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Use of Topical Timolol Maleate as Re-Epithelialization Agent for Treatment of Recalcitrant Wounds of Varying Etiologies

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INTRODUCTION

Chronic wounds are a source of severe morbidity to patients.[1] Wounds are also a notable burden on the healthcare system, with reported prevalence of 4.64% in the U.S. and an annual cost of over 25 billion dollars.[2,3] Moreover, wounds are associated with >15% of all skin disease-related deaths[4] and the 5 year mortality of some chronic wounds is greater than that of many cancers.[5]

Re-epithelialization and dermal proliferation are defining features of wound healing and must occur in order to restore the barrier function of the skin. However, many wounds fail to progress through these phases.[6] Recent in-vitro and in-vivo studies[7-11] as well as case reports,[12-15] have demonstrated that beta-adrenergic receptor blockade with the β1/ β2 antagonist, timolol, either systemically administered or topically applied, promotes re-epithelialization and overall healing in chronic wounds. Timolol is a well-known, readily available and generic drug that has a cost ranging from 3 to 21 dollars.[16]

Herein, we report a retrospective analysis of the effectiveness of topically applied timolol on 55 chronic wounds of varying etiologies in 39 patients.

METHODS

This multi-center, retrospective case-series study was conducted at the Wound Centers at the University of Miami Health System in Miami, Florida and Wound Clinics at the Veterans Affairs Health Care System of Northern California in Mather, California. We identified all wound patients from 2016 to 2018 who received treatment with topical timolol maleate 0.5% (timolol) for at least 4 weeks. Timolol drops at a dose of 1 drop per cm^2 of wound area were instilled with dressing changes twice a day, once a day or every other day. Some patients received continuous application of timolol via a delivery system (Acton™ Topical Deliver System, Aplion Wound Care Technology®, Murfreesboro, TN). Timolol therapy was combined with standard of care. From the patient's medical records, we collected clinical data about wound etiology, duration, location, size and treatments as well as all other relevant history. Healing outcomes were classified into three categories: healed, defined as complete re-epithelialization of the wound and closure, improved, defined as decreasing wound size area, and worsening, defined as increasing wound size area. This study was deemed exempt from the institutional review board at the University of Miami.

RESULTS

There was a total of 39 patients (32 males and 7 females) and 55 chronic wounds. Of the 55 wounds, the majority were venous leg ulcers (VLUs) (n=30) followed by traumatic wounds (n=8) as well as other wound etiologies (Table 1). The median duration of the wound before treatment was 118 days. Following treatment with topical timolol for different durations (Table 2), 34 wounds had healed, and 15 wounds decreased in wound area (Table 1). Two of the VLUs had increasing wound area.

In the healed group, the median treatment duration was 89.5 days, and the median healing rate was -0.25 cm^2/week (Table 2). The majority of the wounds had timolol applied daily (Figure 1). Out of the 34 healed wounds, 3 wounds occurred at locations other than the leg (1 diabetic foot ulcer (DFU), 1 abdominal post-surgical wound). Etiologies in this subgroup included mostly VLUs and traumatic wounds (Table 1).

In the group with decreasing wound area, 8 were still receiving treatment at the time of analysis. The median treatment duration was 112 days, median healing rate was -0.24 cm^2/week and the median percent decrease in wound size was 62% (Table 2). While the majority of wounds occurred on the leg (n=11), 4 occurred in other locations such as the scalp, abdomen and foot. The majority of the wounds in this subgroup were VLUs (Table 1).

In the group that showed no change in wound area, the median treatment duration was 76 days (Table 2). Etiologies are noted in table 1. Of note, there was difficulty evaluating the efficacy of the treatment in the patient with compulsive skin picking due to continued self-excoriations.

In the group with increasing wound area, the median treatment duration was 157 days, the median rate of wound area increase was 0.17 cm^2/week, and the median percent increase was 193% (Table 2). The etiologies in this subgroup were all VLUs.

Venous Leg Ulcer

Since the majority of the wounds were VLUs, we conducted a sub-analysis of this cohort. The median healing rate amongst the VLUs was -0.19 cm^2/week and the median percent reduction was 100% (Figure 2).

