http://dx.doi.org/10.1590/1981-86372022002720200152

| REVISÃO

Application of ozonized oils in human body and oral cavity systems

Aplicação dos óleos ozonizados nos sistemas do corpo humano e cavidade oral

Anildo Alves de **BRITO JÚNIOR**[1] 0000-0002-7752-3104

Jaiza Kênsuly Moura Pinheiro **CARNEIRO**[1] 0000-0002-5439-3175

Júlia Vianna Neri Andrade **REIS**[1] 0000-0003-1805-0949

Tiago José Silva **OLIVEIRA**[1] 0000-0003-0080-9152

Juliana Borges de Lima **DANTAS**[1] 0000-0002-9798-9016

ABSTRACT

Wound healing consists of the coordinated cascade of molecular, cellular and biochemical events that interact for tissue recomposition to occur. The interruption or failure in this process can trigger the delay in closing the lesion and chronic wounds, in addition to infections. Ozonotherapy proves to be a promising alternative, capable of assisting in the tissue healing process, modulating the inflammatory response and preventing the development of infection. To present, analyze and discuss, through a literature review, the use of Ozonated Oils in the repair of the human body and oral cavity systems. SciELO, LILACS, MEDLINE and PubMed databases were consulted, using the following DeCS / MeSH descriptors: "ozonized oil"; "healing"; "ozone therapy". After crossing, counting and analyzing, 13 studies were selected, according to the theme. Due to the complementary theoretical foundation, 44 articles were included, totaling 57 articles used for the present review. Ozonized Oil therapy has beneficial clinical effects, which include antimicrobial, immunostimulating, analgesic, antipoxic, immunoregulatory and antioxidant action. Topical management of ozone has proven effective for the treatment of foot-and-mouth disease, gingivitis, ulcers and dermatitis, contributing to pain relief and healing. Infectious skin diseases, abscesses, allergic skin diseases, scaly erythema, psoriasis and palmoplantar pustulosis are pathological conditions, the treatment of which benefits from the application of Ozonized Oils. Ozonized Oils have been shown to be effective in the treatment of local infections, when in appropriate formulations and in controlled cases, being a promising therapeutic alternative for tissue repair.

Indexing terms: Healing. Oil. Ozone.

RESUMO

A cicatrização de feridas consiste na coordenada cascata de eventos moleculares, celulares e bioquímicos que interagem para que ocorra a recomposição tecidual. A interrupção ou falha nesse processo, pode desencadear o atraso no fechamento da lesão e feridas crônicas, além de infecções. A Ozonioterapia revela-se uma alternativa promissora, capaz de auxiliar no processo de cicatrização tecidual,

[1] Faculdade Adventista da Bahia - FADBA, Departamento de Odontologia. BR-101, km 197, Capoeiruçu, 44300-000, Cachoeira, Bahia, Brasil. Correspondence to: JBL Dantas. E-mail: <judyborges@gmail.com>.

How to cite this article

Brito Júnior AA, Carneiro JKMP, Reis JVNA, Oliveira TJS, Dantas JBL. Application of ozonized oils in human body and oral cavity systems. RGO, Rev Gaúch Odontol. 2022;70:e20220027. http://dx.doi.org/10.1590/1981-86372022002720200152

modular a resposta inflamatória e prevenir o desenvolvimento de infecção. Apresentar, analisar e discutir, através de uma revisão da literatura, o emprego de Óleos Ozonizados no reparo dos sistemas do corpo humano e cavidade oral. Bases de dados SciELO, LILACS, MedLine e PubMed foram consultadas, utilizando os seguintes descritores DeCS/MeSH: "ozonized oil"; "healing"; "ozone therapy". Após cruzamento, apuração e análise, foram selecionados 13 estudos, de acordo com a temática. Devido à fundamentação teórica complementar, 44 artigos foram incluídos, totalizando 57 artigos utilizados para a presente revisão. A terapia com Óleo Ozonizado possui efeitos clínicos benéficos, que incluem ação antimicrobiana, imunoestimulante, analgésica, antipóxica, imunorreguladora e antioxidante. O manejo tópico de ozônio revelou-se eficaz para o tratamento de estomatite aftosa, gengivite, úlceras e dermatite, contribuindo para o alívio da dor e cicatrização. Doenças infecciosas da pele, abscessos, doenças alérgicas da pele, eritema escamoso, psoríase e pustulose palmoplantar são condições patológicas, cujo tratamento é beneficiado pela aplicação de Óleos Ozonizados. Os Óleos Ozonizados têm demonstrado eficácia no tratamento de infecções locais, quando em formulações apropriadas e em casos controlados, sendo uma alternativa terapêutica promissora para o reparo de tecidos.

Termos de indexação: *Cicatrização. Óleo. Ozônio.*

INTRODUCTION

The skin is a vital organ that acts as a barrier against the external environment. Having a similar structure, the oral mucosa represents a coating that protects the region against obstacles from the external environment. Bathed in salivary fluid, it consists of a stratified squamous epithelium, with underlying submucosal (connective) tissue. Unlike the skin, the oral mucosa has no direct attachments. Internal or external disruption to the integrity of the skin, mucosa, membrane, tissues, and organs is referred to as a wound. This can be triggered through a variety of mechanisms including thermal, physical, chemical, and biological agents [1].

