Original Research Paper

Surgery

EFFECTIVENESS OF AUTOLOGOUS PLATELET RICH PLASMA ON CHRONIC NONHEALING WOUNDS

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ABSTRACT

Background: In developing countries like India chronic non healing wounds serves as a huge burden to the patient, family and economy. Platelet extract such as autologous platelet rich plasma (PRP) are known to deliver various growth factor (GFs) which acts locally and accelerates the process of wound healing.

Methodology: A prospective interventional study on patients admitted as inpatients for management of chronic non healing wounds to the Department of General Surgery, Dr.D.Y.Patil Hospital, Kolhapur, from May 2015 to May 2017. A total of 62 patients were under the study.

Results: Autologous PRP showed accelerated and better healing rates. The mean reduction in area was statistically significant in the study group. There were no adverse effects or reactions seen with the use of autologous platelet gel among the study group.

Conclusions: PRP is a simple, safe and effective method of wound care in management of chronic non healing wounds.

KEYWORDS : Platelet rich plasma, chronic non healing wound, growth factors.

Introduction

Chronic wounds are those which show no measurable healing progress i.e. 20 to 40% reduction in the area even after 2 to 4 weeks of optimal treatment, resulting in rise of morbidity and mortality and constituting a significant burden on the economy of health-care system due to requirement of long term care.(1) Chronicity of wounds is associated with various factors. Majority of these nearly 90% are as a consequence of progression from diabetic wound, pressure sores and venous ulcer.(2) Approximately 80% of lower limb amputations are anteceded by foot ulcers. One of the considerable causes of non-traumatic amputation worldwide is diabetes, with frequency being 15 times higher than in non - diabetic population.(3)

The natural process of wound healing in the body involves an intricate and dynamic cascade of events following surgical intervention, trauma, etc., leading to the rehabilitation of injured tissues. These cascades of events activated by tissue injury are composed of four overlapping but well elucidated phases: hemostasis, inflammation, proliferation and remodeling. Chronic wounds however, may be due to delay in any of the four phases of wound healing, but most frequent derangement occurs in the inflammatory or the proliferative phases derangements(4) or have unusually elevated or depressed levels of cytokines, growth factors, or proteinases.

The conventional treatment includes advance therapeutics with drugs (such as antibiotics), intense local dressings, numerous surgical debridement, compression bandage and various surgical interventions/ reconstructions such as skin grafting. Despite of all these measures, they fail to provide the necessary growth factors that can regulate restorative healing process.(5, 6) Therefore, it is highly recommended to understand the underlying cause to promote speeding of wound healing which would enhance the quality of life, productivity at work and lessen the economic impact on health care system. Management of chronic wounds requires a multidisciplinary team of surgeons, diabetic physician, orthopedic surgeon, podiatrist, vascular surgeons, orthotics', neurologist, physiotherapist and social workers as it is associated with a multitude of factors.

Our country India, as a developing nation has a large rural population with compromised mobility and finds it difficult to commute to a hospital due to poor socioeconomic status and lack of awareness of wound care, eventually leading to chronicity of wounds and thereby rise in morbidity and mortality. Thereby, it is important to reach out to the patients with a wound care method like autologous PRP that has a significant economic impact by reducing number of dressings, hospital stay, convenient to the patients, safe, readily available, easy to prepare and inexpensive.

Aims and Objectives:

a) To study the effectiveness of Autologous Platelet Rich Plasma (PRP) on chronic non- healing wound.
b) To determine acceleration of wound healing by autologous PRP.
c) To assess the time taken for complete wound closure using autologous PRP.
d) To determine the advantage of use of autologous PRP to conventional dressing.

Materials and Methods:

A prospective randomized control study was carried out on 62 patients who presented to the surgical OPD of Padmashree Dr.D.Y.Patil Hospital and Research Institute, Department of General Surgery between May 2015 to May 2017.

Inclusion Criteria: Patients of age above 18 years presenting with chronic non healing wound >4weeks with wound area >16cm^2, hemoglobin >10mg/dl and clinically non infected.

Exclusion criteria: Patients who are not willing for follow up, platelet count > 1lakh/ μL, pregnancy, severe wound infection with osteomyelitis, cellulitis, ischemia or gangrene and any skin malignancy. Receiving systemic corticosteroids, or immunosuppressive agents, hyperbaric oxygen therapy (HBOT), electro-stimulation, growth factors or, any cell or tissue derived products for wounds during the 30 day preceding screening.

After admission detailed history and thorough clinical examination was done followed by routine and necessary investigations. Preceding screening, the wound was prepared by thorough cleansing and debridement of necrotic tissue and antibiotic regime as per culture and sensitivity of the wound. Enrolled patients were randomly assigned in a 1:1 ratio to have dressing of the wound done either with autologous PRP and conventional (povidone–iodine 5% and normal saline) dressing. One group with Autologous PRP weekly dressing and monitoring was done whereas in the other group with conventional method daily dressing and weekly monitoring was done.

