

# Efficacy of Activated Autologous Platelet-Rich Plasma in The Treatment of Diabetic Foot Ulcers: An Evidence-Based Brief Literature Review

Dewi Fibrini\*, Achsanuddin Hanafie



Use your smartphone to scan this QR code and download this article

## ABSTRACT

Diabetic foot ulcers (DFUs) present as a debilitating complication of diabetes which approximately affects 6.3% of the global population with significant mortality and morbidity consequences. While one-third of the total cost of diabetic care goes to DFU treatment, 20% patients have unresolved ulcers, while 40% experience recurrency within one year. To improve outcomes in DFU care, activated autologous platelet-rich plasma (aaPRP) is one of the adjuvant therapies proposed. Containing various bioactive proteins such as growth factors and cytokines, it possesses regenerative, antioxidative, anti-inflammatory, and antimicrobial qualities. Furthermore, its autologous nature, which also makes it a highly safe treatment modality, is also promising. In this review, we shortly discuss the pathogenesis of DFUs and how aaPRP may tackle the key players in DFU pathogenesis to improve wound healing outcomes. We also briefly describe its method of isolation and the current views on aaPRP from an authoritative source. Lastly, we summarize the existing evidence for the utilization of aaPRP in the treatment of DFUs.

**Key words:** Diabetic Foot Ulcer, Activated Autologous Platelet-Rich Plasma, Adjuvant Therapy

## INTRODUCTION

Diabetic foot ulcers (DFUs) present as a debilitating complication of diabetes which affects 6.3% of the global population. Some of the cited risk factors of DFUs include lower body mass index, longer diabetic duration, hypertension, and a history of smoking<sup>1</sup>. It is a severe public health issue which carries with it an immense burden of mortality and morbidity. Approximately 90,000 amputations are performed annually as a result of DFUs. This condition may also lead to other complications such as infection, abscesses, and pedal deformities<sup>2</sup>. The mortality rate associated with the development of DFUs is approximately 5% in 1 year, and 42% in 5 years<sup>3</sup>.

Given the current standard practices in the management of DFUs, it is estimated that DFU treatment takes up one-third of the total cost of diabetic care, which translated to US \$176 billion in 2012. In spite of this high cost, around 20% of DFU patients have wounds that remain unhealed after one year. Besides resolution, recurrence is also an issue as about 40% of patients experience recurrency within one year<sup>3</sup>.

To improve outcomes in DFU care, several adjuvant therapies have been proposed, among which is activated autologous platelet-rich plasma (aaPRP). Advances in biotechnology has enabled scientists to uti-

lize platelets for a wide array of uses, such as alopecia<sup>4</sup>, tendinopathy<sup>5</sup>, and even cytokine storm<sup>6-8</sup>. By processing blood samples from the patients, various bioactive proteins which possess favorable properties such as regeneration, can be released<sup>3,9</sup>. This review aims to evaluate the current evidences regarding the efficacy of aaPRP in the treatment of DFUs.

## REVIEW

For this review, the author assembled credible literature from various databases such as PubMed, Scopus, and Wiley Online Library. The keywords used were “Diabetic Foot Ulcer”, “Platelet-Rich Plasma”, and “Platelet-Rich Plasma AND Diabetic Foot Ulcer”. Afterwards, the author handpicked relevant articles to be included in the review. Articles not written in English or that were deemed to be irrelevant were excluded.

In the end, a total of 50 studies were referenced in this review, of which eight are research articles focused on presenting current evidence of aaPRP in the treatment of DFUs. The other 42 are mainly review articles referenced in other parts of this article.

## Pathogenesis of Diabetic Foot Ulcers

The pathogenesis of DFUs is complicated as it is a combination of several risk factors, such as peripheral

Faculty of Medicine, Universitas Prima Indonesia, Jl. Ayahanda No. 68a, Medan, Indonesia, 20118

### Correspondence

Dewi Fibrini, Faculty of Medicine, Universitas Prima Indonesia, Jl. Ayahanda No. 68a, Medan, Indonesia, 20118

Email: dewifibrini@gmail.com

### History

- Received: Jan 31, 2022
- Accepted: May 11, 2022
- Published: June 30, 2022

DOI : 10.15419/br.v8i1.788



### Copyright

© Biomedpress. This is an open-access article distributed under the terms of the Creative Commons Attribution 4.0 International license.



Cite this article : Fibrini D, Hana ie A. Efficacy of Activated Autologous Platelet-Rich Plasma in The Treatment of Diabetic Foot Ulcers: An Evidence-Based Brief Literature Review. *Biotech. Res.* 2022; 8(1):788.

neuropathy, peripheral vascular disease, pedal deformity, trauma, weakened resistance to infection, and more<sup>2</sup>. Here, some of the most dominant risk factors in the pathogenesis of DFU are discussed.

