



ROLE OF PLATELETS DERIVE GROWTH FACTOR IN MANAGEMENT OF DIABETIC FOOT

General Surgery

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ABSTRACT

BACKGROUND: Recently the world health organization reported a global prevalence of diabetes exceeding 300 million people, predicting a further 60-70% increase by the year 2030, which means India alone will have 100 million people by the year 2030. Wound healing is problematic in diabetic patients. Encouraging results have shown that PDGF application is better than good wound care alone. But the evidence to demonstrate the safety and efficacy of PDGF in diabetic ulcer is scanty.

METHODS: The present study was carried out at Govt. Medical College & Hospital, Bettiah, W. Champaran, Bihar for a period of one year, where 50 patients with diabetic foot ulcers were included in the present study. Prospective randomized controlled trial was designed for the study. 50 patients with diabetic foot ulcers admitted in surgery wards at GMC, Bettiah, W. Champaran, Bihar over a period from July 2020 to June 2021. 25 patients were in the study/treatment group, 25 patients were in the control group.

RESULTS: Out of 50, patients, 25 took treatment in the form of conventional normal saline dressings and 25 took treatment with rh - PDGF dressing once a day. Glycaemic control and adequate control of infection was maintained in both the groups. X-ray foot was taken for all patients and bony involvement was excluded. The initial area measurement was taken on day 01 and final area measurement on day 15 was taken on transparent sheet. Planimetry was used to measure the outcome that is the target ulcer area using a transparent graph sheet. Results were calculated by using student 't' test.

CONCLUSIONS: The wounds in the study group treated with rh-PDGF dressing contracted more than the wounds in the control group (38.55% Vs 12.79%; $P \leq 0.001$ - statistically significant) which indicates rh-PDGF dressing is an effective modality to facilitate wound contraction in patients suffering from diabetes. Rh-PDGF dressing is found to be more effective, safe promoter of wound healing and can be used as an adjunct to saline dressing for healing of diabetic wounds and healing of ulcers receiving PDGF was significantly faster as compared to ulcers receiving placebo.

KEYWORDS

Diabetic foot ulcer, PDGF, Ulcers, Wound healing

INTRODUCTION

Diabetes mellitus (DM) is a state of chronic hyperglycaemia producing complication like neuropathy, nephropathy and retinopathy. It is also a strong co-factor in causing atherosclerotic disease dyslipidemia. The microvascular and macrovascular complication prevalence is 46% and 64% respectively. Diabetes is the commonest cause of non-traumatic lower extremity amputations. It has been reported that annually, about 1 to 4 percent of those with diabetes develop a foot ulcer. Invariably the diabetic foot ulcers are chronic ulcers that are resistant to heal because of multidrug resistant organism growth and microvascular complications.^{1,2}

Recent advances in concept of wound healing and the factors and cell types involved in wound healing have opened a pathway for curing chronic ulcers. PDGF is one among the growth factors important in angiogenesis and regeneration that is used in treating chronic ulcers. PDGF are derived from platelets which contain alpha and beta granules. The rh-PDGF is produced by recombinant DNA process by inserting the human gene for the B chain of the Growth Factor in the yeast *saccharomyces cerevisiae*.³ According to WHO report India today heads the world over 32 million diabetic patients.⁴

Recently WHO reported a global prevalence of diabetes exceeding 300 million people, predicting a further 60 - 70 % increase by the year 2030 which means India alone will have 100 million people by the year 2030.⁵ As a result, a parallel increase in the incidence of diabetic lower extremity ulcer is expected. At-least 15% people with diabetes eventually develop a lower extremity ulcer of some sort.⁶ Numerous risk factors for the development of foot ulcers has been suggested, the most important being peripheral sensory neuropathy followed by peripheral vascular disease. The proportion of neuropathic, neuroischemic and purely ischemic lesions in diabetes is 54%, 34% and 10% respectively.⁷

Over 100 physiological factors contribute to wound healing in individuals with diabetes. These include decreased or impaired growth factor production, angiogenesis response, macro phage function, collagen accumulation, epidermal barrier function, quantity of

granulation tissue, keratinocyte and fibroblast migration and proliferation, number of epidermal nerves, bone healing and balance between the accumulation of ECM components and their remodeling by the MMP's. The main reason is related to the loss of balance between metalloproteinase (MMPs) and MMP inhibitor.⁸

