



COMPARATIVE STUDY BETWEEN CONVENTIONAL AND PLATELET RICH PLASMA TREATMENT FOR NON-HEALING DIABETIC FOOT ULCERS

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Introduction -

Non healing diabetic lower limb ulcers are a major complication in patients with diabetes. About 15% of diabetic patients develop chronic ulcer and about 25% of those undergo amputation.^(1,2) In non healing diabetes ulcers healing process is impaired due to vascular and neurological disorders, also due to deficiency of growth factors.⁽³⁾

The patient's initial management included measures to optimize glycemic status as well as extensive debridement, antibiotic treatment based on wound cultures, use of moist dressings and off loading.

Platelet-rich plasma (PRP) is defined as a portion of the plasma fraction of autologous blood having a platelet concentration above baseline⁽⁴⁾.

PRP serves as a growth factor agonist. It contains a high level of platelets and a full complement of clotting and growth factors.

The growth factors support healing by suppress cytokine release and limit inflammation, attracting un-differentiated cells in the newly formed matrix, improve tissue healing and regeneration, new capillary growth, and accelerate epithelialization in chronic wounds.⁽⁵⁾

The purpose of this study is to evaluate how autologous platelet-rich plasma (PRP) affects wound healing of chronic, non-healing diabetic wounds in lower limb as compared to conventional dressings.

Methods -

Total 58 patients were included in the study with lower limb ulcer more than 4 weeks. Using a pretested and predesigned pro forma the study population was randomized into either study group or control group. Out of 58 patients, 29 took treatment in the form of conventional dressings in form of chlorine water dressing and 29 took treatment with Platelet rich plasma dressing. Photographs of the ulcers before and after the dressings were taken & wound area was recorded by Measuring length x width. Platelet rich plasma dressing was done once weekly. The outcome that is the area of the target ulcer was measured by using a metric tape and formation of granulation tissue over the ulcer bed.

All patients with non healing chronic diabetic lower limb ulcers of ≥ 4weeks duration were included in study. Ulcers of non-diabetic aetiology, gangrene in ulcer or on any part of the foot and Patient with active malignancy or bleeding disorders were excluded from study.

For conventional dressing, the ulcer was cleaned with normal saline and chlorine water soaked gauze piece was kept over the ulcer which was covered with pad and roller bandage. For platelet rich plasma dressing, the ulcer was cleaned with Normal Saline. Platelet rich plasma was prepared from patients blood and applied over the wound area once weekly. At the end of treatment the wounds in both the groups were inspected and the wounds were compared based on the following parameters. They are:

1. To compare the rate of wound healing in either group.
2. To compare the granulation tissue formation in either group.

Preparation of the platelet rich plasma⁽⁶⁾ –

PRP was prepared by collecting 20 cc of fresh blood in sodium citrate containing test tube under proper aseptic precaution. The tubes were rotated in a centrifugation machine at 1500 revolutions per minute for 10 minutes. The first centrifugation is called “soft spin”, which allows blood separation into three layers, namely bottom layer (RBC, 55% of total volume), top most (plasma layer, platelet poor plasma PPP, 40% of total volume) and an intermediate layer (PRP, 5% of total volume) called the “buffy coat”. Buffy coat and PPP was separated in other test tube and underwent a second centrifugation at 2500 revolution per minute for 15 minutes, called “hard spin”. After second centrifugation the platelets (PRP) settled at the bottom of the tube. The upper layer containing PPP was discarded and the lower layer of PRP was loaded in a syringe containing calcium chloride (1 part calcium chloride and 9 parts of PRP) as an activator.⁽⁷⁾ A total volume of 2-3 cc was injected at wound site. Blood samples sent for platelet count before and after centrifugation showed a count of 1.65 lac cells/mm³ before and 7.62 lac cells/mm³ after second centrifugation.(Figure 1)

Data Management and Analysis: - Categorical variables will be presented in number and percentage (%) and continuous variables were presented as mean and standard deviation (SD). Normality of data were tested by Kolmogorov-Smirnov test. If the normality is rejected then non parametric test were used. Quantitative variables were compared using Mann-Whitney Test between two groups and Wilcoxon signed ranks test for paired data. P value of <0.05 will be considered statistically significant.

Results -

The present study was carried out with an aim to compare autologous PRP with conventional treatment methods for chronic non healing diabetic lower limb ulcers.

Mean age in PRP group was 46.13 year (range 17 – 75 year) and in control group was 47.72 year (range 30- 65 years). There were a total of 21 males and 8 females in case group and 23 males and 6 females in control group. There were a total of 25 post cellulitic and 4 post traumatic wounds in case group and 24 post cellulitic and 5 post traumatic wounds in control group.

In this study foot dorsal surface and plantar surface (46.6 %) was the most common site for ulcers followed by leg (34.5%) and ankle (19%). The average wound size in case study was 37.74cm². After 1 week of PRP treatment, ulcer size had reduced to average of 35.81 cm². After 2 weeks of PRP treatment, ulcer size had reduced to average of 27.21 cm². After one week of PRP treatment, ulcer size had reduced by 1 % (range 0% - 75%) & 44.21% (range from 8% – 100%) of their original size after 2 weeks.