### Diabetic Peripheral Neuropathy

Diabetic peripheral neuropathy (DPN) afflicts up to 66% and 59% of patients with type 1 and type 2 diabetes respectively<sup>10</sup>. The peripheral neurons supplying the feet are the longest cells in the body. As such, they require adequate vascularization, mitochondrial support, and glucose and lipid metabolism, all of which are unfortunately disturbed in diabetes<sup>11</sup>. As pointed out by experimental and clinical studies, the most important factor in DPN pathogenesis is hyperglycemia-mediated cellular-injury<sup>12,13</sup>. Other factors include obesity, dyslipidemia, impaired neurotrophic support, and microangiopathy which induce oxidative stress, mitochondrial dysfunction, and inflammation<sup>11</sup>.

One way that hyperglycemia causes DPN is through the polyol pathway, wherein excessive intracellular neuron glucose is converted into sorbitol, and into fructose afterwards by aldose reductase and sorbitol dehydrogenase, respectively. The ultimate increase in polyol flux causes intracellular hyperosmolarity and compensatory efflux of osmolytes, one of which is myoinositol, resulting in phosphatidylinositol depletion, hindering the creation of adenosine triphosphate. All of these processes result in Na<sup>+</sup>/K<sup>+</sup> ATPase and protein kinase C dysfunction, impaired axonal transport, and neuronal breakdown. Furthermore, the reduction of glucose to sorbitol is also associated with NADPH consumption (which is required for the glutathione regeneration), thus contributing to oxidative stress<sup>14,15</sup>.

In the presence of excess glucose, shunting to the hexosamine pathway also occurs. The conversion of fructose-6-phosphate to glucosamine-6-phosphate by glutamine: fructose-6-phosphate amidotransferase increases the production of uridine diphosphate N-acetylglucosamine that in turns modifies the transcription factor Sp1. This change results in altered gene expression and protein function. Consequently, vascular dysfunction, inflammation, and oxidative occurs<sup>11,16</sup>.

Other than that, in a hyperglycemic environment, proteins, lipids, and nucleic acids can undergo irreversible non-enzymatic reactions which form advanced glycation products. These group of molecules, which are generated via the attachment of reactive carbohydrate groups, tend to affect cellular function

negatively by disrupting the biological tasks of proteins. They also bind to their receptors which activates the intracellular signaling pathway, leading to inflammation, oxidative, stress, and nuclear degradation. This chain of events results in vascular dysfunction and deficits in neuronal conduction<sup>11,14</sup>.

### Peripheral Vascular Disease

Peripheral vascular disease (PVD) is an important risk factor in the development of DFU. PVD is responsible for about 50% of DFU cases and 70% of deaths in type 2 diabetes<sup>2</sup>. Hyperglycemia, dyslipidemia, and insulin resistance found in diabetics foster the development of PVD through mechanisms similar to those found in coronary artery disease<sup>17</sup>. The risk of PVD is higher and earlier in patients with diabetes due to endothelial dysfunction, vascular smooth muscle dysfunction, inflammation, and hypercoagulability. Other than diabetes, high blood pressure, smoking, dyslipidemia, and older age are also risk factors for PVD<sup>18</sup>. Diabetics, who are at a higher risk of atherosclerosis, may suffer from acute or chronic ischemia which may also lead to gangrene. Poor peripheral vascularization also leads to a poor wound healing process and further worsens the situation.<sup>2</sup> Once it has become a neuroischemic ulcer, the wound is at risk of infection. While ischemia is not an independent risk factor for amputation, simultaneous occurrence with infection greatly increases the risk of limb amputation. Deep infections, such as osteomyelitis or soft tissue infection, are an immediate cause of amputation in 25 — 50% of DFU cases<sup>19</sup>.

### Other Risk Factors

Other risk factors identified by studies include Charcot joints, a long duration of diabetes, retinopathy, nephropathy, hypertension, overweight, obesity, low socioeconomic level, foot deformity, and a history of previous DFUs and/or amputation<sup>20-22</sup>.

### Current Views on Platelet-Rich Plasma in the Treatment of Diabetic Foot Ulcers

In 2016, the Wound Healing Society released guidelines regarding the treatment of DFUs<sup>23</sup>. The guidelines cover diagnosis, offloading, infection control, wound bed preparation, dressings, surgery, adjuvant agents, and the prevention of the recurrence of DFU. The evidence-based guidelines used well-controlled animal studies in addition to clinical studies. The strength of the evidence was then listed as Level I, Level II, or Level III with Level I evidence being the strongest due to being derived from meta-analyses or

multiple laboratory studies supported by at least two clinical series.

Of interest to this review is the “Adjuvant Agents” section of the guidelines which cover topical, device, and systemic adjuvants for the treatment of DFUs. Level I evidence (very strong) was given to the application of platelet derived growth factor (PDGF)<sup>24-26</sup>, cytokines, and growth factors<sup>27-29</sup> to improve DFU healing outcomes.

However, the guidelines stated that a systematic review of multiple trials studying the effects of aaPRP in the treatment DFU have not demonstrated improvements in the wound healing effects<sup>30</sup>. This finding is contradictory to the current literature and principles regarding the mechanism of aaPRP which will be discussed in a later part of this manuscript.

