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### Efficacy of autologous platelet rich plasma (prp) dressings in diabetic foot ulcers: An observational study

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#### Abstract

**Background:** Diabetic foot is a frequent site for complication in DM. The present study was conducted to evaluate the efficacy of PRP dressings in DFUS at different intervals after such dressings.

**Materials & Methods:** The present study was conducted on 25 patients with DFU of either gender (males- 15, females- 10). Wound size as per maximum length and breadth and area were recorded and three PRP dressings were given on day 0, second after first week, and third after 3rd week. On 2nd week, and 4th, 5th, 6th and 8th weeks only saline dressings were applied at weekly intervals. On every dressing patient's wound was measured in maximum length and maximum breadth by using a scale and area was calculated. % area of wound closure was calculated as- Area of previous day- Area of next day/ Area of previous day X 100. Assessment of the DFUs was done as per University of Texas classification of DFU.

**Results:** Age group 30-50 years had 9 (36%), 51-70 years had 15 (60%) and 71-90 years had 1 (4%) patients. Duration of wound was 4-6 weeks seen in 1 (4%) patients, 6-8 weeks seen in 17 (68%) and >8 weeks seen in 7 (28%) patients. The difference was significant ( $P < 0.05$ ). Comparison of wound dimension at day 0, after 1 week, 2 weeks, 3 weeks, 4 weeks, 5 weeks, 6 weeks and 8 weeks. There was significant difference in all parameters at different intervals ( $P < 0.05$ ).

**Conclusion:** We can conclude that successful treatment of chronic diabetic foot ulcers remains challenging for clinicians. The enhancement of wound healing through the application of PRP is supported by basic scientific and clinical studies

**Keywords:** Diabetic, Foot, Platelet rich plasma, Ulcer

#### 1. Introduction

Diabetes mellitus (DM) is one of the most deceitful diseases that affect more than 371 million people all over the world in 2012; by 2030 this will rise to 552 million. Diabetes mellitus is a clinical syndrome characterized by hyperglycemia caused by absolute or relative deficiency of insulin<sup>[1]</sup>. Diabetes mellitus is of two types. Type 1 DM was previously known as insulin dependent diabetes mellitus (IDDM). It is a T-cell mediated autoimmune disease involving destruction of the insulin secreting beta cells of islet of langerhans of pancreas. Hyperglycemia accompanied by the classical symptoms of DM occurs only when 70-80% of beta cells have been destroyed. Type 2 DM was previously termed as non insulin dependent diabetes mellitus (NIDDM). It is more complex than type 1<sup>[2]</sup>.

Diabetic foot is a frequent site for complication in DM. Ulceration occurs as a result of trauma in the presence of neuropathy and/or peripheral vascular disease with infection as a secondary phenomenon following disruption of the protective epidermis<sup>[3]</sup>. The disease often leads to the development of serious health threatening complications. Of all diabetic complications, diabetic foot syndrome (DFS) is one of the most devastating<sup>[4]</sup>.

Use of autologous platelet-rich plasma (PRP) in the form of local application obtained by centrifugation of whole blood and addition of an activator, clotting agent is designed for the creation of local conditions favourable to healing processes. PRP is defined as plasma fraction of autologous blood with a platelet count concentrated above the baseline. It is a repository of growth factors, cytokines, adhesion molecules and clotting agents, and leukocytes. It is very useful in DFU<sup>[5]</sup>. The present study was conducted to achieve wound healing in diabetic foot

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ulcers and to evaluate the efficacy of PRP dressings in DFUS at different intervals after such dressings.

