Available online at www.sciencedirect.com
ScienceDirect
journal homepage: www.elsevier.com/locate/burns

A topical aqueous oxygen emulsion stimulates granulation tissue formation in a porcine second-degree burn wound

Jie Li *, *Yan-Ping Zhang*, *Mina Zarei*, *Linjian Zhu*, *Jose Ollague Sierra*, *Patricia M. Mertz*, *Stephen C. Davis*

Department of Dermatology and Cutaneous Surgery, University of Miami Miller School of Medicine, Miami, FL 33136, USA

ARTICLE INFO

Article history:
Accepted 22 November 2014

Keywords:
Topical aqueous oxygen emulsion
Burn wound
Granulation tissue formation
Collagen
VEGF

ABSTRACT

Background: Oxygen is an essential substance for wound healing. Limited studies have shown that topical oxygen can influence healing. This study evaluated the effects of a Topical Oxygen Emulsion (TOE) on burn wound healing.

Methods: A porcine second-degree burn wound model was used in the study. Burn wounds were randomly assigned to TOE, vehicle control, and no-treatment (air) groups. Effects of TOE on the granulation tissue formation and angiogenesis were studied using hematoxylin and eosin histological analysis. Protein production and gene expression of types I and III collagen and vascular endothelial growth factor (VEGF) were determined using immunofluorescent staining and Reverse Transcription and Polymerase Chain Reaction (RT-PCR), respectively.

Results: The TOE treated wounds exhibited better angiogenesis and granulation tissue formation by histology examination. The immunofluorescence staining and RT-PCR analysis demonstrated that protein production and mRNA expression of VEGF and collagen III were significantly higher in TOE treatment group than vehicle alone and air control groups, while there was no significant difference in the level of collagen I.

Conclusions: Our data demonstrate that TOE enhances burn wound healing via stimulating the expression of VEGF and type III collagen and strongly indicates the potential use of TOE in wounds.

© 2014 Elsevier Ltd and ISBI. All rights reserved.

1. Introduction

Second-degree burn wounds involve destruction of the entire epidermis and a substantial part of the dermis and healing depends on the depth of injury. Although superficial burns can re-epithelialize fairly rapid with minimal scarring, deeper second-degree and third-degree burn can take a few weeks to heal and tend to form more severe scars [1].

* *Corresponding author at*: Department of Dermatology and Cutaneous Surgery, University of Miami Miller School of Medicine, 1600 NW 10th Avenue, Miami, FL 33136, USA. Tel.: +1 305 243 3365; fax: +1 305 243 6191.
E-mail address: jli@med.miami.edu (J. Li).

Abbreviations: CNS, central nervous system; HBOT, hyperbaric oxygen therapy; H&E, hematoxylin and eosin; IF, immunofluorescent; PBS, phosphate buffered saline; PO2, oxygen pressure; OCT, optimal cutting temperature; RT-PCR, Reverse Transcription and Polymerase Chain Reaction; TOE, Topical Oxygen Emulsion; TOT, topical oxygen therapy; VEGF, vascular endothelial growth factor.

http://dx.doi.org/10.1016/j.burns.2014.11.016
0305-4179/© 2014 Elsevier Ltd and ISBI. All rights reserved.

Please cite this article in press as: Li J, et al. A topical aqueous oxygen emulsion stimulates granulation tissue formation in a porcine second-degree burn wound. Burns (2014), http://dx.doi.org/10.1016/j.burns.2014.11.016

The tissue repair process requires an increased metabolic activity of a variety of cells, resulting in a high oxygen demand. Therefore, oxygen delivery is a critical element for the wound healing. The oxygen availability to the wound is a rate-limiting step in early repair [2,3]. There is a hypoxic gradient across the partially vascularized wound and the center is more hypoxic with respect to the edge and surrounding tissues. In addition, it has been shown that the percentage of revascularization correlates well with the magnitude of the hypoxic gradient [4].

A variety of studies have been shown that increased oxygen tension in a wound promotes wound healing by stimulating several processes, including collagen production [5] and blood vessel formation [6,7]. Different therapies have been attempted to increase local oxygen supply to wounds and accelerate wound repair such as systemic hyperbaric oxygen therapy (HBOT) and topical oxygen therapy (TOT). HBOT can be effectively applied to treat wounds, especially in hypoxic tissues [8], but it is relatively costly. Patients scheduled for HBO therapy need a careful pre-examination and monitoring. The predominant complication is represented by pressure equalization problems within the middle ear. Serious complications including barotrauma of the nasal sinuses and oxygen toxicity of the CNS rarely occur [9].

In a case-series study TOT has shown no detrimental effects on wounds and showed beneficial indications in promoting wound healing [2]. However, TOT has limited ability to penetrate the skin. The ideal topical oxygen agent would provide sufficient quantities of oxygen to a wound after application and be non-toxic to the skin to accelerate local tissue repair [10].