### Isolation of Activated Autologous Platelet-Rich Plasma

There exists an abundance of aaPRP technique presented in the literatures but the end products are generally classified, as proposed by Ehrenfest *et al.*,<sup>31</sup> as follows:

- Pure platelet-rich plasma or leukocyte-poor platelet-rich plasma
- Leukocyte- and platelet-rich plasma
- Pure platelet-rich fibrin or leukocyte-poor platelet-rich fibrin
- Leukocyte- and platelet-rich fibrin

Current evidence shows that aaPRP preparations rich in leukocytes may impede healing by inducing inflammation through pro-inflammatory cytokines<sup>32</sup>.

One example of aaPRP isolation involves drawing whole blood from a patient through venipuncture into sodium citrate tubes, centrifuging the blood, separating the supernatant, and centrifuging it again at a faster speed. After this step, the lower-third of the tube will be considered platelet-rich plasma and the rest platelet-poor plasma, which would later be removed. The final product is a concentrated pellet of platelets suspended in plasma<sup>9</sup>.

### Current Evidence for The Utilization of Autologous Platelet-Rich Plasma in Diabetic Foot Ulcers

#### Principles and Molecular mechanism

In DFUs, the lack of oxygen and nutrients hinders the epithelial cells from expressing growth factors, such as vascular endothelial growth factor (VEGF) and PDGF, which are vital for wound healing<sup>33,34</sup>. With

over 1500 bioactive factors found in platelets, aaPRP contains the much-needed growth factors which are beneficial for DFUs as the wounds are deficient in growth factors. Some examples of the growth factors include PDGF which has the ability to enhance collagen synthesis, fibroblast chemotaxis, and proliferation; VEGF which stimulates angiogenesis, the mitosis of endothelial cells, and increases vascular permeability; and the insulin-like growth factor (IGF) which promotes cell growth and differentiation, and also works with PDGF to stimulate collagen synthesis. Other growth factors such as transforming growth factor- $\beta$  (TGF- $\beta$ ), epidermal growth factor (EGF), and fibroblast growth factor (FGF) are also present<sup>35</sup>. A summary of the regenerative functions of some of the bioactive molecules found in aaPRP are summarized in **Table 1**.

Other than growth factors, a diverse array of cytokines are also present in platelets. These proteins can help in the modulation of inflammation and regeneration. Some of these include interleukin (IL)-1 receptor antagonist, IL-10, tumor necrosis factor- $\alpha$ , IL-12p70, and IL-8<sup>44,45</sup>. For example, IL-10 is an anti-inflammatory cytokine that is essential in wound healing as observed in models of fetal wound healing. The tissue repair observed is also not limited to cutaneous but also tendon and myocardial regeneration. Clinical trials have also demonstrated that wounds treated with IL-10 have less scarring<sup>46</sup>.

In the case of leukocyte-rich aaPRP, it is hypothesized that the neutrophils present in the preparation may impede wound healing by releasing massive amounts of reactive oxygen species and nitric oxide. While these secretions are useful to prevent microbial infection and to clear debris from the wound, the high number of leukocytes may delay wound healing. One possible way to control the amount of neutrophils in leukocyte-rich aaPRP is to increase the number of macrophages in the preparation as they induce neutrophil apoptosis. Nevertheless, leukocyte-rich aaPRP has its own uses and benefits outside the context of DFU healing<sup>35</sup>.

#### Safety and Accessibility

Accessibility-wise, the study carried out by Linertová, *et al.*<sup>41</sup> in Spain deemed that aaPRP treatment for DFUs is a cost-saving alternative in most cases when obtained manually as opposed to commercial kits when compared to usual care.

aaPRP is highly safe for human administration. Being autologous in nature, aaPRP has virtually no chance for rejection, cross-contamination, or disease transmission. Safety-wise, there is no rejection and it is

**Table 1: Brief summary of the functions of select bioactive molecules** <sup>33-35</sup>

Bioactive molecule	Function
Platelet Derived Growth Factor (PDGF)	Collagen synthesis, bone cell proliferation, macrophage activation, fibroblast activity
Vascular Endothelial Growth Factor (VEGF)	Promotes angiogenesis, endothelial cell replication and migration, stimulates macrophage and neutrophil chemotaxis
Insulin-like Growth Factor (IGF)	Promotes cellular growth and differentiation, stimulates collagen synthesis with PDGF
Transforming Growth Factor- $\beta$ (TGF- $\beta$ )	Collagen synthesis, promotes angiogenesis, inhibition of osteoclast activity, stimulates leukocyte chemotaxis
Fibroblast Growth Factor (FGF)	Stimulates the replication of mesenchymal cells, chondrocytes, and osteoblasts, also chondrocyte and osteoblast differentiation

