

## Original Research Article

# Efficacy of platelet rich plasma in chronic leg ulcers: a prospective randomized controlled study

Naphisabet Wanniang\*, Pankaj Shukla, Varadraj V. Pai

Department of Dermatology, Venereology and Leprology, Goa Medical College, India

**Received:** 06 November 2020

**Accepted:** 11 December 2020

**\*Correspondence:**

Dr. Naphisabet Wanniang,

E-mail: [naf.wann@gmail.com](mailto:naf.wann@gmail.com)

**Copyright:** © the author(s), publisher and licensee Medip Academy. This is an open-access article distributed under the terms of the Creative Commons Attribution Non-Commercial License, which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

### ABSTRACT

**Background:** Chronic wounds affect approximately 1-2% of the population in Europe and the United States. Platelet rich plasma (PRP) has emerged as an effective, inexpensive, minimally invasive treatment modality for chronic leg ulcers. Objective of the study was to evaluate the efficacy of PRP, and to compare the effectiveness of PRP to regular antiseptic dressing in the management of chronic leg ulcers.

**Methods:** A hundred patients with chronic leg ulcers of more than 6 weeks duration were randomized into two groups (PRP and conventional dressing group). Patients in the PRP group received weekly intradermal injections of PRP to the wound in addition to conventional daily dressings till complete healing of the ulcers or up to a maximum of 6 weekly PRP sessions. Percentage of improvement in the area and volume of the ulcers were recorded. Patients were followed up at 1 month post PRP treatment.

**Results:** The mean reduction in the area of the ulcers at the end of 6 weeks was 66.39% in the PRP group and 28.6% in the control group. The mean reduction in volume of the ulcers at the end of 6 weeks was 71.80% and 37.88% in the case and control group respectively. At the end of 1 month post treatment follow-up, 74% and 10% of the ulcers treated with PRP and with conventional dressing respectively showed complete healing.

**Conclusions:** Leg ulcers treated with PRP showed a significantly higher reduction in the area and volume of the ulcers compared to ulcers treated with conventional moist dressing.

**Keywords:** Chronic ulcers, Platelet rich plasma, PRP

### INTRODUCTION

A chronic wound is defined as wounds which does not progress through the normal phases of wound healing i.e. homeostasis, inflammation, proliferation and remodelling phase, in a timely and orderly manner, thus failing to achieve a sustained anatomical and functional result.<sup>1</sup> The major causes of leg ulcers are venous ulcers, diabetic ulcers and trophic ulcers. In Europe and the United States, approximately 1-2% of the population suffer from chronic wounds.<sup>2</sup> In India, the prevalence of chronic wounds was reported to be 4.5 per 1000 population which accounts for a great deal of patient morbidity and thus has been aptly labelled as a 'silent epidemic'.<sup>3,4</sup> Wound management has

evolved with time and in modern literature, emphasis is made on occlusive antiseptic dressings which provides moisture to the wound.<sup>5</sup> Even with the advances in wound management, the cost for the management of chronic leg ulcers still poses an enormous economic burden to medical services in the west as well as elsewhere in the world.<sup>6</sup> There is a dire need for therapeutic options that work rapidly, are more cost effective and are relatively simpler to apply. Platelet rich plasma (PRP) has emerged as a promising, inexpensive, minimally invasive modality in the treatment of chronic leg ulcers which have failed to show signs of healing with conventional therapies. In light of the above, it was thought worthwhile to compare the efficacy of PRP to conventional treatment i.e. moist

antimicrobial dressing in the management of chronic leg ulcers.

### **Objectives**

The aim of the study was to assess the efficacy of PRP in chronic leg ulcers. The study also compared the effectiveness of PRP to regular moist antimicrobial dressing in the management of chronic leg ulcers.

### **METHODS**

This was a prospective, open labelled, randomized, non-blinded, case control study comparing the efficacy of PRP to conventional wound dressing in the treatment of chronic leg ulcers of various aetiologies done over a period of 12 months (January 2017-December 2017) in the department of dermatology, venereology and leprology, Goa Medical College. After obtaining ethical clearance from the institute's ethics committee, a total of 100 patients with chronic leg ulcers were recruited for the study. Patients with leg ulcers of more than 6 weeks duration, with normal blood parameters (haemoglobin, total count, platelet count) of all age group and both sexes were included in the study. Pregnant patients, hepatitis B, C and human-immunodeficiency virus (HIV) positive patients, patients on immunosuppressants, patients with bleeding disorders and patients not consenting to the study were excluded.