Tissue repair is a biological event where the organism seeks to regenerate damaged tissues, thereby restoring functional and anatomical integrity. This process is influenced by factors such as the characteristics of the harmful agent, extent of tissue damage, and properties of the tissue involved. For example, small wounds tend to regenerate; however, large wounds often involve deeper layers of tissue, and the mechanism of repair is more complex [2].

The environmental conditions in which tissue injury occurs can directly impact the healing process. For example, the repair of wounds in the oral cavity is influenced by the properties of the saliva and also the number and type of microorganisms present. Studies have shown that the oral cavity has the capacity to house 800-1000 different species of bacteria. This wide microbial diversity includes bacteria, archaea, viruses, fungi, and various micro-eukaryotes. Often, the clinical treatment of the oral cavity is challenging due to the ability of these microorganisms to develop infections. Staphylococcus aureus is the most common bacterium in wound infections. As microorganisms interfere with and delay the tissue repair process, the control of microorganisms has become a widely used clinical strategy [3].

The search for therapies that promote tissue repair is the subject of much research. Pharmacotherapy is a commonly used option; however, adverse side effects and the ability to select resistant microorganisms stimulates the need to search for alternative treatments [2]. Nonsteroidal anti-inflammatory drugs and antimicrobials are examples of agents that can assist in the healing process, as they speed up tissue repair, modulate the inflammatory response, and prevent the development of infectious conditions. Studies have also examined the formulations of ozonized oils and their therapeutic action in tissue repair [4].

A significant property of ozonized oils is their oxidative potential, however, they can also have a bactericidal action against bacteria, without developing bacterial resistance, and this is coupled with low toxicity to cells in the body [5]. In addition to being a non-invasive therapy, they have the capacity to enhance the oxygenation of the site through exposure to ozone (O3) and stimulate the release of growth factors by activating nuclear factor kappa B (NF-κB). Ozonized oils are also able to act in conjunction with the immune system by improving the local inflammatory response, through release of growth factors such as epidermal growth factor (EGF), transforming growth factor β (TGF-β), transforming growth factor, vascular endothelial growth factor (VEGF), and platelet-derived growth factor (PDGF) [5,6].

There is growing interest in the use of alternative therapies rather than conventional drug therapy. Ozonized solutions act as excellent therapeutic alternatives for tissue repair and offer antimicrobial effects. The aim of the present study is to analyze and discuss the use of ozonized oils in the repair of tissues and oral cavity systems, through a narrative review of the literature. As it is evident that for repair to occur, there is a need to eliminate microorganisms, we also address the antimicrobial effect of this therapeutic agent.

METHODS

This is a descriptive, exploratory study developed through a critical review of the literature. A detailed database search of the proposed theme was carried out through: Scientific Eletronic Libray Online (SCIELO), Latin American and Caribbean Literature in Sciences (LILACS), Online Search and Analysis of Medical Literature of the United States of America (MedLine), and PubMed. The DeCS / MeSH descriptors used during the literary search were: "ozonized oil", "healing", and "ozone therapy," since the primary objective was to evaluate the effect of this therapeutic agent on tissue repair.

The electronic search and article selection took place between October 2019 and April 2020. After selecting the keywords, the VHL (Virtual Health Library) was cross-checked with the Boolean AND / OR operators of the descriptors as follows: "oil" AND "ozone," in which articles were obtained from the MedLine, PubMed, LILACS, and BBO databases. In SciELO, the descriptor "Ozonized Oil" was used, as the platform allows the search for articles using descriptors in Portuguese. The studies obtained in each database are described in tablel 1.

Table 1 – Crossings of the DESC/MESH descriptors and the number of studies found in the databases.

Data Base	Crossing	Simple Identified	Excluded	Final Sample
SciELO	Óleo Ozonizado	2	1	1
MedLine	Oil AND ozone OR healing AND ozone therapy	19	17	2
LILACS	Oil AND ozone OR healing AND ozone therapy	6	6	0
BBO	Oil AND ozone OR healing AND ozone therapy	1	1	0
PubMed	Oil AND ozone OR healing AND ozone therapy	23	13	10
Total	–	51	38	13

During the investigation, we identified 13 articles whose titles and keywords referred to the theme "Ozonized Oil,". All of these studies had an emphasis on tissue repair, especially in the human body and oral cavity systems. For all studies related to the proposed theme, the titles, abstracts, and texts were read in full. Two reviewers screened the articles. Both had access to the same references, however, they selected them independently.

To refine the search, the inclusion criteria were: articles with complete texts that addressed the proposed theme, publications made in the last 10 years, and without definition of language. The types of work defined for analysis were systematic reviews, meta-analyses, *in vivo* studies, and clinical trials. The exclusion criteria were: articles duplicated on different bases, articles that did not provide the full text, *in vitro* studies, and theses. Additionally, studies that were not related to the research topic were excluded. The inclusion of the articles followed previously established criteria in order to refine the indexed publications.

The theoretical foundation model was structured and based on titles and subtitles that addressed the stages of tissue repair, general properties of ozone therapy, characteristics of ozonized oils on tissue repair, and the use of ozonized oils in the human body and cavity systems. Due to the complementary theoretical foundation, 44 additional articles were included, totaling 57 articles used for the present review.

