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REVIEW ARTICLE

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Liquid ozone therapies for the treatment of epithelial wounds: A systematic review and meta-analysis

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Abstract

Ozonated water and ozonated oils are emerging as potential therapies for wound care, but their efficacy has not been appropriately evaluated. The aim of this systematic review and meta-analysis was to evaluate the therapeutic potential of topical ozone in the treatment of mammalian wounds. A structured search of five scientific databases returned a total of 390 unique studies. Of these, 22 studies were included in this review. Four studies provided enough data to be included in a meta-analysis evaluating the time to complete wound healing. All studies were randomised controlled trials of humans or other mammalian animals that reported clinical signs of wound healing. Each study was critically analysed by a six-point assessment of the risk of bias. Wounds treated with topical ozone had a greater reduction in wound size than similar wounds treated with controls or standard of care in all studies. Those treated with ozonated liquids also had a shorter time to wound healing by approximately one week. In conclusion, topical ozone contributed to enhanced wound healing in all studies. While additional human experiments would be helpful to quantify ozone's specific effects on wound healing compared to standard treatment, topical ozone should be considered as part of an overall wound management strategy.

KEYWORDS

dermatologic agent, efficacy, ozone, topical administration, wound healing

Key Messages

- ozonated water and ozonated oils are emerging internationally as potential treatments for epithelial wounds
- in this systematic review designed to evaluate the efficacy of ozonated liquids in wound healing, 22 studies of human and mammalian animal wounds were evaluated and synthesised
- wounds treated with topical ozone solutions were consistently found to have a greater reduction in size and a faster time to complete wound healing compared to controls

1 | INTRODUCTION

The German chemist Christian Friedrich Schönbein is considered the "father of ozone". In 1840, after passing an electrical discharge through water, he noted an unusual odour that he later called "ozone" after the Greek word "ozein" which translates as, "to smell".[1] Soon after its discovery, ozone was recognised as a potent oxidant with significant disinfectant properties. In the late 1800 s, ozone was used to disinfect operating rooms and to treat and purify water.[1] By the turn of the century, Australian-German surgeon Erwin Payr was the first to apply ozone therapy to medical conditions on a larger scale, utilising topical ozone as a disinfectant during World War I to treat purulent wounds.[1] Nikola Tesla (1856-1943) was the first to patent an ozone generator in the United States and founded the Tesla Ozone Co., manufacturer of ozone generators for medical use. Tesla was also the first to ozonate olive oil, which has since been used for various topical medical applications.[2]

Modern ozone applications include medical treatments, water purification, food processing, medical disinfection, and various industrial applications. Many of these applications leverage ozone's ability to oxidise phospholipids, glycoproteins, and lipoproteins resulting in the inactivation of bacteria, viruses, and other microorganisms.[3,4]

Topical forms of ozone therapies, including either ozonated oils or ozonated water, have gained broad interest and growing usage globally as a component of wound treatment strategies. Different manufacturing techniques produce ozonated oils and ozonated water.[5] While ozonated oils result in a more stable compound, ozonated water is easily produced at the time of use, and its clinical efficacy is achieved rapidly, often within seconds.[5] The potential therapeutic uses of topical ozone include chronic refractory wounds, diabetic foot ulcers, and venous stasis ulcers.[6-10] Although physicians around the world accept topical ozone as a viable treatment option, many still consider topical ozone treatments as "alternative or complementary medicine" despite growing support from the literature.[11]

Acute and chronic wound management places a substantial burden on healthcare systems.[12,13] Conditions such as diabetes, obesity, vascular disease, and the persistent problem of wound infection contribute to clinical, social, and economic challenges. Acute skin lesions that evolve into chronic non-healing wounds result in exponentially higher morbidity, mortality, and cost. Nussbaum and colleagues found that in 2014 alone, chronic wounds affected 15% of Medicare recipients in the United States (8.2 million) and contributed to a total Medicare spending estimate ranging from $28.1 to $96.8 billion.[14] These economic analyses do not account for the loss of productivity and other effects on families and communities. As a result, clinicians and researchers continue to look for ways to favourably impact the healing of wounds to shorten healing times, promote better patient outcomes, and lower overall costs.[15]

Topical ozone is an attractive therapeutic option for treating acute and chronic wounds due to its potent antimicrobial properties, favourable immunologic effects, and improved oxygenation potential while demonstrating a favourable safety profile.[14,16] Several systematic reviews attempt to summarise the therapeutic use of ozone for wounds; however, they include small sample sizes, inconsistent formulations of ozone, or contain high levels of bias.[7-9] There is a need to critically evaluate topical ozone as a treatment option for dermatological wounds. Therefore, we performed a systematic review that seeks to evaluate the therapeutic potential of topical ozone in the treatment of mammalian wounds.