### **Data collection**

After procuring a written informed consent from the patients, a thorough evaluation was done to help delineate the aetiology of the leg ulcers. Emphasis was laid on the duration of the ulcer, comorbidities (diabetes, hypertension), previous treatment history, presence of systemic illness and associated complaints. Clinical examination was carried out on each patient. Points taken into consideration were the site and size of the ulcers, presence or absence of discharge, pain/tenderness, peripheral pulsation, presence or absence of varicosities, surrounding skin changes and testing for sensation for temperature, pressure and touch was also carried out.

### **Randomization**

Patients satisfying the inclusion criteria were randomized into a PRP group or control group (conventional moist dressing without PRP) by using random number table by Chakrabarty.<sup>7</sup>

### **Procedure**

Since secondary bacterial infections are common in tropical climates like ours, all patients received a course of antibiotics based on the bacteriogram prior to the study. This would eliminate any secondary infection that could potentially hamper the wound healing. All wounds were cleaned with normal saline. Patients in the control group were advised to continue with their daily moist

antibacterial dressing of their wound. They were also instructed to follow up every week for 6 weeks. Patients in the PRP group received weekly intradermal injections of PRP around the wound margin in addition to conventional dressing.

### **Preparation of PRP**

Manual preparation of PRP using "double centrifugation method" was used for the study. Under aseptic precaution, 10 ml of venous blood was collected from a peripheral vein into sodium citrate bulbs (9:1). The collected blood was centrifuge in a centrifugation machine (REMI R86) at 400 g (3000 rpm)×10 mins which causes the red blood cells to settle at the bottom while the platelets and plasma are seen in the topmost portion. The plasma thus obtained was again centrifuge in a plain bulb without anticoagulant at 800 g (4000 rpm)×10 mins. The second centrifuge separates the plasma into two layers, an upper portion of platelet poor plasma and a lower portion of platelet rich plasma. Only the lower 1/3rd portion was collected in a tuberculin syringe containing 0.3 ml of 10% calcium chloride which activates the platelets prior to the PRP dressing. Intradermal/subcutaneous injections were given around the four quadrants of the wound using a 23 gauge needle attached to the 1 ml syringe containing the activated PRP. The gel formed after activation was also applied to the wound bed and covered by a non-absorbable dressing material. The injections were repeated weekly up to complete healing of the wound or up to a maximum 6 injections whichever was earlier. Patients were followed up for 1 month after the last PRP session.

### **Method of assessment**

The efficacy of the therapy was measured by the percentage reduction in the area and volume of the ulcers at every weekly visit and at follow up. The length and breadth of the ulcer was measured at the 12-6 o'clock and 3-9 o'clock position respectively using a measuring tape. The depth of the ulcer was measured using a non-malleable sterilized millimetre probe at the deepest point of the ulcers. The area of an ellipse was used to represent the area of the ulcers as the cutaneous wound shape closely resembles the shape of the ellipse. Area was calculated as=length×breadth×0.7854 (area of an ellipse) and volume=area×depth. Similar studies on cutaneous ulcers have used this formula for obtaining the area of the wound.<sup>8</sup> The quality of the wound was assessed using the diabetic foot infection (DFI) wound score at baseline, at week 6 and at 1 month follow up.<sup>9</sup>

### **Statistical analysis**

Statistical analysis of the data was done using statistical package for the social sciences (SPSS), version 14.0. The area and volume of the ulcers were expressed as mean±standard deviation (SD). The categorical data between the two groups such as age, sex were compared using Pearson chi square test. The statistical significance

of the mean area and volume between the two groups was assessed using the independent 't' test. All the tests were two tailed and a p value of 0.05 was considered as statistically significant.

## RESULTS

A total of 100 patients of chronic leg ulcers of various aetiologies fulfilling the inclusion criteria were selected for the study. A total of 50 patients received weekly PRP injections on the ulcers while 50 others were advised for daily antiseptic dressing. The baseline parameters of the PRP and control group are shown in Table 1. The aetiologies of chronic leg ulcers recruited in our study is shown in Figure 1. Our study showed that the mean area and volume of ulcers gradually decrease at successive

weeks, more so in the PRP group than control group (Figure 2 and 3). A higher percentage of improvement in the area and volume of the ulcers was noted in the PRP group than the control group (Table 2). There was a statistically significant improvement in both the area and volume of the ulcers treated with PRP (p value <0.001), and a significant reduction in the DFI wound score from baseline to week 6 which continued to decrease even at 1 month follow up (p value=0.04, 0.017 respectively) (Table 3). The status of the ulcers at 1 month follow up post treatment is shown in Table 4.