State of art

Steps of tissue repair

Wound healing consists of a coordinated cascade of molecular, cellular, and biochemical events. Baron [6] described this detailed healing process in three sequential phases: inflammation, proliferation, and remodeling.

Inflammatory stage

The first phase of the healing process occurs immediately after the tissue injury through the activation of platelets and coagulation mediators. A blood clot forms, and in addition to limiting blood leakage, it acts as a temporary matrix that facilitates the migration of cells to the wound. Activation of the coagulation cascade, parenchymal cells, and the complement system, together with the release of growth factors, produces numerous vasoactive mediators and chemotactic factors that assist in the recruitment of innate immune cells to the site of tissue damage [6].

Neutrophils are the predominant cells at the beginning of the inflammatory phase. Involved in the phagocytosis of necrotic tissue and bacterial particles, they also provide a source of reactive oxygen species (ROS), creating a hostile environment for bacteria. Neutrophils also stimulate the production of pro-inflammatory cytokines and the proliferation of keratinocytes. From 24-72 h after tissue damage, there is a migration of monocytes, which differentiate into macrophages upon reaching the tissue space. These cells have phagocytic activity, and remove foreign bodies and debris from the wound. In addition, macrophages act as antigen presenters and produce growth factors (PDGF, VEGF, TGF-β, and fibroblast growth factor) necessary to stimulate the formation of granulation tissue. These cells maintain homeostasis inside the wound, preventing a pathological inflammatory response [7].

The inflammatory phase persists for a period of 3-10 days. The characteristic events of this phase are translated into the cardinal signs of inflammation: heat, redness, pain, tumor, and loss of functio [6].

Proliferative stage

The proliferation stage occurs from 4-12 days after the injury and is characterized by an increased number of fibroblasts, a phenomenon known as fibroplasia. These cells are responsible for collagen deposition, producing a sealing structure over the damaged area, and the consequent formation of the new extracellular matrix. Fibroblasts also secrete keratinocyte growth factor derivatives, and endothelial cells produce VEGF. The angiogenesis stage then begins, in which new blood vessels originate from pre-existing vessels. Angiogenesis promotes the supply of oxygen and nutrients to the tissue and subsequently leads to re-epithelialization, where the outer layer of the wound recovers [6, 7].

Remodeling step

Remodeling is the last stage of healing, characterized by the reorganization of connective tissue, collagen remodeling, and the beginning of the contractile response. This phase begins two to three weeks post injury. However, even after one year, the wound may maintain disorganized collagen fibers. This healing process involves a complex system of biological events due to the restoration and maintenance of tissue integrity [6].

The remodeling process of collagen fibers is marked by the conversion of type III collagen to type I collagen. The metalloproteinases MMP2, MMP3, and MMP14 are expressed in the remodeling of this scar tissue. Approximately 30 days after tissue damage, a balance between collagen types I and III is achieved. For this healing process to be successful, it is necessary for the tissue to be well oxygenated, since poor oxygenation can trigger local cell damage from

excessive tension in the distal tissues [7]. The result of the interaction between the extracellular matrix, supported by active fibroblasts, from the edges to the wound bed, will decide the scar quality [2].

Although the description of the three phases is carried out sequentially, it should be noted that tissue repair occurs through a complex and continuous mechanism, which begins immediately after tissue injury, with the phases of inflammation, proliferation, and remodeling occurring in an overlapping and even parallel nature [6]. The interruption or failure of this healing process can trigger a delay in closing the lesion or chronic wounds, which can lead to the production of excessive scar tissue, hypertrophic scars, or keloids. It is worth mentioning that infectious processes interfere with the quality of healing, with the need for alternative therapies that promote antimicrobial and healing actions, concurrently [3].

Ozone Therapy

Ozone is a triatomic allotropic form of oxygen (O3), in which the three atoms are organized into a cyclic structure. It has a unique odor, with an oxidizing and unstable characteristic, which is up to 10 times more soluble in water compared to the diatomic form of oxygen (O2). O3 is a molecule that decomposes naturally due to its high reactivity and instability, which makes storage difficult. The rate of ozone depletion can interfere with the presence of water, temperature, and the presence of ions. Reduced ionic concentrations and low temperatures increase their half-life, while high concentrations of ions and high temperatures decrease its half-life [8].

The local administration of ozone therapy can be performed either with a mixture of water and gas, or with ozonized oils. Several types of infections, such as bladder, vaginal, and rectal, are effectively treated with ozone therapy. Other injuries such as ulcers, burns, infected or clean wounds can be treated with topical application of the ozonated substance. When properly used, ozonized oils are an excellent therapeutic alternative [5,9,10].

Ozonized Oils

The use of ozonated solutions, especially oils, is easy to implement. This is because, in addition to presenting topical use, and unlike gas systems, oils do not develop a risk of inhalation. It is also worth noting that when the gas is solubilized through vehicles, it becomes more effective for application on the skin, as its effectiveness is increased in a humid environment [1,3,6].

