

USE OF AUTOLOGOUS PLATELET RICH PLASMA (PRP) IN DIABETIC FOOT
ULCERS TREATMENTDr. Hemant Sharma*¹ and Anubha Sharma²¹Senior Resident, Department of Orthopaedics, Government Medical College, Amritsar.²Senior Resident, Department of Paediatrics, Government Medical College, Amritsar.

*Corresponding Author: Dr. Hemant Sharma

Senior Resident, Department of Orthopaedics, Government Medical College, Amritsar.

Article Received on 01/12/2018

Article Revised on 22/12/2018

Article Accepted on 13/01/2019

ABSTRACT

Introduction: India has 42 million cases of Diabetes, the most common complication of which is Diabetic Foot Ulceration (DFU), affecting 15% of Diabetic population in a lifetime. Non-healing diabetic foot ulcers and the resulting potential amputations present significant costs to the healthcare system and reduce patient's quality of life. Emerging cellular therapies such as platelet – rich plasma (PRP) can have an adjunctive role in a standardized, quality treatment plan. PRPT is a rich source of locally active (bioregulating) growth factors and cytokines that improve conditions of wound healing. The aim of this study was to assess wound healing in DFU treated by PRP. **Materials and Methods:** The study was conducted in Orthopaedics Department, Guru Nanak Dev Hospital, Govt. Medical College, Amritsar from December 2016 to July 2017 on patients admitted for treatment of diabetic foot ulcers. 12 patients of either sex with diabetic foot were selected. PRP dressings were done on 1st, 7th and 21st day, saline dressings were done in the rest of the week and results seen at the end of 8th week. **Conclusion:** Autologous PRP holds great potential in treatment of diabetic ulcer of foot with re-epithelialization at a faster pace possibly due to various growth factors present in it, although duration of wound and grade of wound are also important prognostic factors.

KEYWORDS: DFU: Diabetic foot ulcer, PRP: Platelet rich plasma, wound healing.

INTRODUCTION

India with approximately 42 million cases is ranked first in the list of the ten nations most affected with diabetes.^[1,2] Among diabetes mellitus related complications, foot ulceration is the most common, affecting approximately 15% of diabetic patients during their lifetime.^[2] This can be attributed to several social and cultural practices such as barefoot walking, inadequate facilities for diabetes care and education, and poor socioeconomic conditions.^[3] Limb amputation has a major impact on the individual, not only in distorting body image, but also with regard to loss of productivity, increasing dependency, and costs of treating foot ulcers if patients require inpatient care.^[4] Sporadic qualitative research suggests that diabetic foot ulceration has a profound social impact with patients reporting stigma, social isolation, loss of social role, and unemployment.^[5]

Frykberg et al^[6] cites a 1998 study of 67,000 diabetes-related lower extremity amputation (LEA) and a similar study that resulted in a total of 984,000 hospital days, each length of stay averaging 15 days. Nonhealing diabetic foot ulcers and the resulting potential amputations present significant costs to the healthcare system and reduce patient quality of life. The goal of

diabetic foot ulcer treatment is to obtain wound closure as expeditiously as possible. Accepted therapeutic objectives and standards of care for diabetic foot ulcers include wound debridement, pressure relief in the wound area, appropriate wound management (eg, moist wound healing), infection management, ischemia management, medical management of comorbidities, and surgical management as needed.^[6] Emerging cellular therapies such as platelet – rich plasma (PRP) can have an adjunctive role in a standardized, quality treatment plan. Autologous Platelet rich plasma, including multiple growth factors, have been used to treat wounds since 1985. Following a rapid release of growth factors, platelets contained in PRP synthesise and secrete their additional quantities for subsequent 7 days. After that time, the healing function is taken over by macrophages. Experimental and clinical studies demonstrated the most profound accelerating effect on wound healing in the 3rd week after PRP application.^[7,8]

In vivo prospective controlled studies as well as retrospective and cost effectiveness studies documenting the effect of this therapy have been published.^[9-11] *In vitro* research has shown that platelets contain

components and properties for wound healing,^[12] likewise, plasma contains fibrin matrix.^[13]

In 2001, Margolis^[14] published a retrospective study analyzing the treatment results of 26,599 patients with diabetic neuropathic foot ulcers who had been treated with an autologous platelet releasate. The results suggest that platelet releasate provided with standardized care was more effective than standard care alone.

