

Bacterial Colonization of Leg Ulcers and Its Effect on Success Rate of Skin Grafting

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Conflict of interest: Nil

Abstract:

Background and Aim: A number of studies have emphasized the relevance of qualitative rather than quantitative bacteriology, whereas others have done the opposite. However, the impact of bacteria in a wound is determined by three primary factors: bacterial load, pathogenicity of the bacteria, and host immune response. As a result, in this investigation, a quantitative bacteriological profile of granulating wound beds that were prepared for grafting was performed in order to determine the effect of bacterial bioburden on the success rate of Split Thickness Skin Grafting.

Material and Methods: The current investigation was organized and carried out in the department of general surgery, in collaboration with the department of microbiology at the medical college and affiliated hospital. Before collecting the samples using standard proforma, a comprehensive history was documented. All patients were thoroughly evaluated, and their haemoglobin and random blood sugar levels were measured. The skin graft take up evaluation will be performed on the eighth post-grafting day to determine if the graft has taken up. On the tenth post-skin grafting day, the donor site dressing was removed and evaluated for symptoms of infection as well as whether or not the donor site was healing.

Results: The study included 200 patients with chronic non-healing ulcers who met the inclusion criteria and were prepared for split thickness skin graft surgery. Of the 200 patients, 154 were male and 46 were female. Sixty-four patients out of 200 had pre-operative quantitative culture results showing bacterial counts $> 10^5$ CFU/g of tissue, indicating considerable bacterial development. Four patients out of the 64 had isolated mixed flora. Bacterial counts $< 10^4$ CFU/g of tissue were seen in fourteen cases.

Conclusion: The outcomes of skin grafting in individuals with persistent lower limb ulcerations may be further improved by excluding these two categories, treating underlying venous and arterial issues, treating according to the swab findings, and closely monitoring these patients. These strategies merit more research. The bacterial bio-burden and pathogenicity in the wound determine the impact of bacteria on wound healing.

Keywords: Bacterial Colonization, Lower Limb, Skin Grafting, Ulcer.

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Introduction

Chronic non-healing wounds impose a considerable burden on healthcare systems while also causing morbidity and death among patients. Non-healing ulcers lower people's morale and cause them to retreat from society and employment. It impacts patients' quality of life as well as their productivity at work.

In India, an estimated 2.8 million people suffer from chronic wounds each year, with treatment costs in the billions of rupees. A community-based epidemiological study of wounds in India found the frequency of chronic wounds at 4.5 per 1000 people. [1,2]

Split skin transplant is made up of epidermis and varying amounts of dermis. It is simple and rapid to

cover a vast surface area of wounds, with the added benefit of a shorter healing period and hospital stay, as well as less donor site morbidity. It is an efficient approach for treating big ulcers of various causes.

The majority of the necessary conditions for effective skin transplantation are well understood. They include a sufficiently vascularized recipient bed, a good graft, precise approximation, and graft immobilisation in regard to the ulcer. Even if the essential conditions are satisfied, the transplant may fail due to bacterial infection. [3,4]

Different organisms cause grafting to fail by producing proteolytic enzymes that breakdown the essential fibrin scaffold, resulting in the failure of

the applied skin grafts. Beta haemolytic streptococci and *Pseudomonas* are known to impair the success rate of skin grafts. Factors such as ageing and diabetes are known to impact wound healing and are being studied. [5,6]

A number of studies have emphasized the relevance of qualitative rather than quantitative bacteriology, whereas others have done the opposite. However, the impact of bacteria in a wound is determined by three primary factors: bacterial load, pathogenicity of the bacteria, and host immune response. As a result, bacteriological culture of chronic non-healing wounds in preparation for skin grafting should be conducted using a tissue biopsy sample or a deep wound swab. [7,8] As a result, in this investigation, a quantitative bacteriological profile of granulating wound beds that were prepared for grafting was performed in order to determine the effect of bacterial bioburden on the success rate of Split Thickness Skin Grafting.

