TREATMENT OUTCOME OF AUTOLOGOUS PLATELET-RICH PLASMA VERSUS CONVENTIONAL THERAPY AMONG PATIENTS WITH CHRONIC VENOUS LEG ULCERS

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Abstract
Objective: To compare the outcome of treatment with autologous platelet-rich plasma (PRP) versus conventional therapy for patients with chronic venous leg ulcers.
Methods: This randomized controlled trial was conducted at the department of Dermatology, Jinnah Postgraduate Medical Centre, Karachi from 1st September 2022 to 25th May 2023. Patients aged 30-80 years of either gender having chronic venous leg ulcer were enrolled. Patients were randomly allocated to Autologous PRP and conventional therapy group. PRP was applied after proper surgical debridement and was dressed with a non-absorbent dressing once weekly for 6 weeks. Conservative therapy was performed by compression using graduated elastic stockings below the knee and dressing using Saline and Vaseline gauze weekly for 6 weeks. Outcome was assessed in terms of complete healing, length of ulcer, and width of ulcer at 6 weeks.
Results: Of 96 patients, the mean age was 59.17 ±7.01 years. There were 53 (55.2%) females and 43 (44.8%) males. A significantly lower mean length of ulcer at 6 weeks was observed in PRP than conventional therapy patients, i.e., 2.11 ±0.88 vs. 2.97 ±0.84 (p-value <0.001). Moreover, a significant mean difference of width of ulcer at 6 weeks was observed in PRP than conventional therapy patients, i.e., i.e., 1.36 ±0.42 vs. 1.54 ±0.39 (p-value 0.037). Moreover, a significantly higher frequency of complete healing was observed in PRP than conventional therapy, i.e., 26 (54.2%) and 7 (14.6%) respectively (p-value <0.001).
Conclusion: A better outcome of PRP was observed compared to conventional therapy in patients with chronic venous leg ulcers.

Keywords: Outcome, Autologous platelet-rich plasma (PRP), Conventional Therapy, Chronic Venous Leg Ulcers

INTRODUCTION:
Chronic venous insufficiency (CVI) is one of the most common diseases worldwide. Its prevalence ranges from 10% to 30%. It is a progressive disease caused by venous hypertension secondary to

venous reflux or obstruction.[1-2] Clinical presentations of CVI range from mild skin trophic changes to non-healing venous ulcers. [3] The prevalence of CVI is highest in Western countries where it already consumes up to 2% of healthcare. [4] The symptoms are often ignored by the patient until it gets debilitating, hindering one's day-to-day social and occupational activities. Patients usually present late with advance form of disease.

Venous leg ulcers (VLUs) are defined as open lesions between the knee and ankle joint that occur in the presence of venous disease. [5] Vascular ulcers are a common problem, with a frequency of about 1%-2% in the general population. [6] They can be venous, arterial, or both, with the most common presentation being venous accounting for 80%-90% of all vascular ulcers. [7] Another factor that influences the development of venous leg ulcers is calf muscle pump failure. Calf pump failure arises from paralysis, immobility, sleeping in a chair with legs dependent for long periods of time, and fixed ankle joints. The calf muscle, through contraction and relaxation, aids in the flow of blood back to the heart through the veins. Failure of this mechanism causes stasis of blood and increased venous pressure. [3]

PRP is a volume of autologous plasma that has a platelet concentration above baseline i.e., five times more than normal platelet counts.[8] PRP enhances wound healing by promoting the healing process by seven growth factors present in it. They are platelet derived growth factor, fibroblast growth factor, vascular endothelial growth factor, epidermal growth factor, transforming growth factor. These growth factors are important in modulating mesenchymal cell recruitment, proliferation and extra-cellular matrix synthesis during the healing process.[9] The use of PRP has dramatically increased in the fields of orthopaedics, spine surgery, reconstructive plastic surgery, oral and maxillofacial surgery, and dermatological indications. Nonetheless, its use in the treatment of wounds is not as widespread as in other fields. [10]

