Use of Ozonized Oil in Chronic Wounds of Lower Limbs: Preliminary Results

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Abstract. A randomized, double-blind clinical trial approved by the Research Ethics Committee (no. 4,246,236) was conducted in which elected patients had chronic wounds in the lower extremities. Participants aged 55 to 70 years, of both genders, with wounds on the lower limbs for more than 12 weeks of vascular, diabetic or traumatic etiology. 08 patients were randomly assigned in two groups: Group 1 – patients with chronic wounds received treatment with ozonated sunflower oil, concentration of 600 m^2/kg; (milliequivalent/kg) and Group 2 – patients with chronic wounds who underwent treatment with traditional sunflower oil. All patients, when necessary, before starting treatment received debridement necrotic tissues was performed. To perform the bandage, the wound was first cleaned with 0.9% saline solution and then ozonized or traditional sunflower oil were administered throughout the wound bed, and then covered with sterile dry gauze, padding in bandage, and fixed with adhesive. All patients received the same procedure three times a week for 12 weeks. The results showed that the wounds of group 1, ozonized sunflower oil, improve in the aspects of the wounds with the presence of granulation tissue and absence of signs of infection and occurred reduction or complete repair of wounds. In the conventional curative group, with non-ozonized traditional sunflower oil, one wound completely repaired, however the other lesions continued with seroso type exudation, with the presence of edema at the wound site, presence of sign and signs of infection. In view of the above, it was observed that the use of ozonized sunflower oil promoted bactericidal and restorative effects.

Keywords: Treatment · Wound · Ozonized oil · Repair · Tissue damage · Sunflower oil

1 Introduction

The structural and physiological rupture of the integumentary system is called a wound [1, 2]. Immediately after the injury of the tissue integumentary, begins a complex repair process, which includes the interaction of a series of biological events and phenomena capable of stimulating the process of tissue repair, namely inflammation: proliferation,

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proliferation [3]. Nonetheless, in the course of this process, changes may occur that culminate in repair deficiency and, consequently, in the delay or even in the absence of tissue repair, thus, as wounds become chronic [1]. The bandage is the standard treatment of these wounds, its main objective is to provide a physical barrier of temporary protection; absorb wound drainage; and provide the moisture needed to optimize re-epithelialization [4]. Ozone therapy has been an adjunct wound treatment, contributing to tissue repair in an economically viable, noninvasive way and without side effects [5].

The ozone molecule has antimicrobial effect without causing resistance [6]. In addition, the literature shows that ozone therapy improving the inflammatory response by stimulating the synthesis and release of cytokines and growth factors such as epidermal growth factor (EGF), platelets derived growth factor derivative (PDGF), growth transformer factor (TGF) and vascular endothelial growth factor (VEGF), growth transformer factor (TGF) and vascular endothelial growth factor (VEGF) [7–9]. Ozone can used in different form, such as gaseous or dissolved in water or oil (ozonized solutions) [9]. The vantage of ozonized oil is that expenses can reach a decrease proportional to 25% if equated with antibiotic expenses [8, 10].

In addition, ozonized oil has anti-inflammatory and analgesic effects, increasing the release of growth factors capable of contributing to tissue repair [12]. Although ozonized solutions are auxiliary in tissue repair, there is no standardized protocol for the treatment of skin wounds or even comparative studies to evaluate the advantages and disadvantages of ozonized oil. Therefore, this study aimed to evaluate and compare the effects of ozonized sunflower oil with traditional sunflower oil in the treatment of chronic wounds.

2 Material and Methods

A randomized, double-blind clinical trial approved by the Research Ethics Committee (no. 4,246,236) was conducted in which elected patients had chronic wounds in the lower extremities. Eight participants were selected by the means of active search in all health units of the municipality and dissemination of the project to the community by pamphlet and social network. After surveying the candidates participating in the research, they were evaluated to verify whether they were eligible using the following criteria: Inclusion: Patients aged 30 to 70 years, both genders, with wounds on the lower limbs more than 12 weeks of vascular, diabetic or traumatic etiology. Exclusion: Bedridden patients, neoplasms, leprosy, patients with neurological problems, pregnant patients, patients with HIV (Human Immunodeficiency Virus).

Patients were randomly assigned in two groups: Group 1 – Patients with chronic wounds received treatment with ozonized sunflower oil, concentration of 600 m^2/kg; (milliequivalent/kg); Group 2 – Patients with chronic wounds who underwent treatment with traditional sunflower oil.

2.1 Intervention

All patients, when necessary, received adequate debridement of necrotic tissues. To perform the dressing, the first wound was cleaned with 0.9% saline solution and then

ozonized sunflower oil or traditional sunflower oil was administered throughout the wound bed, and then covered with sterile dry gauze, padding in bandage and fixed with adhesive. All patients received the same procedure three times a week for 12 weeks.