In the multicentre study by Villela and Santos, and de Leon *et al.*^[15,16] on a group of 200 patients with 285 wounds, PRP could restart the healing process in the majority of cases. Rapid treatment response was observed in 275 of 285 wounds, and the size of the reply was high with reported statistically significant outcomes. Carter *et al.*^[17] completed a meta-analysis on the use of PRP on wounds healing patients with DFU, which led to the conclusion that autologous PRP gel promises as an effective treatment for severe DFU. Dougherty^[18] statistically analysed efficacy versus cost and quality of life of a patient with DFU treated with PRP compared to conventional or other alternative therapeutic methods. Cost analysis completed in the group of over 200 thousand patients involved quality of life, recurrence, necessary amputation, and mortality. The study demonstrated the highest cost effectiveness with the consideration of quality of life for patients treated with PRP. According to the analysts, PRPT is potentially the most attractive alternative for DFU, that may reduce the cost burden and health effects of non-healing DFU.

Therefore, PRP seems to be an effective, if not the most effective and safe preparation used for therapy of DFU. PRP is an autologous product, therefore constitutes no risk of viral hepatitis or HIV infection. Observed abnormal reactions to clotting activators are very rare.

PRP is a rich source of locally active (bioregulating) growth factors and cytokines that improve conditions of wound healing. Relatively simple and cheap production of PRP argues for continued interest in that adjunct method. It seems that specific cellular therapy constitutes an additional and valuable option in therapy of DFU resistant to the conventional therapy.

Therefore the aim of this study was to achieve wound healing in Diabetic foot ulcers which are otherwise very difficult to heal with routine dressing, as well as to evaluate the efficacy of PRP dressings in DFUS at different intervals after such dressings.

MATERIAL AND METHODS

Sampling

The study was conducted in Orthopaedics Department, Guru Nanak Dev Hospital, Govt. Medical College, Amritsar from December 2016 to July 2017 on patients admitted here for treatment of diabetic foot ulcers. 12 patients of either sex with diabetic foot were selected for this study. The informed consent of each patients was

taken on the prescribed proforma. Patient with liver cell failure, severe cardiomyopathy, any major lower limb amputation, bleeding or platelet disorder, low immunity or corticosteroid therapy were excluded from the study.

RESEARCH METHOD

After institutional ethics board clearance, volunteer participants were examined. Wounds' size as per maximum length and breadth and area were recorded and three PRP dressings were given - first on day 0, second after first week, and third after 3rd week. On 2nd week, and 4th, 5th, 6th and 8th wk, only saline dressings were applied at weekly intervals. On every dressing patient's wounds were measured in maximum length and maximum breadth by using a scale and area was calculated.

Assessment of the DFUs was done as per University of Texas classification^[18] of DFU (annexure 1). Patient were also reviewed by MEDICINE Department for control of DM.

Preparation of Autologous Platelet Rich Plasma

On the day of the treatment, the patient was taken to the Blood Bank, GND Hospital, Amritsar. A triple blood bag 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 1st blood bag. The bag was kept at room temperature (20-22 degrees 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 degrees C for 5 minutes. This separated the whole blood into red blood cell concentrate at the bottom and plasma above.

4/5th of the plasma was separated into the 2nd 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 degrees C for 10 minutes after balancing accurately. The plasma separated into an upper layer of platelet poor plasma (PPP) and platelet concentrate (PRP) below. The PPP layer was expressed into the 2nd satellite bag, double sealed, separated and kept aside.

The PRP (platelet concentrate) extract in the 1st satellite bag was approximately 12- 15 ml. This bag was sent to the OT immediately, where it was kept at room temperature before use.