Material and Methods

The current investigation was organized and carried out in the department of general surgery, in collaboration with the department of microbiology at the medical college and affiliated hospital. The trial was scheduled to last one year. The research comprised patients who were admitted to the general surgery department after being scheduled for a split thickness skin transplant. The ethics committee was notified about the study, and an ethical clearance certificate was acquired prior to its initiation.

The exclusion and inclusion criteria are as follow:

1. Patients with chronic, non-healing ulcer wounds, such as pressure ulcers, neuropathic ulcers, burns ulcers, traumatic ulcers, and diabetic ulcers, are prepared for split thickness skin grafting.
2. Patients who had ulcers at numerous places and were scheduled for split thickness skin grafting were included individually.

Exclusion conditions

1. Patients under 18 years old.
2. Patients with chronic non-healing ulcer wounds are ready for full thickness skin grafts or flaps.

Relevant clinical data were acquired from participants who met the inclusion criteria. Before collecting the samples using standard proforma, a comprehensive history was documented, including name, age, gender, current symptoms, prior history of any co-morbid illness, recent antibiotic therapy, and surgical information. The patients' vital signs were recorded. In the majority of instances, the ulcer size exceeded 5cm².

All patients were thoroughly evaluated, and their haemoglobin and random blood sugar levels were measured. Anaemic patients received blood transfusions as needed, and skin grafting was performed only once haemoglobin levels had been restored to normal. Diabetics received anti-diabetic therapy, and skin grafting was performed only once blood glucose levels were controlled. All patients with low blood protein levels were given a high protein diet and any required albumin infusions; skin grafting was performed only after hypoproteinemia had been corrected.

Infectious situations necessitated careful wound debridement and the administration of appropriate antibiotics; skin grafting was performed only when the ulcer established good healthy red granulation tissue and minimum exudates. Skin grafting was performed on all patients once the lesion had grown healthy red granulation tissue with no pus discharge or slough. To ensure maximum coverage, a graft from the thigh was fenestrated and put on an ulcer with healthy red granulation tissue in one or more pieces. Swabs were obtained 48 hours before to split thickness skin grafting, as well as on the fifth post-operative day. Swabs were grown, and bacterial isolates were identified using established methods.

A skin grafted location is considered healed if the graft take-up is greater than 95%. The skin graft take up evaluation will be performed on the eighth post-grafting day to determine if the graft has taken up. On the tenth post-skin grafting day, the donor site dressing was removed and evaluated for symptoms of infection as well as whether or not the donor site was healing. All patients were released from the hospital only when the skin graft seemed stable and full movement had been restored. Follow up All patients were followed for up to 12 weeks.

The essential data was obtained, and the statistical significance was discovered using the analysis SPSS version 17.0.

Results

The study included 200 patients with chronic non-healing ulcers who met the inclusion criteria and were prepared for split thickness skin graft surgery. The patients' ages ranged from 20 to 70 years, with a mean age of 40 years for those who were ready for the procedure.

The majority of the patients belonged to the age group of 31 to 60 years. Of the 200 patients, 154 were male and 46 were female. The majority of patients who were ready for split thickness skin grafting had traumatic ulcers (116 patients), followed by diabetic ulcers (44 patients).]

Sixty-four patients out of 200 had pre-operative quantitative culture results showing bacterial counts > 10⁵CFU/g of tissue, indicating

considerable bacterial development. Four patients out of the 64 had isolated mixed flora. Bacterial counts $< 10^4$ CFU/g of tissue were seen in fourteen cases. As a result, in pre-operative quantitative culture, 82 bacterial isolates were collected. *Pseudomonas aeruginosa* and *Staphylococcus aureus* were the most frequently isolated pathogens

in pre-operative quantitative culture. When pre-op quantitative bacterial count was less than 10, there was less graft absorption. The link between pre-operative bacterial count and graft absorption rate was statistically significant, as evaluated by "Independent sample T test". p-value is less than 0.05.