2.2 Clinical Assessment

The patient's anamnesis was performed by applying the patient data collection instrument, which included information on the main complaint, history of family disease, main comorbidities, lifestyle, blood glucose measurement and blood pressure measurement. Next, the clinical evaluation of the lesions in which the following clinical aspects were evaluated was performed: wound site, tissue characteristics, exudated amount, edge characteristics, injured skin, based on the protocol used by Campos et al. [13]. These data were obtained before starting the intervention in all participants involved in the research and in cases where the wounds did not repair after 12 weeks of treatment. Clinical evaluation was performed again. As the wound areas were photographed at the beginning and end of treatment using a digital camera.

2.3 Microorganism Analysis

The analysis of microorganisms was performed by collecting the swab from the lesion(s) in the first treatment session. The patient who still presented lesions after 12 weeks was collected in a new swab analysis. The material was collected by an adequate and qualified professional for this procedure, respecting the asepsis techniques and biosafety standards.

3 Results

The study was conducted in 08 patients of both genders, being 5 women and 3 men aged 55 to 70 years, 50% of term venous wounds, 37.5% diabetic and 12.5% traumatic and in relation to wound time, all were more than 12, in the evaluation of weeks of comorbidity weeks the patients evaluated fear diabetes, hypertension and venous insufficiency, only 1 patient did not present comorbidity.

During the treatment, 3 wounds had total repair, two belonging to the group receiving ozonized sunflower oil closed and one receiving conventional dressing. As initial morphological alterations in both groups showed that at the beginning of treatment they presented alterations, characterized by irregular edges, total loss of skin thickness with extensive destruction, muscle damage or support structures, intense edema, presence of exudate, macerated and presence of sides. After treatment, Fig. 1 shows complete repair or reduction of wounds, improvement in wound aspects with the presence of granulation tissue and absence of signs of infection.

Figure 2 represents the wounds belonging to group 2, conventional dressing group with non-ozonized sunflower oil. After the 12-week period, one wound completely noticed, however the too many lesions continued with serosotype exudation, with the presence of edema at the wound site, presence of crumbles and signs of infection.

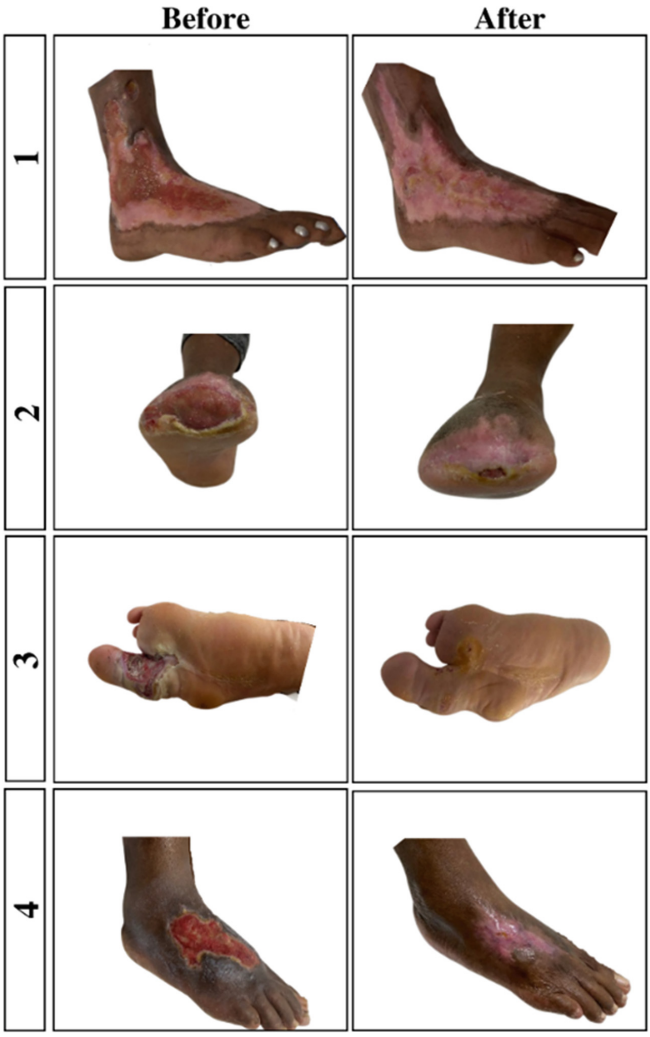

Fig. 1. Wounds treated with ozonized sunflower oil – Group 1

Due to the skin lesions presenting clinical signs of infection samples were collected to identify microorganisms. The results show that in the 8 wounds treated in this study, 13 species of bacteria were identified (Table 1).

The patients of group 1, ozonized sunflower oil, showed greater presence of microorganisms in the lesions, the patients had more than 1 species of bacteria, than the wounds belonging to group 2, traditional sunflower oil.

The most detected species of bacteria in the groups were *Pseudomonas aeruginosa* 38.4%, *Klebsiella pneumoniae* 23.7% and *Klebsiella oxytoca*, *Morganella*, *Enterococcus faecalis*, *Proteus mirabilis* and *providencia stuartii,* 7.6% with one.

