

## **Efficacy and Safety Of 0.25% Timolol Gel in Enhancing Full Thickness Skin Grafts Healing and Cosmetic Outcomes: A Randomized, Controlled Trial**

### **Background and Significance**

The role of topical beta-blockers in promoting wound healing is currently emerging in the international literature<sup>1-3</sup>.  $\beta_2$ -Adrenergic receptors (B2AR) are the only subtype of beta-adrenoceptors expressed on skin<sup>4-6</sup>. They can be found in secretory coil of apocrine glands, keratinocytes, fibroblasts and melanocytes. The distribution of these receptors provides insight on dermatological disorders that may be affected by  $\beta$ -blockers. Keratinocyte migration occurs by the facilitation of chemotaxis, the polarization of cells, and activation of extracellular signal-related kinases essential in the signaling of promigratory pathways. The B2AR activation inhibits keratinocyte migration by activating the serine/threonine phosphatase 2A, which downregulates phosphorylation of extracellular signal-related kinases necessary for migration. Therefore, B2AR antagonists prevent the phosphorylation of phosphatase 2A and have the downstream effect of extracellular signal-related kinase promotion, inducing a promigratory pathway in keratinocytes<sup>4-6</sup>. Keratinocyte migration also occurs by galvanotaxis, a phenomenon in which cells migrate in response to electric stimuli. Keratinocytes can be stimulated to migrate with the formation of electrical poles and the application of electrical fields. The B2AR antagonists improve the ability of keratinocytes to respond to such migratory cues, whereas the B2AR agonists decrease keratinocytes' ability to respond, further implicating the use of topical timolol for recalcitrant wounds<sup>4-6</sup>. Angiogenesis and dermal fibroblast proliferation are also regulated by B2ARs. The B2AR antagonists have been found to promote angiogenesis in chick chorioallantoic membrane assays and *in vivo* murine wound models. Dermal fibroblast migration is also increased (by 27%) when exposed to B2AR antagonists, and epidermal differentiation is improved with B2AR antagonists and  $\beta_1$ - and  $\beta_2$ -receptor antagonists<sup>5,6</sup>.

Full-thickness skin grafts (FTSG) are one of the most commonly performed procedures in dermatologic, plastic and burn surgery. Various experimental approaches to optimize the healing of FTSG receiving sites have been described<sup>7-10</sup>; however, no clearly superior and easily applicable method has gained wide acceptance in daily practice.

As indicated by preliminary evidence in other wound healing endeavors<sup>1-6</sup>, 0.25% timolol gel may represent a commercially available, safe and simple, painless and relatively inexpensive treatment for improving healing of FTSG receiving site, as well as for improving cosmetic long term outcomes.

## **Specific Aims**

To assess the efficacy and safety of topically applied 0.25% timolol gel in promoting wound healing in full-thickness skin graft (FTSG) receiving sites, sized  $\leq 6$ cm and located anywhere on the body except the lower limbs, versus standard of care (SOC) by:

1. Evaluating healing in response to treatment with 0.25% topical timolol gel versus SOC in terms of wound surface area at the receiving site of a FTSG at 7 days.
2. Evaluating healing in response to treatment versus SOC in terms of rate of successful (completely healed) graft using a blinded physician assessment score at the receiving site of a FTSG at 7 days.
3. Evaluating patient discomfort during the healing process by means of a patient pain VAS
4. Determining the side effects associated with 0.25% timolol gel versus SOC

## **Subject Selection**

Patients will be recruited at the Mohs and Dermatologic Surgery Center. Any patients who are potentially eligible to participate in the study will be initially approached by either a treating physician (other than the PI) or a nurse or a resident/fellow to discuss potential participation. All patients evaluated at the Mohs and Dermatologic Surgery Center who need to undergo a surgical intervention including a reconstructive phase with a FTSG will be eligible for participation in this study. The patients will be encouraged to make further inquiries about the study if they are interested. Written informed consent will be obtained prior to initiating the treatment.