In the healed VLU group, the median treatment duration was 101 days and the median healing rate was -0.19 cm^2 per week (Table 2).

In the decreasing wound area group, the median treatment duration was 192 days. median healing rate was -0.24 cm^2 per week and the median percent reduction was 62% (Table 2).

None of the VLU wounds went unchanged after treatment. However, the 2 wounds that worsened were VLUs.

In both the healed and decreasing wound area groups, the healing rate was fastest amongst the wounds that received continuous timolol application or every day application, followed by every other day application and twice a day application (Table 3). Additionally, the highest percent reduction in wound size was in continuous application followed by every day application.

TABLE 1.

Wound Diagnoses and Counts

Diagnosis	Patients	Wounds	Healed Wounds	Improved Wounds	Worsening Wounds	Unchanged Wounds
VLU	15	30	22	6	2	0
Traumatic	8	8	5	2	0	1
Pyoderma gangrenosum (PG)	3	3	1	2	0	0
Diabetic foot ulcer	2	2	1	1	0	0
Malignancy (non-HIV KS)	1	1	0	1	0	0
Radiation dermatitis	2	2	0	1	0	1
Post-surgical	2	2	1	1	0	0
Post-surgical + radiation	1	1	0	1	0	0
Graft versus host disease	1	1	1	0	0	0
Neuropathic trauma	1	1	1	0	0	0
Pressure	1	1	1	0	0	0
Vasculitis	1*	1	1	0	0	0
Bullous pemphigoid	1	1	0	0	0	1
Compulsive skin picking	1	1	0	0	0	1
Total	**39**	**55**	**34**	**15**	**2**	**4**

Numbers of patients, wounds and outcomes after treatment with topical timolol maleate 0.5% (KS – Kaposi sarcoma * - patient also had separate PG counted above)

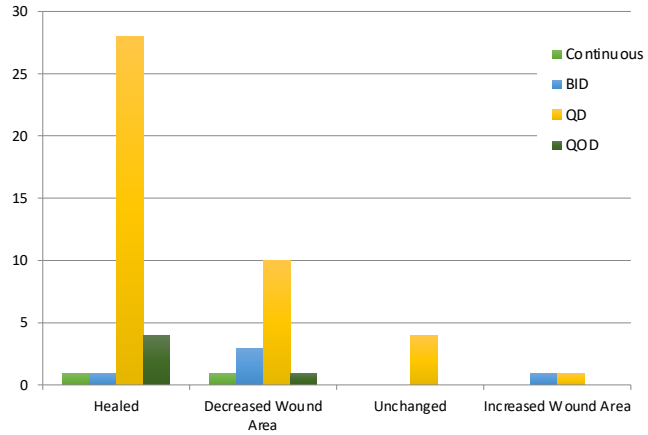
TABLE 2.

Results By Healing Group

Outcome	Median Treatment Duration (days) (avg, std, min, max)	Median Healing Rate (cm2/week) (avg, std, min, max)	Median Change in Wound Size (%) (avg, std, min, max)
Healed	89.5 (109, 78, 14, 336)	-0.25 (-0.66, 1.3, -0.03, -7.23)	-100
Decreasing wound area	112 (168, 188, 21, 628)	-0.24 (-0.72, 1, -0.002, -3.4)	-62 (-55, 29, -2.5, -94)
Unchanged wound area	76 (85, 32, 68, 131)	0	0
Increasing wound area	157 (157, 34, 133, 181)	0.18 (0.18, 0.19, 0.05, 0.31)	193 (193, 61, 150, 236)
Total VLU	124 (145, 112, 28, 538)	-0.19 (-0.4, 0.5, .31, -2.2)	-100 (-73, 75, 236, -100)
VLU healed	101 (124, 84, 28, 336)	-0.19 (-0.42, 0.84, -0.04, -2.2)	-100
VLU decreasing wound area	192 (217, 187, 42, 538)	-0.24 (-0.52, 0.56, -0.05, -1.3)	-61.7 (-63.2, 20.6, -33.8, -93.5)

Results of the median treatment duration, healing rate and change in percentage wound size per healing group. The average (avg), standard deviation (std), minimum (min) and maximum (max) are also reported.