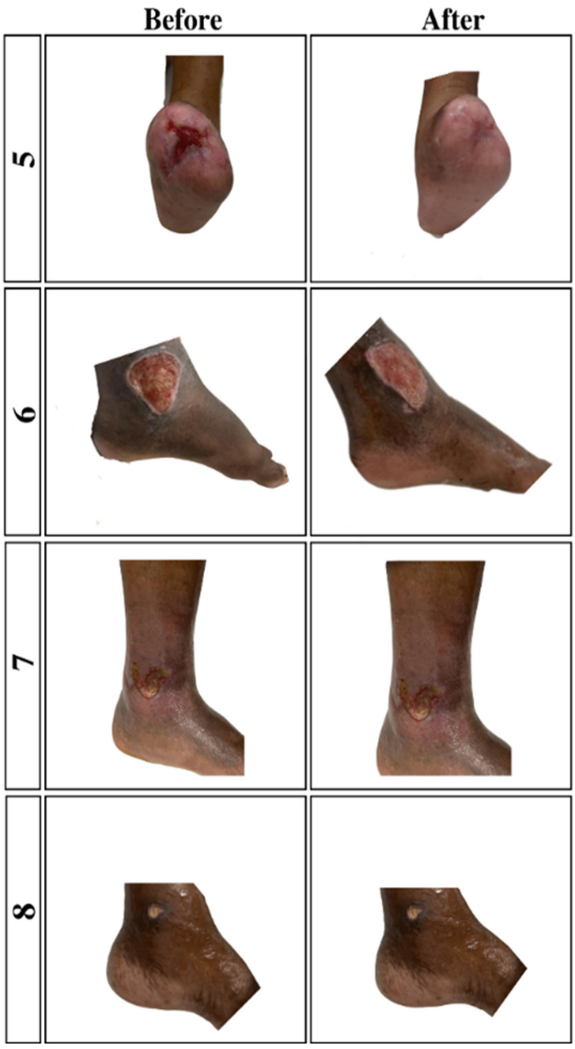

Fig. 2. Wounds treated with traditional sunflower oil – Group 2

After the 12-week period, the wounds that did not fully notice were reevaluated and it was observed that two patients belonging to group 1, ozonized sunflower oil, still had an injured area, as wounds were not infected. However, as wounds belonging to group 2, conventional dressing with traditional sunflower oil, still presented the presence of the same bacteria initially identified (Table 2).

Table 1. Microorganisms present in wounds before treatment

Patient	Bacterium	Level
Group 1		
1	*Morganella Morganii*	Abundant
	Klebsiella pneumoniae	Abundant
	Enterococcus faecalis	Abundant
2	*Mirabilis*	Abundant
	Klebsiella pneumoniae	Lightweight
	Pseudomonas aeruginosa	Lightweight
3	*Pseudomonas aeruginosa*	Lightweight
	Klebsiella pneumoniae	Lightweight
4	*Oxytouch klebsiella*	Lightweight
	Pseudomonas aeruginosa	Lightweight
Group 2		
5	*Pseudomonas aeruginosa*	Lightweight
6	*Stuartii Provides*	Lightweight
7	Negative	–
8	*Pseudomonas aeruginosa*	Lightweight

Table 2. Microorganisms present in wounds after treatment

Patient	Bacterium	Level
Group 1		
1	Negative	–
2	Negative	–
3	No injury	–
4	No injury	–
Group 2		
5	No injury	–
6	*Stuartii Provides*	Lightweight
7	Negative	–
8	*Pseudomonas aeruginosa*	Lightweight

4 Discussion

The present study observed that the use of ozonized sunflower oil promoted bactericidal effect and stimulated the process of skin repair of chronic wounds. These findings are in accordance with the study by Zanardi et al. [14] that observed, *in vitro*, the bactericidal effect of ozonized sesame oil in Gram-positive and Gram-negative bacteria (*Staphylococcus aureus*, *Enterococcus faecalis*, *Pseudomonas aeruginosa*, *Escherichia coli* and *Candida albicans*), which are often detected in wounds in humans. Furthermore, the authors declare that the application of ozonized oil is very promising in a variety of skin and mucosal infections, due to its bactericidal effect, and also warn that, before the application of ozonized oil, the damaged surface of the skin should be cleaned with the removal of necrotic tissue and excess liquid exudate.

Similarly, in an *in vivo* study, it was observed that the use of camellia oil ozonized in excisional wounds on the back of rats accelerated the process of skin repair [15].

Positive results were observed in clinical studies, Campanati et al. [16] compared the use of ozonized oil with hyaluronic gel in the treatment of 30 patients with second-degree skin burn. Each skin burn was subdivided into two symmetrical parts, one part was treated with occlusive dressing of ozonized oil and the lateral part of the lesion was treated with topical application of hyaluronic acid in gel, once a day, for 12 weeks. The authors found that all treated lesions improved regardless of the treatment used, and that ozonated oil was as effective as hyaluronic acid in improving erythema, tension, itching and burning sensation reported by patients.

In view of the above, the use of ozonized sunflower oil may be an alternative treatment for chronic and/or infected skin lesions.

5 Conclusions

The present study demonstrates the potential reparative effect of ozonized sunflower oil in the treatment of chronic wounds and promoted the bactericidal effect on treated wounds.

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Conflict of Interest. The authors declare that they have no conflict of interest.

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