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ORIGINAL ARTICLE

Effect of a Topical Gel Based on Adelmidrol + Trans-Traumatic Acid in the Treatment of Diabetic Foot Ulcers: An Open-Label Study

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Background: Diabetic foot ulceration is a severe complication of diabetes characterized by chronic inflammation and impaired wound healing. This study aims to evaluate the effect of a medical device gel based on Adelmidrol + Trans-traumatic acid in the healing process of diabetic foot ulcers.

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Methods: Thirty-seven diabetic patients with foot ulcers of mild/moderate grade were treated with the gel applied daily for 4 weeks on the affected area. The following parameters were evaluated at baseline and weekly: a) wound area, measured drawing a map of the ulcer then calculated with Photoshop6 tools, b) clinical appearance of the ulcer, assessed recording the presence/absence of dry/wet necrosis, infection, fibrin, neoepithelium, exudate, redness, granulation tissue.

Results: Topical treatment led to progressive healing of diabetic foot ulcers with a significant reduction of the wound area and an improvement in the clinical appearance of the ulcers. No adverse events treatment-related were observed.

Conclusions: The results of this open-label study show the potential benefits of Adelmidrol + Trans-traumatic acid topical administration to promote re-epithelialization of diabetic foot ulcers. Further studies need to confirm the observed results.

The diabetic foot ulcer is a debilitating complication of diabetes mellitus, with a 25% lifetime incidence in patients with both type 1 and 2 diabetes (1). Foot ulceration associated with impaired wound healing produces chronic wounds that impair the life quality of diabetic patients, leading to hospitalization and often resulting in lower extremity amputations (2). Evidence shows that diabetes is characterized by a systemic and cutaneous chronic inflammatory state, which contributes to impaired wound healing (3–5). A substantial role in this phenomenon is played by mast cells (MCs). MCs are immune cells that mature in tissues. In

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normal conditions, they can contribute to skin injury repair, increasing acute inflammation, enhancing new epidermis regeneration, and new blood vessel formation, through the release of several mediators (6–9). However, MCs have also been associated with aberrant healing, as shown by the altered number of mast cells found during the remodelling phase in the skin injury repair in streptozotocin-induced diabetes mellitus in mice (10). An increase in the number of degranulated MC has been reported in both human and mouse diabetic unwounded skin (11). Noteworthy, an improvement of wound healing in diabetic mice has been reported after systemic MC stabilization with disodium cromoglycate (11) and following the topical treatment of diabetic mice with an indole-type MC stabilizer, either before or after wound induction (12). In line with these results, photobiomodulation, a treatment with known MCs stabilizer properties (13), after surgery, significantly improved the tensiometric properties of wound healing alongside with a decrease of total and degranulated MCs number during the remodelling step in a diabetic rat model (14).

Adelmidrol is a diethanolamide derivative of azelaic acid, with amphipathic properties, which make it particularly suitable for topical use. Adelmidrol has been shown to increase the endogenous level of palmitoylethanolamide (PEA)(15), a physiological MC modulator, endowed with anti-inflammatory, and pain-relieving effects (16–18). Thus, Adelmidrol application is able to enhance the intrinsic cellular protection mechanism of the skin (15). This assumption is supported by the finding that its use in canine skin wounds reduces the degranulation of cutaneous mast cells (19), increases the percentage of re-epithelialization, and leads to an

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increase in the density of elastic fibres (20). Finally, the ultrasonographic evaluation of the experimental wounds showed that the topical application of Adelmidrol significantly reduces the wound volume, thus improving wound healing over time (21).

Trans-traumatic acid, also named "plant wound hormone", is a monounsaturated dicarboxylic acid produced by enzymatic degradation of fatty acids (linoleic and linolenic), and many plants physiologically secrete it in response to various types of damage (22–24). Experimental evidence has shown that Trans-traumatic acid can induce proliferation and differentiation of plant tissues and stimulate the post-traumatic cicatricial response. Moreover, it can favor the repair of animal tissues, increasing their cellular proliferation (25) with an increase in the re-epithelialization and growth of fibroblasts in the injured areas (26). This pro-re-epithelizing effect was attributed to the ability of the Trans-traumatic acid to favor the convergence of keratinocytes starting from the margins of the lesions, thus accelerating the reconstruction of epithelial integrity (25).

The purpose of the present investigation is to verify whether the topical application of a medical device gel based on the combination of Adelmidrol + Trans-traumatic acid allows an improvement in re-epithelialization and cicatrization of diabetic foot ulcers.

Materials and Methods

This open-label study was conducted at the outpatient Diabetic Foot Clinic of the Division of Endocrinology, Diabetes and Metabolism, Azienda Ospedaliera Universitaria Integrata, Verona.

