

REVIEW ARTICLE

Investigating Effectiveness of Topical Autologous Platelet-rich Plasma as Prophylaxis to Prevent Wound Infection: A Systematic Review and Meta-analysis

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ABSTRACT

Autologous platelet-rich plasma (PRP) was reported as having potent antimicrobial properties. However, the literature showed conflicting results. Therefore, we aim to investigate the effectiveness of topical autologous PRP as prophylaxis to prevent wound infection. We searched major electronic databases such as MEDLINE, EMBASE, CENTRAL, and Web of Science to identify RCT studies regarding this topic. The selection of included studies followed the PRISMA guidelines. We included ten RCTs comprising 1257 participants. In general, PRP showed no effect in reducing the risk of wound infection (RR: 0.84; 95% CI: 0.66–1.06; $p=0.14$). However, subgroup analysis based on wound characteristic showed that PRP significantly reduced wound infection risks in acute wounds (RR: 0.76; 95% CI: 0.58–0.99; $p=0.04$). Meanwhile, activation of PRP had no effect in reducing wound infection risks ($p=0.77$). In conclusion, we suggest routine autologous topical PRP application in acute wound care due to PRP antimicrobial properties and regenerative potential.

Keywords: Platelet-rich plasma, Topical administration, Wound infection, Antimicrobial agent, Systematic review

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INTRODUCTION

A wound is defined as any disintegration of the skin or mucous membrane, with or without tissue loss underneath the skin(1). Wounds are classified as chronic wound when they fail to establish a normal anatomical and functional state within three months period or may not heal at all(2). Despite advancements in medical technology, chronic wounds remain a healthcare burden. In 2012-2013, the NHS (UK) reported there were 2.2 million patients suffering from chronic wounds, costing 5.3 billion of the national expenditure(3). The USA spent \$25 billion annually to treat more than 6 million patients with wound-related complication(4). Normal wound healing process consists of three stages: inflammatory, proliferative, and remodelling(5). The

inflammatory stage is marked by vascular spasm to minimise blood loss and platelet aggregation. In the proliferative phase, a new blood vessel (angiogenesis) must be built in order to provide oxygen and nutrient to the new granulation tissue. Myofibroblasts enable the wound to contract as the new tissues and reduce the wound gap, followed by resurfacing of epithelial cells which marks the end of the proliferative phase(6). Remodelling phase is a phase where the collagen III is remodelled to stronger collagen I(5). This phase usually begins around 21 days after injury and can continue for years. However, the physiological wound healing process could be hindered by infection(7). Bacteria can release toxin and substances which will result in an elongation of the inflammatory phase, causing the wound to enter a chronic state and unable to recover(8). In addition, the prolonged inflammatory phase will cause more deposition of matrix metalloproteases (MMP) around the injury site. MMP is a protease which is able to degrade the extracellular matrix, making the wound harder to heal(9).

Platelet-rich plasma (PRP) is a blood product which has a relatively high concentration of platelet in plasma compared with whole blood. PRP is known by a lot of names such as platelet releasate, platelet gel, and platelet-derived growth factor. Platelet possesses alpha granules, which could release myriad of growth factors and mediators such as PDGF, b-FGF, VEGF, EGF, IGF-1, and TGF- β 1 upon activation. As a result, PRP could stimulate and accelerate healing in the various pathway(10). Moreover, alpha granules also secrete complement factors and chemokines toll-like receptor. Chemokines toll-like receptor provides protection against microorganism by inducing Reactive Oxygen Species and Reactive Nitrogen Species production(11). In addition, PRP has abundant proteins called Platelet Microbicidal Peptides. There are myriad variants of PMP such as NAP-2, CXCL-4, and CXCL-8(12). Those growth factors are displayed not only remarkable antimicrobial effect against plethora bacteria such as *S. aureus* and *E. coli* but also exhibit effective antifungal effect against *C. neoformans*(13). Several in vitro studies reported that PRP has significant antimicrobial activity. PRP could inhibit numerous bacteria we commonly encountered in a clinical setting such as *E. coli*, *E. faecalis*, *K. pneumonia*, MRSA, MSSE, and *P. aeruginosa* (14). Study in animal trials also showed PRP quicken re-epithelialisation speed and healing rate in MRSA-infected wound(15). Even though PRP is proven to have potent antimicrobial properties, it is only effective against planktonic bacteria, but not biofilm bacteria(16). Therefore, it is more reasonable to utilise the antimicrobial properties of PRP for preventive measure rather than curative. PRP can be activated by thrombin or calcium chloride to enhance its microbial properties. Activated PRP exhibits better antimicrobial efficacy due to the faster release of growth factor inside the platelet (17).

There is not yet a systematic review and meta-analysis which evaluated specifically on topical PRP effectiveness in reducing the risk of wound infection. There were several studies which investigated the PRP effect on wounds(18-20). However, their clinical outcomes were mainly wound healing, complete epithelialisation, or wound healing rate. In addition, those reviews do not focus on topically applied PRP. Therefore, the main purpose of this systematic review and meta-analysis was to evaluate the potency of the antimicrobial effect of topical autologous PRP treatment for preventing wound infection while also evaluating PRP regenerative potential in addition.

METHODS

Eligibility criteria

Type of studies

We only included Randomised Controlled Trials (RCTs) comparing PRP and other wound care treatment. Studies without information about the wound infection proportion after interventions, cross-over clinical trial,

cluster randomised trial were excluded. There was no language or timeline restrictions.

Type of participants

All people aged 18 years or above with wound injury from any cause were included. Patients who consume systemic immunosuppressant drugs, pregnant women, and patients with pre-existing wound infection prior to intervention were excluded. Patients with autoimmune disease, haematological abnormalities, HIV/AIDS, Systemic Lupus Erythematosus, scleroderma, and chronic liver failure were also excluded.

Type of intervention

We only included studies comparing topical autologous PRP with other wound care treatment, such as placebo or standard care. Interventions given via injection or any means other than topical were excluded.

Type of outcome measure

Outcome measures selected were in accordance with prior studies on how to define wound infection and surgical site infection such as clinically diagnose from sign and symptom, laboratory test, wound culture test, and scoring system(21).