The response to PRP varied according to the aetiology of the ulcers, with diabetic ulcers showing the best response. (Table 5). Ulcers of shorter duration responded better to PRP as shown in Figure 4.

**Table 1: Baseline parameters in PRP and control group.**

Parameters	PRP group	Control group
Age in years (SD)	52.3 (14.6)	56.4 (13.7)
Sex distribution (M:F)	1.8	1.5
Mean duration of ulcers (months) (SD)	9 (10.3)	3.9 (2.1)
Mean area in cm <sup>2</sup> (SD)	8.32	6.9
Mean volume in cm <sup>3</sup> (SD)	1.52	2.24
DFI score	10.68	11.78

**Table 2: Percentage improvement of area of the ulcers.**

	% improvement in the area of the ulcers		% improvement in the volume of the ulcers	
	PRP group (%)	Control group (%)	PRP group (%)	Control group (%)
No improvement	4	18	6	12
0-20	8	22	6	22
20-40	4	20	6	16
40-60	8	22	10	18
60-80	24	10	12	20
80-100	48	8	64	12

**Table 3: Outcome parameters.**

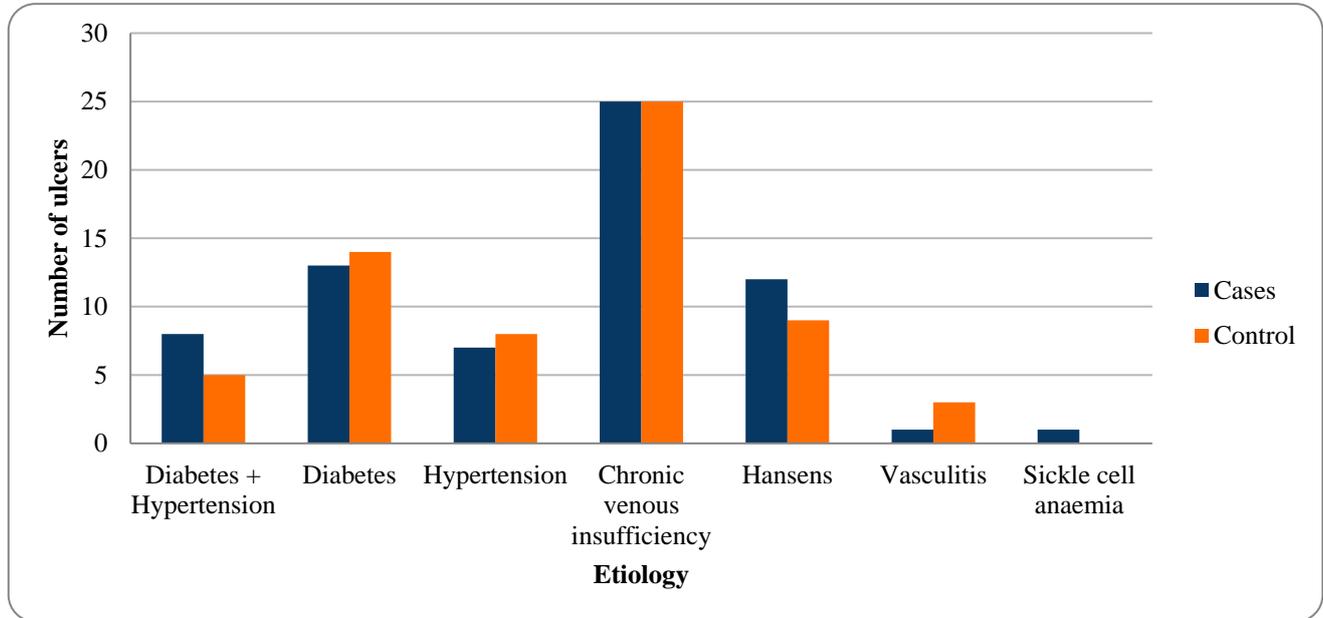
Outcome parameters	PRP group	Control group	P value
Area reduction in cm <sup>2</sup> at week 6	4.96	1.91	0.029
Area reduction in % at week 6	66.4	28.6	<0.001
Volume reduction in cm <sup>3</sup> at week 6	0.96	0.84	0.005
Volume reduction in % at week 6	71.8	37.9	<0.001
DFI score at week 6	6.98	9.22	0.004
DFI score at 1 month follow up	6.28	8.32	0.017