### Materials & Methods

On the day of surgery patients taken to blood bank. A triple bag (figure 1) was taken and 48 ml of CPD (citrate phosphate dextrose) was removed and discarded leaving just 14 ml of CPD in the bag. 100 ml of patient's whole blood was drawn by a clean, single venipuncture into the 1<sup>st</sup> blood bag. The bag was kept at room temperature (20-22 °C) before preparing platelet concentrate for not more than 6 hours. The bag was kept in the bucket of refrigerated centrifuge (Heraeus Cryofuge 6000i) and balanced accurately and centrifuged at 2000 rpm at 22 °C for 5 minutes. This separated the whole blood into red blood cell concentrate at the bottom and plasma above. 4/5<sup>th</sup> of the plasma was separated into the 2<sup>nd</sup> satellite bag, double sealing the tubing between the primary bag and the satellite bag. The primary bag with RBC concentrate was separated and kept aside. The remaining 2 satellite bags were again centrifuged at 4000 rpm at 22 °C for 10 minutes after balancing accurately. The plasma got separated into an upper layer of platelet poor plasma (PPP) and platelet concentrate (PRP) below. The PPP layer was expressed into the 2<sup>nd</sup> satellite bag, double sealed, separated and kept aside. The PRP (platelet concentrate) extract in the 1<sup>st</sup> satellite bag (figure 2) was approximately 12- 15 ml. PRP was removed from blood bag using aseptic technique and was put in a sterile container (figure 3a). Wound (figure 3b) was cleaned with normal saline. Then sterile gauze was soaked in PRP (figure 3c) and applied over the wound area (figure 3d) & dressing was done. Patient was advised to walk with partial

weight bearing. Dressing was opened after one week when similar dressing with PRP was done again. Then one week later simple normal saline dressing was done & one week later 3<sup>rd</sup> PRP dressing was given. Then at weekly interval normal saline dressings was done upto 6 weeks and then at 8 weeks. The present study was conducted in Orthopaedics Department, Guru Nanak Dev Hospital, Govt. Medical College, Amritsar on patients admitted for treatment of diabetic foot ulcers. It comprised of 25 patients of either gender (males- 15, females- 10). All were informed regarding the study and written consent was obtained. Ethical clearance was obtained from ethical committee.

In this study, 25 patients were examined carefully. Wound size as per maximum length and breadth and area were recorded and three PRP dressings were given on day 0, second after first week, and third after 3rd week. On 2nd week, and 4th, 5th, 6th and 8th weeks only saline dressings were applied at weekly intervals. On every dressing patient's wound was measured in maximum length and maximum breadth by using a scale and area was calculated.

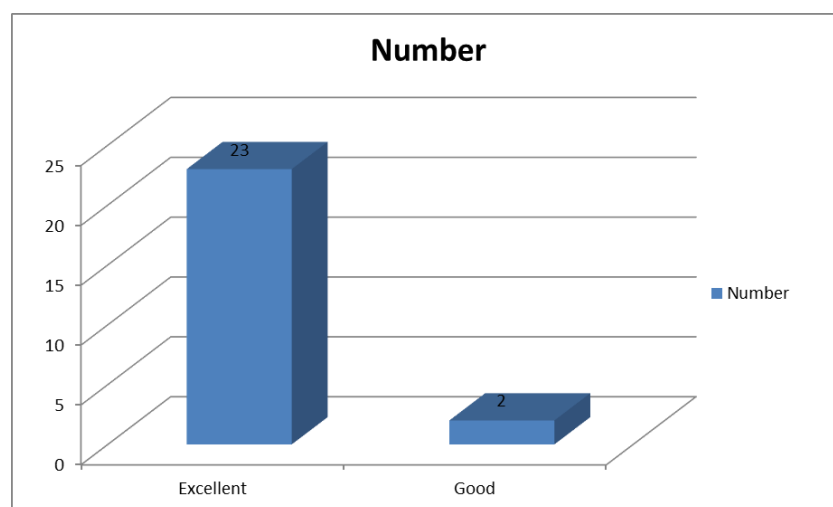
% area of wound closure was calculated as- Area of wound at previous dressing- Area of wound at present dressing/ Area of wound at day 0 X 100. Assessment of the DFUs was done as per University of Texas classification of DFU. Grading of results was 90-100% healed- excellent, 70-90% healed- good, 50-70% healed- fair and less than 50% healed- poor. Results thus obtained were subjected to statistical analysis for correct inferences. P value less than 0.05 was considered significant.