FIGURE 1. Frequency of treatment with timolol in each outcome group.



Frequency of topical timolol application broken down by healed, decreasing wound area, unchanged wound area and increasing wound area. BID = twice a day QD = once a day QOD= every other day

TABLE 3.

Dose Response in the VLU Group			
	Average healing rate (cm2/week)	Wound area percent reduction	Average days treated with timolol
Continuous	-1.21*	93.5*	56*
BID	-0.15	79	172
QD	-0.49	95	154
QAD	-0.18	67	46

The average healing rate, wound area percent reduction and average days treated with topical timolol by groups treated with topical timolol either continuously, twice a day (BID), once a day (QD) and every other day (QAD).
*only one patient

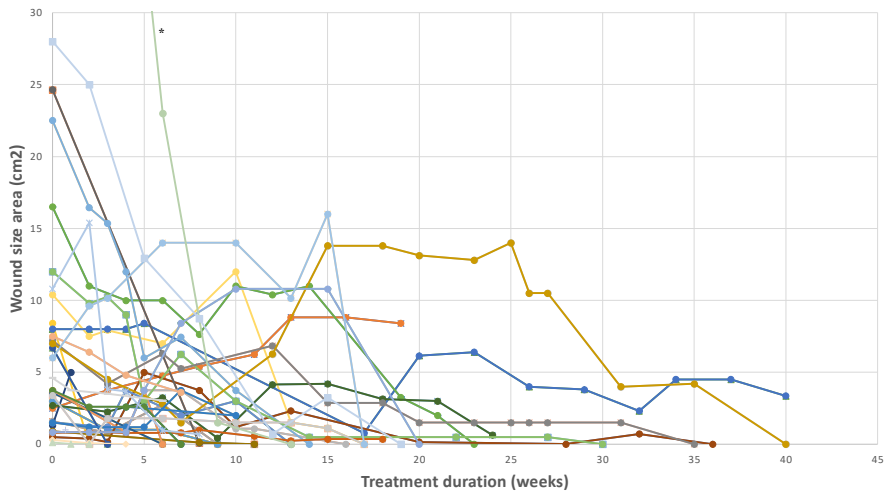
DISCUSSION

Chronic non-healing wounds represent a significant burden on the healthcare system with over 25 billion dollars spent per year in the treatment of these recalcitrant wounds.[1] It is estimated that this number will increase significantly due to rising levels of obesity, diabetes and an aging population.[2,3] Importantly, it is a source of morbidity and mortality for those suffering from non-healing wounds.[1] At the same time these wounds are often unresponsive to standard therapy and therefore building a body of evidence for adjunct therapies is crucial to augment this gap in practice. The overall effect of timolol to improve healing in different wound types suggests a direct effect on stalled migration of keratinocytes for wound re-epithelialization, and supports the use of timolol as an adjunct for wound healing. Indeed, in our centers timolol is used often for well-granulating wounds that appear stalled in the epithelialization stage as well as other wounds and herein we report our continued success.

An essential step in the wound healing process is re-epithelialization, which occurs when keratinocytes migrate, proliferate and differentiate at the wound surface. Recalcitrant wounds that fail to epithelialize are most often due to delays in migration. In order for keratinocyte migration to take place, they must break cell-cell contacts, polarize, and initiate migration by reorganizing their cytoskeletal structure.[6] Without this process a wound will remain unhealed. As a result, much effort has been made to elucidate pathways and agents that favor re-epithelialization as an important adjunctive treatment in promoting wound healing.[17]

One such treatment that has been shown to be effective has been β-adrenergic receptor blockade using timolol. β2-adrenergic receptors (β2-AR) are found on keratinocytes,[18,19] dermal

FIGURE 2. Rate of healing in the VLU group.