**Table 2: Summary of current evidence in this literature review of aaPRP for treating DFU**

Author (year)	Intervention	Control	Outcome
<b>Parameter: Efficacy on Wound Healing</b>			
Driver, <i>et al.</i> (2006) <sup>36</sup>	aaPRP gel dressing	Saline gel dressing	Significantly higher healing rate and shorter time-to-heal in intervention
Ahmed, <i>et al.</i> (2017) <sup>37</sup>	aaPRP gel activated with thrombin	Antiseptic ointment gel	Significantly better healing rate and less infection in intervention group
Gupta, <i>et al.</i> (2021) <sup>38</sup>	aaPRP dressing and total-contact casting	Saline dressing and total-contact dressing	Intervention is no more efficacious than control in treating diabetic foot
Yarahmadi, <i>et al.</i> (2021) <sup>39</sup>	aaPRP fibrin glue dressing along with oral vitamin E and vitamin C	aaPRP fibrin glue dressing	Significant reduction of wound size in both groups. Significant decrease in inflammatory and oxidative markers in intervention group
<b>Parameter: Safety and Accessibility</b>			
Karina, <i>et al.</i> (2021) <sup>40</sup>	Intravenous aaPRP	N/A	No observed adverse event (allergic reaction, infection, coagulation)
Linertová, <i>et al.</i> (2021) <sup>41</sup>	Usual care + manually obtained aaPRP vs. usual care + kit-obtained aaPRP vs. usual care		PRP for the management of DFU may be cost-effective or cost-saving if implemented appropriately
<b>Parameter: Others</b>			
Hassanien, <i>et al.</i> (2020) <sup>42</sup>	Perineural aaPRP injection	Medical treatment	Significant improvement in pain and numbness in DFU patients treated with intervention
Nolan, <i>et al.</i> (2020) <sup>43</sup>	aaPRP + fat graft vs. fat graft vs. usual care		Better fat graft survival and neovascularization in aaPRP + fat graft group. No clinical difference between the three groups.

minimally invasive. Interestingly, Karina, *et al.*<sup>40</sup> formulated an aaPRP preparation protocol that is also suitable for systemic administration via intravenous infusion. The study also proved that aaPRP prepared with such a technique is devoid of leukocytes and platelets after the last stage of activation. Additionally, the aaPRP in the study was also administered through a blood transfusion set to eliminate any potential cellular debris. Out of the more than 600 patients, none reported any adverse effects related to the systemic aaPRP administration.

### Current Evidence and Study Results

A double-blind randomized controlled trial (RCT) in 2021 by Yarahmadi *et al.* found that aaPRP administration in a fibrin glue dressing (along with oral vitamin E and C supplementation) has the capability to help in DFU wound healing. This is evidenced by a significant wound size reduction and decrease in prooxidants compared to the control group<sup>39</sup>. This study results prove that aaPRP can decrease the immense oxidative stress brought about by hyperglycemia. On a side note, another RCT involving perineural aaPRP injection for DPN also showed an improvement in pain and numbness<sup>42</sup>. This encouraging result for improved peripheral nerve function may suggest that the benefits of aaPRP in DFU also extend beyond its antioxidative and anti-inflammatory properties.

Ahmed *et al.*<sup>37</sup> also found that aaPRP in gel form is superior to a local antiseptic dressing in terms of healing rate and infection prevention in clean ulcers. In a previous canine study, aaPRP preparations have exhibited antimicrobial activity against methicillin-resistant *Staphylococcus aureus*<sup>47</sup>. This antimicrobial property may be attributed to the presence of a chemokine (C-C motif) ligand (CCL)-3, CCL5, and chemokine (C-X-C motif) ligand (CXCL)-1. Along with CCL3, CCL5, and CXCL1, transforming growth factor- $\beta$ 1 is also present and reported to be bacteriostatic. Furthermore, synergistic activity between aaPRP and antibiotics have also been observed<sup>48</sup>.

However, a recent RCT concluded that a PRP dressing is no more efficacious than a placebo of normal saline in DFU managed with total-contact casting<sup>38</sup>. The results of said study contradicts an older RCT done in 2006 by Driver *et al.*<sup>36</sup> which found that 81.3% patients in the DFU group treated with aaPRP gel saw wound resolution compared to 42.1% patients in the saline gel group. The difference in administration and the presence of a total-contact cast may play a role in the difference of results; this is to be explored in future studies.

Studies combining aaPRP to enhance the regenerative potential and graft survival rates of mesenchymal stem cells also exist. The combination of aaPRP with stromal vascular fraction (SVF) may be implemented as a non-animal source of growth factors to support stem cell proliferation and differentiation<sup>49</sup>. A murine study of wound healing has also procured promising results, with the group of rats treated with SVF and aaPRP having the most accelerated wound healing compared to aaPRP or SVF alone<sup>50</sup>. An RCT further confirmed this dynamic between aaPRP and stem cells. Histological analyses demonstrated improved neovascularization and graft survival in DFU wounds, although these results are not accompanied by a shortened wound healing time<sup>43</sup>.

The findings of this literature review have been summarized in **Table 2**.

## CONCLUSIONS

Along with the current principles of treatment, *i.e.*, offloading, infection control, wound bed preparation, and others, aaPRP poses as an attractive adjunct treatment to DFU due to its efficacy, as well as its economic and safety profiles. As shown by its ability to improve wound closure, reduce oxidative stress, improve peripheral nerve function, prevent debilitating deep tissue infections, and overall improve safety, more clinicians should test aaPRP as an adjunct or alternative treatment for refractory DFUs. Future studies should explore the optimal form and dosage of aaPRP for DFU treatment.