Primary outcome

- The proportion of wound infection

Secondary outcome

- Wound size-reduction
- The proportion of healed wound

Search method for identification of studies

Electronic searches

The author built a robust search strategy with the appropriate keyword and medical subject heading terms (Appendix 1-4) in consultation with a medical librarian in our institution. This search strategy was built based on the PICO (Patient, Intervention, Comparison, Outcome) concept which recommended by Cochrane Handbook for Systematic Reviews of Intervention version 5.1.0.

P: Adult with wound problem

I: Topical platelet-rich plasma (PRP)

C: Placebo or standard treatment/care

O: Proportion of infected wound, proportion of healed wound, and wound size-reduction

The literature search was conducted in the following electronic databases:

- Medline: 1900 to present (July, 14th 2020)
- Embase: 1900 to present (July, 14th 2020)
- CENTRAL: All years
- Web of Science: 1900 to present (July, 14th 2020)

Reference list of both included and excluded study on this review were checked for additional relevant references.

Data collection and analysis

Selection of studies

Literature obtained from literature research were downloaded and exported to Rayyan (22). After duplicates removal, two independent reviewers (IIP and BDV) screened the titles and abstracts. Potentially eligible studies were read in full-text to select the included studies based on our eligibility criteria. Exclusion reasons after full-text reading and details of the workflow are illustrated in the PRISMA flow diagram. Any discrepancies that occurred were resolved with discussion.

Data extraction and management

Data from eligible studies were extracted and recorded in a table created from Microsoft Word. Data extracted from the eligible studies were author & publication year, country, study design, unit of randomisation, unit of analysis, setting, follow-up duration, enrolment period, number of participant, characteristic of participant, inclusion criteria, exclusion criteria, wound characteristic, type of intervention, number of events, mean of the outcome, and standard deviation in each group.

Assessment of risk of bias in included studies

The potential risk of bias of individual study was assessed by the Cochrane Risk of Bias (ROB) tool 2 (23).

Measurement of treatment effect

Dichotomous data were calculated and presented as a risk ratio, whereas continuous data were calculated and presented as mean difference. In order to observe the treatment effect more comprehensively, outcome measures were combined with meta-analysis and analysed with taking particular subgroup into consideration. The uncertainty of effect was described as 95% confidence interval (CI).

Unit of analysis issue

This review evaluates the patient with a wound problem treated with topical PRP or standard care/placebo. Unit of analysis in this review is patient instead of amounts of wounds. Thus, cross-over trials were excluded to prevent the needs of complicated analysis. Data from eligible multiple arm trial were included only if they were relevant to the review focus.

Dealing with missing data

In case of absence of dichotomous data, other data such as percentage, risk ratio, and odds ratio were obtained as a substitute. For continuous data, missing data were retrieved by using alternative data such as median, standard error, p-value, and interquartile range.

Assessment of heterogeneity

Statistical heterogeneity (inconsistency) was measured with Chi-square and I^2 test. Chi-square was used to detect whether the differences in outcome is caused

by pure chances, whereas the I^2 test was used to identify variability in the results. In the Chi-square test, heterogeneity can be concluded if it has p-value < 0.1. Meanwhile, a maximum tolerable value of I^2 is 40% and a value of less than 40% is regarded as insignificant and can be ignored (24).

Assessment of reporting biases

Reporting (publication) bias is analysed with a funnel plot when a minimum of ten studies are included; otherwise, the power is too low. If the funnel plot illustrates asymmetrical shape, we should suspect the presence of reporting bias. The author did not perform the funnel plot since studies included in quantitative synthesis were fewer than ten.

Data synthesis

Statistical analysis in this study was performed with Review Manager 5. Both fixed-effect models and random effect models were performed in this study. Fixed effect model was preferred when there is no significant heterogeneity suspected.

Subgroup analysis and investigation of heterogeneity

We performed two different subgroup analysis on the primary outcome: based on the characteristic of the wound (acute or chronic) and characteristic of intervention (activated or non-activated PRP). Subgroup analyses were not conducted on the secondary outcome due to a limited number of studies.

Sensitivity Analysis

To investigate the robustness of this review, the author prespecified a sensitivity analysis to investigate the effect of excluding studies with a high risk of bias. The author also repeated primary meta-analysis in case the range of values for decisions was uncertain by using alternative measures of effect size and statistical model.

Summary of Findings table

Summary of Findings table is presented using GRADE (Grades of Recommendation, Assessment, Development and Evaluation Working Group) approach, as in accordance with Cochrane recommendation. Besides the quality of evidence, the GRADE approach also provides information about the magnitude of the effect and whether their impact remains significant despite confounding removal.

RESULT

Description of studies

Result of the search

The initial search yielded 3004 articles. After 925 duplicates removal, 2079 studies were screened for its relevance. After titles and abstracts screening, the author obtained 47 potentially eligible studies. Following the full-text reading, 37 studies are excluded for various reasons explained in Fig. 1. Ultimately, qualitative

analysis in this study is based on ten RCTs, whereas nine RCTs are included in the meta-analysis (Fig. 1).

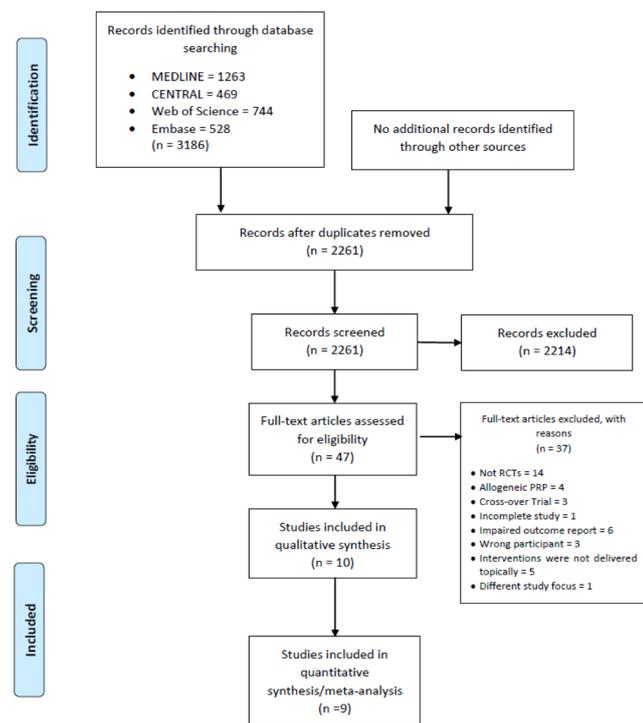


FIG. 1: Nine RCTs are included in the meta-analysis

Included studies

Ten RCTs were included in this review(25-34). There is no cluster trial and cross-over trial included in this review. One trial was a one-arm trial(32), and the other nine were a two-arm trial.