PRP Dressing Procedure

PRP was removed from blood bag using aseptic technique and was put in a sterile container. Wound was cleaned with normal saline and scrapped & pressed to achieve control of bleeding if any. Then sterile gauze was soaked in PRP and applied over the wound area &

dressing was done. Patients was advised to walk with partial weight bearing. Dressing were opened after one week when similar dressing was done again. Then one week later simple normal saline dressing was done & then a week later 3rd PRP Dressing was done which was followed by saline dressings every week till 8th week. At each dressing, area of the wound was measured & change was recorded.

RESULTS

Initially, 12 patients provided informed consent forms and participated in active screening (see Figure 1). Of these patients, 2 were dropped from the study due to failure to meet the inclusion/ exclusion criteria. Ultimately 10 patients were enrolled.

Age and Sex distribution of the group (Table 1 and Table 2), shows that majority of the patients (90%) fall in the age range of 50-70 years. The males outnumbered females with a ratio of 3:2.

Table 1.

Age (Range in Years)	No. of Patients	Percentage
50-60 years	5	50%
61-70 years	4	40%
Above 70 years	1	10%

Table 2.

Sex	No. of patients	Percentage
Male	6	60%
Female	4	40%

It was also seen that the right foot was more commonly involved than the left foot. In 60% of the cases the right foot was involved as compared to 40% with the left.

Table 3.

Foot	No. of patients	Percentage
Right	6	60%
Left	4	40%

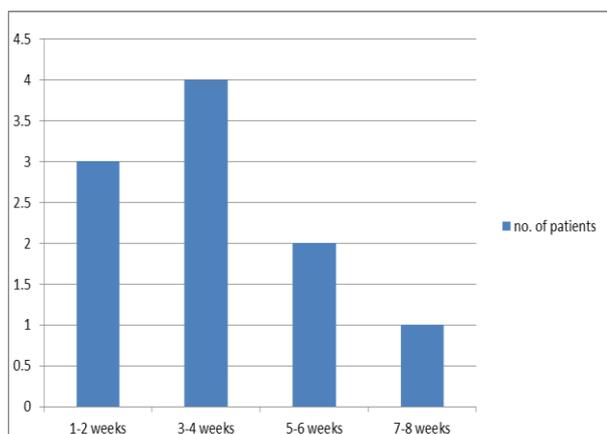


Fig. 1: Duration of wound.

The time of presentation among the patients with the wounds varied in duration, however 40% of the patients presented within 3-4 weeks of the wound, followed by 3 patients who presented within the first two weeks of the injury. (Graph1).

The wound of the patients were also divided into various grades according to Texas classification. About 40% of the patients who presented fell in grade 2, followed by 30% in grade 1 (graph 2). Only one patient had a grade 3 wound.

No. of patients in various grades according to the Texas classification

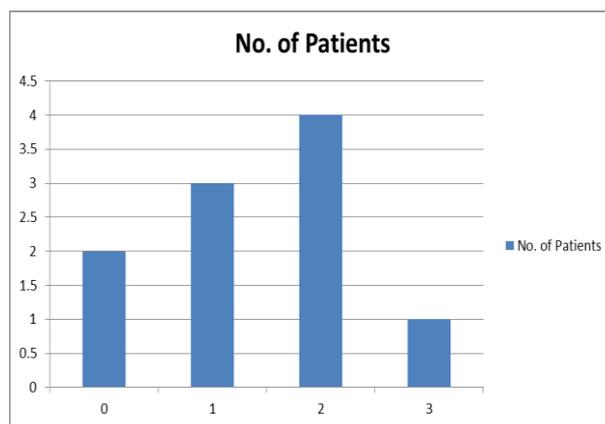


Fig. 2: Grade according to Texas Classification.

The results were graded according to the following scale at the end of 8th week of dressing

Grading of results

- 90 - 100 % healed.....Excellent
- 70 - 90 % healed.....Good
- 50 - 70 % healedFair
- Less than % 50% healedPoor

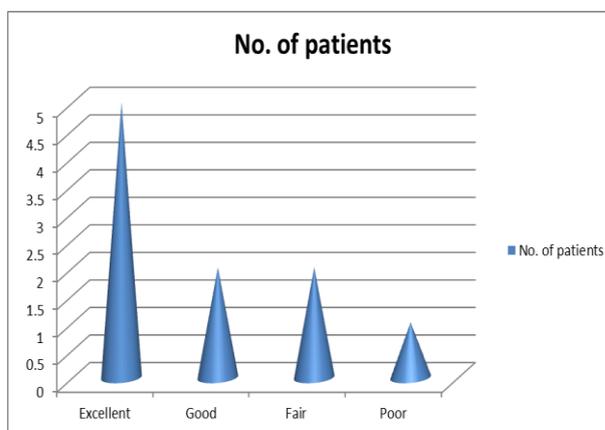


Fig. 3: Grade of wound healing.