The rationale of the current is that venous ulcers are the most common form of leg ulcers which have a significant impact on quality of life and work productivity. In addition, the costs associated with the long-term care of these chronic wounds are substantial. Conventional therapies such as dressings, surgical debridement, compression bandage and even skin grafting cannot provide satisfactory healing since these treatments are not able to provide necessary growth factors that can modulate healing processes. Autologous PRP is a safe, simple, affordable and less expensive procedure in the treatment of chronic ulcers with reportedly good results. Since, it is an autologous method, it is biocompatible and safe. The aim of the current study was to compare the outcome of treatment with autologous PRP versus conventional therapy for patients presenting with chronic venous leg ulcers. Data from this would help in establishing it as a treatment of choice thereby lead to reduction in cost and benefit the patient both financially and psychologically.

Methods

This randomized controlled trial was conducted at department of Dermatology, Jinnah Postgraduate Medical Centre, Karachi, Pakistan from 1st September 2022 to 25th May 2023. Ethical approval was obtained prior to the commencement of the study (**IRB #NO.F.2-81/2021-GENL/249/JPMC**), also approved on **ClinicalTrials.gov PRS with ID No NCT05974982**. Moreover, signed informed consent was obtained from all study participants prior to the enrollment of the study.

All patients were enrolled through non-probability consecutive sampling. The inclusion criteria were patients aged 30-80 years of either gender who were presented with chronic venous leg ulcer. However, patients with history of peripheral arterial disease, acute venous thromboembolism, platelet count <150,000, bleeding disorders, osteoporosis, peripheral neuropathy, and congestive cardiac failure, chronic liver disease, asthma, COPD, and stroke were excluded. In addition, pregnant women were also excluded from the study.

Brief history of demographic information such as age, gender and place of residence were noted. Patients were examined by dermatologist with over 10 years of experience. Then the lesions length and width were measured by a ruler and documented and photographed by the camera of the researcher. Wound area was calculated using the formula for an ellipse: Length × width × 0.7854

(an ellipse is closer to a wound shape than a square or rectangle that would be described by simple length × width).

The sample size for the study was 96 patients (48 in each group). The sample size was calculated by using the open epi sample size calculator where, Alpha=5%, Power of the test 1-beta=80, taking width of ulcer at 6 weeks (1.04±1.27 cm vs 1.63±0.69 cm).[11]

Chronic venous leg ulcer was labeled as positive on the basis of the presence of leg ulcers for more than 6 months of duration who have not received any previous treatment, having wound size less than 10 cm^2, and ankle/brachial index of >0.80, indicating adequate arterial perfusion.

Smoking was labeled as "Yes" if patient currently or has history of smoking 10 or more cigarettes per day for at least 5 years or 5 or more cigarettes per day for at least 10 years otherwise was labeled as "No". Whereas respondents who said they were physically active for 4 or less hours/week were labeled 'inactive' and others as active.

Patients were randomly allocated using sealed opaque envelop bearing A= Autologous platelet-rich plasma group and B= Conventional therapy group. Aseptic precautions 20 ml of venous blood was drawn of patients who were in group A and added to a test tube containing acid citrate dextrose in a ratio of 9:1 (blood: Acid citrate dextrose), centrifuged at 5000 rpm for 15 min to separate the red blood cells from the platelets and plasma. Then the supernatant and the buffy coat composed of platelets and plasma were collected and centrifuged again at 2000 rpm for 5-10 min. The bottom layer about 1.5 ml was taken and 10% calcium chloride was added (0.3 ml for 1 ml of PRP). Then the activated PRP was applied to the wound after proper surgical debridement and was dressed with a non-absorbent dressing (paraffin gauze). This process was repeated once weekly for 6 weeks. Patients in group B were treated conservatively by compression using graduated elastic stockings below the knee and dressing using Saline and Vaseline gauze weekly for 6 weeks. Outcome was assessed in terms of complete healing was defined as 100% epithelialization, length of ulcer at 6 weeks, and width of ulcer at 6 weeks.