Settings

The studies were conducted in the USA(32, 34), Spain(27), UK(28), Lithuania(31), France(33), Norway (35), Egypt (36), China (30), and Greece(29). All of the studies were conducted in a hospital setting except for one study conducted in a heart clinic setting(26).

Patients

Total patients included in this review were 1257 patients (male/female distribution: 731/526), comprised of 631 participants from the control group and 626 participants from the treatment group aged 18 years old or older and had a wound-related issue. Based on wound characteristic, 677 participants suffered a chronic wound, while the other 580 suffered an acute wound.

Interventions

All trials compared topical PRP as an intervention against standard care or placebo. There were 488 participants who received activated PRP, 138 who received non-activated PRP, and 631 who received standard care/placebo. PRP activation was mostly done by adding Calcium Chloride or Thrombin. None of the activation PRP processes involved alcohol. Only two studies included in this review used topical non-activated PRP

as an intervention. Follow-up duration ranged from 3 weeks until 12 weeks.

Risk of bias in included studies

Fig. 2 depicts a summary of the author’s judgement on ten included RCTs in this review. In summary, nine of ten studies included in this review are judged to have a moderate risk of bias by the author, while only one is deemed a high risk of bias. High-bias study is excluded from the meta-analysis.

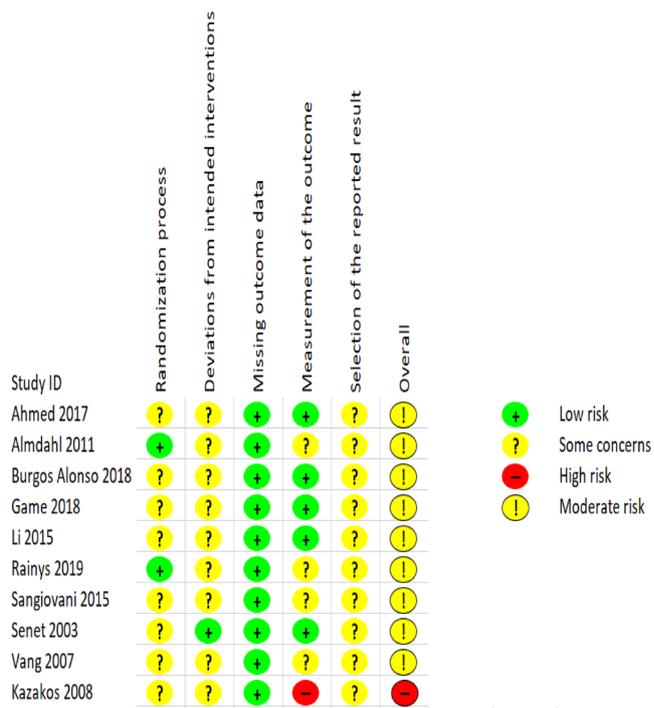


FIG. 2: Nine RCTs are included in the meta-analysis

Effect of Intervention

All ten trials included in this study contributed data to the primary outcome. However, only five studies contributed data to a secondary outcome.

Primary outcome

Qualitative Synthesis

Overall, data from included trials showed PRP group has less wound infection occurrence compared to the control group, which is 90 out of 626 participants against 108 out of 631 participants. However, the distribution of the data is quite varied, leading to a different conclusion in several studies. In a study conducted by Ahmed et al., PRP is more favourable on reducing potential wound infection compared to standard care. Out of 28 patients in the control group, six of them suffered from wound infection in the following twelve weeks. Meanwhile, wound infection in participants of the PRP group only two out of 28. This difference is statistically significant (p-value: 0.011)(25). Same results were reported in three studies(28, 30, 33). In those studies, they reported a proportion of wound infection in the PRP group is less

compared to the control group. However, the results were not statistically significant. Game et al. reported 51 events of infection from a total of 132 patients in the PRP group; while in the control group, the wound infection events were 63 out of 134 with p-value 0.208. Four other studies in this review reported a similar number of wound infections between PRP and control group(27, 29, 31, 37) while two other studies reported standard care to have less event of wound infection(26, 32). However, this result is not statistically significant.

This result discrepancy could be a result of the difference in wound characteristic and intervention type. If we look closer to the characteristic of the intervention, eight studies used activated PRP(25-27, 30-32, 37) and only two studies used non-activated PRP(28, 33). Opinions are divided among researcher. A study reported it is not necessary to activate PRP. In fact, non-activated PRP showed better result compared to activated PRP(38). Contrarily, another study conducted in 2017 reported superiority of activated PRP. It stated that activated PRP released more growth factors, thus increasing PRP regenerative potential(39). In terms of wound characteristic, five studies observed chronic wounds and the other five studies observed acute wounds. Characteristic of the wounds might play a key factor for determining antimicrobial activity of PRP. Difference growth factor secreted between the acute wound and chronic wound might result as completely different environment, which might alter the pattern of microbial organism growth. Further quantitative analysis needs to be done in order to investigate the true effect of PRP antimicrobial activity.

Quantitative Synthesis

Even though ten studies were included in qualitative synthesis, quantitative synthesis in this review only included nine trials (Fig. 3A and Fig. 3B). Due to its high risk of bias, one study was excluded in a meta-analysis(29). Based on author interpretation of the forest plot, PRP is more favourable than standard care in preventing wound infection (RR: 0.84; 95% CI: 0.66–1.06), shown by the black diamond lie on the left side. Both groups had 599 participants but have a different number of events. PRP group had 90 wound infection occurrences while it was 108 occurrences in the standard care group. However, this result was not statistically significant, as it has p-value was higher than 0.05 (p-value: 0.14). In summary, PRP had no additional benefits when it applied topically to prevent wound infection. Therefore, the author did a subgroup analysis to investigate the PRP antimicrobial effect further and in more details. In addition, the primary outcomes included in this review had some concern of risk of bias made which make the quality of evidence relatively low.