Chagas & Mira [11] evaluated the biological effects of ozonized oil on the healing of dermal wounds induced in animals. They described that for the oil production method the following materials are required: an ozone generator, a source of medical oxygen, a glass container with a lid, and extra virgin vegetable oil without preservatives. It is necessary that the ozone generator is connected to an energy source, and the medical oxygen. The ozone generator must also be connected to the glass bottle (with oil inside) sealed and with an outlet connected to an ozone catalyst. To start the production of ozonized oil, the oxygen valve is opened followed by the initiation of generator to the stabilizer at full power. It is common that inside the glass container, some bubbles begin to form, which represents the beginning of the process of ozonization of the oil. It is recommended to leave each mL of the substance for one min in the ozonation process [9,10].

Many types of oils can serve as vehicles, however olive and sunflower oils are mainly used. The acyclic and unsaturated hydrocarbons, also called alkenes (alkenes) present in vegetable oils, lead to the formation of the 1,2,4-trioxolane portion, which represents the active form of ozone in these substrates. The presence of the trioxolane ring in plant matrices has the role of generating compounds responsible for the tissue repair process when the oil is applied at the site of the wet wound or ulcer [9]. Oil therapy can reach an effective potential through the presence of double bonds of unsaturated fatty acids that would be ozonated. Following the reaction of ozone with oil, double ozone

oxides bind to an oxygen atom, characterizing the ozone bubbling process. This process produces peroxides, and the degree of ozonation is determined by the quantification of these [9,10,12].

Ozonized oils were initially used in Cuba to treat ulcers, viral, bacterial, and parasitic infections, using topical and oral application routes. There is no defined protocol for the ozonation of oils; however, the literature states that if the ozonation is prolonged, beyond two days, the oil will solidify. This process results in 160 mg of ozone per gram of oil. To keep the product in good condition for up to two years, it is recommended to store it under refrigeration and dilute it in vegetable oil when using [4].

To obtain a well-ozonized oil, some characteristics must be considered. The first is that when the oil is ozonated, it loses its original color, becoming colorless. The second characteristic is the increase in oil density according to the proportion of exposure to ozone, becoming more viscous than it previously was. Finally, the ozonated product must be stored in a dark glass container and sealed completely. The disadvantage of this storage is that it is not possible to visualize the quality of the product and can only be determined upon opening the container. The product can also be evaluated by smell. It should also be noted that perfumes can change the color of the oil, and therefore, it is important that this change is explained by the manufacturer [9,12].

Since the industrialization of ozonized oil, some parameters regarding the quality of the derivatives have been observed. Firstly, the type and quality of ozone generators must be regulated by recognized bodies. Secondly, ozonation conditions should be assessed with respect to reactors, time, quality, and type of material, whether there is presence of water or even catalysts. Finally, the effectiveness of the ozonator is vital as it controls gas flow and the O3 concentration. Additionally, medical O2 should be used concomitantly, as it increases the efficiency of ozonation. This is mainly due to 78% nitrogen being present in unsaturated substrates in ambient air, which can lead to the production of potentially toxic nitrate by-products [12].

It is essential that the vegetable oils undergo controlled oxidation during the manufacturing process so the reaction with ozone takes place under pre-established conditions. Active components resulting from ozonation are hydroperoxides and other lipid peroxidation products, which have non-specific germicidal peculiarities, and are involved in tissue healing and repair processes. Because of these characteristics, oils are valuable in treating ulcers, septic processes, local infections, and fistulas [9].

Among the various types of oils that can be ozonated, the literature suggests that sesame oil may be beneficial. This has been highlighted due to the use of it by the pharmaceutical industry, where its useful chemical composition achieves the balance between oleic acid and linoleic acid. Clinical studies have shown that ozonized oils have several antimicrobial therapeutic properties, as well as the ability to stimulate tissue repair [9,10].

Effects of ozonized oils on human tissue

Ozonized oil has a range of beneficial clinical effects, including antimicrobial action, immunostimulating, analgesic, antipoxic, immunoregulatory, antioxidant, epigenetic, vasodilation, and biosynthesis [13-23]. One of the most striking properties is a strong oxidizing action in the destruction of microorganisms. When properly applied, ozone therapy is able to eliminate pathogens and, following the release of oxygen, activates the proliferation of fibroblasts and keratinoblasts, initiating the construction of the intercellular matrix, with consequent tissue healing. Therefore, in contaminated lesions, this therapeutic agent is able to acts in a twofold nature to destroy microorganisms and the consequently accelerate tissue repair [10-12].

Ozonized oils have an active molecule in their chemical structure that has been widely applied to treat more than 50 conditions, including skin diseases [10]. For the treatment of psoriasis, this oil has shown an equivalent and, in some parameters, superior efficacy in relation to topical corticosteroid preparations. This effectiveness justified the ability of oils to differentiate keratinocytes by increasing the transcription of the KRT10 gene, mediated by the tumor protein (Tp63), with improvement in the clinical signs of psoriasis [12,14].

The application of these oils to ulcers can lead to healing and restore the remodeling of scar tissue. One study found that ozone therapy with oils healed ulcers in the elderly and diabetics in less time, with no local or generalized side effects [9]. This performance is based on the presence of the 1,2,4-trioxolane molecule, which, when in contact with the ulcer, decomposes slowly, with the generation of local oxygen and hydrogen peroxide (H2O2), together with ROS and a trace of oxidation product lipids (4-HNE). These reaction products generate prolonged disinfection and stimulation of

Table 2 – Distribution of use of ozonated oils in human body systems.