Granulation tissue started appearing in the wound at a mean of 1.48 weeks in PRP group as compared to mean of 2.21 weeks in control group. After six weeks of follow up all patients of PRP dressings healed completely in comparison to control group (72.4 %). Complete healing of lower limb wounds after PRP treatment took 4 weeks as compared to 7.03 weeks in control group.

Granulation tissue started appearing in the wound at a mean of 1.48 weeks in PRP group as compared to mean of 2.21 weeks in control group. (Table 1)

Wound size reduced completely in PRP group at 6 week in comparison to mean of 84.48 % reduction in control group at 6 week follow up. (Table 2)

After 6 weeks of follow up all patients of PRP dressings healed completely in comparison to control group (72.4 %). (Table 3)
Complete healing of lower limb wounds after PRP treatment took 4 weeks as compared to 7.03 weeks in control group. (Table 4) (Figure 2,3,4)

Discussion -

Management of chronic non healing ulcers is depends upon multiple factors and is regulated by interactions between a large number of cell types, extracellular matrix proteins and mediators such as cytokines and growth factors. If these interactions are not balanced, it may result in a chronic wound. High bacterial count leads to a prolonged inflammatory response with high levels of cytokines. This leads to increased production of matrix metallo-proteases. Increased matrix metallo-protease activity results in uncontrolled breakdown of extracellular matrix and growth factors.⁽⁸⁾

The average wound size in PRP group was 37.74cm². After 1 week of PRP treatment, ulcer size had reduced by an average of 1%. After 2 weeks, the size decreased to an average of 46 % of their original size. In conventional dressing group initial wound size was 33.96 cm², which became 34.69 cm² after 1 week and 30.99 cm² after 2 week. Thus, after PRP treatment there is improvement in rate of healing in the second week onwards. Similar study was conducted by Gurgun et al. (2007)⁽⁹⁾, who showed comparable results. The average wound size in our study was 37.74cm² (range 1 cm² - 216 cm²) compared to his study in which average wound size of 6.7 cm² (range 0.4 cm² - 22.3 cm²), 13 patients with 14 recalcitrant leg and foot ulcers of various etiologies were included in his study.

After 6 weeks all wounds healed completely in PRP group in which 20 cases healed by PRP dressing and 3 cases required SSG cover while in control group 27.6% patients had residual wounds. Faster healing in the PRP group could be due to the increased growth factors present in the PRP as it showed increased platelet counts.

Blood samples sent for platelet count before and after centrifugation showed a count of 1.65 lac cells/mm³ before and 7.62 lac cells/mm³ after second centrifugation. According to study by Yavuz Kececi et al⁽¹⁰⁾, the mean platelet count in the whole blood was 2.60×10^5 (0.6×10^5)/ μ L and increased in the PRP groups, ranging from 4.98×10^5 (0.3×10^5)/ μ L to 9.76×10^5 (0.3×10^5)/ μ L.

Crovetti et al⁽¹¹⁾, McAleer et al⁽¹²⁾, and Steenvorde et al⁽¹³⁾ reported wound closure rates in recalcitrant ulcers between 37.5% and 66%. Cervelli et al (2009)⁽¹⁴⁾ observed that 16 of 20 chronic lower-extremity ulcers re-epithelialized during an average of 9.7 weeks, with PRP application.

In our study granulation tissue started appearing in the wound at a mean of 1.48 weeks (range 1 week – 2 weeks) in PRP group as compared to mean of 2.21 weeks (range 1 week – 4 weeks) in control group. Thus, PRP promotes faster granulation tissue formation and enhances wound healing. In conventional dressings group granulation started appearing at mean of 2.21 weeks

According to Bo Jørgensen et al⁽¹⁵⁾ application of Vivostat® PRF® is repeated with one week's intervals in conjunction with normal change of dressings. Usually, granulation is initiated within the first two to three applications, but it is recommended to continue treatment for altogether 5-6 weeks.

In our study complete healing of lower limb wounds after PRP treatment took 4 weeks (ranges 2 weeks – 6 weeks) as compared to 7.03 weeks (ranges 4- 20 weeks) in control group.

Driver et al⁽¹⁶⁾ carried out the first prospective, randomized, controlled multicenter trial in the United States with the use of autologous PRP for the treatment of chronic diabetic foot ulcers and included 72 patients. They compared the effectiveness of autologous PRP gel to that of

normal saline for 12 weeks and found that 68.4 percent (13/19) of patients in the PRP group and 42.9 percent (9/21) in the control group had wounds that healed. Wounds in the PRP group healed after a mean of 42.9 days (SD 18.3) vs. 47.4 days (SD 22.0) in the control group.

Conclusions -

Granulation starts appearing in the a mean period of 1.48 weeks in wounds treated with PRP as compared to mean of 2.21 weeks with conventional dressings. Complete healing of lower limb wounds after PRP treatment takes 4 weeks as compared to 7.03 weeks in control group. Study suggests that Platelet rich plasma dressing significantly increases granulation tissue formation and wound healing in chronic non healing diabetic lower limb ulcers.

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