Secondary outcome

Qualitative Synthesis

Out of ten trials included in this studies, there were

only two studies which reported proportion of healed wound (25, 28) and only three studies reported wound size reduction (25, 27, 31). There are 322 participants involved in the two studies which reported a proportion of healed wound outcome, which consist of 160 participants from the intervention group and 162 participants from the control group. The result from both studies was consistently reported that PRP had an additional benefit over standard care to increase wound healing. After 12 weeks follow up, Ahmed et al. reported a proportion of healed wound in both PRP and standard care. The result is 24 patients wound were healed from a total of 28 patients in the PRP group, while only 19 patients wound were healed from a total of 28 patients in the control group. This difference in this study is statistically significant (p-value: 0.041). PRP's regenerative potential superiority is also shown in the study conducted by Game et al. Following 12 weeks period of follow-up, 27 wound patients from PRP group were reported to achieve full recovery while only 17 out of 134 patients managed to achieve full recovery in the control group. However, the difference in this study was not statistically significant (p-value: 0.082). Even though the same trend of superiority was shown by both studies, there is a significant difference in wound healed percentage even though the follow-up period were the same. Both studies used the same population, which is a diabetic wound patient. However, there is a source that might explain this difference. Game et al. used Non-activated PRP for intervention, while Ahmed et al. used Activated PRP. Further analysis is needed to further investigate this result.

In three studies which investigate wound size reduction, the result is also consistent. PRP was reported able to add additional benefit when it applied topically to reduce wound size. Ahmed et al. reported wound size in the PRP group ranged from 2.2–10.2 cm² while the control group ranged from 2.47–11.55 cm². This result was obtained after twelve weeks follow-up period with reasonably similar baseline characteristic in both groups. Other study conducted by Burgos-Alonso et al. reported PRP averagely reduced wound size about 3.9 cm² after nine weeks, while standard care only reduced 3.2 cm² on average. However, the difference was not statistically significant (p-value: 0.6818). Rainys et al. also reported a similar result of PRP regenerative potential. In the PRP group, nine out of 35 patients achieved full re-epithelialisation; while in the control group, only six out of 34 patients managed to achieve full re-epithelialisation. Even though there is a difference, it is also not significant like the previous study conducted by Burgos-Alonso et al. (p-value: 0.417). There are 133 total participants combined from both groups, consist of 66 participants from the PRP group and 67 participants from the control group. Combining the results from those studies could provide a clearer understanding of PRP regenerative potential and its effect on wound healing. Therefore, further analysis is needed to investigate the

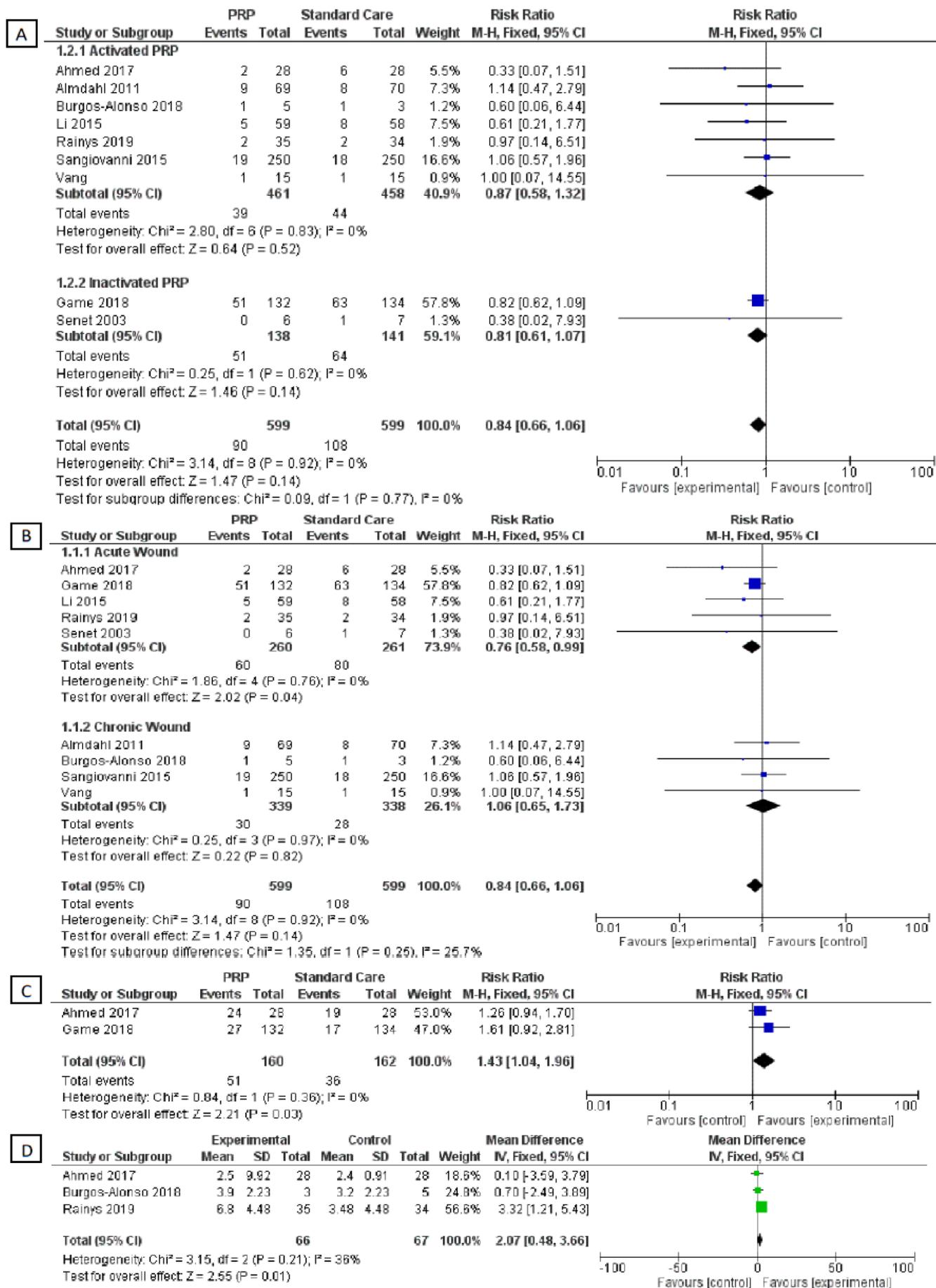


FIG.3: The generated forest plot

difference, whether it is statistically significant or not.