## ABBREVIATIONS

**aaPRP:** Activated autologous platelet-rich plasma  
**CCL:** Chemokine (C-C motif) ligand  
**CXCL:** Chemokine (C-X-C motif) ligand  
**DFU:** Diabetic foot ulcer  
**DPN:** Diabetic peripheral neuropathy  
**IL:** Interleukin  
**PVD:** Peripheral vascular disease  
**PDGF:** Platelet derived growth factor  
**RCT:** Randomized controlled trial  
**SVF:** Stromal vascular fraction  
**VEGF:** Vascular endothelial growth factor

## ACKNOWLEDGMENTS

None.

## AUTHOR'S CONTRIBUTIONS

All authors equally contributed to this work, read and approved the final manuscript.



## FUNDING

None.

## AVAILABILITY OF DATA AND MATERIALS

Not applicable.

## ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Not applicable.

## CONSENT FOR PUBLICATION

Not applicable.

## COMPETING INTERESTS

The authors declare that they have no competing interests.

## REFERENCES

- Zhang P, Lu J, Jing Y, Tang S, Zhu D, Bi Y. Global epidemiology of diabetic foot ulceration: a systematic review and meta-analysis. *Annals of Medicine*. 2017;49(2):106–16. PMID: 27585063. Available from: [10.1080/07853890.2016.1231932](https://doi.org/10.1080/07853890.2016.1231932).
- Noor S, Zubair M, Ahmad J. Diabetic foot ulcer: A review on pathophysiology, classification and microbial etiology. *Diabetes & Metabolic Syndrome*. 2015;9(3):192–9. PMID: 25982677. Available from: [10.1016/j.dsx.2015.04.007](https://doi.org/10.1016/j.dsx.2015.04.007).
- Everett E, Mathioudakis N. Update on management of diabetic foot ulcers. *Annals of the New York Academy of Sciences*. 2018;1411(1):153–65. PMID: 29377202. Available from: [10.1111/nyas.13569](https://doi.org/10.1111/nyas.13569).
- Khatu SS, More YE, Gokhale NR, Chavhan DC, Bendsure N. Platelet-rich plasma in androgenic alopecia: myth or an effective tool. *Journal of Cutaneous and Aesthetic Surgery*. 2014;7(2):107–10. PMID: 25136212. Available from: [10.4103/0974-2077.138352](https://doi.org/10.4103/0974-2077.138352).
- Prodromos CC, Finkle S, Prodromos A, Chen JL, Schwartz A, Wathen L. Treatment of Rotator Cuff Tears with platelet rich plasma: a prospective study with 2 year follow-up. *BMC Musculoskeletal Disorders*. 2021;22(1):499. PMID: 34051761. Available from: [10.1186/s12891-021-04288-4](https://doi.org/10.1186/s12891-021-04288-4).
- Karina K, Rosliana I, Rosadi I, Sobariah S, Christoffel LM, Novariani R. Phase I/II Clinical Trial of Autologous Activated Platelet-Rich Plasma (aaPRP) in the Treatment of Severe Coronavirus Disease 2019 (COVID-19) Patients. *International Journal of Inflammation*. 2021;2021:5531873. PMID: 34306612. Available from: [10.1155/2021/5531873](https://doi.org/10.1155/2021/5531873).
- Karina K, Christoffel LM, Novariani R, Rosadi I, Rosliana I, Rosidah S, et al. The Effect of Intravenous Autologous Activated Platelet-Rich Plasma Therapy on "Profibrotic Cytokine" IL-1 $\beta$  Levels in Severe and Critical COVID-19 Patients: A Preliminary Study. *Sikavitsas VI, editor. Scientifica (Cairo) [Internet]*. 2021;2021:1–7. Cairo: Scientifica; 2021. Available from: [10.1155/2021/9427978](https://doi.org/10.1155/2021/9427978).
- Karina K, Christoffel LM, Novariani R, Rosadi I, Rosliana I, Rosidah S. Case Report on Adjunct Intravenous Autologous Activated Platelet-Rich Plasma Therapy in Severely Ill COVID-19 Patients. *Biomedical Research and Therapy*. 2021;8(10):4614–9. Available from: [10.15419/bmrat.v8i9.697](https://doi.org/10.15419/bmrat.v8i9.697).
- Dhurat R, Sukesh M. Principles and methods of preparation of platelet-rich plasma: A review and author's perspective. *Journal of Cutaneous and Aesthetic Surgery*. 2014;7(4):189–97. PMID: 25722595. Available from: [10.4103/0974-2077.150734](https://doi.org/10.4103/0974-2077.150734).
- Iqbal Z, Azmi S, Yadav R, Ferdousi M, Kumar M, Cuthbertson DJ. Diabetic Peripheral Neuropathy: Epidemiology, Diagnosis, and Pharmacotherapy. *Clinical Therapeutics*. 2018;40(6):828–49. PMID: 29709457. Available from: [10.1016/j.clinthera.2018.04.001](https://doi.org/10.1016/j.clinthera.2018.04.001).