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Patients with a diagnosis of type 1 or type 2 diabetes who have the complication of foot ulcerative lesions were enrolled according to the following criteria. **Inclusion criteria:** age ≥ 18 years; presence of foot ulcers of grade 1 A/B/C, 2 A/B according to the Texas Wound Classification System (27); reliability to compliance. **Exclusion criteria:** presence of moderate/severe vascular pathology: ABI (ankle/brachial index) < 0.70 , TcPO₂ (tension cutaneous oxygen) < 30 mmHg (28); presence of moderate/severe infection according to the PEDIS scale; presence of gangrene/dry eschar.

This study was performed following the Good Clinical Practice guidelines and the principles of the Declaration of Helsinki of 1975 and was notified to the departmental Review Board.

Patients were enrolled in the period between May 2017 and August 2018. After expressing informed consent, patients were treated with 1 topical application a day of a gel-based on Adelmidrol + Trans-traumatic acid [Nevamast® gel, Epitech Group SpA, Saccolongo, Italy], on the affected area, after careful cleansing of the wound, for 4 weeks.

The following parameters were evaluated for each patient: a) wound area, measured drawing a map on a polyurethane film grid, which was scanned and calculated with Photoshop CS6; b) clinical appearance of the ulcer, assessed recording the presence/absence of dry/wet necrosis, infection, fibrin, neoepithelium, exudate, redness, granulation tissue. Both the wound area and clinical appearance of ulcers were evaluated weekly. Complete healing was defined as 100% epithelization, and in this case, a value of 0 was attributed to the wound area.

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The offloading device adequacy used by the patient was also assessed weekly. The offloading device was evaluated "adequate" if it was a knee-high device, with an appropriate foot-device interface, worn correctly and continuously all-day-long (29).

Statistical analysis

Statistical analysis to evaluate the variation of ulcer area over time was performed using the generalized linear mixed model (GLMM). Variables such as gender, age, BMI, diabetes duration, type of diabetes, wound duration, presence of neuropathy, vascular complications, offloading were included in the model as covariates. The wound area is given as mean $\pm$ standard error (SE). A responder analysis was also performed. For this purpose, as minimum efficacy criteria (MEC), it was established the corresponding minimum percentage reductions in the wound area. In particular, a patient was considered a responder if the wound area decreased by at least 30%, 50%, or 70% after 4 weeks of treatment (30,31). Percentages of patients reaching the established MEC values were calculated.

The typical ulcer signs were quantified calculating percentages of patients with dry/wet necrosis, infection, fibrin, neoepithelium, exudate, redness, and granulation tissue at each follow-up.

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Results

Thirty-seven diabetic patients with foot ulcers were considered for the study. Demographic data and ulcer details of the patients are described in table 1. Eleven patients dropped out for different reasons: 2 for stationary symptoms, 1 patient for family reasons, 1 patient died of infarction, other 7 patients left because they recovered before the end of the study. Data collected for drop-out patients were considered for the analysis. All uncollected data were deemed to be missing.

Statistical analysis with GLMM showed a significant variation of the mean wound area over time ($p < 0.05$), moving from a value of $77.5 \pm 14.9 \text{ mm}^2$ at baseline to a value of $36.7 \pm 10.2 \text{ mm}^2$ at week 4 (Figure 1, Figure2). Post-hoc analysis carried out between each follow-up and its previous one showed a significant reduction ($p < 0.05$) between baseline and week 1. None of the covariates considered affected the wound area reduction.

Percentages of responder patients for each established MEC at the end of the treatment are given in Table 2.

Concerning changes in ulcer appearance, presence of dry/wet necrosis, infection, fibrin, exudate, redness decreased in patients between baseline and week 4, while the presence of neopithelium and granulation tissue increased between baseline and week 4 (Figure 3).

Percentages of patients presenting the typical clinical features of diabetic ulcers at each follow up are reported in Table 3.

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Discussion

This study shows an improvement in wound healing of diabetic foot ulcers in response to the topical treatment with a gel-based on Adelmidrol + Trans-traumatic acid. The wound area significantly decreases over one month of treatment, and in particular, the effect is already significant after the first week of treatment (Figure 1, Figure 2). At the same time, the percentage of patients with signs of poor wound bed status (presence of dry/wet necrosis, infection, fibrin, exudate, redness) at the end of the treatment, decreases when compared with baseline, while the percentage of patients with signs of healing wound bed status (presence of neoepithelium and granulation tissue) increases by the study end (Figure 3). Our results are in line with a preclinical study showing that Adelmidrol + Trans-traumatic acid can accelerate the healing process in diabetic mice after wound induction, compared with animals vehicle, which shows impaired healing (32).

The diabetic foot usually affects patients with a long duration of illness, with different comorbidities and complex to manage clinically. Moreover, the complexity of this pathological condition also depends on several variables such as the presence of neuropathy, vascular complications, but also inadequate offloading, and other factors such as age, gender, BMI, type of diabetes, diabetes duration, wound duration that can influence ulcer manifestation. From our statistical analysis, none of the variables mentioned above affects wound area reduction. The main limitations of the study are the open-label design with the lack of blindness, small patient sizes, which increase the difficulty in estimating the effect of the treatment.