Quantitative Synthesis

In the secondary outcome of quantitative synthesis (Fig. 3C and Fig. 3D), there are four trials included (25, 27, 28, 31). The reason being is only those four trials were provided secondary outcome data. In a proportion of healed wound forest plot (Fig. 3C), the graph clearly depicted that PRP is significantly beneficial compared to standard care (p-value: 0.03). Patients who treated with PRP were 1.43 times more likely to achieved wound recovery in the same period of follow up (95% CI: 1.04–1.96). PRP regenerative effect was further evaluated based on wound size reduction. Wound size-reduction forest plot (Fig. 3D) illustrated that PRP is more favourable compared to standard care in terms of reducing wound size (MD: 2.07; 95% CI: 0.48–3.66). This difference is also statistically significant (p-value: 0.01). In summary, PRP has a significant regenerative effect to enhance wound healing. Therefore, the author felt there is no necessity to perform subgroup analysis for the secondary outcome.

Heterogeneity

There is no reported imprecision and inconsistency in the results (p-values given for Chi-square were all > 0.1 and I2 test results were all < 40%).

Subgroup analysis

The author performed subgroup analysis to investigate topical PRP antimicrobial potential based on treatment type (Activated PRP vs Non-activated PRP) and wound characteristic (Acute wound vs Chronic Wound).

Treatment type: Activated PRP vs Non-activated PRP

This subgroup analysis purpose was to investigate the effectiveness of activated PRP and Non-activated PRP in preventing wound infection if they were applied topically. Both activated PRP and inactivated PRP produced a similar result in terms of preventing wound infection. There were seven studies which compared Activated PRP with standard care (25-27, 30-32, 37).

On the other hand, there were only two studies which compared Non-Activated PRP with standard care (28, 33). In studies which compared Activated PRP with standard care, the author found 39 cases of wound infection from a total of 461 patients in the experimental group and 44 cases of wound infection from a total of 458 patients in the control group. Meanwhile, in studies which compared Non-Activated PRP with standard care, the author found 51 wound infection occurrences from a total of 138 patients in the intervention group and 64 wound infection occurrences from a total of 141 patients in the control group. From the illustration depicted in Fig. 3A, we can conclude that both Activated PRP (p-value: 0.52) and Non-Activated PRP (p-value: 0.14) have no significant effect in reducing the risk of wound infection. There was no heterogeneity found in Activated PRP and Non-Activated PRP subgroup analysis.

Wound Characteristic: Acute Wound vs Chronic Wound

This subgroup analysis aim was to analyse further the effectiveness of topical PRP antimicrobial activity on the acute wound and chronic wound (Fig. 3B). There are 521 participants with an acute wound which consist of 260 participants in the PRP group and 261 participants in the control group. Wound infection occurrences in the PRP group and control group were 60 and 80, respectively. Meanwhile, there are 677 participants with a chronic wound. Out of 677 participants, 339 were treated with PRP. Thirty people from the PRP group developed wound infection while only 28 people from the control group developed wound infection. It revealed that PRP could decrease the risk of wound infection in acute wound significantly (p-value: 0.04). When applied topically, PRP could decrease the risk of getting wound infection 0.76 lower (95% CI: 0.58–0.99) compared to the standard care. In other words, patients who treated with standard care are 1.3 times more likely to get wound infection compared to patients who treated with PRP. We found dissimilar result on chronic wound case where it showed almost no effect (RR: 0.106; 95% CI: 0.65–1.73; p-value: 0.82). There was no heterogeneity found in acute wound and chronic wound subgroup

PRP compared to standard care for preventing wound infection					
Patient or population: preventing wound infection					
Setting: clinical intervention					
Intervention: PRP					
Comparison: standard care					
Outcomes	No of participants (studies) Follow up	Certainty of the evidence (GRADE)	Relative effect (95% CI)	Anticipated absolute effects	
				Risk with standard care	Risk difference with PRP
Proportion of wound infection (wound infection)	1198 (9 RCTs)	⊕⊕⊕○ LOW	RR 0.96 (0.66 to 1.39)	80 per 1,000	3 fewer per 1,000 (27 fewer to 31 more)
Proportion of healed wound (Healed wound)	322 (2 RCTs)	⊕⊕⊕○ LOW	RR 1.31 (1.03 to 1.62)	222 per 1,000	69 more per 1,000 (7 more to 138 more)
Wound size reduction (Wound size)	133 (2 RCTs)	⊕⊕⊕○ LOW	-	The mean wound size reduction was 1	MD 2.07 higher (0.48 higher to 3.66 higher)

*The risk in the intervention group (and its 95% confidence interval) is based on the assumed risk in the comparison group and the relative effect of the intervention (and its 95% CI).

CI: Confidence interval; RR: Risk ratio; MD: Mean difference

GRADE Working Group grades of evidence
High certainty: We are very confident that the true effect lies close to that of the estimate of the effect
Moderate certainty: We are moderately confident in the effect estimate: The true effect is likely to be close to the estimate of the effect, but there is a possibility that it is substantially different
Low certainty: Our confidence in the effect estimate is limited: The true effect may be substantially different from the estimate of the effect
Very low certainty: We have very little confidence in the effect estimate: The true effect is likely to be substantially different from the estimate of effect

FIG.4: The GRADE approach

analysis.

Summary of findings

Based on the Cochrane group recommendation, we summarised and appraised the quality (strength) of our evidence with the GRADE approach (Fig. 4).

DISCUSSION

The meta-analysis on Activated PRP and Non-Activated PRP showed no difference with meta-analysis result prior to subgroup analysis. There was no substantial heterogeneity found in both. The generated forest plot (Fig. 3A) also suggested there was no statistically significant subgroup effect (p-value: 0.52). In other words, PRP activation did not alter PRP antimicrobial activity. The author found different cases when did a subgroup analysis based on wound characteristic. The generated forest plot (Fig. 3B) illustrated that PRP was an effective intervention when it is applied on the acute wound (p-value: 0.04), but it is not as effective when it was applied on a chronic wound (p-value 0.82). There was no heterogeneity found in both analyses.