Table 1: Distribution of type of ulcers among study population n= 200

Type of the ulcers	No. of patients
Diabetic ulcer	112
Post cellulitis	22
Traumatic ulcer	14
Burns ulcer	26
Pressure sore	4
Total	200

Table 2: Bacteriological Profile of pre-operative quantitative bacterial culture

Type of isolate	Organism	Number of organisms
Gram positive	<i>Staphylococcus aureus</i>	18
Gram negative	<i>Pseudomonas aeruginosa</i>	18
	<i>Pseudomonas species</i>	7
	<i>Klebsiella pneumonia</i>	10
	<i>Escherichia coli</i>	6
	<i>Proteus mirabilis</i>	9
Total		68

Discussion

The gold standard for treating skin abnormalities brought on by persistent, non-healing wounds is split thickness skin grafting. A well-vascularized, non-infected wound bed is essential before grafting, despite the fact that the outcome of skin transplants is dependent on several parameters. In order to assess the impact of preoperative bacterial bioburden on the influence of graft absorption rate, preoperative bacterial culture of the pre-grafted wound bed and semi-quantitative bacterial culture of the post-operative wounds were obtained for this study.[9,10]

According to research by Kontiainen et al. [11], 60% of the pathogens identified from lesions in skin and soft tissue were streptococcus and *S. aureus*, while 10% of lesions were caused by *pseudomonas* and other bacteria. According to Halbert et al. [12], the most frequently isolated organisms were *S. aureus* (48%), mixed coliforms (28%), *Pseudomonas aeruginosa* (21%), and anaerobes (17%).

The impact of the pre-operative bacterial count on the graft uptake result was examined in this study. The mean graft uptake percentage in the current research was 74.09% for patients with CFU/g of tissue $> 10^5$, and 95% for patients with CFU/g of tissue $< 10^4$ in pre-operative quantitative culture. Thus, it was shown that graft absorption was statistically significantly lower when preoperative bacterial culture was $> 10^5$ CFU/g of tissue. P value less than 0.05.

The mean graft absorption percentage in this study, when pre-grafted wound was infected with *Pseudomonas aeruginosa* was 61.65% and for *Staphylococcus aureus* was 69.42%. Similar results were seen in a research by Gilliland et al., wherein wound beds infected with *Staphylococcus aureus* had a mean graft absorption rate of just 75%. [7,13,14]

In a post-operative semi-quantitative culture of 156 patients, 124 individuals showed bacterial growth that was considered substantial, as indicated by bacterial counts of at least 3+. One patient had growth with a 2+ bacterial count. The most typically isolated pathogens in post-operative semi-quantitative culture were *Staphylococcus aureus* and *Pseudomonas aeruginosa*.

Out of the 200 participants in the trial, 50 individuals had diabetes. The patient's pre-operative diabetes state and graft absorption failure had a statistically significant connection (p value < 0.05). Compared to patients without diabetes, those with diabetes had a mean graft uptake rate of 79.54% as opposed to 89.55%. This is comparable to a research by Bosman et al. where diabetes was found to be a significant factor negatively affecting the absorption of the transplant. Patients with diabetes have a 4.06 times higher risk of post-operative complications following STSG than patients without diabetes, according to a review paper by Ryan J. Donegan et al.

Prior to surgery, the ulcers in the current research had been present for an average of 3.5 months

(range: 1 month to 6 months). The proportion of graft uptake and ulcer length was negatively correlated. The proportion of graft uptake decreased as ulcer length increased.

Conclusion

The outcomes of skin grafting in individuals with persistent lower limb ulcerations may be further improved by excluding these two categories, treating underlying venous and arterial issues, treating according to the swab findings, and closely monitoring these patients.

These strategies merit more research.

The bacterial bio-burden and pathogenicity in the wound determine the impact of bacteria on wound healing.

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