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Furthermore, the small ulcer size considered at baseline led in some cases to rapid healing, which causes an early exit of some patients from the study, therefore, increasing missing data. Notably, treatment with Adelmidrol + Trans-traumatic acid doesn't show adverse events, is considered easy to use, and shows potential efficacy in diabetic foot ulcer healing. According to a recent study, the achievement of a 50% reduction in wound area observed over 4 weeks of treatment is a robust predictor of long term healing prognosis (30,31). In our study, 41.5% of patients obtained more than 50% of reduction in wound area at the end of the treatment, thus encouraging further prospective studies with a double-blind design, larger patient numbers, a larger size of the ulcer at baseline to confirm our findings. Moreover, long-term treatment period would allow appreciating safety and efficacy of this topical gel better. The results obtained in this study on diabetic foot ulcers, a chronic inflammatory condition characterized by an abnormal increase in MC degranulation (11,12,14), and the already reported ability of Adelmidrol to increase PEA levels (15), a physiological MC modulator known to promote chronic inflammation resolution(16–18), prompt us to sustain the potential contribution that Adelmidrol could provide to the healing processes. Therefore, topical application of Adelmidrol in association with Trans-traumatic acid represents a new therapeutic opportunity for this complex syndrome. Since the manifestations of the diabetic foot derive from specific problems of the lower limbs such as ischemia and neuropathy, that often intersect biomechanical issues, infection, and wound repair, a multidisciplinary approach is always needed. In particular, topical therapy has to involve an in-

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depth evaluation of all the variables that influence wound healing, which has to be followed by the application of specific diversified local treatments, in order to guarantee the most significant healing of the foot and minimize recurrences. Further prospective randomized studies are needed to confirm the results obtained and optimize the effect of the treatment studied.

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Figure 1 Foot wound area over time in diabetic patients receiving topic treatment based on Adelmidrol + Trans-traumatic acid

Data are given as mean $\pm$ standard error (SE). The mean wound area decreases significantly over time ($p < 0.05$); post-hoc analysis showed a significant reduction between baseline and week 1 ($p < 0.05$). Statistical analysis was performed with GLMM.

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Figure 2: Diabetic foot ulcers examples of two patients (P.L. and H.L.), at baseline (A, C) and after 4 weeks of topic treatment based on Adelmidrol + Trans-traumatic acid (B, D)

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Figure 3 Typical clinical features of diabetic ulcers, at baseline and after 4 weeks of topic treatment based on Adelmidrol + Trans-traumatic acid

Percent of patients with dry/wet necrosis, infection, fibrin, exudate, redness decreased between baseline (T0) and week 4 (T4); percent of patients with neoepithelium and granulation tissue increased between baseline (T0) and the week 4 (T4) of treatment.

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Table 1 Demographic and treated ulcer details of the patients treated with Adelmidrol + Trans traumatic acid

Number of patients	37
Gender Male/Female	27/10
Age (mean $\pm$ SD)	69 $\pm$ 10.8
BMI (mean $\pm$ SD)	27.5 $\pm$ 4.6
Type 1 diabetes (%)	8.1
Type 2 diabetes (%)	91.9
Diabetes duration (mean $\pm$ SD)	18.4 $\pm$ 11.8
Wound duration	
$\leq$ 30 days	51.4
>30 days	48.6
Smoker (%) - current or past	18.9
Presence of neuropathy (%)	91.9
Previous foot ulcer (%)	56.8
Ulcer area (mm ²)	77.5
University of Texas grade (%)	
1	70.3
2	29.7
University of Texas stage (%)	
A	37.8
B	2.7
C	59.4
Concomitant pathologies(%)	
hypertension	62.2
IMA	16.2
Ictus	8.1
retinopathy	40.5
Presence of vascular complications (%)	
Yes	75.7
No	24.3
Diabetes therapy (%)	
diet	5.4
hypoglycemic agents	43.2
insulin	37.8
Offloading (%)	
adequate	45.9

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inadequate	51.3
missing	2.7

S.D.= standard deviation

**Table 2 Percent of responders with a wound area reduction of at least 30%, 50% or 70%,
calculated at week 4 after Adelmidrol + Trans-traumatic acid treatment**

Wound Area Reduction (%)	Responders Patients at Week 4 (%)
>30	45.5
>50	41.5
>70	31.4

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Table 3 Patients presenting the typical clinical features of diabetic ulcers at each follow-up

n= total number of patient

Clinical features of diabetic ulcers	Percent of patients				
	Baseline	Week 1	Week 2	Week 3	Week 4
	n=37	n=34	n=31	n=30	n=27
Dry necrosis	2.7	5.9	6.5	6.7	0.0
Infection	5.4	2.9	0.0	0.0	3.7
Fibrin	64.9	64.7	54.8	43.3	22.2
Neo-epithelium	5.4	52.9	74.2	73.3	81.5
Wet necrosis	2.7	0.0	0.0	0.0	0.0
Exudate	45.9	20.6	12.9	23.3	25.9
Granulation tissue	54.1	73.5	77.4	76.7	70.4
Redness	18.9	8.8	3.2	0.0	3.7