The author also did a meta-analysis to investigate PRP regenerative potential. There are two studies which reported a proportion of healed wound and three studies which reported wound size reduction. Based on the quantitative analysis, the proportion of healed wound in the PRP group was significantly greater than the wound care group. The author recorded 51 out of 160 participants in the PRP group had complete wound recovery while only 35 from 162 participants in the control group had complete wound healing (Fig. 3C). This difference was also statistically significant (p-value: 0.03). The author also did an analysis to further investigate PRP regenerative potential by doing an analysis with different outcome measure, which is wound size reduction. PRP showed similar superiority compared to standard care, and the result was also statistically significant (MD: 2.07; 95%CI: 0.48–3.66) (Fig. 3D).

Currently, there is no systematic review which specifically investigates the effectiveness of topical PRP to reduce the risk of wound infection. There is one review which investigates the effectiveness of autologous PRP for treating chronic wounds. However, the review focus lay on PRP regenerative capability instead of antimicrobial potency(19). Furthermore, the review unable to draw a conclusion due to sparseness and low quality of existing trials. Even though it was full of uncertainty, the review concluded PRP was not effective as an intervention to enhance wound healing, particularly in a chronic wound. This finding was contradictive with what the author concluded based on ten trials included in this review. The author found the proportion of healed wound in the PRP group was significantly higher than those who received standard care treatment. In addition,

the author also used a different method to measure PRP regenerative capability, which is by measuring wound size reduction. The author found quite a similar result. Participants who treated with PRP had significantly smaller wound size compared to participants who received standard care treatment. Therefore, the author concluded PRP has a positive impact on wound healing. However, the trials included in the meta-analysis which investigate PRP regenerative ability have one common thing, which is all of those trial population was patients with a chronic wound. Therefore, it is not wise to generalise this conclusion on a patient with an acute wound problem. Even though there is a study which has a different conclusion and results with this review, there is also a study which draws a similar conclusion and results with this review(18). The study conducted by Carter et al. was concluded that PRP has a positive effect on wound healing.

The primary aim of this study was to investigate topical autologous PRP to reduce the risk of wound infection. The author found that PRP only effective as prophylaxis for wound infection in acute wound population, but not chronic wound. The author found similar results about PRP antimicrobial effect in several studies. A study reported PRP provided additional benefit in exterminating infection caused by a microorganism(40). This contention also supported by another study which strongly suggested that PRP is an effective intervention as antimicrobial drugs against several bacteria such as MSSA, MRSA, Group A Streptococcus and Neisseria Gonorrhoea in both in vitro and in vivo setting(41). However, a more specific study reported PRP is only effective against bacteria in planktonic form. PRP was reported as having little effect in exterminating bacteria in biofilm form(16). Those studies could explain the results found by the author in this review. The author found PRP only effective in an acute wound, but not chronic wound. A discrepancy of PRP effects on acute and chronic wound might be related to biofilm formation.

Biofilms are defined as three-dimensional colonies of bacteria enveloped in an extracellular matrix called extracellular polymeric substance (EPS)(42). Biofilm started from planktonic bacteria which attach to a surface and then multiplied. Multiplied bacteria then forming a colony and later secrete EPS after reached a condition called quorum sensing. EPS provided not only a physical barrier, but also allow free transfer of genetic material, oxygen, and nutrients between bacteria in a biofilm. Furthermore, biofilm also allows free transfer of antibiotic resistance gene-carrying plasmids, making biofilm eradication more complicated(43). Based on studies conducted by Pham et al., PRP ineffectiveness against biofilm was highly likely due to the existence of EPS which enhance bacterial defence system. Biofilm can form within the first 24 hours after planktonic bacteria attachment(44).

A chronic wound is the discontinuation of skin which do not heal/require a long period of time to heal and often recurring. Specifically, the chronic wound is defined as skin and soft tissues injuries which do not recover to their functional and anatomical state at least in one month(45). Within one month, the wound might get infected by planktonic bacteria which transform to a biofilm. We also have to keep in mind that not all of the wound infection shows a typical symptom. There are a lot of cases of a wound which do not show any sign of infection but has a positive bacterial swab test. Pre-existing biofilm might be present in a chronic wound, hence making PRP antimicrobial properties ineffective.

It is also interesting to look further on studies with an acute wound in this review. There are four studies which observe a patient with an acute wound. The acute wound could be further categorised as a post-surgical and post-traumatic wound. Participants from three out of four studies which observe a patient with acute wound were categorised as a patient with a post-surgical wound. Surgery room setting was maintained sterile; thus, the infection rate was controlled well. There is a little chance of the presence of pre-existing bacteria prior to application of PRP in those studies. Therefore, PRP treatment in those populations was effective. There is still a need for further research to investigate whether PRP antimicrobial activity in reducing the risk of wound infection is effective only on post-surgical wound population or on any acute wound population (35). In this review, the author also found there is no statistically significant difference in outcome whether PRP activation process was conducted or not. The possible explanation for this phenomenon is due to the presence of natural PRP activator such as thrombin in the wound environment. Inflammatory agents could activate resident thrombocytes which later release growth factors such as thromboxane A-2. This cascade activation will finally release thrombin which is an important factor in activating platelet. This finding also supported by previous studies which reported an insignificant effect of PRP activation (38, 39).

Strength, limitation, and future directions

Our literature search was conducted based on robust search strategy without language or timeline restrictions. Methodology and principle used in this review followed the Cochrane recommendation. Therefore, this review provided enough information to answer the research question based on evidence in the current time. As illustrated in the Summary of Finding table (Fig. 4), the assessment of evidence quality was conducted in accord with the GRADE method. There were no imprecisions, inconsistencies, and indirectness issue identified by the author. The author did not find clinical nor methodological heterogeneity. However, the author strongly suspected that risk of bias might have an important effect that could lead to alteration in an estimate of effect. Furthermore, the author found various

outcome measurement method in studies included in this review. While all of them might be legitimate, the difference in outcome measurement method can lead to bias outcome.

Nevertheless, this review provided valuable information about several important factors that might affect intervention effect in the clinical setting such as age, population characteristic, details of the intervention, setting, etc. Evidence collected in this review showed a promising result of PRP application to prevent wound infection in an acute wound. Trials in this review were conducted in various continents (America, Europe, Africa, and Asia). Therefore, it is reasonable to say the result from this review could be applied to various population from different races. However, all of the countries which conducted the trials were a well-developed country. It might not be feasible to apply in the under-developed country due to limitation in medical technology. In addition, all of the studies settings were in hospital or secondary/tertiary level health care. It needs further consideration to applied PRP for wound care in a primary care setting. Another limitation of this review is its applicability on children and patients with comorbidities. Further research is needed to apply this intervention in those population.