### Results

**Table I:** Comparison between different time periods

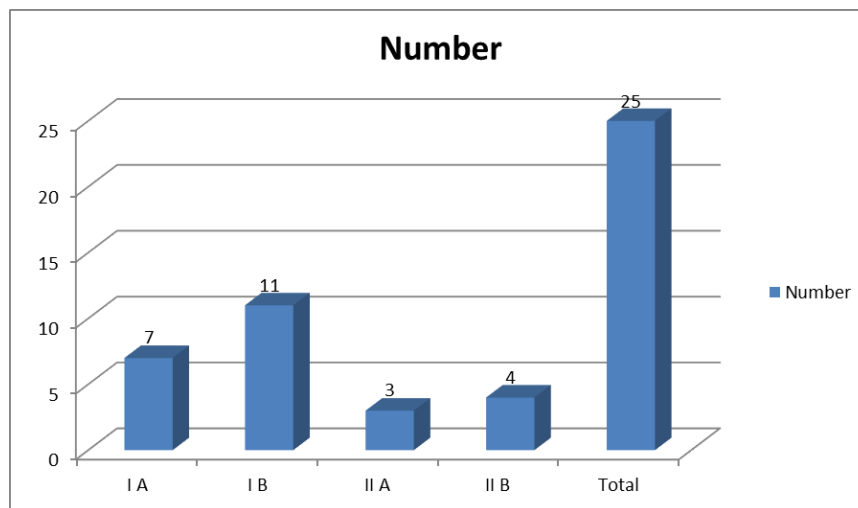
Parameters	Day 0	After 1 week	2 week	3 week	4 week	5 week	6 week	8 week	P value
Length	6.5	5.6	5.03	4.3	3.3	2.22	1.4	0.5	0.01
Breadth	5.5	4.6	3.9	3.5	2.4	1.6	1.02	0.27	0.001
Area of wound	36.2	27.4	17.5	14.1	8.2	1.8	1.1	1.2	0.012
% of wound closure	0	23.7	17.9	20.2	15.5	12.5	5.3	6.9	0.001

Table III shows comparison of wound dimension at day 0, after 1 week, 2 weeks, 3 weeks, 4 weeks, 5 weeks, 6 weeks and 8 weeks. There was significant difference in all parameters at different intervals ( $P < 0.05$ ).



**Graph I:** Grading of results

Graph I shows that 23 showed excellent and 2 showed good results.



**Graph II:** Texas grading in patients

Graph II shows that shows Texas grading. Maximum number of patients had I B (11) followed by I A (7), II B (4) and IIA (3). The difference was significant ( $P < 0.05$ ).

### Discussion

Chronic non-healing leg ulcer is a major health problem. Chronic wounds come with cost and morbidity for the patients and society also.<sup>6</sup> There were 14 (60%) males and 10 (40%) females in present study. Abdelaziz *et al.*<sup>[7]</sup> conducted a study on 27 males and 23 females. We found that maximum number of patients had Texas grade II B (9) followed by I A and II A (6 each). Tran *et al.*<sup>[11]</sup> found Texas grade IA in 4 and IIA in 2 patients. In present study, the mean age of patients was  $54.9 \pm 4.5$  years (males- 57.3 years and females- 52.6 years). The mean age of patients in study by Atef<sup>[8]</sup> was  $48 \pm 7.38$  years. We found that at day 0, the mean length of wound was 5.5 cm, breadth was 5.5 cm and area was 36.2 cm.<sup>2</sup> Babeai *et al.*<sup>[9]</sup> found that mean length and breadth was 5.6cm and 5.8 cm respectively. The mean length of wound after 1 week was 5.68 cm, breadth 4.67cm and area was 27.4 cm.<sup>2</sup> The percentage of wound area closure was 23.7%. Hadi *et al.*<sup>[10]</sup> found 20.1% wound closure after 1 week.