**Table 4: Status of the ulcers at 1 month follow up.**

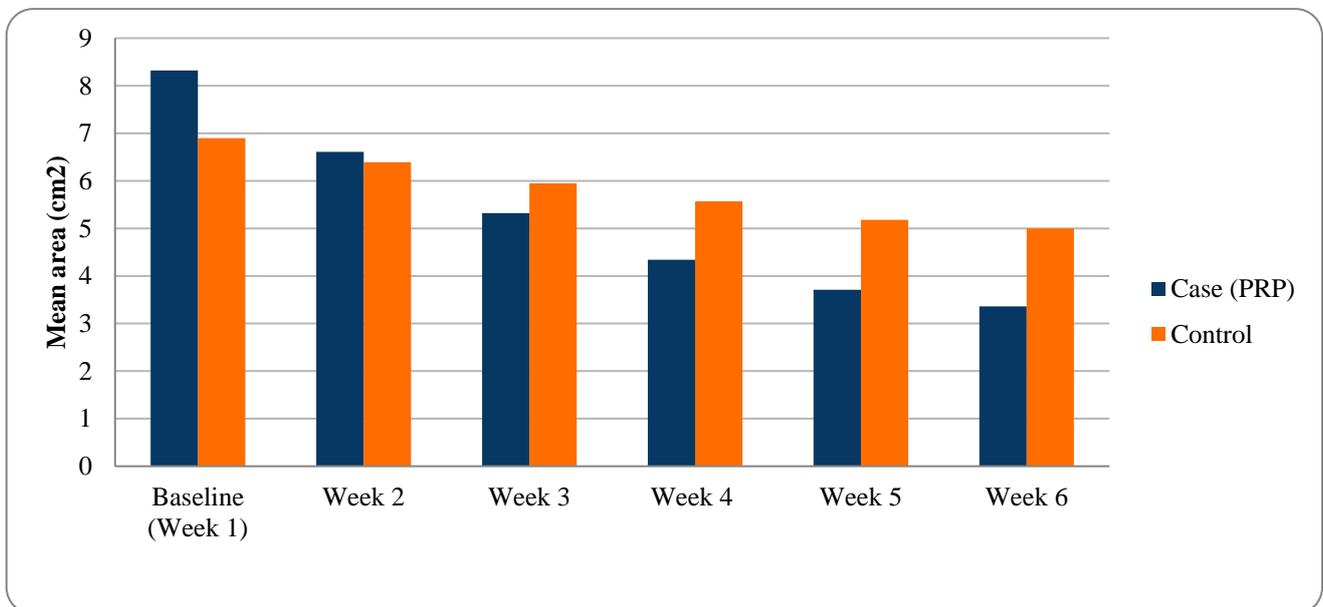
At 1 month follow up	Case (PRP group)	Control	Percentage	
			Case (%)	Control (%)
Complete healing	37	5	74	10
Partial improvement	5	18	10	36
No improvement	6	25	12	50
Worsened	2	2	4	4

**Table 5: Response of ulcers of various aetiologies to PRP at the end of 6 weeks.**

Ulcers of various etiologies	% reduction in area	% reduction in volume
DM + HTN	77.98	86.14
DM	82.95	88.26
HTN	72.2	81.83
CVI	57.61	64.36
Hansens	69.79	72.9



