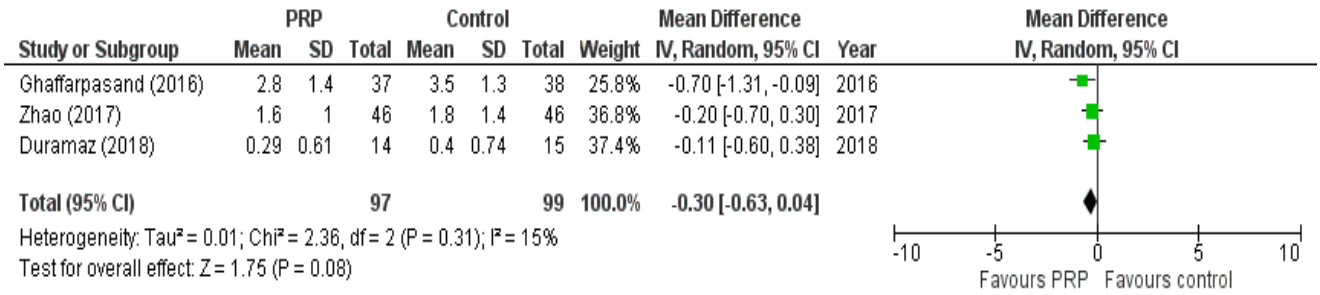

Fig. 4 Forrest plot of post-operative visual analogue scale (VAS)

control in pooled data analysis. The PRP application was used as an adjuvant therapy to surgical treatment in all studies. Studies have shown benefits in PRP application in both atrophic and hypertrophic fracture non-union included in this study. Other factors related to fracture union such as gender, age, smoking habit, high body mass index, and mechanism of fracture was addressed in several studies minimize any confounding factor to the intervention.(5,7,9–11) Most of the studies applied directly to the fracture site during surgery with onsite PRP preparation during surgery. Activation of PRP were common in several studies using thrombin, tranexamic acid, calcium gluconate, 10% calcium chloride, batroxobin or combination of those solutions as platelet activator.(6,7,10,11) Previous studies has shown PRP ability to induce the release of vascular endothelial growth factor (VEGF), transforming growth factor-β1 (TGF-β1), platelet derived growth factor AB (PDGF-AB), interleukin 1β (IL-1β), and tumor necrosis factor α (TNF-α).(12) PRP activation will increase the secretion of the required growth factors and cytokines thus enhance the ability of fracture healing. The presence of growth factors provided, support regeneration on the fracture site. This complies to the diamond concept of fracture healing, which refers to the requirement of growth factors, osteogenic cells, osteoconductive matrix as three biological properties along with and mechanical stability to ensure an optimum fracture healing.(4,13)

The application of PRP is particularly useful in oligotrophic or atrophic fracture non-union where there are inadequate biological factors surrounding the fracture site.(4) This study also showed benefits of PRP application in hypertrophic fracture non-union

as one study showed significant number of subjects with hypertrophic non-union.(9) The application of PRP provides necessary growth factors and cytokines in the fracture site which improves fracture healing rate and time.(14) However, surgical intervention in PRP application also considered important as surgical intervention will induce inflammatory process in the fracture site.(12) Thus the application of PRP is more effective with other inflammatory present in the fracture site. There is no study that recommends PRP application or injection as an individual intervention for fracture non-union.

There was no significant difference in term of VAS improvement in PRP applied patients compared to control. All studies reported an improvement of VAS in both groups. Similar results were also reported in other previous studies. Meta-analysis by Manini et al. Also found no statistically significant pain reduction by VAS between PRP group and control in spinal fusion surgery. (15) It was originally believed that activity of platelets during early inflammatory process corelates indirectly to reduction of pain.(9) This statement has not been able to be proven across literatures, even has been suggested otherwise.

The only study assessing patients' functional capacities was not able to prove any difference between two groups. All patients in both PRP and control group achieved complete union with no complications, similarly. At the same time, PRP also didn't cause longer operation time nor surgical morbidity in any patient. Sheth et al. reported a meta-analysis on PRP use for various orthopedics indications, from which no significant superiority of functional improvement can be seen on PRP group compared to control group.(16)

This study indicates PRP as one of the easiest autologous agents to acquire and still possess superior healing properties. As a blood-derived preparation, PRP is readily available from the patients themselves with and easy to process compared to other autologous agents, such as bone marrow.(4)

Strength and limitation

This is the first systematic review and meta-analysis evaluating PRP application for fracture non-union, exclusively on clinical settings. This study also accentuates the possibility of mass application of PRP for fracture non-union in the near future, given into consideration the vast involvement of stem cells therapy for fracture non-union and PRP itself as one of the most easily-obtained and straightforward stem cell option.

The risks of bias in this study varied with several studies with high risks of bias and one study with low risks of bias. There were two clinical studies without comparison included in this study with this study which showed high rate of fracture union in PRP application.(6,8)

CONCLUSION

The application of PRP with surgical intervention is effective to increase fracture union rate and shorten fracture union time in the management of fracture non-union. However, the improvement of VAS and functional score could not be concluded in this study due to limited of evidence. Supported by the overall result from this study, PRP treatment is highly considered as an adjuvant for fracture non-union management in clinical settings. Further research should be conducted to evaluate its clinical safety and compare PRP treatments' superiority with other biological adjuvants.

ACKNOWLEDGEMENT

Not applicable. We declare there was no conflict of interests.

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