Healing of the wound was graded into four grades namely excellent, good, fair and poor. About half of the sample patients (50%) had excellent healing, rest (40%) had good to fair healing whereas only one patient had poor healing.



Fig. 4: Chronic Diabetic wound in 55 year old female after debridement.



Fig. 5: Platelet rich plasma soaked gauze dressing in the same patient.



Fig. 6: Improved soft tissue coverage and good signs of healing with wound contraction at 6 weeks.

DISCUSSION

This was an observational study to appreciate the effect of platelet rich plasma in wound healing of diabetic foot ulcer patients. 9 out of 10 patients included in the study belonged to the age group of 50-70 years which is the time during which the complication of type 2 diabetes mellitus such as foot ulcers usually manifest. There were higher no. of males than females possibly because in our rural set up men turn up for medical attention more easily than women. Right foot was more commonly involved than the left mainly due to the fact that right being the dominant foot gets injured more frequently, similar finding was given by study of Driver *et al*^[19] in whose study there were 23 right feet as compared to 17 left.

The duration after the inception of the wound to its presentation also seems to matter as later the patient came for medical attention, higher was the grade of his/her wound, as it could be understood that out of three patients who had wound duration of 5-8 weeks two had Texas grade 2 wounds and one had grade 3. Earlier the patient came for treatment better was the result. Seven out of ten patients had excellent to good healing which meant 70-100 % closure. These patients presented within first four weeks of their wound and had Texas grade 2 or lower. Driver *et al*^[19] (2006) also reported better results with PRP than control group, when standardized for size, the proportion of completely healed wounds was 13 out of 16 (81.3%) and eight out of 19 (42.1%) in PRP gel and control treatment groups, respectively ($P = 0.036$, Fisher's exact test). The two patients with fair wound healing had high grade infection of their ulcer which was managed by meticulous debridement and antibiotic coverage. All the patients with greater than 50% wound closure had good diabetic control, only one patient who had grade 3 wound with long duration (>8 weeks) of wound had poor healing. This patient also had poor diabetic control.

This observation from our study concurred with findings made by Margolis *et al*^[14] in 2002 who stated that wound duration and grade of wound are important factors in prognosis of wound healing, and if this is not taken into account it can lead to a selection bias which was the reason for not so good result of PRP dressings conducted in his study.

CONCLUSION

Autologous PRP holds great potential in treatment of diabetic ulcer of foot with re-epithelialization at a faster pace possibly due to various growth factors present in it, although duration of wound and grade of wound are also important prognostic factors. However, better statistical support of this theory could have been established with a larger sample size.

Annexure 1: University of texas classification of diabeic foot ulcer.^[4]

	Grade-0	Grade-1	Grade-2	Grade-3
Stage-A	Preulcerative or postulcerative lesion completely epithelialized	Superficial wound, not involving tendon, capsule or bone	Wound penetrating to tendon or capsule	Wound penetrating to bone or joint
Stage-B	Infection	Infection	Infection	Infection
Stage-C	Ischemia	Ischemia	Ischemia	Ischemia
Stage-D	Infection and ischemia	Infection and ischemia	Infection and ischemia	Infection and ischemia