2 | MATERIALS AND METHODS

2.1 | Search strategy

The Cochrane Collaboration's *Handbook for Systematic Reviews of Interventions* was used as a guideline for procedures in this review.[17] To identify studies on the use of topical ozone in wound healing, a comprehensive search strategy was developed and executed in five[5] databases for all years up to August 2021: Medline (Ovid), Embase, Cochrane Library, Web of Science Core Collection, and Google Scholar. The search consisted of keywords and controlled vocabulary used in the title and the abstract. The terms used were associated with ozone in its topical form of oil, water, aqueous, or liquid, combined with terms related to skin injuries and wounds. Limits were added to the searches to retrieve English-language and randomised controlled trial studies. In addition, we conducted analogous searches in Google Scholar to discover additional relevant grey literature. Finally, the results from all databases used were aggregated in Endnote and de-duplicated using the Covidence review manager software (Covidence, RRID:SCR_016484) for further screening. All searches in this study were developed and executed by a medical librarian (Mirian Ramirez), and the full search strategies can be found in the Appendix A.

2.2 | Inclusion and exclusion criteria

Randomised controlled trials involving the use of topical ozonated water or oil on mammalian tissue were included. Additionally, we required that articles report

clinical markers of wound healing, including wound size, time to complete healing, number of wounds fully healed in a specific time interval, rate of healing, rate of infection, amputation rate, or grossly observed granulation tissue formation. Those that reported microscopic or histological markers alone were not included. We were most interested in comparing topical ozone treatment to the standard of care for wound management defined by the authors. We did not discriminate based on the location or time frame of the study; however, studies were required to be in English for evaluation. Pain, inflammation, and other subjective or indirect markers of wound healing were not considered.

2.3 | Selection process

The four authors involved in screening articles (Daniel J. Romary, Sarah A. Landsberger, Brian R. Leon, and K. Nicole Bradner) participated in two rounds of training with a sample of 20 articles in each round to attain high inter-rater reliability prior to beginning the article screening process. To assess agreement amongst the four raters, interclass correlations (ICC) were calculated using SPSS statistical package (IBM SPSS Statistics v. 27, RRID: SCR_019096, 2020). The interclass correlation coefficient using a two-way mixed-effects model with an absolute agreement based on average measures indicated excellent inter-rater reliability, ICC = 0.92 (95% CI: 0.85-0.97).

Using the inclusion/exclusion criteria in Covidence, the authors first screened study titles and abstracts and then evaluated full-text studies for inclusion. Two authors reviewed each study at all stages, with a third resolving any disputes.

2.4 | Data extraction & risk of bias assessment

The authors collectively extracted basic information and result from the studies. Additionally, the authors evaluated the risk of bias relative to desired outcomes using the Cochrane Collaboration's risk of bias tool, assigning low, high, or unclear risk for the following categories: random sequence generation, allocation concealment, blinding of participants and personnel, blinding of outcome assessment, incomplete outcome data, selective reporting, and other bias.[18] Two authors independently assessed the risk of bias for each article, employing group consensus to resolve any disputes. For consistency, any study design that had multiple groups on the same person/animal (ie, within-subject trial), was given a "low risk" rating in the random sequence generation category given that auto-controlling is an alternative to treatment randomization in minimising selection bias.