We observed that diabetic foot ulcer (DFU) after 2 weeks, the mean length of wound was 5.03 cm, breadth 3.9 cm and area was 17.5 cm.<sup>2</sup> The percentage of wound area closure was 17.9%. Tran *et al.*<sup>[11]</sup> in their study found that the average time for completed healing was 7.1 weeks. In present study, diabetic foot ulcer (DFU) after 3 weeks, the mean length of wound was 4.31 cm, breadth 3.5 cm and area was 14.1cm.<sup>2</sup> The percentage of wound area closure was 20.2%. There was significant difference in length of wound, area of wound. This is similar to the results of Knighton *et al.*<sup>[12]</sup> who found 18.4% wound closure after 3 weeks. We found that diabetic foot ulcer (DFU) after 4 weeks, the mean length of wound was 3.3 cm, breadth 2.4 cm and area was 8.2 cm.<sup>2</sup> The percentage of wound area closure was 15.5%. Our results are in agreement with Manish *et al.*<sup>[13]</sup> who observed 16% wound closure in 4<sup>th</sup> weeks. We observed that the diabetic foot ulcer (DFU) after 5 weeks, the mean length of wound was 2.2 cm, breadth 1.6 cm and area was 1.8 cm.<sup>2</sup> The percentage of wound area closure was 12.5%. Martinez *et al.*<sup>[14]</sup> found 95% total reduction in size of DFU after 5 weeks. We observed the diabetic foot ulcer (DFU) after 6 weeks, the mean length of wound was 1.4 cm, breadth 1.02 cm and area was 1.17 cm.<sup>2</sup> The percentage of wound area closure was 5.3%. We found significant reduction in size of ulcers after 6 weeks. Murtuzua

*et al.*<sup>[15]</sup> found that 14 out of 15 patients DFU revealed after 6 weeks using PRP dressings.

In this study we found that the diabetic foot ulcer (DFU) after 8 weeks, the mean length of wound was 0.5 cm, breadth 0.27 cm and area was 1.2 cm.<sup>2</sup> The percentage of wound area closure was 6.9%.

We observed significant difference in wound dimension at day 0, after 1 week, 2 weeks, 3 weeks, 4 weeks, 5 weeks, 6 weeks and 8 weeks. Sayedh *et al.*<sup>[16]</sup> in their study found that the mean follow-up period was 12 weeks. The estimated time of wound healing was 12 weeks for 82.5% of the patients in group A and 97.5% of the patients in group B; the PRP group was found to be more effective with fewer complications. Similarly, Vickie *et al.*<sup>[17]</sup> found that the average wound area closure rate per day was 0.051 cm<sup>2</sup> for the PRP gel group versus 0.054 cm<sup>2</sup> for the control group. In the majority wound dataset, the wound area closure per day was 0.042 cm<sup>2</sup> for the PRP gel group and 0.043 cm<sup>2</sup> for the control group.

We observed that in 23 (92%) results were excellent and in 2 (8%) good. We compared wound dimension at day 0, after 1 week, 2 weeks, 3 weeks, 4 weeks, 5 weeks, 6 weeks and 8 weeks. There was significant difference in all parameters at different intervals ( $P < 0.05$ ). Vickie *et al.*<sup>[17]</sup> in their study found that in 82% there was excellent results, in 13% good and in 5% fair.

DFUs seriously affect the life quality of diabetic mellitus patients. Although there are some breakthroughs in the diabetic mellitus foot care to reduce the DFUs, diabetic mellitus patients faced with amputation.



**Fig 1.** PRP concentrates





Fig 2. Autologous PRP concentrates



Fig 3a. Gauze and PRP in sterile container



Fig 3b. Wound area after scraping



Fig 3c. Gauze soaked in PRP



Fig 3d. Application of PRP gauze on wound

Case 1.



Fig 4 a & 4 b: PRP DRESSING APPLIED



Fig 4c: NS DRESSING APPLIED



Fig 4d: NS DRESSING APPLIED

Case 2.



Fig 5a & 5b: PRP DRESSING APPLIED



Fig 5c & 5d: NS DRESSING APPLIED

### Conclusion

We can conclude that successful treatment of chronic diabetic foot ulcers remains challenging for clinicians. The enhancement of wound healing through the application of PRP is supported by basic scientific and clinical studies. Research has shown that PRP are responsible for actively extruding growth factors, which initiate soft tissue healing and recruitment of stem cell. Platelet rich plasma is safe and effective method for treatment of DFUs.

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