Data were analyzed on SPSS Version 20. Mean and standard deviations was calculated for the quantitative variables like age, length of ulcer and width of ulcer at baseline and at 6 weeks and duration of chronic venous leg ulcer. Frequencies and percentages were calculated for qualitative variables like gender, place of residence, smoking status, physical inactivity, family monthly income status, occupational status, educational status and outcome. The chi-square test was applied to see the association of complete healing with group. Moreover, mean difference of length and width before and after the treatment were explored using paired t-test and mean difference of length and width in between group were explored using independent t test.

Results

Of 96 patients, the mean age was 59.17 ±7.01 years. There were 67 (69.8%) patients with ≤60 years and 29 (30.2%) with >60 years of age. Gender distribution showed that 53 (55.2%) patients were females and 43 (44.8%) was males. The mean duration of leg ulcer was 8.96 ±1.20 months. Smoking history was observed in 30 (31.3%) patients. There were 34 (35.4%) employed participants. Whereas 33 (34.4%) of the patients were illiterate.

The mean age was significantly higher among patients who were in PRP group as compared to those who were in conventional group, i.e., 61.22 ±6.26 vs. 57.13 ±7.19 (p-value 0.004). Whereas other baseline characteristics such as gender (p-value 0.837), residence (p-value 0.404), total monthly family income (p-value 0.525), smoking status (p-value 0.1860, physical activity (p-value 0.102), and occupational status (p-value 0.671) were not statistically significant in between both groups. (Table 1)

A significant mean difference of length of ulcer was observed at baseline and at 6 weeks, i.e., 4.67 ±0.64 vs. 2.54 ±0.96 (p-value <0.001, 95% CI 1.90 to 2.37). Moreover, a significant mean difference of width of ulcer was observed at baseline and at 6 weeks, i.e., 2.88 ±0.90 vs. 1.45 ±0.41 (p-value <0.001, 95% CI 1.90 to 2.37). Similar findings were observed when stratified on the basis of PRP and conventional therapy. (Table 2)

Though the mean difference of length of ulcer and width at baseline was non-significant between PRP and conventional therapy whereas at 6 weeks both length of ulcer (p-value <0.001) and width (p-value 0.037) were found significantly lower among patients who were in PRP group as compared to those who were in conventional group. (Table 3)

A significantly higher frequency of complete healing was observed in patients who were in PRP group as compared to those who were treated with conventional therapy, i.e., 26 (54.2%) and 7 (14.6%) respectively (p-value <0.001). (Figure 1)

Table 1: Comparison of baseline characteristics of the patients in between groups (n=96)

	Total	PRP (n=48)	Conventional Therapy (n=48)	
	mean ±SD	mean ±SD	mean ±SD	p-value
Age, years	59.17 ±7.01	61.22 ±6.26	57.13 ±7.19	0.004$^\beta$
Duration of leg ulcer, months	8.96 ±1.20	9.08 ±7.19	8.83 ±0.88	0.312$^\beta$
Total monthly family income, rupees	57,718 ±9,398	56,541 ±9697.4	58,895 ±9037.1	0.222$^\beta$
	n (%)	n (%)	n (%)	p-value
Age, years				
≤60	67	28 (41.8)	39 (58.2)	0.014$^\yen$
>60	29	20 (69.0)	9 (31.0)	
Gender				
Male	43	22 (51.2)	21 (48.8)	0.837$^\yen$
Female	53	26 (49.1)	27 (50.9)	
Residence				
Rural	58	27 (46.6)	31 (53.4)	0.404$^\yen$
Urban	38	21 (55.3)	17 (44.7)	
Total monthly family income, rupees				
≤55,000	35	19 (54.3)	16 (45.7)	0.525$^\yen$
>55,000	61	29 (47.5)	32 (52.5)	
Smoking Status				
Smoker	30	18 (60.0)	12 (40.0)	0.186$^\yen$
Non-Smoker	36	30 (45.5)	36 (54.5)	
Physical Activity				
Yes	50	29 (58.0)	21 (42.0)	0.102$^\yen$
No	46	19 (41.3)	27 (58.7)	
Occupational status				
Employed	34	18 (52.9)	16 (47.1)	0.671$^\yen$
Unemployed	62	30 (48.4)	32 (51.6)	
Education				
Illiterate	33	25 (75.8)	8 (24.2)	<0.001$^\yen$
Literate	63	23 (36.5)	40 (63.5)	