Authors / Year	Methodology	Results
Menéndez et al. [16]	Randomized controlled phase III clinical trial. The effectiveness of ozonized oil in the treatment of Tinea pedis was evaluated. Ozonized oil was compared with 2% ketoconazole cream (Nizoral®, Janssen, São José dos Campos, SP, Brazil) in 200 people, with allocation of 100 patients in each group. Administration took place twice a day for 6 weeks.	All injuries regressed. From a clinical and mycological point of view, a success rate of 75 and 81% was achieved for ozonized oil and ketoconazole, respectively, with no statistically significant difference. None of the groups showed side effects. After 6 months of treatment, there was no recurrence in the ozone group. Ozonized oil proved to be an effective and low-cost medication.
Menéndez et al. [17]	Randomized phase III clinical trial for the treatment of onycosis. A total of 400 outpatients were randomly allocated to 2 groups: Experimental, treated with ozonized oil twice a day, and the control, treated twice a day with 2% cream ketoconazole (Nizoral®, Janssen, São José dos Campos, SP, Brazil), for 3 months.	In the group that used ozonized oil, 90.5% of the patients exhibited complete remission. In the control group, 13.5% achieved complete regression and 59% maintained the initial clinical aspect. One year later, the experimental and control group had 2.8% and 44.4% of relapses, respectively. Topical ozone therapy has shown superior efficacy in the treatment of onycosis. No side effects were observed.
Campanati et al. [18]	A total of 30 patients with 2nd degree skin burns in the reepithelialization phase were included. The burn was subdivided into 2 symmetrical parts: 1 st treated with exclusive application of ozonized oil and the 2nd part, used hyaluronic acid gel, daily, for 12 weeks. All patients underwent clinical evaluation and videocapillaroscopy at the beginning of the study, 6 and 12 weeks later.	Regardless of the treatment used, all injuries showed improvement. Ozonized oil was as effective as hyaluronic acid in improving tension, itching, erythema and burning sensation signaled by patients. However, ozonized oil appears to be more effective than hyaluronic acid in reducing hyperpigmentation after injury ($p<0.05$).
Solovăstru et al. [19]	Randomized controlled study with 29 patients who had chronic venous ulcers for less than 2 years. Ozonized Oil 9.6% O_3 and α-bisabolol (Azexin, Alfa Wassermann, Bologna, Italy) were administered in the Experimental group or cream of vitamin A and E, talc and zinc oxide, in the control group for 30 days. On days 0, 7, 14 and 30, ulcers were measured. The area of the wound and the speed of healing were calculated.	The group that received the formulation with ozonized oil and α-bisabolol showed complete healing of the ulcers (25% *vs* 0%). In addition, the ulcers in the Experimental group had a noticeable reduction in the wound, with 34%, 59% and 73% after 7, 14 and 30 days of ozone administration, respectively ($p<0.05$).
Laureti et al. [20]	A total of 26 patients with Crohn's disease and residual fistula with minimal drainage were included in the study. Patients were treated with ozonized oil. This agent was administered daily for a period of 1 to 6 months	Residual healing of the fistula occurred in 65.4% (17/26) of the patients, with an average follow-up time of 23.81 (range, 2 to 42) months.
Lu et al. [21]	Randomized blind study with 60 patients with Tinea pedis, who were allocated to 2 groups. In the control group, Naftinfin Hydrochloride (Chongqing Hua Bang Pharmaceutical Ltd., China) and ketoconazole cream (Chongqing Hua Bang Pharmaceutical Ltd., China) were applied once a day. The ozone group was treated with an ozonized water bath and subsequent topical application of ozonized oil, once a day. The treatments took place for 4 weeks. At the end of the 1st, 2nd and 4th week, clinical and laboratory information was collected.	Within 4 weeks of therapy, 6 patients in the control group were positive by mycological examination, while 1 patient using ozone was positive, with no statistical difference between the groups ($p>0.05$). Changes in signs and symptoms at the end of the 1st, 2nd and 4th week were noted and did not show any significant difference between the groups, in the 3 different moments ($p>0.05$). No observations regarding side effects were reported.
Quin et al. [22]	Randomized clinical study with 60 children with atopic dermatitis, who were allocated to the experimental group (n=30) and the control group (n=30). Hydrotherapy was performed 3-5 times a week and ozonized oil was applied twice a day. In the control group, washing was performed with warm running water and application of base oil (Nanhaizhi Medical Technoly Co., Ltd) and moisturizer, if necessary. The treatment lasted 2 weeks.	Inflammation in the skin reduced and erosions healed after 3-5 days of topical ozone application. The effective rates were 80% in the Experimental group and 20% in the control group in the respective treatments.

Source: Own authorship.

fibroblast and keratinoblast proliferative activity. It is worth noting that, before applying the oil, the surface of the skin is clean in order to remove necrotic tissue, loose deposition of fibrin, pus, and excess liquid exudates. Generally, ozonized oil is applied twice a day to skin ulcers [7, 9].

Therapy based on ozonated oil preparations can act as a potent antibacterial agent by reducing the amount of Staphylococcus species, and can restore the balance of microbiota in the skin. The oil can act by selecting beneficial microbial species, such as Acenetobacter, which improves the conditions of injured skin [10]. Restoration of the epidermal microbiota helps to repair the function of the skin barrier. In addition, ozone therapy also acts more effectively as an antiseptic agent than topical antibiotics, as it reduces the risk of bacterial resistance [10,15].