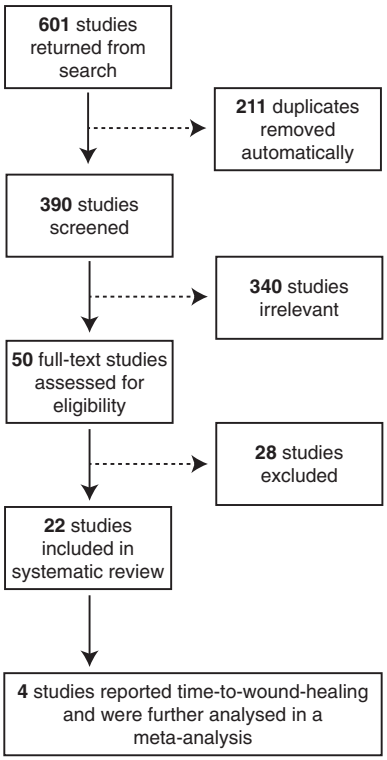

FIGURE 1 PRISMA diagram for study selection

2.5 | Statistical methods

A meta-analysis was performed on all studies that reported time to wound healing. The analysis was completed in Stata (Stata 17, StataCorp) using a random-effects model with empirical Bayesian methods. Given that all studies reported the time in days, raw (unstandardized) mean difference was used to calculate effect sizes. A funnel plot was also produced with this analysis to evaluate publication bias. Other outcome metrics, including wound size, were not evaluated in meta-analyses due to the heterogeneity of study designs, number of experimental groups, outcome metrics, and reporting timelines.

3 | RESULTS

3.1 | Literature search

The literature search returned 390 studies for screening (Figure 1). Following title and abstract screening, 340 studies were deemed irrelevant, as they did not meet pre-determined study criteria. Of the 50 articles that were

TABLE 1 Characteristics of 22 studies included in the systematic review

Author(s)	Country	Subjects	Type of ozone	Experimental Groups	Summary of Methods	Outcome variables
Canpolat 2021[35]	Turkey	21 rats (4 wounds per rat)	Ozonated oils. (Sesame, Nigella sativa, Hypericum perforatum)	Within-subject trials: 1. Ozonated sesame oil 2. Ozonated nigella oil 3. Ozonated hypericum perforatum oil 4. Control--no treatment	Ozonated oil applied 2x/day for 21 days. Rats divided into 3 groups euthanized at 7, 14, and 21 days respectively.	Time to complete wound healing
de Araujo 2017[19]	Brazil	5 horses (4 wounds per horse)	Ozonated andiroba oil and ozonated saline solution	Within-subject trials: 1. Ozonated andiroba 2. Ozonated saline solution 3. Pure andiroba oil 4. Saline solution	Wounds created and differential treatments applied. Horses' wounds treated daily and evaluated twice a day until complete healing of wounds occurred	Wound size, time to complete wound healing
Dharmavaram 2015[29]	India	30 Patients of both genders >16 yrs with 1 to 5 ulcers, with duration of less than 48 h	Ozonated oil	1. Ozonated oil 2. Sesame oil 3. Distilled water	Wounds treated 4x/day for 5 days.	Wound size
di Filippo 2020[20]	Brazil	8 horses with 8 wounds each (2 wounds to each leg); 4 horses per group	Ozonated sunflower seed oil	1. Wounds on two limbs treated with saline. 2. Wound on one limb treated with pure sunflower oil 3. Wound on one limb treated with ozonated sunflower seed oil	Treatments applied daily for 21 days.	Wound size; time to wound healing
Elshenawie 2013[34]	Egypt	30 (15 per group) diabetic patients (aged 21-60), any grade of foot ulcer	Ozonated olive oil	1. Ozonated olive oil dressings 2. Control dressings	Ozone group--ozone dressings daily for 5 weeks; control group--control dressings for 5 weeks	Time to complete wound healing; signs of wound healing
Filippi 2001[30]	Germany	30 patients (ages 18-35) with 3 wounds of the oral mucosa	Ozonated water (11-12 μg/mL)	1. Ozonated water 2. Water 3. No treatment	Treatments administered for 2, 4, and 7 days (10 per group).	Wound size
Hu 2019[31]	India	136 patients with diabetic foot ulcers (68 per group)	Ozonated water	1. Vacuum-assisted closure (VAC) and ozone water flushing 2. VAC only	Wounds assessed at 1, 2, 3, and 1 month during the treatment	Time to wound healing; wound size

TABLE 1 (Continued)