$^\beta$Independent t-test applied, $^\yen$Chi-Square test/Fisher-Exact test applied. The p-value of ≤0.05 was considered as significant

**Table 2: Mean difference of length of ulcer and width at baseline in between group (n=96)**

	Length, cm			Width, cm		
	mean ±SD	p-value	95% CI	mean ±SD	p-value	95% CI
Total (n=96)						
Baseline	4.67 ±0.64	<0.001	1.90 to 2.37	2.88 ±0.90	<0.001	1.90 to 2.37
At 6 weeks	2.54 ±0.96			1.45 ±0.41		
PRP (n=48)						
Baseline	4.75 ±0.60	<0.001	2.35 to 2.93	2.87 ±0.91	<0.001	1.23 to 1.79
At 6 weeks	2.11 ±0.89			1.36 ±0.42		
Conventional Therapy (n=48)						
Baseline	4.60 ±0.67	<0.001	1.31 to 1.94	2.89 ±0.90	<0.001	1.31 to 1.94
At 6 weeks	2.97 ±0.84			1.54 ±0.39		

$^\beta$Dependent t-test applied, p-value of ≤0.05 was considered as significant

Table 3: Mean difference of length of ulcer and width in between group (n=96)

	PRP (n=48)	Conventional Therapy (n=48)	
	mean ±SD	mean ±SD	p-value
Length of ulcer at baseline, cm	4.75 ±0.60	4.60 ±0.0.68	0.267
Width of ulcer at baseline, cm	2.87 ±0.91	2.89 ±0.90	0.911
Length of ulcer at 6 weeks, cm	2.11 ±0.88	2.97 ±0.84	<0.001
Width of ulcer at 6 weeks, cm	1.36 ±0.42	1.54 ±0.39	0.037

$^\beta$Independent t-test applied, p-value of ≤0.05 was considered as significant
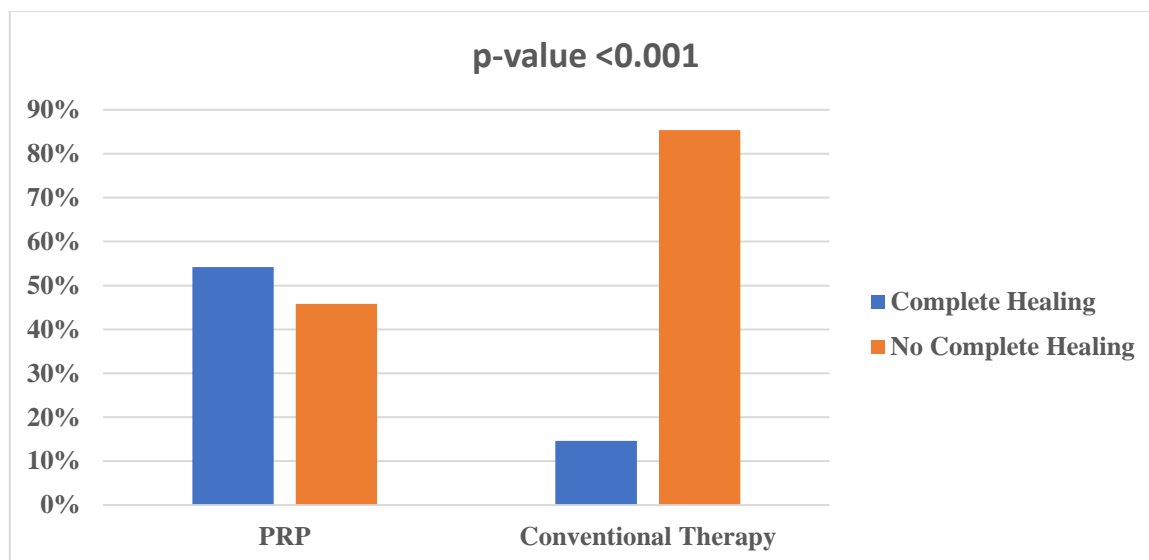
Figure 1: Comparison of complete healing with respect to group (n=96)

