



Effectiveness of Amnion on Chronic Wounds Vs Saline Dressing

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ABSTRACT

Introduction: Chronic wounds have deleterious effects on patient health and quality of life. Human placental membranes consist of amnion and chorion which are conjoined and avascular structures. These membranes have abundant growth factors and cytokines and extracellular matrix which can induce angiogenesis and dermal fibroblast proliferation and recruit mesenchymal stem cells involved in wound repair supplying an ideal environment for wound closure. **Aims & Objectives:** To compare the effectiveness of human amnion vs. normal saline dressing in chronic wounds. **Place and duration of study:** This study was conducted at Shaikh Zayed Medical Complex for a period of 6 months. **Material & Methods:** This randomized control trial was carried out in the Department of Plastic Surgery at Shaikh Zayed Hospital, Lahore from 20-08-2019 to 20-02-2020. 60 patients of both genders, 18 - 65 years of age having ulcers of more than six weeks duration and larger than 10 x 10 cm were inducted using non-probability consecutive sampling technique. They were divided into two groups of 30 patients each. Group A (Amnion) and Group B (Saline Dressing). Patients were assessed pre-treatment, wound dressings applied daily and followed up for 6 weeks for efficacy of dressing based on reduction in wound size and its relation to wound chronicity. Data was entered and analyzed using SPSS version 20. **Results:** There were 27 (90%) male and only 3 (10%) females patients in group A and 21 (70%) male and 9 (30%) females patients in group B. Mean age of the patients was 45.60±12.51 years for group A and 44.20±11.65 years for group B. 27 (90%) patients demonstrated good efficacy of treatment at six weeks in group A as compared to 5 (17%) patients in group B which is statistically significant (P 0.041). Significant healing was noted in amnion treated chronic wounds of up to 21 weeks duration. **Conclusion:** Our randomized control trial shows that amniotic membrane dressing are beneficial in the treatment of chronic wounds as compared to normal saline dressings.

Keywords: Chronic wounds, Amniotic fluid, Saline dressing.

INTRODUCTION

Chronic wounds, which do not show signs of healing within 30 days impose a significant and often underappreciated burden on the individual, the healthcare system and the society as a whole. They are associated with numerous deleterious effects on patient health and quality of patient life.¹ Major factors leading to wound chronicity are ischemia, prolonged or excessive inflammatory phase that degrade and inactivate components of extracellular membrane and growth factors necessary for normal cell functions, reperfusion injury, drug resistant microbial colonization, systemic illnesses, age and repeated physical trauma. Chronic wounds can be broadly classified into vascular ulcers, diabetic ulcers and pressure ulcers. Chronic diabetic foot ulcerations (DFUs) are a devastating complication

of uncontrolled diabetes.² Chronic ulcers are related to uncontrolled diabetes. Diabetic foot ulcers and ulcers due to chronic venous insufficiency (venous leg ulcers [VLUs]).³ can persist for years and tend to recur. Chronic leg wounds resulting from surgical procedures are due to underlying medical conditions like diabetes or peripheral vascular disease that leads to poor wound healing. Co-morbidities that contribute to poor wound healing include hypertension, obesity, diabetes and dyslipidemia.⁴

The non-healing DFUs along with consequences like infection, limb amputation and increased risk of myocardial infarction can increase the risk of mortality.^{5,6} Chronic VLUs are susceptible to microbial infections, which leads to delayed healing and results in complications like cellulitis and sepsis.⁷

The human amniotic membrane grafts were first used as a biomaterial for reconstructive surgery by Davis in 1910. The early use of amniotic membranes focused on treating ulcers and skin burns. At the end of the 1990s, processing and long-term storage like cryopreservation and dehydration of amniotic membrane steered usage in wound care, spinal and reconstructive surgery.⁸ In reconstructive surgery, amniotic membranes have been used for diverse areas such as the cervix, ureter, nerve regeneration, finger tips and deep periodontal defects.^{9,10} Human placental membranes are composed primarily of amnion and chorion, which are two avascular but conjoined layers.¹¹ These membranes are rich in extracellular matrix proteins and growth factors & cytokines which can induce angiogenesis and dermal fibroblast proliferation and recruit mesenchymal stem cells involved in wound repair and regeneration^{12,13}, supplying an ideal environment for epidermis formation and wound closure.