A study comparing a synthetic antifungal and ozonized oil for the treatment of "Tinea pedis," concluded that most antifungals led to side effects, especially when used over long periods. However, ozonized oil proved to be safer, more effective, and a low-cost antimycotic therapy and an alternative to conventional treatment [15]. Table 2 shows the results of studies that evaluated the performance of ozonized oil in various systems of the human body through tissue repair directly or indirectly, in which there was initially antimicrobial action.

Infectious skin diseases, abscesses, allergic diseases (atopic dermatitis, eczema, urticaria), scaly erythema, psoriasis and palmoplantar pustulosis are conditions which benefit from the application of ozonized oils [11,13,16]. The effectiveness of this therapy for wound healing, recovery of ulcers, and excellent antimicrobial activity against protozoan parasites, such as Giardia duodenalis and Leishmania major, is also noteworthy. Treatment with oil can be either monotherapy or in combination with other medications [15]. For patients with diabetes mellitus and atherosclerosis, the benefits of ozonized oils can represent an integrative therapy for the treatment of tissue injuries. Other potential topical applications are ocular use and antifungal vaginal formulations [23].

Properties of ozonized oils on the oral cavity

There are few studies in the literature that address the use of ozonized oil in the oral cavity, as well as its exact mechanism of action in injuries of different origins located in this anatomical site [24-30]. According to Ripamonti [24], topical management of ozone therapy can be effective in the initial stages of wound healing. The authors further confirmed that ozone therapy acted positively in the cascade of early tissue healing in secondary palatal wounds, increasing the expression of VEGF. It is suggested that further studies are developed to clarify the additional effects of topical ozone therapy with regard to application in different soft tissue healing methods.

For conditions such as foot-and-mouth stomatitis, gingivitis, ulcers, and dermatitis, the use of ozonized oil can help relieve pain and accelerate the tissue repair process. To improve periodontal health and assist in the healing of oral lesions and wounds, the application of these oils has been shown to be a promising and effective alternative treatment [25,26].

In dentistry, the application of ozonized oils has been following the transformations and future therapeutic advances [23,26]. The beneficial properties of ozonized formulations for the treatment of oral cavity lesions suggest that these substances are incorporated as an adjunctive therapy or even an alternative to conventional therapy [24-30]. Table 3 shows studies that have shown the effectiveness of using ozonized oils for patients' oral conditions.

Topical application of ozonized vegetable oils, with short-term subcutaneous coadministration of the immunosuppressive agent cyclosporine A, improve osseointegration, leading to accelerated bone neoformation around dental implants. In the field of oral lesions, the literature shows regression of lesions of cold sores, aphthous ulcerations, oral candidiasis, and angular cheilitis, as well as reduction of the burning sensation of oral lichen planus following treatment with ozonized oil. This antimicrobial and healing potential suggests the effectiveness of ozone therapy as a therapeutic agent for various disorders in the oral cavity [24-30].

Table 3 – Therapies with ozonated oils with an emphasis on oral cavity and interconnected systems.

Authors / Year	Methodology	Results
Ripamonti et al. [24]	Preliminary prospective study of phase I-II in 10 subjects undergoing treatment with bisphosphonates diagnosed with osteonecrosis. Ozonized oil was applied to wounds ≤2.5cm. Each patient was treated with a maximum of 10 local applications of the oil, 1 time every 3 days, with each application lasting 10 minutes.	The lesions healed completely, between 3 and 10 applications. No surgical intervention was necessary. The radiographic examination found no residual bone lesions after therapy. It was also observed success and tolerability to ozone in the form of oil.
Hernández et al. [27]	Phase III randomized clinical trial. The sample included 90 patients with pericoronitis. In the experimental group, ozonized oil was applied 3 times a day, for 48 or 72 hours. In the control group, trichloroacetic acid was applied, antibiotic therapy depending on the type of pericoronitis and relief of trauma in the region (extraction of the antagonist or reduction of cusps).	The effectiveness, with statistical significance (p=0.004) of the ozonized oil in the treatment of pericoronitis was verified in relation to the control group.
Patel et al. [28]	Randomized study with 18 individuals, who were divided into a control group (n=10) and an ozonized oil group at a concentration of 14 Ig/mL (n=8). A free gingival graft surgical procedure was performed, with the application of 2 mL of oil to the palatal wounds daily for 1 week. Clinical and cytological analysis was performed at 7 different times.	The group treated with ozonized oil, when compared to the control, showed significant improvements (p≤0.05) in wound size on days 5, 7, 14, 21 and 28 and higher cytological results (p≤0.001) on days 7, 14 and 21 and in the 2nd and 3rd month, after the surgical procedure.
Patel et al. [29]	Randomized triple-blind placebo controlled study with 24 subjects with localized gingival recession, who were treated surgically, and allocated to an ozonized oil group at a concentration of 14Ig/mL (n=12) and a control group (n=12). After 24 hours, from the 3rd, 7th, 14th and 21st day and 2, 3, 8 and 18 months after the operation, cytological parameters were recorded.	The results showed that there was a significant evolution (p<0.001) in epithelial repair on the 7th, 14th and 21st days and 2, 3 and 8 months after the operation in the ozonized oil group compared to the control group.
Huang et al. [30]	A total of 60 Herpes Zoster patients were allocated to two groups: control; oral use of valacyclovir, 0.3g per day, 3 times a day, associated with lowlevel laser and topical mupirocin ointment 2%, 2 times a day; ozone group followed the same treatment protocol, except replacing the mupirocin ointment with topical use of ozone, which included the daily hydrotherapy associated with ozonized oil, twice a day. Symptoms, adverse reactions and discoid cells were observed on days 0, 3, 7 and 14.	On the 7th day of treatment, the discoid cells in the ozone group regressed, with a statistically significant difference compared to the control group (p<0.05). The pain scores in each situation between the two groups was statistically significant (p<0.05). Clinical results showed 100% effectiveness in the Ozone group and 86.7% in the Control group, with statistical difference (p<0.05).