Author(s)	Country	Subjects	Type of ozone	Experimental Groups	Summary of Methods	Outcome variables
Kim 2009[21]	Korea	8 guinea pigs (4 wound sites per animal = 32 wounds)	Ozonated olive oil	1. Ozonated oil (on 2 Wounds per animal, n = 16) 2. Pure olive oil (n = 8) 3. No treatment (n = 8)	Treatment applied to each wound daily followed by dressing. Wounds measured over 11 days.	Wound size
Kim 2020[38]	Korea	80 patients (40 per group) >18 and < 65	Ozonated water concentration (1 ppm, 2 and 4 ppm)	1. Sitz bath with ozonated water 2. Sitz bath with ordinary tap water.	Sitz bath daily	Time to wound healing
Menendez 2002[39]	Cuba	200 patients <15 yrs with tinea pedis (100 per group)	Oleozon oil	1. Oleozon topical 2. Ketoconazole cream 2% (Nizoral)	treatment 2x/day ×6 weeks	Time to complete wound healing
Pai 2014[22]	India	24 rats (6 per group)	Ozonated sesame oil	1. High dose ozonated sesame oil 2. Low dose ozonated sesame oil 3. Framycetin 1% w/w cream 4. Sesame oil	Treatments applied 1x/day for 11 consecutive days.	Wound size
Patel 2011[32]	India	22 patients of both genders (aged 20-40) with palatal wounds	Ozonated olive oil (14 μg/mL)	1. Ozonated olive oil 2. Pure olive oil	Standard size palatal graft/wound created. Topical solution applied to wound daily for one week. Wound size assessed for 28 days.	Wound size
RizaoĞLu 2018[23]	Turkey	21 rats (4 wounds per rat)	Ozonated sesame oil, ozonated hypericum perforatum oil, and ozonated nigella	1. Untreated group 2. Ozonated sesame oil 3. Ozonated hypericum perforatum oil 4. Ozonated nigella sativa oil	All oils applied 2x/day for 7, 14, or 21 days (7 rats per group)	Wound size
Roshan 2018[36]	Syria	36 rats (6 per group)	Ozonated sunflower oil and ozonated linseed oil	1. Sunflower oil 2. Linseed oil 3. Ozonated sunflower oil 4. Ozonated linseed oil 5. Positive control (Mebo ointment) 6. Negative control (no treatment)	Oils applied topically ×2 daily	Time to complete wound healing
Sanguanini 2020[24]	Brazil	48 rats (12 per group)	Ozonated water and ozonated sunflower oil	1. Ozonated water (4.5 μg/mL) 2. Ozonated sunflower oil (418 mEq/kg; 50 uL) 3. Normal saline	Treatments were performed daily. 3 animals each	Wound size

(Continues)

TABLE 1 (Continued)

Author(s)	Country	Subjects	Type of ozone	Experimental Groups	Summary of Methods	Outcome variables
				4. 0.2% allantoin cream	euthanized and assessed on days 3, 8, 15, and 22.	
Sghaireen 2020[40]	Saudi Arabia	28 patients with a wound on each side of mouth, ages 18 to 75 years	Ozonated water (0.05 ppm)	Within-subjects trials 1. Ozone water irrigation 2. Control (saline irrigation).	Both treatments applied and repeated after 3 days.	Signs of wound healing
Solovastru 2015[33]	Spain	29 patients >18 yrs with chronic venous leg ulcers <2 years duration (15 in study group and 14 in control)	Ozonated oil	1. A-bisabolol spray + ozonated oil 2. Control cream (vitamin A & E, talc and zinc oxide)	Daily application for 30 days.	Wound size; time to complete wound healing
Vahlepi 2020[25]	Indonesia	50 rats (10 groups/ 5 per group	Ozonated aloe vera oil (concentrations of ozone of 600, 1200, 1800 mg/mL)	1. Control (aloe vera only) 2. Control (gentamicin) 3 to 5. Ozonated aloe vera oil with concentrations of ozone of 600, 1200, 1800 mg/mL 6. Aloe vera only 7. Gentamicin 8 to 10. Ozonated aloe vera at concentrations of 6 001 200, and 1800 mg/mL	5 groups (1 to 5) treated for 3 days then assessed; 5 groups (6 to 10) treated for 7 days then assessed.	Wound size
Valacchi 2011[26]	South Korea & Italy	36 mice (9 mice in ozone experiment)	Ozonated sesame oil (45 mg/L)	Within-subject trials 1. Pure sesame oil 2. Ozonated oil, low peroxide value 3. Ozonated oil, medium peroxide value 4. Ozonated oil, high peroxide value 5. No treatment 6. No treatment	Treatments applied topically 2x/day up to 14 days.	Wound size
Xiao 2017[27]	China	24 mice (12 per group)	Ozone oil (400 ɥl) contains 99% ozonide, superoxide, and Camellia oil	1. Control group 2. Ozone treated (ozone applied with a swab once every 2 days for 12 days)	Study group treated for 12 days once every 2 days. Area of the wounds measured every 2 days.	Wound size
Ye 2016[37]	China	30 rats (10 per group)	Ozonated water	1. Control (saline) 2. Ozonated water 3. Chlorhexidine	Wounds treated daily for 21 days.	Time to wound healing