Presently the diabetic foot ulcers treated with some physical therapies such as vacuum assisted closure, high voltage pulsed current electrical stimulation, hyperbaric oxygen therapy (HBOT), negative pressure wound therapy (NPWT), some biological therapies were also evaluated in diabetic foot ulcer treatment.^{9,10,11} Some growth factors such as Epidermal growth factors, granulocyte colony stimulating factor, nerve growth factor, vascular endothelial growth factor activated platelet rich plasma were evaluated in diabetic foot ulcers.¹²

Platelet derived growth factor PDGF is a dimeric protein, composed of 2 disulfide - linked polypeptide chain. It exists in 3 different isomers, the heterodimer PDGF - ab consisting of an a and b chain, and two homodimers, consisting 2a or 2b chains (pdgf - aa and pdgf - bb) it has been shown in preclinical and clinical studies to promote the formation of granulation tissue and to stimulate wound healing.¹³ Microscopic examinations of the wound treated with topical PDGF showed a marked increased intensity of the inflammation phase of the wound healing cascade characterized by an increased presence of neutrophils, monocytes and fibroblast. It is hypothesized that PDGF positively promotes angiogenesis indirectly through its activities on other inflammatory cells.

Encouraging results have shown, that PDGF is better than good wound care alone.¹⁴ The average time for healing was shorter with greater reduction in ulcer size.¹⁵ Clinical trials conducted in western countries have demonstrated the safety and efficacy of PDGF in the management of diabetic foot ulcer but only few trials are conducted in India hence the need for this study in our setup.

Aim of the study was to evaluate the efficacy of PDGF over saline dressing in healing of diabetic ulcers of the foot, to compare and analyze the distribution of diabetic ulcers of the foot with age, sex,

location of the ulcer (plantar or dorsum).

MATERIAL AND METHODS

Prospective randomized controlled trial design was done for study. 50 Patients with diabetic foot ulcers admitted in surgery wards at GMC, Bettiah, W. Champaran, Bihar over a period from July 2020 to June 2021. 25 patients were included in study/treatment group. 25 patients were in control group.

Inclusion criteria were patients with diabetes, wagner's stage I, II, III target ulcer more than 4 week duration. Exclusion criteria were radiological evidence of under lying osteomyelitis, ulcers resulting from any other cause (e.g. electrical, chemical, radiation etc.) any concomitant disease (example connective tissue disease), any medication affecting healing (e.g. steroid), pregnant women, ankle brachial index <0.4, poor nutritional status, (<6.5gms% total proteins and albumin <3.5 gm%). Ulcers were defined as break in continuity of epithelium of skin and mucus membrane. The lower extremity neuropathic ulcers were randomized. If the patient had one ulcer it was randomized either for treatment group or control group.

If the patient had two ulcers one was randomized for treatment group and the other for control group before randomization the target was debrided. Eligibility for randomization was; full medical history, complete examination, radiographs and doppler of lower extremity with other relevant investigations. Once eligibility was confirmed, particulars of target ulcers like surface area were measured. The ulcer was classified according to Wagner's grading. Thereafter these ulcers were randomized to ulcer treated with placebo gel, ulcer treated with PDGF gel.

Both placebo gel and PDGF gel were provided by the same manufacturers and had similar packing. The wounds were covered with approximately 1.5 mm layer of PDGF gel and moist saline dressing. Adequate control of infection was done by giving oral or injectable antibiotics and debridement done where required. The intended period of treatment was 6 month/ complete healing which ever was earlier.

At each follow up visit at an interval of 1 week for 8 weeks and then every 2 weeks till 12 weeks and after that every 4 weeks for 24 weeks, area of target ulcer was assessed clinically for granulation, percentage decreased in size and culture sensitivity.

Association between drug used and wound healing was calculated using the Chi square test. Comparison of all the other discrete variables was done using Chi square test. Statistical significance was determined by P-value <0.05.

Using a pretested and predesigned proforma the study population was randomized into either study group or control group using an open label randomization technique. Out of 50, patients, 25 took treatment in from of conventional normal saline dressings and 25 took treatment with rh - PDGF dressing once a day. Glycemic control and adequate control of infection was maintained in both the groups. If culture grows organism, both control and study group cases would be treated with antibiotics as per culture sensitivity report. X-ray foot was taken for all patients and bony involvement was excluded. The initial area measurement was taken on day 01 and final area measurement on day 15 was taken on transparent sheet. Plannimetry was used to measure the outcome that is the target ulcer area using a transparent graph sheet. Results were calculated by using student 't' test.