- Sloan G, Selvarajah D, Tesfaye S. Pathogenesis, diagnosis and clinical management of diabetic sensorimotor peripheral neuropathy. *Nature Reviews Endocrinology*. 2021;17(7):400–20. PMID: 34050323. Available from: [10.1038/s41574-021-00496-z](https://doi.org/10.1038/s41574-021-00496-z).
- Feldman EL, Callaghan BC, Pop-Busui R, Zochodne DW, Wright DE, Bennett DL. Diabetic neuropathy. *Nature Reviews Disease Primers*. 2019;5(1):41. PMID: 31197153. Available from: [10.1038/s41572-019-0092-1](https://doi.org/10.1038/s41572-019-0092-1).
- Callaghan BC, Little AA, Feldman EL, Hughes RA. Enhanced glucose control for preventing and treating diabetic neuropathy. *Cochrane Database of Systematic Reviews*. 2012;6(6). PMID: 22696371. Available from: [10.1002/14651858.cd007543.pub2](https://doi.org/10.1002/14651858.cd007543.pub2).
- Hosseini A, Abdollahi M. Diabetic neuropathy and oxidative stress: therapeutic perspectives. *Oxidative Medicine and Cellular Longevity*. 2013;2013:168039. PMID: 23738033. Available from: [10.1155/2013/168039](https://doi.org/10.1155/2013/168039).
- Oates PJ. Aldose reductase, still a compelling target for diabetic neuropathy. *Current Drug Targets*. 2008;9(1):14–36. PMID: 18220710. Available from: [10.2174/138945008783431781](https://doi.org/10.2174/138945008783431781).
- Brownlee M. Biochemistry and molecular cell biology of diabetic complications. *Nature*. 2001;414(6865):813–20. PMID: 11742414. Available from: [10.1038/414813a](https://doi.org/10.1038/414813a).
- Thiruvoipati T, Kielhorn CE, Armstrong EJ. Peripheral artery disease in patients with diabetes: Epidemiology, mechanisms, and outcomes. *World Journal of Diabetes*. 2015;6(7):961–9. PMID: 26185603. Available from: [10.4239/wjcd.v6.i7.961](https://doi.org/10.4239/wjcd.v6.i7.961).
- Radha TP, P S A, Annamalai S. Diabetes Mellitus and Peripheral Vascular Disease. *Int J Contemp Med Res*. 2020;7(7):11–3. Available from: [10.21276/ijcmr.2020.7.7.26](https://doi.org/10.21276/ijcmr.2020.7.7.26).
- Lepäntalo M, Apelqvist J, Setacci C, Ricco JB, de Donato G, Becker F. Chapter V: diabetic foot. *European Journal of Vascular and Endovascular Surgery*. 2011;42:60–74. PMID: 22172474. Available from: [10.1016/S1078-5884\(11\)60012-9](https://doi.org/10.1016/S1078-5884(11)60012-9).
- Al-Rubeaan K, Derwish MA, Ouizi S, Youssef AM, Subhani SN, Ibrahim HM. Diabetic foot complications and their risk factors from a large retrospective cohort study. *PLoS One*. 2015;10(5):e0124446. PMID: 25946144. Available from: [10.1371/journal.pone.0124446](https://doi.org/10.1371/journal.pone.0124446).
- Yazdanpanah L, Shahbazian H, Nazari I, Arti HR, Ahmadi F, Mohammadianinejad SE. Incidence and risk factors of diabetic foot ulcer: A population-based diabetic foot cohort (ADFC study)-two-year follow-up study. *International Journal of Endocrinology*. 2018;2018:7631659. PMID: 29736169. Available from: [10.1155/2018/7631659](https://doi.org/10.1155/2018/7631659).
- Cardoso HC, Zara AL, Rosa SS, Rocha GA, Rocha JV, de Araújo MC. Risk Factors and Diagnosis of Diabetic Foot Ulceration in Users of the Brazilian Public Health System. *Journal of Diabetes Research*. 2019;2019:5319892. PMID: 31612148. Available from: [10.1155/2019/5319892](https://doi.org/10.1155/2019/5319892).
- Lavery LA, Davis KE, Berriman SJ, Braun L, Nichols A, Kim PJ. WHS guidelines update: diabetic foot ulcer treatment guidelines. *Wound Repair and Regeneration*. 2016;24(1):112–26. PMID: 26663430. Available from: [10.1111/wrrr.12391](https://doi.org/10.1111/wrrr.12391).
- Steed DL. Clinical evaluation of recombinant human platelet-derived growth factor for the treatment of lower extremity ulcers. *Plastic and Reconstructive Surgery*. 2006;117(7):143–9. PMID: 16799381. Available from: [10.1097/01.prs.0000222526.21512.4c](https://doi.org/10.1097/01.prs.0000222526.21512.4c).
- Bhansali A, Venkatesh S, Dutta P, Dhillon MS, Das S, Agrawal A. Which is the better option: recombinant human PDGF-BB 0.01% gel or standard wound care, in diabetic neuropathic large plantar ulcers off-loaded by a customized contact cast? *Diabetes Research and Clinical Practice*. 2009;83(1):e13–6. PMID: 19081156. Available from: [10.1016/j.diabres.2008.10.005](https://doi.org/10.1016/j.diabres.2008.10.005).