Source: Own authorship.

FINAL CONSIDERATIONS

Considering the number of products available for the treatment of wounds, it is relevant that the selection of these agents is carried out based on evidence. Factors such as the general condition of the patient, location and type of the wound, availability and access to the desired treatment, and patient preference must be considered.

The topical application of ozone therapy, through the formulation of ozonized oils, has been shown to be effective in the treatment of local infections, as well as in the acceleration of tissue repair, and is a promising alternative for the treatment of several conditions involving skin and the oral cavity. In addition, ozone therapy has provided evidence for its use in topical administration and has an appropriate safety profile, especially when administered in the form of ozonized oil. However, changing treatments requires scientific support and must also be associated with technologies available in the market. For this reason, more studies need to be carried out in an attempt to discover the exact mechanism of action in tissue repair, as well as to obtain adequate protocols.

Collaborators

AA Brito Júnior collaborated with data analysis, wrote the text and contributed to the formatting of the same. JKMP Carneiro contributed to the writing text and bibliographic research. JVNA Reis contributed to the revision and formatting of the article. TJS Oliveira contributed to a final review of the article. JBL Dantas participated to the all stages of the scientific article and its final review

for publication. All authors made substantial contributions to the conception, design of the work, acquisition, analysis, interpretation of data, approval of the final version to be published; and are responsible for all aspects of the work, ensuring that issues related to the accuracy or integrity of any part of the article are properly investigated and resolved.