Group A: Dressing of wound weekly with Autologous Platelet Rich Plasma.
Group B: Conventional daily dressing (povidone-iodine 5% and normal saline).

Steps of PRP preparation:
Step 1: Collection of blood: Under all aseptic precautions, 20 to 30 ml of whole blood was drawn by performing a venipuncture from the antecubital region and collected in anticoagulated vaccutainer tubes (heparin bulb). The tubes were shaken thoroughly to ensure mixing of anticoagulant with drawn blood.

Step 2: Separation of platelets: The collected blood was placed in the portable centrifuge and subjected for the first spin at 1400 rpm for 15 minutes which separates the blood into 3 layers (bottom –RBC's, middle-buffy coat, top-plasma).The plasma containing platelets was collected using a pipette and transferred to another bulb.

Step 3: Preparation of PRP: The collected plasma was subjected for second spin at 2100 rpm for 15 minutes which separates it into upper 2/3rd volume of platelet poor plasma (PPP) which is discarded and lower 1/3rd yields concentrated platelet rich plasma(PRP). The PRP was aspirated and kept aside for use over the wound.

Intervention:
In group A, autologous PRP was applied over the wound by using insulin /tuberculin syringe over a cleaned wound. The application was made in a spraying technique for equal and proper spread of the autologous PRP over the wound which formed a layer. The wound was covered with nonadherent, soothing and minimal pain at dressing change material marketed as Bactiguage followed by a layer of sterile dressing pad and bandaging was done. While group B, povidone-iodine IP 5% and normal saline 0.9% was used for daily dressing of the wound in traditional manner.

Weekly assessment and evaluation of the wound during hospital stay and after discharge weekly follow-up for 21 weeks or complete wound closure, whichever is early. Prompt clinical evaluation was done if infection was suspected or diagnosed and relevant microbiologic samples were sent for culture & sensitivity and treated with antibiotics.

The progress of wound healing were assessed by measuring length and width of the wound by using a measuring tape and photographs were taken at the beginning and weekly for a period of 21 weeks or complete wound healing, whichever is earlier.The secondary end point of the study was achievement of complete wound closure with the aid of skin grafting in either group. 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).[6]

The data thus collected was analyzed to determine the effectiveness of autologous PRP on chronic non healing wounds.

Observation and Results:
a) Sex Incidence (Table 1)

	No. of cases	Percentage
Male	42	67.74
Female	20	32.26%
Total	62	100%

b) Age incidence (Table 2)

Age Group In Years	Total Male And Female	%
21-30	2	3.22%
31-40	9	14.51%
41-50	18	29.04%
51-60	15	24.19%
61-70	11	17.75%
71-80	6	9.68%
81-90	1	1.61%
Total	62	100%

c) Predisposing factors (Table 3)

Predisposing factors	No. of patients	Percentage
Infection	28	45.16%
Diabetes	15	24.19%
Trauma	10	16.14%
Varicose Vein	4	6.45%
PVD	2	3.22%
Pressure	3	4.84%
Total	62	100%

d) Division of autologous PRP and conventional dressing in group

Group I and group II respectively (Table 4)

	Male	%	Female	%	Total male and female	%
Group "A" autologous PRP dressing	19	30.65%	13	20.97%	32	51.62%
Group "B" conventional dressing	23	37.10%	7	11.29%	30	48.38%
Total	42	67.74%	20	32.26%	62	100%

e) Rate of wound healing (Table 5)

Rate of wound healing	Group A No. of patients	Percentage	Group B No. of patients	Percentage	Total No. of patients	Percentage
0-2.5	6	18.80%	11	36.70%	17	27.42%
2.6-5	19	59.40%	12	40%	31	50%
5.1-7.5	2	6.20%	5	16.70%	7	11.29%
7.6-10	3	9.40%	2	6.60%	5	8.06%
10.1-12.5	2	6.20%	0	0%	2	3.23%
Total	32	100%	30	100%	62	100%

f) Time taken for reduction in wound area (Table 6)

Percentage of reduction of area of wound	Mean duration in weeks		t value	p value
	Group A	Group B		
25%	2.025±0.86 (n=32)	3.026±0.94 (n=30)	4.34	<0.0001**
50%	3.12±1.24 (n=30)	4.97±1.39 (n=25)	5.11	<0.0001**
75%	4.62±2.00 (n=27)	6.67±1.51 (n=24)	4.13	<0.0001**
90%	6.00±2.70 (n=26)	8.2±1.97 (n=24)	3.29	<0.0019**
100%	7.92±3.48 (n=26)	10.41±2.48 (n=24)	2.93	<0.0053**

g) Duration of hospital stay (Table 7)

		Duration in days	P value
	Mean	Standard deviation	
Group A	26.9	12.9	0.0009
Group B	39.7	9.2	

Figure1:Showing complete epithelization of wound after 4 weeks of PRP dressing.