TABLE 1 (Continued)

Author(s)	Country	Subjects	Type of ozone	Experimental Groups	Summary of Methods	Outcome variables
Yuniati 2021[28]	Indonesia	60 rats with induced diabetes (10 per group)	Ozonated coconut oil (3360 ppm)	1. Negative control (no diabetes; gentamycin) 2. Positive control (diabetes; gentamycin) 3. P1 (virgin coconut oil) 4. P2 (90-min ozonated coconut oil) 5. P3 (7-hrs ozonated virgin coconut oil) 6. P4 (14-hrs ozonated virgin coconut oil)	Wounds treated 1x daily for 14 days. Wounds assessed on day 1, 3, 5, 7, and 14.	Wound size

subject to full-text review, an additional 28 studies were excluded because of incorrect study design, ozone type, or outcome variables. As a result, 22 studies published between 2002 and 2021 were included in this systematic review.

3.2 | Study characteristics

The study details and outcomes of the 22 papers included in this systematic review are listed in Table 1. The 13 animal studies involved 371 animals [rats (n = 290), mice (n = 60), guinea pigs (n = 8), horses (n = 13)] and the 9 human studies involved a total of 585 participants. In the treatment condition, 5 studies used ozonated water, 15 used ozonated oils and 2 used both ozonated oil and water. The ozonated oils studied included aloe vera oil (n = 1), sesame oil (n = 4), camellia oil (n = 1), sunflower oil (n = 3), coconut oil (n = 1), olive oil (n = 3), hypericum perforatum oil (n = 2), nigella sativa oil (n = 2), andiroba oil (n = 1), linseed oil (n = 1) and unspecified oil (n = 3). Comparison groups included pure oil (n = 13), no treatment or saline only (n = 14), and/or standard treatment (n = 12). Standard treatment approaches included chlorhexidine, a-bisbolol spray, gentamicin, ketoconazole cream 2%, Mebo ointment, allatoin cream 0.2%, framycetin cream 1%, sitz baths, or vacuum-assisted closure. Regarding outcomes, 15 studies reported data on wound size, 10 studies reported on time to complete wound healing and 2 studies reported other clinical signs of wound healing.

3.3 | Impact of ozonated oils/water on wound size

3.3.1 | Animal studies

Ten studies examined the impact of ozonated oils/water on wound size. (Table 2) Three studies were within-subject trials of ozonated oils/water compared to control or standard treatment[19-21] and seven studies were between-subjects trials of ozonated oils and/or ozonated water compared to control or standard treatments.[22-28]

Each of the three within-subject design studies found that wounds treated with ozonated oils were significantly smaller than wounds treated with pure oil and/or saline ($P < 0.05$). In a study of ozonated andiroba oil and ozonated saline used to treat wounds on horses, wounds treated with ozonated andiroba oil were significantly smaller than wounds treated with ozonated saline, andiroba oil, and saline solution by day 14 ($P < 0.05$).[19] In a similar study on horses, wounds treated with ozonated