Patients will be excluded for the following reasons:

- 1) Age less than 18 years of age
- 2) Pregnant women (a pregnancy urine test will be performed to woman able to become pregnant at our clinic before enrollment)
- 3) (Use of systemic drugs that can impede wound healing, such retinoids or immune-suppressive drugs)
- 4) Severe coagulation disorders
- 5) Severe, uncontrolled systemic comorbidities, such as diabetes, arthritis, etc.
- 6) Hypersensitivity to 0.25% timolol gel
- 7) Not willing to provide written informed consent

Patients will be included if they meet the following criteria:

- 1) Age  $\geq 18$  years of age
- 2) Undergoing a procedure which results in the need of a FTSG
- 3) Willing to provide written informed consent

## **Subject Enrollment**

The principal investigator or a co-investigator will inform patients about the study during their initial consultation at the Mohs and Dermatologic Surgery Center, Dana-Farber/Brigham and Women's Cancer Center at Faulkner Hospital. The patients will be encouraged to make further inquiries about the study if they are interested. Informed consent will be obtained prior to initiating the treatment. If patients are interested in taking part in the study, the investigator will meet with them in private to review the consent form and address any study-related questions.

## **Study Procedures**

The study protocol will begin the day of surgery. Eligible patients will be assigned by computer-based randomization to case (0.25% timolol gel) or control (SOC) group and treated as follows.

### Receiving site of FTSG/case group:

- 1) During surgery: application of 0.25% timolol gel (2 drops per cm<sup>2</sup>) on wound bed before FTSG is placed
- 2) During surgery: application of 0.25% timolol gel (2 drops per cm<sup>2</sup>) over FTSG after inseting of the graft
- 3) After bolster removal (7 days): daily cleansing and daily Vaseline application for 4 weeks as per SOC

### Receiving site of FTSG/control group:

- 1) FTSG surgery as per SOC
- 2) After bolster removal (7 days): daily cleansing and daily Vaseline application for 4 weeks

Subjects will be followed up at 7 days post-surgery.

Standardized pictures will be taken. Wound surface area assessed by histogram planimetry<sup>11</sup>; rate of successful graft, as well as wound infection rate and reoperation rate will be assessed by a blinded physician. Subject "pain perception VAS" will be recorded 7 days after the surgery. Subject and physician will record any side effects occurring during the study period. Patients will be asked about their overall satisfaction with the wound healing process, the worst pain that they can recall and when was it after the surgery, and any other comments or concerns, including any side-effects or difficulties during the study period that they may want to share with us.

## **Biostatistical Analysis**

Following sample size calculation, using the two samples with percentage values analysis, the study will enroll 58 subjects, randomized into the two study groups (29 subjects for each group) to detect a 35% difference in terms of complete FTSG take (based on the FTSG score scale) with a 90% power ( $\alpha=0.05$ ). If 20% of subjects agree to participate, enrollment will be complete within approximately 15 months.

### **Risks and Discomforts**

The risks and discomforts of the topical application of 0.25% timolol gel include erythema, itching, irritant or allergic contact dermatitis in case of subject sensitization. Other discomforts and risks, including pain and infection, are implied in the surgical procedure patients have undergone (in both groups).

Patients assigned to the Case Group might incur in an increased risk of wound infection because they will not apply Vaseline, which might better protect the wound against infection and/or other contaminants, while maintaining moisture.

Risks to confidentiality will be minimized by limiting access to patient identifiers to study staff only. Data analysis will be conducted on de-identified data.

### **Potential Benefits**

The expected benefits of topical 0.25% timolol gel dressings in FTSG when compared to SOC include: quicker healing, improved rate of successful grafting, better long term scarring and thus cosmetic outcome, improved patient satisfaction, decreased patient pain during the post-surgical healing phase, and decreased number of post-surgical complications and reoperation rate.

### **Monitoring and Quality Assurance**

The study coordinator and the principal investigator will review the study data on an ongoing basis to ensure data quality and compliance with IRB approved protocol. Any deviations from the IRB approved protocol will be reported to the Partners IRB Office in a timely manner. Data will be reviewed regularly by the principal investigator and study coordinator to identify and resolve discrepancies. Subject identifiers will only be accessible to study staff and all data will be de-identified before review and analysis. Any deviations from the IRB approved protocol will be reported to the Partners IRB Office in a timely manner.

Risks to confidentiality will be minimized using coded data and controlling access to the study database. Only study staff will have access to subject identifying information.

Unanticipated problems involving risks to subjects or others will be submitted through Insight within 5 working days/7 calendar days of the date the investigator first becomes aware of the problem. For adverse events (AEs), any unanticipated untoward or unfavorable medical occurrence, including abnormal sign, symptom, or disease, that indicates that the research places subjects at increased risk of physical or psychological harm than previously known/recognized will be submitted through Insight/eIRB. For non-AEs, unanticipated incidents, experiences, information, outcomes, or other problems that indicate that the research places subjects at an increased risk of physical, psychological, economic, legal, or social harm than was previously known or recognized will also be submitted through Insight.

## References

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