Rate of healing of each venous leg ulcer (VLU) after the initiation of treatment with topical timolol. *Wound size area for this wound begins at 94.1 cm² at initiation of topical timolol

fibroblasts[20] and melanocytes.[21] Specifically, keratinocytes were discovered to have β2-ARs as the major class of expressed adrenergic receptors.[22]

Initial in-vitro and in-vivo studies have demonstrated that the activation of β2-AR prevents migration of keratinocytes through various signaling mechanisms.[23] Additionally, the activation of β2-AR prevents the polarization of the keratinocyte and cytoskeleton organization that are key steps to initiate migration.[24] Interestingly, keratinocytes were also shown to express enzymes required for catecholamine synthesis, suggesting an autocrine signaling mechanism that activates the β2-AR in the event of wounding.[9]This is in addition to circulating catecholamines that are present under organismal stress conditions, such as wounding.[24]

Logically, subsequent studies examined the converse paradigm, and tested whether blockade of the receptor using β2-AR antagonists could act as a pro-motogenic agent to bring about wound healing. These reports demonstrated that β2-AR antagonists, such as timolol, enhanced wound healing by increasing the rate of keratinocyte migration.[8] Furthermore, β2-AR antagonists accelerated skin re-epithelialization in a human skin model of a chronic wound.[9]

Timolol, a non-specific β blocker, has been anecdotally reported to safely promote re-epithelialization in chronic wounds.[12-14] In a recent retrospective case-controlled study timolol improved the healing of chronic leg ulcers.[25] However, many of these reports have focused on the use of timolol as a re-epithelialization agent solely for the treatment of VLUs.

In our analysis, we found topically applied timolol to be effective in healing recalcitrant wounds of varying etiologies. Specifically, timolol was effective in healing challenging wounds such as radiation dermatitis, pyoderma gangrenosum and malignancy related wounds amongst others.

Notably, the chronicity of many of these wounds was profound with a median duration of 118 days before treatment with timolol versus 89.5 days with timolol. Moreover, many of these wounds had demonstrated recalcitrance even when standard care and other advanced therapies such as skin substitutes were used by expert clinicians in interdisciplinary wound centers. Importantly, there were no reports of adverse reactions to timolol's use.

Within the VLU subgroup we found that 28 of the 30 venous leg ulcers responded to treatment with timolol, as has the previous observational study.[12] However, the novel findings here are the dose dependency of the response, whereby the highest rate of healing was in the patient that had continuous application of timolol. Yet, we found the slowest healing rate to be amongst those that applied timolol twice a day. These findings are likely related to the effectiveness of compression in the patient that received continuous timolol. By having timolol continuously applied to the wound, it afforded the patient the ability to remain in compression and not have to change the dressing as often. However, those that applied timolol twice a day probably did not achieve optimal compression.

Interestingly, and consistent with our previous experience,[12] time to response with timolol is about 3 months. This highlights the need to recognize that timolol is an adjunct to therapy at the current paradigm of use. Future research should focus on testing different dosing regimens and timolol concentrations. Regardless, this information is very useful for the design of future studies of timolol treatment as current healing outcomes in clinical trials of wounds end at 12 weeks. Alternatively, this may suggest that timolol should be introduced earlier in the treatment plan, perhaps before the pathologically hyperproliferative wound edge is established. Future clinical investigations into the use of timolol for chronic wounds should allow sufficient follow up time to capture the full healing effect and include early intervention arm. This need for randomized controlled trials with excellent confounder exclusion criteria can be highlighted by our results whereby, the overwhelming majority of the patients saw improvement or healing of their recalcitrant wounds, while the two patients who demonstrated worsening of their wounds had additional complex comorbidities and medical therapy that may have complicated, delayed or prevented wound healing.

Some limitations to this analysis are the lack of gender diversity, due to the majority of the patients being treated at the Veterans Association Health Care System where most patients are male. However, VLUs in men are more difficult to heal[26] and therefore our success is encouraging as a treatment option for this difficult population. Additionally, the uncontrolled nature of this study limited our ability to comment on causality.

Even with these limitations, our data suggests that topical timolol may be an effective and safe treatment for chronic wounds, especially those that seem to be stalled in the re-epithelialization stage. Often, chronic wounds need multiple modalities in addition to standard of care to bring about healing. Timolol is an inexpensive and effective treatment in the armamentarium of the wound healing clinician.

DISCLOSURES

The authors of this case series have no conflict of interest to declare.

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