REFERENCES

1. Qin R, Steel A, Fazel N. Oral mucosa biology and salivary biomarkers. Clinics in Dermatology. 2017;35:477-83. https://doi.org/10.1016/j.clindermatol.2017.06.005
2. Zhang S, Chen C, Ying J, Wei C, Wang L, Yang Z, Qi F. Alda-1, an Aldehyde Dehydrogenase 2 Agonist, Improves Cutaneous Wound Healing by Activating Epidermal Keratinocytes via Akt/GSK-3β/β-Catenin Pathway. Aesthetic Plast Surg. 2020; 44(3):993-1005.
3. Sun Y, Ogawa R, Xiao B, Feng Y, Wu Y, Chen LH, et al. Antimicrobial photodynamic therapy in skin wound healing: A systematic review of animal studies. Int Wound J. 2019; 17(2):285-99. https://doi.org/10.1111/iwj.13269
4. Sanguanini RC. Efeitos da água ozonizada e do óleo ozonizados no reparo tecidual de feridas cutâneas experimentalmente induzidas em ratos. Goiânia: UFG; 2019.
5. Borges GÁ, Elias ST, Silva SM, Magalhães PO, Macedo SB, Ribeiro AP, Guerra EN. In vitro evaluation of wound healing and antimicrobial potential of ozone therapy. J Craniomaxillofac Surg. 2017;45(3):364-70. https://doi.org/10.1016/j.jcms.2017.01.005
6. Baron JM, Glatz M, Proksch E. Optimal Support of Wound Healing: new insights. Dermatology. 2020;236(6):1-8.
7. Childs DR, Murthy AS. Overview of Wound Healing and Management. Surg Clin North Am. 2017;97(1):189-207. https://doi.org/10.1016/j.suc.2016.08.013
8. Qin S, Cheng L, Selorm AL, Yuan F. An Overview of Ozone research. J Adv Oxid Technol. 2018; 21(1):297-302.
9. Zeng J, Lu J. Mechanisms of action involved in ozone-therapy in skin diseases. Int Immunopharmacol. 2018;56:235-41. https://doi.org/10.1016/j.intimp.2018.01.040
10. Zeng J, Dou J, Gao L, Xiang Y, Huang J, Ding S, et al. Topical ozone therapy restores microbiome diversity in atopic dermatitis. Int Immunopharmacol. 2020;80:106191. https://doi.org/10.1016/j.intimp.2020.106191
11. Chagas LH, Mira A. Efeito do óleo ozonizado em lesões cutâneas em ratos. Rev Cultivando Sabor. 2015;(Edição Especial):168-81.
12. Gao L, Dou J, Zhang B, Zeng J, Cheng Q, Lei L, et al. Ozone therapy promotes the differentiation of basal keratinocytes via increasing Tp63-mediated transcription of KRT10 to improve psoriasis. J Cell Mol Med. 2020;24(8):4819-29. https://doi.org/10.1111/jcmm.15160
13. Monzillo V, Lallitto F, Russo A, Poggio C, Scribante A, Arciola CR, Colombo M. Ozonized Gel Against Four Candida Species: a pilot study and clinical perspectives. Materials. 2020;13(7):1731. https://doi.org/10.3390/ma13071731
14. Tan L, Huang J, Lu J. Clinical efficacy of ozonated oil in the treatment of psoriasis vulgaris. Med Sci. 2018;43(2):173-178.
15. Aghaei M, Aghaei S, Sokhanvari F, Ansari N, Hosseini SM, Mohaghegh MA, et al. The therapeutic effect of ozonated olive oil plus glucantime on human cutaneous leishmaniasis. Iran J Basic Med Sci. 2019;22(1):25-30. https://doi.org/10.22038/ijbms.2018.29232.7064
16. Menéndez S, Falcón L, Simón DR, Landa N. Efficacy of ozonized sunflower oil in the treatment of tinea pedis. Mycoses. 2002;45(8):329-32. https://doi.org/10.1046/j.1439-0507.2002.00780.x
17. Menéndez S, Falcón L, Maqueira Y. Therapeutic efficacy of topical OLEOZON® in patients suffering from onychomycosis. Mycoses. 2011;54(5):272-7. https://doi.org/10.1111/j.1439-0507.2010.01898.x
18. Campanati A, De Blasio S, Giuliano A, et al. Topical ozonated oil versus hyaluronic gel for the treatment of partial- to full-thickness second-degree burns: A prospective, comparative, single-blind, non-randomised, controlled clinical trial. Burns. 2013;39(6):1178-83.
19. Solovăstru LG, Stîncanu A, De Ascentii A, Capparé G, Mattana P, Vâţă D. Randomized, controlled study of innovative spray formulation containing ozonated oil and α-bisabolol in the topical treatment of chronic venous leg ulcers. Adv Skin Wound Care. 2015;28(9):406-9.
20. Laureti S, Aborajooh E, Mattioli B, Poggioli G. Treatment of minor dehiscence after endorectal advancement flap in perianal Crohn's fistulas with ozonized oil NOVOX®. Tech Coloproctol. 2015;20(2):139-40. https://doi.org/10.1007/s10151-015-1404-4
21. Lu J, Guo M, Ligui H, Wu K, Xiang Y, Huang J, Gao L. Efficacy of combination of ozonated water with oil for treatment of tinea pedis. Zhong Nan Da Xue Xue Bao Yi Xue Ban. 2018;43(2):147-51. Chinese. https://doi.org/10.11817/j.issn.1672-7347.2018.02.007
22. Qin G, Huang J, Pan Y, Xiang Y, Ou C, Huang J, et al. Topical ozone application: An innovative therapy for infantile atopic dermatitis. Med Sci. 2018;43(2):163-7. https://doi.org/10.11817/j.issn.1672-7347.2018.02.010
23. Ugazio E, Tullio V, Binello A, Tagliapietra S, Dosio F. Ozonated oils as antimicrobial systems in topical applications: their characterization, current applications, and advances in improved delivery techniques. Molecules. 2020;25(2):334.
24. Ripamonti CI, Cislaghi E, Mariani L, Maniezzo M. Efficacy and safety of medical ozone (O(3)) delivered in oil suspension applications for the treatment of osteonecrosis of the jaw in patients with bone metastases treated with bisphosphonates: preliminary results of a phase I-II study. Oral Oncol. 2011;47(3):185-90. https://doi.org/10.1016/j.oraloncology.2011.01.002

25. Khatri I, Moger G, Kumar NA. Evaluation of effect of topical ozone therapy on salivary Candidal carriage in oral candidiasis. Indian J Dent Res. 2015; 26(2):158-62.

26. Anzolin AP, da Silveira-Kaross NL, Bertol CD. Ozonated oil in wound healing: what has already been proven? Med Gas Res. 2020;10(1):54-9. https://doi.org/10.4103/2045-9912.279985

27. Hernández FM, Ibarra MDM, Valdés MRG. Evolución clínica de las pericoronaritis tratadas con OLEOZON® en urgencias estomatológicas. Rev Méd Electrônica. 2010;32(2):8-16.

28. Patel PV, Kumar V, Kumar S, GD V, Patel A. Therapeutic effect of topical ozonated oil on the epithelial healing of palatal wound sites: a planimetrical and cytological study. J Invest Clin Dent. 2011;2(4):248-58. https://doi.org/10.1111/j.2041-1626.2011.0 0072.x

29. Patel PV, Kumar S, Vidya GD, Patel A, Holmes JC, Kumar V. Cytological assessment of healing palatal donor site wounds and grafted gingival wounds after application of ozonated oil: an eighteen-month randomized controlled clinical trial. Acta Cytol. 2012;56(3):277-84.

30. Huang J, Huang J, Xiang Y, Gao L, Pan Y, Lu J. Topical ozone therapy: An innovative solution to patients with herpes zoster. Zhong Nan Da Xue Xue Bao Yi Xue Ban. 2018;43(2):168-72.

Received on: 26/8/2020
Final version resubmitted on: 17/12/2020
Approved on: 26/1/2021

Assistant editor: Marcelo Sperandio