MATERIAL AND METHODS

This randomized control study was carried out in the Department of Plastic Surgery Shaikh Zayed Medical Complex, Lahore from 20.08.2019 to 20.02.2020. Patients of both genders between 18 to 65 years of age having ulcers more than six weeks duration and larger than 10 x 10 cm were included. Patients receiving radiation or chemotherapy, known or suspected malignancy were excluded. Two groups A and B were included in the study by non-probability consecutive sampling technique. The calculated sample size of 60 patients having 30 patients in each group with a 95% confidence level was included. After the approval by Hospital Ethical Committee informed consent was taken from all 60 patients. A detailed history, general examination and investigations were carried out. Laboratory testing and imaging were performed when indicated. Human placenta was used as the amniotic source in the study. Rigorous serological screening for human immunodeficiency virus, Hepatitis B, Hepatitis C, human T-cell lymphotropic virus and tuberculosis was carried out for patients registered for elective Cesarean Section. After consent from mother placenta was taken and thoroughly washed with normal saline then amnion separated from chorion, and was placed in sterilized container having normal saline, glycerin, and Inj. Augmentin and Geneticin. Group A patients with chronic non healing wounds who had undergone wound debridement and dressings previously, had daily amnion applied on their wounds. Patients weekly follow up was done

thereafter and complete immobilization and leg elevation was advised. Group B patients had only Normal saline dressing daily on their wounds and healing was assessed in terms of wound size and time of wound healing. Both groups were assessed after 6 weeks for comparison in wound healing and Efficacy of treatment. Efficacy of treatment was defined as assessment after 6 weeks with more than 80% clinically assessed wound coverage and epithelialization.

Statistical analysis:

Data was entered and analyzed using SPSS version 20. The quantitative data of patients like age, wound size, chronicity of wound and time of wound healing presented as Mean±SD. Frequency and percentage were calculated for gender, age and chronicity of wound. Chi-square test was further applied taking p value ≤0.05 as significant.

RESULTS

Total 60 patients were enrolled. The largest age group comprised of patients in 41-60 years of age. Mean age of the patients was 45.60±12.51 years for group A and 44.20±11.65 years for group B with p value >0.05 (Table-1). In our study there were 27 (90%) male patients and only 3 (10%) were female patients in group A and 21 (70%) male patients and 9 (30%) were female patients in group B (Table-2). Twenty five (83%) patients were having chronic ulcers between 6-14 weeks duration while there were 17 (57%) patients in group B. There were 3 (10%) and 5 (17%) patients in group A and group B respectively with 15-21 weeks duration wounds. Only 2 (7%) patients were present in group A for >21 weeks chronic ulcers while there were 8 (26%) patients in group B. (Table-3).

Frequency of efficacy with healing and wound coverage at 6 weeks was observed in 27 (90%) patients in group A compared to 5 (15%) patients in group B, which is statistically significant (p <0.05) (Table-4).

In relation to chronicity of wounds and efficacy of treatment at 6 weeks, 24 patients (80%) in Group A having chronic ulcers of 6-14 weeks duration and 5 (17%) patients in Group B had healed. Regarding more chronic ulcers having 15-21 weeks duration all 3 (10%) patients in group A responded to amniotic membrane dressing while none responded in group B which is statistically significant (P 0.041). Ulcers having >21 weeks of wound did not heal in both groups (Table-5).

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Age in years	Group A (n=30) Amnion		Group B (n=30) Saline Dressing	
	No.	%	No.	%
20 – 40	10	33.0	12	40.0
41 – 60	17	57.0	16	53.0
>60	3	10.0	2	7.0
Mean±SD	45.60±12.51		44.20±11.65	

Table-1: Age Distribution of Patients in Both Groups (n=60)

Sex	Group A		Group B	
	No.	%	No.	%
Male	27	90.0	21	70.0
Female	3	10.0	9	30.0
M:F ratio	9.0:1		2.33:1	

Table-2: Sex Distribution of Patients in Group A and B (n=60)

Chronicity of wound	Group A		Group B	
	No.	%	No.	%
6 – 14	25	83.0	17	57.0
15 – 21	3	10.0	5	17.0
>21	2	7.0	8	26.0
Mean±SD	11.63±4.35		14.10±5.06	

Table-3: Frequency of Chronicity of Wound in Weeks in Both Groups (n=60)

Efficacy at 6 Weeks	Group A		Group B	
	No.	%	No.	%
Yes	27	90.0	5	17.0
No	3	10.0	25	83.0

Table-4: Efficacy of treatment at 6 Weeks in Group A and B (n=60)

Chronicity of Wound (weeks)	Group A		Group B	
	Yes	No	Yes	No
6 – 14	24(80%)	1(3%)	6 – 14	24(80%)
15 – 21	3(10%)	0	15 – 21	3(10%)
>21	0	2(7%)	>21	0