26. Blume P, Driver VR, Tallis AJ, Kirsner RS, Kroeker R, Payne WG. Formulated collagen gel accelerates healing rate immediately after application in patients with diabetic neuropathic foot ulcers. *Wound Repair and Regeneration*. 2011;19(3):302–8. PMID: 21371164. Available from: [10.1111/j.1524-475X.2011.00669.x](https://doi.org/10.1111/j.1524-475X.2011.00669.x).
27. Hanft JR, Pollak RA, Barbul A, van Gils C, Kwon PS, Gray SM. Phase I trial on the safety of topical rhVEGF on chronic neuropathic diabetic foot ulcers. *Journal of Wound Care*. 2008;17(1):30–2. PMID: 18210954. Available from: [10.12968/jowc.2008.17.1.27917](https://doi.org/10.12968/jowc.2008.17.1.27917).
28. Uchi H, Igarashi A, Urabe K, Koga T, Nakayama J, Kawamori R. Clinical efficacy of basic fibroblast growth factor (bFGF) for diabetic ulcer. *European Journal of Dermatology*. 2009;19(5):461–8. PMID: 19638336. Available from: [10.1684/ejd.2009.0750](https://doi.org/10.1684/ejd.2009.0750).
29. Cruciani M, Lipsky BA, Mengoli C, de Lalla F. Granulocyte-colony stimulating factors as adjunctive therapy for diabetic foot infections; 2013. Available from: [10.1002/14651858.CD006810.pub3](https://doi.org/10.1002/14651858.CD006810.pub3).
30. Martinez-Zapata MJ, Martí-Carvajal AJ, Solà I, Expósito JA, Bolívar I, Rodríguez L. Autologous platelet-rich plasma for treating chronic wounds; 2016. Available from: [10.1002/14651858.CD006899.pub3](https://doi.org/10.1002/14651858.CD006899.pub3).
31. Ehrenfest DMD, Rasmusson L, Albrektsson T. Classification of platelet concentrates: from pure platelet-rich plasma (P-PRP) to leucocyte- and platelet-rich fibrin (L-PRF). *Trends in Biotechnology*. 2009;27(3):158–67. PMID: 19187989. Available from: [10.1016/j.tibtech.2008.11.009](https://doi.org/10.1016/j.tibtech.2008.11.009).
32. Dragoo JL, Braun HJ, Durham JL, Ridley BA, Odegaard JJ, Luong R. Comparison of the acute inflammatory response of two commercial platelet-rich plasma systems in healthy rabbit tendons. *The American Journal of Sports Medicine*. 2012;40(6):1274–81. PMID: 22495144. Available from: [10.1177/0363546512442334](https://doi.org/10.1177/0363546512442334).
33. Tran TD, Le PT, Pham PV. Diabetic foot ulcer treatment by activated platelet rich plasma: a clinical study. *Biomedical Research and Therapy*. 2014;1(2):37–42. Available from: [10.7603/s40730-014-0008-3](https://doi.org/10.7603/s40730-014-0008-3).
34. Chen Z, Fu S, Wu Z, Chen J, Huang Y, Wang Y. Relationship between plasma angiogenic growth factors and diabetic foot ulcers. *Clinica Chimica Acta*. 2018;482:95–100. PMID: 29614308. Available from: [10.1016/j.cca.2018.03.035](https://doi.org/10.1016/j.cca.2018.03.035).
35. Pavlovic V, Ciric M, Jovanovic V, Stojanovic P. Platelet Rich Plasma: a short overview of certain bioactive components. *Open Medicine (Warsaw)*. 2016;11(1):242–7. PMID: 28352802. Available from: [10.1515/med-2016-0048](https://doi.org/10.1515/med-2016-0048).
36. Driver VR, Hanft J, Fylling CP, Beriou JM, Group ADFUS. A prospective, randomized, controlled trial of autologous platelet-rich plasma gel for the treatment of diabetic foot ulcers. *Ostomy/Wound Management*. 2006;52(6):68–70. PMID: 16799184.
37. Ahmed M, Reffat SA, Hassan A, Eskander F. Platelet-Rich Plasma for the Treatment of Clean Diabetic Foot Ulcers. *Annals of Vascular Surgery*. 2017;38:206–11. PMID: 27522981. Available from: [10.1016/j.avsg.2016.04.023](https://doi.org/10.1016/j.avsg.2016.04.023).
38. Gupta A, Channaveera C, Sethi S, Ranga S, Anand V. Efficacy of intralesional platelet-rich plasma in diabetic foot ulcer. *J Am Podiatr Med Assoc*. 2021;111(3):Article\_7. Available from: [10.7547/19-149](https://doi.org/10.7547/19-149).