For saline dressing, the ulcer was cleaned with normal saline soaked gauze piece was kept over the ulcer which was covered with pad and roller bandage.

For rh-PDGF dressing, the infected ulcer was cleaned with normal Saline. Commercially available rh-PDGF_BB gel (0.01) was applied on the gauze piece and put on the ulcer. It was then covered with pad and roller bandage.

The dressings were changed daily morning in either control and study group and appearance of healthy granulation tissue is observed. The initial area and final area of the size of the ulcer are measured on 15th day by planimetry using a transparent graph sheet and subject to statistical analysis.

The following formula was used to calculate % reduction in area of wound after 15 days period in both cases and control groups.

$$\text{Rate of contraction of wound after 15 days of treatment} = \frac{\text{Initial Area} - \text{Final Area}}{\text{Initial Area}} \times 100$$

RESULTS

Table 1 : Age Distribution

Age (years)	No. of cases	Percentage
20-30	02	4
31-40	06	12
41-50	11	22
51-60	19	38
>60	12	24
Total	50	100

In our study it was observed that diabetic foot was commonest in the age group between 51-60 years of age.

Table 2 : Sex Distribution

Sex	No. of cases	Percentage
Male	34	68.00
Female	16	32.00
Total	50	100

In our study it was observed that diabetic foot was more common in the males (68.00%) as compared to females (32.00).

Table 3 : Site of ulcer in the study

Site	No. of cases	Percentage
Plantar	28	56.00
Dorsum	22	44.00
Total	50	100

In our study it was observed that diabetic foot more commonly occurs on the plantar aspect (56%) of the foot as compared to the dorsal aspect (44%).

Table 4 : Onset of diabetic foot ulcers

Type of onset	No. of cases	Percentage
Traumatic	32	64.00
Spontaneous	18	36.00
Total	50	100

Trauma is then most common cause of diabetic foot ulcer (64%) while only 36% were spontaneous in origin.

Table 5 : Anti diabetic agents

Anti-diabetic	No. of cases	Percentage
OHA	9	18.00
Insulin	41	82.00
Total	50	100

In our study most of the participants were taking insulin for glycaemic control.

Table 6 : Wound Contraction

Group	Mean area reduction	±S.D.	Median	p-value
Control	12.79%	2.55	11.81	P<0.001
Study	38.55%	2.52	37.58	

In our study it was observed that mean % of area reduction was higher in study group (38.55%) as compared to the controls (12.79%).

Diabetic foot ulcers in the study group had better mean % of wound contraction of 38.55% as compared to the control group which had mean % of wound contraction of 12.79%, the difference in the mean is 25.76%, which is studied using unpaired student t test was found to be significant (p <0.001).

Table 7 : Time to achieve wound healing

Time to achieve wound healing (weeks)	Drug 1 (Placebo)		Drug 2 (PDGF)		Total
	No. of ulcer	%	No. of ulcer	%	
1-5	5	29.4	10	55.6	15
6-10	3	17.7	6	33.3	9

11-24	5	29.4	2	11.1	7
Did not heal	4	23.5	0	0	4
Total	17	100	18	100	35

At the end of 24 weeks 100% ulcers in group receiving drug 2 (PDGF) achieved score of 1 and in group receiving drug 1 (placebo) 76.4 % ulcers achieved score of 1. Percentage of healing in ulcers was 50 % better in group receiving drug 2 (PDGF).

There was no relation of age, gender, type of diabetes mellitus, duration of diabetes mellitus, regular/irregular treatment of diabetes mellitus, body mass index, ankle brachial index, neurological deficit, type of ulcer, foot deformity, Wagner grade, edema, granulation, infection, duration of ulcer, area of ulcer, absolute lymphocyte count, glycemic control, total protein and albumin with time to achieve wound closure. This means that PDGF was solely responsible for the ulcers to heal 50 % faster.