Discussion:
Age and sex incidence:
In 6 of the Cochrane studies conducted on effect of PRP on chronic wounds, the mean age of the cases and controls were largely in 60-70 years age group as compared to the present study, where the mean age was in the 40-70 years age group. In Sakata study(7), percentage of women participating was 23% and 32.26% the present study. But the present as well as in the remaining studies, the difference in age between cases and controls was found to be statistically insignificant.

Predisposing factor:
In relation to the theories, the leading causes of chronic wound include diabetes, peripheral vascular disease, venous disease and pressure sores, followed by various local and systemic causes. Stadelman has

stated that hypoxia is a significant contributing factor in the formation and failure of healing of vascular ulcers.(8)

According to a study carried out in Germany, venous insufficiency was the dominating causative factor in 47.6% and arterial insufficiency in 14.5%, and 17.6% of ulcers were due to combined arterial and venous insufficiency. Rarer causes included vasculitis (5.1%), exogenous factors (3.8%), and pyoderma gangrenosum (3%).(9)The study of Sakata 2012 observed that the major proportion of the wounds was a complication of DM (73%) followed by arterial ulcers (13%) and pressure ulcer.(7)

Unlike other studies, the present study had a sizable proportion of the wounds of nonspecific infective aetiology – nearly 45.16 % in both cases and controls, followed by DFU.

Rate of wound healing:
The experimental group showed filling up of wound bed with granulation tissue, epithelialization and contraction of wound. In the Krupski trial, the rate of healing in cm2/week was studied as a secondary outcome and the rate of healing in the control group was 1.9 ± 2.7cm2/week. In contrast, the wounds in the experimental group increased in size and thus the values are recorded as negative numbers - 4.3 ±12.2 cm2/ week. Whereas, in Anil and Sindhuri 2016 (10) study the rate of healing in the control group was 85 ± 1.67cm2/ week and platelet group was 11.87 ± 9.71cm2/week. The present study observed that the rate of healing in group A (3.77±1.69cm2/week) was more than in group B (3.40±1.46cm2/week) but statistically insignificant. In the above discussed three trials the percentage of wound area healed was reported as a secondary outcome.

Percentage of reduction in the wound area:
In the present study the time required for reduction in percentage of wound area by 25%, 50%, 75% and 100% was taken as the primary outcome. Knighton (11)and Anitua observed that the mean percentage of surface area reduction at end of 8 weeks of study in the PRP group are 93% ± 17% and 72.94% ± 22.25% respectively, whereas for the control group 41%± 39% 21and 48%± 33.56% respectively. In Anil & Sindhuri(10) study, the platelet group showed 46.95% ± 15.16% percentage reduction whereas the control group showed 2.28% ± 2.54% at the end of two weeks. In the present study percentage reduction of area of wounds per week in group A 14.4±4.04% is observed to more than group B 10.19±3.64%.

Time taken for Complete wound Healing:
All patients in both the group achieved complete wound healing. The mean time taken for complete wound healing was 7.92±3.48 weeks (n=26) in the experimental group A and 10.41±2.48 weeks (n=24) in the observation group B. In 13 patients complete wound healing was achieved by skin grafting, of which 6 belonged to group A and to 7 group B.

Hospital stay:
The present study shows that the mean duration of hospital stay in group B (39.7±9.2 days) was comparatively more in group A (26.9±12.9 days). The time taken for each dressing with autologous PRP was 30 to 40 minutes. No adverse effects were observed with platelet dressing.

The PRP derived from the patient contains natural growth factors in a biologically determined ratio. Because of its autologous origin, it poses no risk of immunogenic reactions or human to human transmission of infection like HIV, Hepatitis B, etc. thereby, serving as a potentially safe, useful and cost effective modality of treatment of chronic nonhealing wound. Despite of its shortcomings, this study indicates that the topical application of autologous PRP is more effective than standard therapy in assisting the healing process of chronic wound.

Conclusion and Summary:
The present prospective randomized comparative study has been conducted on 62 patients presenting with chronic non healing ulcer (> 4weeks) between May 2015 to May 2017 to study the effectiveness of autologous platelet rich plasma on chronic nonhealing wound which revealed that:

* The majority of the patients with chronic nonhealing wounds were in the 40-70 years of age group.

* Nonspecific infection is the most common cause of chronic non healing wound.
* Autologous PRP increases the rate of wound healing as compared to conventional dressing. Further it reduces number of dressings, hospital stay and follow up visits as compared to conventional dressings. These in turn affect the cost incurred in the treatment of chronic wounds by the patient as well as on the economy of health care system.
* Since chronic non healing wound has a multifactorial origin, a multi-disciplinary approach with holistic view forms the backbone for the management of chronic non healing wound and does not solely depend on the type of dressing.
* Although a subjective finding, most patients with autologous PRP dressing reported early with greater degree of mobilization and compliance as compared to conventional daily dressing.

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