Discussion

The study highlighted the findings that compared the effectiveness of PRP therapy with conventional therapy in treating ulcers. The results indicate several positive outcomes associated with PRP therapy. Firstly, the study observed a significantly lower mean length of ulcer at six weeks in patients treated with PRP compared to those receiving conventional therapy. This suggests that PRP therapy may contribute to faster healing and reduction in ulcer size. Various other studies have also reported favorable outcome of PRP for chronic venous leg ulcers.[11-14] Conservative wound treatment for venous leg ulcers requires approximately three month to heal, which has significant personal and financial expenses for the patients. Since the average platelet lifespan is approximately two weeks, it is reported in various studies including recent systematic reviews and meta-analysis that using PRP will need cleaning the wound and repeating the procedure once weekly, lowering the physical and local wound variables that contribute to a delay in wound healing.[11-16]

According to the current study findings, the width of the ulcers also showed a significant mean difference at six weeks between the PRP therapy group and the conventional therapy group. This implies that PRP therapy may have a beneficial effect on reducing the width or breadth of ulcers, further contributing to the healing process. Similar findings were reported in previous studies as well.[17,18]

Lastly, the study found a significantly higher frequency of complete healing in patients treated with PRP compared to those undergoing conventional therapy. This indicates that PRP therapy may have a higher success rate in achieving full recovery from ulcers. In study by Moneib et al., they compared autologous PRP with conventional treatment for patients who had chronic venous leg ulcers. They discovered full responses in 35% of cases vs 0% of cases, a 6-week length of 1.59 cm versus 2.00 cm, and a 6-week breadth of 1.04 cm versus 1.63 cm.[11]

Overall, these findings suggest that PRP therapy may be more effective than conventional therapy in terms of reducing ulcer length and width, as well as increasing the likelihood of complete healing.

The quality of data of autologous PRP for treating chronic venous leg ulcers is low as individual studies are conflicting, with positive results approving this biotechnology and others showing no advantage.[16,19,20]

The findings of the current study could be highlighted in the light of limitations that the study was carried out in a single center healthcare institute with limited number of samples. Due to these limitations of study population, all participants were enrolled through non-probability consecutive samples. Secondly, it was challenging to blind participants or providers to the treatment group since the administration of PRP is visible and distinct from standard care. Third, there is currently no consensus on the optimal protocol for PRP preparation, including variations in platelet concentration, activation methods, and preparation techniques. This lack of standardization can introduce variability in the results and make it difficult to compare findings across different studies. Lastly, chronic venous leg ulcers can be chronic and recurrent, often requiring long-term management. RCTs with relatively short follow-up periods may not capture the full impact of PRP treatment on wound healing, recurrence rates, and other relevant long-term outcomes. Despite of these limitations, this clinical trial is a significant effort in reporting the outcome of PRP in chronic venous leg ulcer patients.

Conclusion

In our cohort of patients with chronic leg ulcers, we observed that those who received PRP treatment showed significant improvements compared to those who received conventional therapy. The PRP-treated group exhibited higher rates of complete healing, reduced ulcer length, and decreased ulcer width after a 6-week period.

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