REFERENCES

- Ramachandran A, Ma RCW, Snehalatha C. Diabetes in Asia. *Lancet*, 2010; 375: 408- 418.
- Shankhdhar KKK, Shankhdhar U, Shankhdhar S. Diabetic foot problems in India: An overview and potential simple approaches in a developing country. *Current Diabetes Reports*, 2008; 8: 452-457.
- Viswanathan V, Shobhana R, Snehalatha C, Seena R, Ramachandran A. Need for education on foot care in diabetic patients in India. *J Assoc Physicians India*, 2009; 47: 1083-1085.
- Shobhana R, Rao PR, Lavanya A, Vijay V, Ramachandran A. Cost burden to diabetic patients with foot complications: a study from Southern India. *J Assoc Physicians India*, 2000; 48: 1147-1150.
- Harrington C, Zagari MJ, Corea J, Klitenic J. A cost analysis of diabetic lower extremity ulcers. *Diabetes Care*, 2000; 23: 1333-1338.
- Robert G. Frykberg, Thomas Zgonis, David G. Armstrong, Vickie R. Driver, John M. Giurini, Steven R. Kravitz, Adam S. Landsman, Lawrence A. Lavery, J. Christopher Moore, John M. Schuberth, Dane K. Wukich, Charles Andersen, John V. Vanore. *Diabetic Foot Disorders: A Clinical Practice Guideline (2006 Revision)*, September–October 2006; 45(5): Supplement, S1-S66.
- Pietramaggiore G, Scherer SS, Mathews JC, et al. Healing modulation induced by freeze-dried platelet-rich plasma and micronized allogenic dermis in a diabetic wound model. *Wound Repair and Regeneration*, 2008; 16(2): 218–225.
- Slesacek T, Paetzold H, Nanning T, et al. Autologous derived, platelet-rich plasma gel in the treatment of nonhealing diabetic foot ulcer: a case report. *Therapeutic Advances in Endocrinology and Metabolism*, 2012; 3(2): 75–78.
- Cenni E, Ciapetti G, Pagani S, Perut F, Giunti A, Baldini N. Effects of activated platelet concentrates on human primary cultures of fibroblasts and osteoblasts. *Journal of Periodontology*, 2005; 76(3): 323–328.
- Kark LR, Karp JM, Davies JE. Platelet releasate increases the proliferation and migration of bone marrow-derived cells cultured under osteogenic conditions. *Clinical Oral Implants Research*, 2006; 17(3): 321–327.
- Knighton DR, Hunt TK, Thakral KK, Goodson WH., III Role of platelets and fibrin in the healing sequence. An in vivo study of angiogenesis and collagen synthesis. *Annals of Surgery*, 1982; 196(4): 379–388.
- Borzini P, Mazzucco I. Platelet-rich plasma (PRP) and platelet derivatives for topical therapy. What is true from the biologic view point? *ISBT Science Series*, 2007; 2(1): 272–281.
- Lacci KM, Dardik A. Platelet-rich plasma: support for its use in wound healing. *Yale Journal of Biology and Medicine*, 2010; 83(1): 1–9.
- Margolis DJ, Kantor J, Santanna J, Strom BL, Berlin JA *Diabetes Care*. Effectiveness of platelet releasate for the treatment of diabetic neuropathic foot ulcers, 2001 Mar; 24(3): 483-8.
- D.L.Villela and V.L.C.G.Santos, “Evidence on the use of platelet-rich plasma for diabetic ulcer: a systematic review,” *Growth Factors*, 2010; 28(2): 111–116.
- J. M. de Leon, V. R. Driver, C. P. Fylling et al., “The clinical relevance of treating chronic wounds with an enhanced near-physiological concentration of platelet-rich plasma gel,” *Advances in Skin & Wound Care*, 2011; 24(8): 357–368.
- M. J.Carter, C. P. Fylling, and L.K. Parnell, “Use of platelet rich plasma gel on wound healing: a systematic review and meta-analysis,” *Eplasty*, 2011; 11: article e38.
- E. J. Dougherty, “An evidence-based model comparing the costeffectiveness of platelet-rich plasma gel to alternative therapies for patients with nonhealing diabetic foot ulcers,” *Advances in Skin & Wound Care*, 2008; 21(12): 568–575.
- Driver VR1, Hanft J, Fylling CP, Beriou JM; Autologel Diabetic Foot Ulcer Study Group. A prospective, randomized, controlled trial of autologous platelet-rich plasma gel for the treatment of diabetic foot ulcers. *Ostomy Wound Manage*, 2006 Jun; 52(6): 68-70, 72, 74.