Topical Oxygen Emulsion (TOE) is a relative new technique which delivers emulsion-containing supersaturated oxygen to a wound and slowly releases additional oxygen. This technology is based on perfluorocarbon droplets being encapsulated within an aqueous continuous phase and has a high oxygen carrying capacity [11].

Our earlier porcine in vivo study found that TOE significantly enhanced the rate of wound re-epithelialization in porcine second-degree burns using a salt-split technique [11]. In this assay, the wounds with surrounding normal skin were excised and incubated in 0.5 M sodium bromide at 37 °C for 24 h, allowing for a separation of the dermis from the epidermis. After the separation, the epidermal sheet was examined macroscopically for defects. The defect was defined as a hole in the area of the wound. Reepithelialization is considered complete if no defect is present or vice versa. However, the effects and mechanisms of TOE on burn wound healing process are still unknown. In this study, we evaluated the effects of TOE on the dermal angiogenesis and granulation tissue formation in porcine second-degree burn wounds. The potential mechanisms involved were explored by measuring the gene expression level with Reverse Transcription and Polymerase Chain Reaction (RT-PCR) analysis of types I and III collagens and vascular endothelial growth factor (VEGF) and their protein production using immunofluorescence staining techniques.

2. Materials and methods

Topical aqueous oxygen emulsion (TOE) and vehicle only emulsion were provided by TherOx Inc. (Irvine, CA, USA) prior to the use. TOE contains super-saturated oxygen which can be delivered topically to a wound [11,12]. The TOE formulation is based on perfluorocarbon droplets being encapsulated within an aqueous continuous phase allowing slow release of oxygen over time. The vehicle is a proprietary oil in water perfluorocarbon emulsion [12]. The oxygen solubility of the perfluorocarbon is relatively high. The dissolved oxygen concentration contained in the topical emulsion is approximately 2.0 ml of O2 (standard temperature and pressure) per ml of emulsion prior to dispersion, which is twenty times greater than water. The oxygen is dissolved into the perfluorocarbon emulsion and stored under pressure in a small dispensing bottle. By maintaining pressure on the emulsion, dissolution and outgassing are prevented during storage and the maximum oxygen concentration is delivered on dispensation. Upon topical applying, the oxygen released from the emulsion sustained for up to 12 h above the atmosphere measured by a transcutaneous oxygen monitor. The topical cream is formulated with biocompatible emulsifying agents to ensure adequate stability of the dispersed perfluorocarbon droplets [11,12].

2.1. Animals, burn wounds, treatments and sample collections

Pigs are an excellent animal model for the evaluation of therapeutic agents for the skin repair due to the similarity in skin histology with humans. There is a strong correlation between pig and human wound-healing studies [13]. Six white, specific pathogen free pigs were used as well-defined porcine models for this study [13,14]. The study was conducted in compliance with the University of Miami's Standard Operating Procedures and approved by the University of Miami Institutional Animal Care and Use Committee [11].

Deep partial thickness second-degree burn wounds, 8.5 mm in diameter by 0.8 mm deep, were made on the anterior two-thirds of each animal skin using cylindrical brass rods which were heated in a boiling water bath to 100 °C. Steam burn on the skin was prevented by drying the rod. The brass rod was held at a vertical position on the skin for 6 s, with all pressure supplied by gravity [11]. Immediately after burning, the roof of the burn blister was removed with a sterile spatula. The burn wounds were made approximately 2 cm from each other and were then randomly assigned to one of three treatment groups: (1) topical aqueous oxygen emulsion (TOE) which contains super-saturated oxygen, (2) topical vehicle only control (Ctr), and (3) no-treatment (air). TOE and vehicle only emulsion were provided in blinded pressurized containers (TherOx Inc., Irvine, CA, USA). The wounds were treated with 200 mg of TOE or vehicle only emulsion or no-treatment twice daily for the first 5 days and once thereafter for a total of 21 days.

On day 0 (prior to wounding) and days 1, 4, 7, 10, 14 and 21 after wounding, excisional biopsies were taken from each treatment group. A total of 114 samples, including six unwounded skin biopsies at day 0 and six wounds (one from each pig) from each treatment at each time point, were collected. Each biopsy was taken in the center of the wounds with adjacent normal skin on each side and divided into three parts. One third of each specimens were immediately placed in cold phosphate buffered saline (PBS) then embedded into Optimal Cutting Temperature (OCT) embedding media and stored in

Please cite this article in press as: Li J, et al. A topical aqueous oxygen emulsion stimulates granulation tissue formation in a porcine second-degree burn wound. Burns (2014), http://dx.doi.org/10.1016/j.burns.2014.11.016

Download English Version:

https://daneshyari.com/en/article/6048694

Download Persian Version:

https://daneshyari.com/article/6048694

Daneshyari.com