**Figure 1: Aetiology of ulcers.**



**Figure 2: Mean area (cm<sup>2</sup>) at successive weeks.**

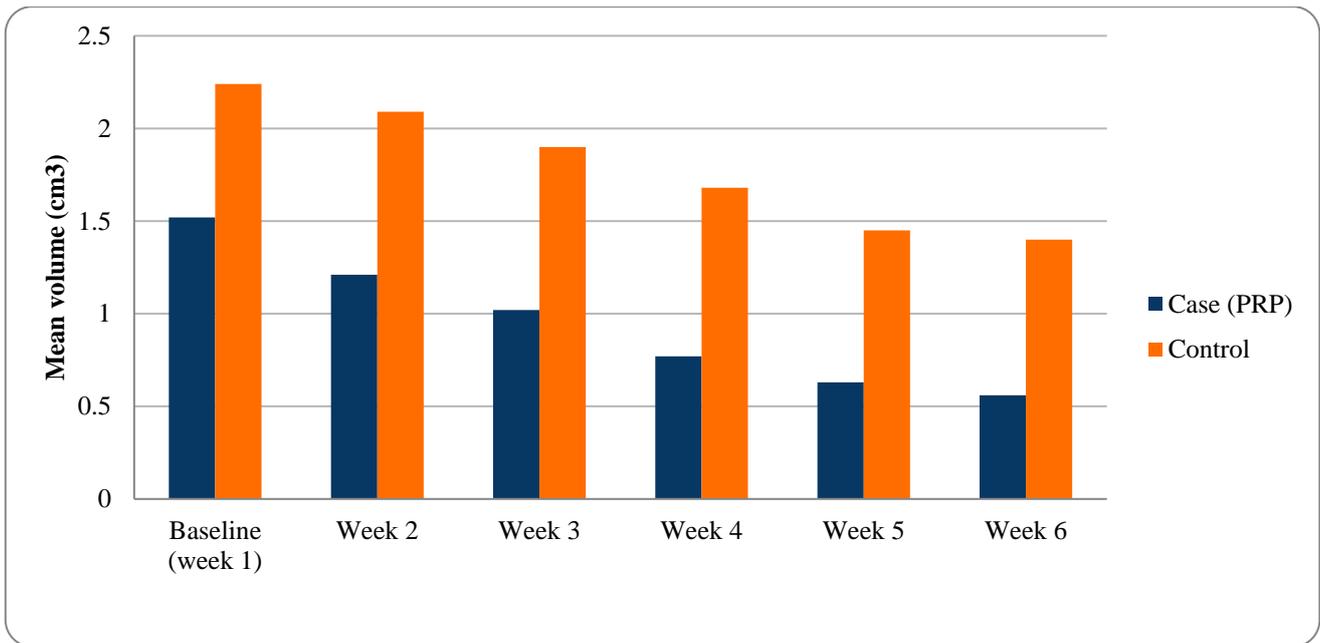


Figure 3: Mean volume (cm<sup>3</sup>) of ulcers at successive weeks.

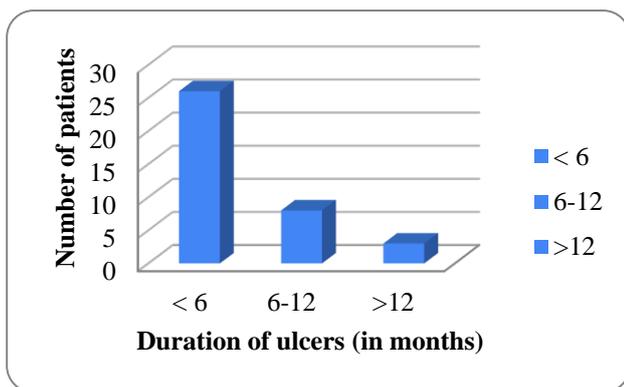


Figure 4: Response to PRP based on duration of ulcers.

## DISCUSSION

Management of chronic ulcers poses an economic burden to patients, society as well as the health services. The chronicity of the leg ulcers is believed to be due to lack of growth factors crucial for wound healing and due to frequent superinfection.<sup>10</sup> PRP has emerged as a therapeutic option aiming to target this aspect of wound healing. It's reported to work by acting as a drug delivery system, delivering various growth factors such as vascular endothelial growth factor (VEGF), transforming growth factor (TGF), insulin-like growth factor (IGF), platelet-derived growth factor (PDGF), epidermal growth factor (EGF) which are released from the alpha granules of the platelets into the wound. These growth factors attract undifferentiated cells in the newly formed matrix of the wound to stimulate cell division, angiogenesis and re-epithelization thereby promoting wound healing.<sup>11,12</sup> In addition to providing growth factors, PRP has been found