DISCUSSION

Diabetic foot ulcers are chronic wounds, with prolonged inflammation phase and shows cessation of epidermal growth. Invariably the diabetic foot ulcers are resistant to heal because of multidrug resistant organism growth and microvascular complications. In the present study it was seen that the incidence of diabetic foot ulcers were more in males [68%] as compared to females (32%). The second national data source, NHDS documented more males suffering from diabetic foot ulcer than females. Diabetic foot ulcers are most commonly seen in 5th decade next common in the sixth decade, 22 % of patients were in fourth decade. Older the patient more the prevalence of having diabetic foot ulcer. In this study patients with vascular complications such as pulse less limb and the patients with osteomyelitis were excluded. In this study, 64% of the ulcers were traumatic in origin trauma being the triggering factors secondary to neuropathy. 36% were spontaneous in origin secondary to blister rupture or unnoticed trivial trauma.

More than half (56%) of the patients had ulcer on the plantar surface of the forefoot and the remaining (44%) had on the dorsum of foot. Study conducted by Edmonds et al. showed more foot ulcers were on plantar and fore foot areas.¹⁶ Most of the diabetic foot ulcers are invariably due to poor foot care and due to gait abnormalities.

Most of the patients (82%) were on insulin for control of sugar whereas only 18% were on oral hypoglycaemic agents. In our study it was observed that participants receiving rh-PDGF dressing had better wound contraction of 38.55. As compared to the group receiving only conventional dressing (normal saline dressing) in whom the mean wound contraction was 12.79% these were found to be statistically significant on unpaired student t test ($p < 0.001$) suggesting that rh-PDGF dressing enhances wound healing in diabetic wounds.

Study have found 12.79% (S.D; 2.55: Median; 11.81) contraction of wounds in the control groups as compared to 38.55% (S.D; 2.52: Median; 37.58) contraction of wounds in study group. Therefore, study groups are having 25.76% more wound contraction as compared to control group. On applying unpaired student t-test $p < 0.001$ which is statistically significant. From our study, we can say that rh-PDGF dressing therapy facilitates wound healing in patients suffering from diabetes mellitus.

Mean age which was in 5th decade in our study was almost similar to most of the studies. Majority of the patients were in the 6th decade according Margolis et al.¹⁷ The lower age of the patients presenting with diabetic lower extremity ulcer can be attributed to life style changes due to urbanization which increases the risk of developing lower extremity diabetic ulcers. Male to female ratio was 1.6:1. This finding was similar to the study done by Hardikar et al.¹⁸ Higher incidence of males over females could be due to the fact that females have a largely indoor existence in our society. Most of the ulcers patients had duration of diabetes < 10 years. This was similar to the study done by Ogbera et al which had average duration of diabetes $8.4 \text{ years} \pm 5.4$ in patients developing diabetic ulcers.¹⁹

A study done by Boyko et al also known as the seattle diabetic foot study showed average duration of diabetes in patients in developing diabetic ulcers to be $12.9 \text{ years} \pm 9.6$.²⁰ This could be attributed to the fact that neuropathy and foot deformity develop after many years of diabetes mellitus which is the leading cause of developing ulcers in diabetes ulcers. Most of the ulcers were neuropathic in this study, our findings are similar to those of Mam et al.²¹ Infection was found to be

more in group receiving drug 2 (PDGF). Our findings were almost similar to the study done by Wieman et al.²²

In spite of more infection in group receiving PDGF all the ulcers healed whereas there was less number of infected ulcers in group receiving placebo. Percentage healing in ulcer was 50% better in group receiving drug 2 (PDGF). Our findings were similar to Wieman et al, Margolis et al, Embill et al, Nagai et al, Lone et al and Hardikar et al.²³⁻²⁶

CONCLUSION

The wounds in the study group treated with rh-PDGF dressing contracted more than the wounds in the control group (38.55% versus 12.79%; $P \leq 0.001$ - statistically significant) which indicates rh-PDGF dressing is an effective modality to facilitate wound contraction in patients suffering from diabetes. Rh-PDGF dressing is found to be more effective, safe promoter of wound healing and can be used as an adjunct to saline dressing for healing of diabetic wounds.

In spite of having many diabetic patients with ulcers presenting in our OPD the sample size appears to be small as compared to generalized population due to exclusion criteria, in order to get almost identical type of ulcers in which no other factors play a role in healing. However healing of ulcers receiving PDGF was significantly faster as compared to ulcers receiving placebo.

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