This review showed that PRP has a potent antimicrobial activity when it is applied topically on the acute wound using autologous PRP. However, further research on PRP optimal dose for preventing wound infection while still maintaining its regenerative as well as using allogeneic topical PRP is needed. In addition, this study only covers PRP topical application on a wound, while another method such as injection could be more effective in reducing the risk of wound infection and provide better wound healing capability. Research related to combination of PRP and EPS-degrading substance should be studied further in the future. Potential EPS degrading substances such as MMP-1 and MMP-9 should be considered to be combined with PRP. This research would be paramount because the only hurdle for PRP to kill biofilm bacteria is biofilm ability to degrade EPS. Furthermore, there is still not yet evidence that reported which type of PRP (among the four subcategories, i.e. P-PRP, L-PRP, P-PRF, and L-PRF)(46)) is the most prominent one for treating wound infection. Lastly, this review was written based on ten included trials which have a moderate risk and high risk of bias. In the future, high-quality primary RCTs are needed to produce a better systematic review.

CONCLUSION

The author found evidence that autologous PRP can significantly reduce the risk of wound infection if it applied topically on the acute wound. However, that is not the case in chronic wound due to the possibility of pre-existing biofilm bacteria. The author also found

that Activated PRP and Non-Activated PRP have no difference in reducing the risk of wound infection. The author recommended adding topical PRP as part of a new standard treatment for acute wound care. Besides its enormous regenerative potential, PRP also has potent antimicrobial activity. The author also suggests forbidding irrational prophylaxis antibiotic application in a clean acute wound.

Based on the evidence in this review, PRP activation was not essential to reduce the risk of wound infection. However, PRP activation might increase the quality of other clinical outcomes, such as wound reduction size and percentage of epithelialisation. Further research in this area is needed to rationalise the utilisation of PRP activation. However, the certainty of evidence in this review is low. Therefore, further good quality RCTs in this aspect are still needed to provide better evidence. As for now, this is the current best evidence which can be used as a foundation to improve wound care guidelines. Inclusion of PRP in the new standard treatment for wound care should be done carefully and started from a small population.

REFERENCES

1. Kujath P, Michelsen A. Wounds - from physiology to wound dressing. *Dtsch Arztebl Int*. 2008;105(13):239-48.
2. Lazarus GS, Cooper DM, Knighton DR, Margolis DJ, Pecoraro RE, Rodeheaver G, et al. Definitions and Guidelines for Assessment of Wounds and Evaluation of Healing. *Archives of Dermatology*. 1994;130(4):489-93.
3. Guest JF, Ayoub N, McIlwraith T, Uchegbu I, Gerrish A, Weidlich D, et al. Health economic burden that wounds impose on the National Health Service in the UK. *BMJ Open*. 2015;5(12):e009283.
4. Sen CK, Gordillo GM, Roy S, Kirsner R, Lambert L, Hunt TK, et al. Human skin wounds: a major and snowballing threat to public health and the economy. *Wound Repair Regen*. 2009;17(6):763-71.
5. Gonzalez ACdO, Costa TF, Andrade Zda, Medrado ARAP. Wound healing - A literature review. *An Bras Dermatol*. 2016;91(5):614-20.
6. Medrado A, Costa T, Prado T, Reis S, Andrade Z. Phenotype characterization of pericytes during tissue repair following low-level laser therapy. *Photodermatology, Photoimmunology & Photomedicine*. 2010;26(4):192-7.
7. Healy B, Freedman A. Infections. *BMJ*. 2006;332(7545):838-41.
8. Guo S, Dipietro LA. Factors affecting wound healing. *J Dent Res*. 2010;89(3):219-29.
9. Menke NB, Ward KR, Witten TM, Bonchev DG, Diegelmann RF. Impaired wound healing. *Clinics in Dermatology*. 2007;25(1):19-25.
10. Le ADK, Enweze L, DeBaun MR, Dragoo JL. Current Clinical Recommendations for Use of Platelet-Rich Plasma. *Curr Rev Musculoskelet Med*. 2018;11(4):624-34.
11. Yeaman MR. Platelets: at the nexus of antimicrobial defence. *Nature Reviews Microbiology*. 2014;12(6):426-37.
12. Tang Y-Q, Yeaman MR, Selsted ME. Antimicrobial Peptides from Human Platelets. *Infection and Immunity*. 2002;70(12):6524.
13. Krijgsveld J, Zaat SA, Meeldijk J, van Veelen PA, Fang G, Poolman B, et al. Thrombocidins, microbicidal proteins from human blood platelets, are C-terminal deletion products of CXC chemokines. *J Biol Chem*. 2000;275(27):20374-81.
14. Cieřlik-Bielecka A, Bold T, Zylkowski G, Pierchała M, Krylikowska A, Reichert P. Antibacterial Activity of Leukocyte- and Platelet-Rich Plasma: An In Vitro Study. *Biomed Res Int*. 2018;2018:9471723-.
15. Farghali HA, AbdElKader NA, AbuBakr HO, Aljuaydi SH, Khattab MS, Elhelw R, et al. Antimicrobial action of autologous platelet-rich plasma on MRSA-infected skin wounds in dogs. *Scientific Reports*. 2019;9(1):12722.
16. Pham TAV, Tran TTP, Luong NTM. Antimicrobial Effect of Platelet-Rich Plasma against *Porphyromonas gingivalis*. *Int J Dent*. 2019;2019:7329103-.
17. Mariani E, Filardo G, Canella V, Berlingeri A, Bielli A, Cattini L, et al. Platelet-rich plasma affects bacterial growth in vitro. *Cytotherapy*. 2014;16(9):1294-304.
18. Carter MJ, Fylling CP, Parnell LKS. Use of platelet rich plasma gel on wound healing: a systematic review and meta-analysis. *Eplasty*. 2011;11:e38-e.
19. Martinez-Zapata MJ, Martiñ-Carvajal AJ, Sola I, Expósito JA, Bohar I, Rodríguez L, et al. Autologous platelet-rich plasma for treating chronic wounds. *Cochrane Database of Systematic Reviews*. 2016(5).
20. Smith OJ, Kanapathy M, Khajuria A, Prokopenko M, Hachach-Haram N, Mann H, et al. Systematic review of the efficacy of fat grafting and platelet-rich plasma for wound healing. *International Wound Journal*. 2018;15(4):519-26.
21. Kim H. Wound Infection. *Arch Plast Surg*. 2019;46(5):484-5.
22. Ouzzani M, Hammady H, Fedorowicz Z, Elmagarmid A. Rayyan—a web and mobile app for systematic reviews. *Systematic Reviews*. 2016;5(1):210.
23. Sterne JAC, Savović J, Page MJ, Elbers RG, Blencowe NS, Boutron I, et al. RoB 2: a revised tool for assessing risk of bias in randomised trials. *BMJ*. 2019;366:l4898.
24. Higgins JPT, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. *BMJ*. 2003;327(7414):557-60.
25. Ahmed M, Reffat S, Hassan A, Eskander F. Platelet-