39. Yarahmadi A, Modagheh MHS, Mostafavi-Pour Z, Azarpira N, Mousavian A, Bonakdaran S. The effect of platelet-rich plasma-fibrin glue dressing in combination with oral vitamin E and C for treatment of non-healing diabetic foot ulcers: a randomized, double-blind, parallel-group, clinical trial. *Expert Opinion on Biological Therapy*. 2021;21(5):687–96. PMID: 33646060. Available from: [10.1080/14712598.2021.1897100](https://doi.org/10.1080/14712598.2021.1897100).
40. Karina K, Ekaputri K, Biben JA, Purwokoto RH, Sibuea TP, Astuti SL. Evaluating the Safety of Intravenous Delivery of Autologous Activated Platelet-rich Plasma. *Journal of Health Sciences (Sarajevo)*. 2021;11(2):61–5. Available from: [10.17532/jhsci.2021.1276](https://doi.org/10.17532/jhsci.2021.1276).
41. Linertová R, Pino-Sedeño TD, Pérez LG, Aragón-Sánchez J, Andia-Ortiz I, Trujillo-Martin M. Cost-effectiveness of Platelet-Rich Plasma for Diabetic Foot Ulcer in Spain. *The International Journal of Lower Extremity Wounds*. 2021;20(2):119–27. PMID: 32037919. Available from: [10.1177/1534734620903239](https://doi.org/10.1177/1534734620903239).
42. Hassanien M, Elawamy A, Kamel EZ, Khalifa WA, Abolfadl GM, Roushdy AS. Perineural platelet-rich plasma for diabetic neuropathic pain, could it make a difference? *Pain Medicine*. 2020;21(4):757–65. PMID: 31298289. Available from: [10.1093/pm/pnz140](https://doi.org/10.1093/pm/pnz140).
43. Nolan GS, Smith OJ, Heavey S, Jell G, Mosahebi A. Histological analysis of fat grafting with platelet-rich plasma for diabetic foot ulcers-A randomised controlled trial. *International Wound Journal*. 2022;19(2):389–98. PMID: 34169656. Available from: [10.1111/iwj.13640](https://doi.org/10.1111/iwj.13640).
44. Cassano JM, Kennedy JG, Ross KA, Fraser EJ, Goodale MB, Fortier LA. Bone marrow concentrate and platelet-rich plasma differ in cell distribution and interleukin 1 receptor antagonist protein concentration. *Knee Surgery, Sports Traumatology, Arthroscopy*. 2018;26(1):333–42. PMID: 26831858. Available from: [10.1007/s00167-016-3981-9](https://doi.org/10.1007/s00167-016-3981-9).
45. Pochini AC, Antonioli E, Bucci DZ, Sardinha LR, Andreoli CV, Ferretti M. Analysis of cytokine profile and growth factors in platelet-rich plasma obtained by open systems and commercial columns. *Einstein (Sao Paulo, Brazil)*. 2016;14(3):391–7. PMID: 27759829. Available from: [10.1590/S1679-45082016AO3548](https://doi.org/10.1590/S1679-45082016AO3548).
46. King A, Balaji S, Le LD, Crombleholme TM, Keswani SG. Regenerative Wound Healing: The Role of Interleukin-10. *Advances in Wound Care (New Rochelle, NY)*. 2014;3(4):315–23. PMID: 24757588. Available from: [10.1089/wound.2013.0461](https://doi.org/10.1089/wound.2013.0461).
47. Farghali HA, AbdElkader NA, AbuBakr HO, Aljuaydi SH, Khat-tab MS, Elhelw R. Antimicrobial action of autologous platelet-rich plasma on MRSA-infected skin wounds in dogs. *Scientific Reports*. 2019;9(1):12722. PMID: 31481694. Available from: [10.1038/s41598-019-48657-5](https://doi.org/10.1038/s41598-019-48657-5).
48. Zhang W, Guo Y, Kuss M, Shi W, Aldrich AL, Untrauer J. Platelet-rich plasma for the treatment of tissue infection: preparation and clinical evaluation. *Tissue Engineering Part B, Reviews*. 2019;25(3):225–36. PMID: 30712506. Available from: [10.1089/ten.teb.2018.0309](https://doi.org/10.1089/ten.teb.2018.0309).
49. Karina K, Rosliana I, Rosadi I, Schwartz R, Sobariah S, Afni I. Safety of technique and procedure of stromal vascular fraction therapy: from liposuction to cell administration. *Scientifica*. 2020;2020:2863624. PMID: 32695550. Available from: [10.1155/2020/2863624](https://doi.org/10.1155/2020/2863624).
50. Karina K, Samudra MF, Rosadi I, Afni I, Widyastuti T, Sobariah S. Combination of the stromal vascular fraction and platelet-rich plasma accelerates the wound healing process: pre-clinical study in a Sprague-Dawley rat model. *Stem Cell Investigation*. 2019;6(18):18. PMID: 31463311. Available from: [10.21037/sci.2019.06.08](https://doi.org/10.21037/sci.2019.06.08).