to possess antimicrobial properties which could help take care of the local infections in the wound.<sup>13,14</sup> The current working definition of therapeutic PRP is platelet concentration of 1 million/ml.<sup>15</sup> Considering that there is a lack of a well-defined standard method of preparing PRP, we chose the double spin method so as to adequately concentrate the platelets in the plasma as done by other studies.<sup>16</sup> We conducted a randomized control study to assess the efficacy of PRP to conventional moist dressing in the treatment of chronic leg ulcers. The two groups were well matched in respect to the age of the patients and sex distribution. There was no statistical difference between the two groups in this aspect. However, the mean duration of the ulcers was significantly higher in the case group than the control group. The median age of our patients was 55.5 years (range 21-85 years). In our study there were 62% males and 38% females. Similar sex distribution was seen by Suthar and Gupta.<sup>17</sup> The mean duration of the ulcers was 6.47 months ranging from 1.5-36 months. Similar duration of ulcers were quoted by Suryanarayan.<sup>8</sup> We observed that venous ulcers (38.17%) was the major cause of leg ulceration followed by diabetic ulcers (30.53%) and trophic ulcers secondary to leprosy (16.03%). Similar etiological distribution for leg ulcerations was reported in the study done by Suthar et al.<sup>17</sup> The most common associated symptoms was discharge from the wound followed by varicosities. Other symptoms were skin changes, pain, paraesthesia, loss of sensation and itching. Similar symptoms were noted by Park et al.<sup>18</sup> In our study, we observed the mean area of the ulcer at baseline was 8.32±12.15 cm<sup>2</sup> and 6.90±6.89 cm<sup>2</sup> in the case and control group respectively. At 6 weeks, the mean area reduced to 3.36±6.7 cm<sup>2</sup> in the case group and to 5.0±5.56 cm<sup>2</sup> in the control group. There was a statistically significant (p<0.001) reduction of 66.4% (case group) and 28.6% (control group) reduction in the mean area at the end of 6

weeks. The mean volume reduction at 6 weeks was observed to be 71.80% and 37.89% in the case and control group respectively which was also statistically significant ( $p < 0.001$ ). Anitua et al reported similar results in their study.<sup>19</sup> In our study, 48% of patients in the PRP group while 8% patients in the control group had >80% improvement in the area of the ulcers at the end of week 6. We also observed 64% and 12% of the patients in the PRP and control group had >80% improvement in the volume of the ulcers at the end of six weeks. We noted a total of 7 (14%) patients in the PRP group achieved complete healing of their ulcers i.e. 100% improvement in the area and volume of the ulcers at week 6 with mean healing time of  $4 \pm 1.29$  weeks. Sacchidanand et al also reported a mean healing time of 5.1 weeks with PRP.<sup>20</sup> Diabetic ulcers showed the highest response rate to PRP i.e. there was an average of 82.95% and 88.26% reduction in area and volume respectively at week 6. Trophic ulcers showed an average of 69.79% reduction in area and 72.9% reduction in volume at the end of six sessions of PRP. Chronic venous ulceration showed the poorest response to PRP. There was only 57.61% and 64.36% average reduction in area and volume at the end of six weeks. In our study, amongst the ulcers that completely healed with PRP, 70.27% were ulcers of shorter than 6 months duration, 32.43% were ulcers between 3 months to one year and 8.11% were ulcers of more than one year duration. This showed that ulcers of shorter duration responded better to PRP treatment. A similar correlation was noted by Suryanarayan et al.<sup>8</sup> The adverse effects noted in the PRP group was pain during the injections of PRP into the wound. The pain was however short lived and subsided after the procedure. In our study, the DFI score was used to assess the quality of the wound before and after treatment. This scoring system takes into account both the qualitative (such as presence of discharge, pain, erythema etc.) as well as quantitative parameters of the wound (area, depth, undermining). Comparing the DFI score between the two groups before and after treatment showed a significantly better improvement in the quality of the wound after PRP injections. This improvement was maintained even at 1 month follow up.

## CONCLUSION

Chronic leg ulcers are a common problem in our part of the country contributing to patient morbidity and loss of countless work hours. PRP has proved to be an effective and safe option for the treatment of chronic leg ulceration not responding to conventional treatment. It is also an inexpensive form of treatment for patients of developing countries not affording expensive and advanced wound care. However additional studies with larger sample size, accurately matched subjects and longer follow period up is required. Standardized methods of preparing PRP needs to be defined for better comparison of the efficacy of the preparations.