- Rich Plasma for the Treatment of Clean Diabetic Foot Ulcers. 2017;38:206-11.
26. Almdahl S, Veel T, Halvorsen P, Vold M, Mølsted P. Randomized prospective trial of saphenous vein harvest site infection after wound closure with and without topical application of autologous platelet-rich plasma. 2011;39(1):44-8.
 27. Burgos-Alonso N, Lobato I, Hernández I, Sebastian K, Rodríguez B, March A, et al. Autologous platelet-rich plasma in the treatment of venous leg ulcers in primary care: a randomised controlled, pilot study. 2018;27:S20-S4.
 28. Game F, Jeffcoate W, Tarnow L, Jacobsen J, Whitham D, Harrison E, et al. LeucoPatch system for the management of hard-to-heal diabetic foot ulcers in the UK, Denmark, and Sweden: an observer-masked, randomised controlled trial. 2018;6(11):870-8.
 29. Kazakos K, Lyras D, Verettas D, Tilkeridis K, Tryfonidis M. The use of autologous PRP gel as an aid in the management of acute trauma wounds. 2009;40(8):801-5.
 30. Li L, Chen D, Wang C, Yuan N, Wang Y, He L, et al. Autologous platelet-rich gel for treatment of diabetic chronic refractory cutaneous ulcers: a prospective, randomized clinical trial. 2015;23(4):495-505.
 31. Rainys D, Cepas A, Dambrauskaite K, Nedzelskiene I, Rimdeika R. Effectiveness of autologous platelet-rich plasma gel in the treatment of hard-to-heal leg ulcers: a randomised control trial. 2019;28(10):658-67.
 32. Sangiovanni T, Kiebzak G. Prospective Randomized Evaluation of Intraoperative Application of Autologous Platelet-Rich Plasma on Surgical Site Infection or Delayed Wound Healing. 2015;37(5):470-7.
 33. Senet P, Bon F, Benbunan M, Bussel A, Traineau R, Calvo F, et al. Randomized trial and local biological effect of autologous platelets used as adjuvant therapy for chronic venous leg ulcers. 2003;38(6):1342-8.
 34. Vang SN, Brady CP, Christensen KA, Allen KR, Anderson JE, Isler JR, et al. Autologous platelet gel in coronary artery bypass grafting: effects on surgical wound healing. *J Extra Corpor Technol.* 39(1):31-8.
 35. Almdahl SM, Veel T, Halvorsen P, Vold MB, Mølsted P. Randomized prospective trial of saphenous vein harvest site infection after wound closure with and without topical application of autologous platelet-rich plasma. 2011;39(1):44-8.
 36. Ahmed M, Reffat SA, Hassan A, Eskander F. Platelet-Rich Plasma for the Treatment of Clean Diabetic Foot Ulcers. 2017;38:206-11.
 37. Vang SN, Brady CP, Christensen KA, Allen KR, Anderson JE, Isler JR, et al. Autologous platelet gel in coronary artery bypass grafting: effects on surgical wound healing. *J Extra Corpor Technol.* 2007;39(1):31-8.
 38. Gentile P, Cole JP, Cole MA, Garcovich S, Bielli A, Scioli MG, et al. Evaluation of Not-Activated and Activated PRP in Hair Loss Treatment: Role of Growth Factor and Cytokine Concentrations Obtained by Different Collection Systems. *Int J Mol Sci.* 2017;18(2):408.
 39. Vahabi S, Yadegari Z, Mohammad-Rahimi H. Comparison of the effect of activated or non-activated PRP in various concentrations on osteoblast and fibroblast cell line proliferation. *Cell and Tissue Banking.* 2017;18(3):347-53.
 40. Varshney S, Dwivedi A, Pandey V. Antimicrobial effects of various platelet rich concentrates-vides from in-vitro studies-a systematic review. *Journal of oral biology and craniofacial research.* 2019;9(4):299-305.
 41. Li H, Hamza T, Tidwell JE, Clovis N, Li B. Unique antimicrobial effects of platelet-rich plasma and its efficacy as a prophylaxis to prevent implant-associated spinal infection. *Adv Healthc Mater.* 2013;2(9):1277-84.
 42. Chen L, Wen Y-m. The role of bacterial biofilm in persistent infections and control strategies. *Int J Oral Sci.* 2011;3(2):66-73.
 43. Stalder T, Top E. Plasmid transfer in biofilms: a perspective on limitations and opportunities. *NPJ Biofilms Microbiomes.* 2016;2:16022.
 44. Chandki R, Banthia P, Banthia R. Biofilms: A microbial home. *J Indian Soc Periodontol.* 2011;15(2):111-4.
 45. Clinton A, Carter T. Chronic Wound Biofilms: Pathogenesis and Potential Therapies. *Laboratory Medicine.* 2015;46(4):277-84.
 46. David MDE, Tomasz B, Allan M, Piero B, Francesco I, Gilberto S, et al. In Search of a Consensus Terminology in the Field of Platelet Concentrates for Surgical Use: Platelet-Rich Plasma (PRP), Platelet-Rich Fibrin (PRF), Fibrin Gel Polymerization and Leukocytes. *Current Pharmaceutical Biotechnology.* 2012;13(7):1131-7.