Table-5: Efficacy of treatment in Group A and B according to Chronicity of Wound at six weeks post-treatment (n=60)



Fig-1: A venous ulcer on left leg after debridement



Fig-2: After 1st amnion placement



Fig-3: After 2nd amnion placement



Fig-4: After six weeks

DISCUSSION

Our study shows that amnion is the best treatment and beneficial for patients of chronic wound healing as mentioned in various studies. The use of placental membranes was first reported in the mid of last century¹⁴. In the beginning, the amnion was mainly used as a treatment for burns, and its use for the treatment of chronic ulcers was limited to few cases.¹⁵ But now, it has been shown that the human amnion and chorionic membrane is helpful for the healing of diabetic foot ulcers (DFUs).¹⁶

About 34% diabetic population may develop lower limb ulcers, and 12% of those will ultimately require lower limb amputation. Conditions associated with diabetes, like neuropathy and poor glycemic controls and peripheral vascular disease, as well as other co-morbidities, also play a role in slowing down the healing process and increase rates of recurrence. An estimated 40% of the diabetic population has a recurrence in 1 year after healing of ulcers. The slow healing process of lower limb ulcers can lead to an increased risk of infection and resultant amputation.¹⁷

In 2011, it was approximately 366 million people, and 7% world population was diabetic out of which 80% of these diabetics were living in developing countries. DFUs are one of the most common and growing complications of diabetes. These are complex chronic wounds having a long-term impact on quality of life, morbidity and mortality.¹⁸ Approximately 25% of diabetic patients develop ulcers of the lower extremity over time.¹⁹ Treatment of these ulcers is challenging as they recur frequently, and the healing process is slow.²⁰

More than 80,000 amputations are reported every year in diabetic patients of United States. 85% of lower limb amputations preceded by ulcers in diabetic patients and it is estimated that 85% of these amputations may be preventable. Sometimes antibiotics may be required along with debridement to control bacterial growth. Antimicrobial for chronic wound include topical antiseptics, topical antibiotics and systemic antibacterial. There is no evidence that one antibiotic or antiseptic is better than other for decreasing time of wound healing. Many latest antimicrobial therapies can be targeted which can be beneficial in wound healing.²¹ The placental membranes as biological dressing for wound has been used since early 1900s. These are rich in collagen matrix containing abundant growth factors, fibroblast, cytokines, viable epithelial cells and mesenchymal stem cells (MSCs).^{21,22}

Another common cause of chronic leg ulcers are venous insufficiency leading to chronic venous

ulcers, most commonly on lower leg. They are difficult to treat and use of amniotic membrane on these wounds shows improved healing.

The mesenchymal stem cells (MSCs) contribute to all phases of the wound healing followed by immunomodulatory capacity, to enter other host cells, and produce multiple growth factors and matrix proteins required for wound healing.²³ Thus, placental allograft with viable cell populations and MSCs gives the opportunity to produce and make proteins available necessary for wound repair.²⁴ placental membranes include regenerative cells and introducing these robust young cells in chronic wound can help in healing in chronic wounds.²⁵

Amniotic membrane has analgesic and bacteriostatic effect, as reported in studies irrespective of preparation method, i.e. fresh, refrigerated or frozen amniotic membrane. Its anti-inflammatory properties are beneficial in chronic wound management, where there is loss of balance between proteases and anti-proteases. In our study mean age of patients was 45.60±12.51 years. 17(57%) patients were between 41-60 years of age. In one of the reported studies by El-Heneidy, the mean age of patients was 32.86±6.94 years which is comparable with our study.²⁷ In another study the mean age was 64 years which is slightly high from our study.²⁸ In a similar study carried out the mean age was 53.5 years in group A and 50.2 years in group B which is comparable with our study.²⁹

In our study 27 (90%) patients were male in group A and 21 (70%) in group B. In comparison, 3 (10%) were females in group A and 9 (30%) were females in group B, with male to female ratio 9.0:1, 2.22:1 respectively which is comparable to one of the studies carried out by Tettelbach 73% of the study population consists of male which were more than females.³⁰ In our study the mean wound size was 13.77±3.32 in group A, and 15.03±4.13cm in group B. In a similar study reported by Stephen the mean wound size was 7.6cm which is comparable with our study.³¹ Our study also found amnion to be more efficacious at treating chronic wounds of 6-21 weeks duration vs saline healing which concurs with recent literature,²⁶ Our study shows that the growth factors present in amniotic membrane speed up the healing process in chronic wounds.

CONCLUSION

This study shows that the human amniotic membrane is beneficial in treating chronic wounds more than normal saline dressing.

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