*Funding: No funding sources*

*Conflict of interest: None declared*

*Ethical approval: The study was approved by the institutional ethics committee*

## REFERENCES

- 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. *Arch Dermatol.* 1994;130:489-93.
- Park J, Hwang S, Yoon IS. Advanced Growth Factor Delivery Systems in Wound Management and Skin Regeneration. *Molecules.* 2017;22:1259.
- Shukla VK, Ansari MA, Gupta SK. Wound Healing Research: A Perspective From India. *Int J Low Extrem Wounds.* 2005;4:7-8.
- Sen CK, Gordillo GM, Roy S, Kirsner R, Lambert L, Hunt TH, et al. Human skin wounds: A major and snowballing threat to public health and the economy. *Wound Repair Regen.* 2009;17:763-71.
- Jones J. Winter's concept of moist wound healing: a review of the evidence and impact on clinical practice. *J Wound Care.* 2005;14:273-6.
- Demidova-Rice TN, Hamblin MR, Herman IM. Acute and Impaired Wound Healing: Pathophysiology and Current Methods for Drug Delivery, Part 1. *Adv Skin Wound Care.* 2012;25:304-14.
- Chakrabarty D. One More Table of Random Two-Digit Numbers. *Int J Adv Res Sci Eng Technol.* 2016;3:13.
- Suryanarayan S, Budamakuntla L, Khadri SS, Sarvajnamurthy S, et al. Efficacy of autologous platelet-rich plasma in the treatment of chronic nonhealing leg ulcers. *Plast Aesthetic Res.* 2014;1:65.
- Lipsky BA, Berendt AR, Cornia PB, Pile JC, Peters EJJ, Armstrong DG, et al. Infectious Diseases Society of America Clinical Practice Guideline for the Diagnosis and Treatment of Diabetic Foot Infections. *Oxf Univ Press.* 2012;54:132-73.
- Yuan T, Zhang CQ, Tang MJ, Guo SC, Zeng BF. Autologous Platelet-rich Plasma Enhances Healing of Chronic Wounds. *Wounds.* 2009;21:6.
- Kakudo N, Kushida S, Minakata T, Suzuki K, Kusumoto K. Platelet-rich plasma promotes epithelialization and angiogenesis in a split thickness skin graft donor site. *Med Mol Morphol.* 2011;44:233-6.
- Alves R, Grimalt R. A Review of Platelet-Rich Plasma: History, Biology, Mechanism of Action, and Classification. *Skin Appendage Disord.* 2018;4:18-24.
- Li H, Li B. PRP as a New Approach to Prevent Infection: Preparation and In vitro Antimicrobial Properties of PRP. *J Vis Exp.* 2013;74:50351.
- Cieřlik-Bielecka A, Bold T, Ziřłkowski G, Pierchała M, Królikowska A, Reichert P. Antibacterial Activity of Leukocyte- and Platelet-Rich Plasma: An In Vitro Study. *BioMed Res Int.* 2018;2018:1-8.

15. Marx RE. Platelet-Rich Plasma (PRP): What Is PRP and What Is Not PRP? *Implant Dent.* 2001;10:225-8.
16. Gopinath VPK, Simi VM, Basheer Ahammed K, Farisa PM, Ali Rishad CM. Intralesional autologous platelet rich plasma therapy in chronic nonhealing cutaneous ulcers: an interventional study from a tertiary care centre in North Kerala. *Int J Res Dermatol.* 2019;5:116.
17. Suthar M, Gupta S, Bukhari S, Ponemone V. Treatment of chronic non-healing ulcers using autologous platelet rich plasma: a case series. *J Biomed Sci.* 2017;24:16.
18. Park KY, Kim IS, Yeo IK, Kim BJ, Kim MN. Treatment of refractory venous stasis ulcers with autologous platelet-rich plasma and light-emitting diodes: a pilot study. *J Dermatol Treat.* 2013;24:332-5.
19. Anitua E, Aguirre JJ, Algorta J, Ayerdi E, Cabezas AI, Orive G, et al. Effectiveness of autologous preparation rich in growth factors for the treatment of chronic cutaneous ulcers. *J Biomed Mater Res B Appl Biomater.* 2008;84B:415-21.
20. Sarvajnamurthy S, Suryanarayan S, Budamakuntala L, Suresh DH. Autologous platelet rich plasma in chronic venous ulcers: Study of 17 cases. *J Cutan Aesthetic Surg.* 2013;6:97.

**Cite this article as:** Wanniang N, Shukla P, Pai VV. Efficacy of platelet rich plasma in chronic leg ulcers: a prospective randomized controlled study. *Int J Res Dermatol* 2021;7:78-84.