TABLE 2 Results of studies evaluating wound size

Author	Day of study	Wound size measurement
Animal studies		
de Araujo 2017[19]	14	Ozonated andiroba oil < ozonated saline, andiroba oil, saline*
di Filippo 2020[20]	14 & 21	Ozonated sunflower oil < pure oil*
Kim 2009[21]	5 & 7	Ozone < pure olive oil group*
Pai 2014[22]	12	High dose ozonated oil < sesame oil*
RizaoĞlu 2018[23]	14	Ozone oils < untreated*
Sanguanini 2020[24]	3 & 8	Ozonated water < ozonated sunflower oil, saline, 0.2% allantoin cream*
Vahlepi 2020[25]	3	Ozonated aloe vera oil 1200 and 1800 mg/mL < aloe vera only*
	7	Ozonated aloe vera [AV] (1800 mg/mL) < AV oil and gentamicin*; ozonated AV (1200 mg/mL) < gentamicin*; ozonated AV 600 mg/mL) < gentamicin*
Valacchi 2011[26]	6	Medium peroxide value ozonated oil < no treatment***
	14	All ozone groups < no treatment*
Xiao 2017[27]	3, 5, & 7	Ozonated oil < control*
	9	Wounds in both groups completely healed
Yuniati 2021[28]	14	14-h ozonated virgin coconut oil (OVCO) < 7-hrs OVCO <90-min OVCO < virgin coconut oil < neg control < pos control (No statistical testing)
Human studies		
Dharmavaram 2015[29]	2	Ozonated oil < sesame oil, distilled water*
	4	Ozonated oil < sesame oil, distilled water*
	6	Ozonated oil, sesame oil < distilled water*
Filippi 2001[30]	2	Ozonated water < water, no treatment*
	4 & 7	Ozonated water = water, no treatment (*ns*)
Hu 2019[31]	7, 14, 21	Ozone + VAC < VAC*
	28	Ozone + VAC = VAC (*ns*)
Patel 2011[32]	5, 7, 14, 21, & 28	Ozonated olive oil < pure olive oil*
Solovastru 2015[33]	7, 14, 30	Ozone + a-bisabolol < control*

Note: *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.
Abbreviation: ns, no significant difference.

sunflower were significantly smaller than wounds treated with pure oil at days 14, and 21 ($P < 0.05$).[20] Moreover, wound contracture was greater in the ozonated sunflower oil group than the pure oil and saline groups on days 7, 14, and 21 ($P < 0.05$).[20] In a study of 32 wounds created on guinea pigs, wound size was significantly smaller on days 5 and 7 in the wounds treated with ozonated olive oil than in wounds treated with pure olive oil ($P < 0.05$).[21]

Similar results to those listed above were found in the seven between-subjects design animal trials of ozonated oils/water on wound size. Three studies assessed the efficacy of ozonated sunflower oil on reducing wound size and found the smaller size of wounds compared to controls.[22,23,26] Three studies assessing the impact of ozone concentration (low, medium, high) on wound size found smaller wound sizes in ozone-treated groups but differed on which ozone concentrations were most effective.[22,25,26] In one study, high dose ozonated sesame oil was associated with a smaller wound size on day 12 as compared to pure oil ($P < 0.05$).[22] Other authors found medium concentrations of ozonated sesame oil were associated with significantly reduced wound size on day 6 as compared to no treatment ($P < 0.001$) and wounds treated with any concentration of ozonated sesame oil were smaller on day 14 than those receiving no treatment ($P < 0.001$).[26] A third study found smaller wound sizes with medium and high concentrations of ozonated aloe vera oil on day 3 and smaller wound sizes with low, medium, and high concentrations on day 7 as compared to controls ($P < 0.05$).[25] Findings from four remaining studies comparing the size of wounds treated with various ozonated oils to control

and standard treatment were somewhat mixed but generally trended towards ozonated liquids producing smaller wound size than comparators.[22,24,25,28] A 2014 study found significantly smaller wounds in rats treated with ozonated sesame oil as compared to pure oil by day 12 ($P < 0.05$) but no difference was found in wound size between rats treated with ozonated oil and those treated with framycetin.[22] In a 2020 study utilising rats, wounds treated with ozonated water or saline were found to be significantly smaller on day 3 and 8 than wounds treated with ozonated sunflower oil, or 0.2% allantoin cream ($P < 0.05$).[24] A 2017 study of wounds on 24 rats found a smaller wound size ratio on days 3, 5, and 7 in rats receiving ozonated oil versus control.[27] Lastly, in 60 rats with induced diabetes, wounds treated over a 14-day period with ozonated coconut oil were significantly smaller than wounds treated with pure coconut oil or gentamycin ointment.[28]

3.3.2 | Human studies

Five studies involving human patients that examined wound size post-intervention were included in this review.[29-33] (Table 2) All five studies reported reduced wound size in the ozonated treatment groups as compared to controls or standard care. Two studies examined the impact of ozonated oil or water on oral wounds.[30,32] A 2001 study compared ozonated water, pure water, or no treatment on three wounds in the oral mucosa in each of 30 patients and found smaller wound size on day 2 in the wounds treated with ozonated water ($P < 0.05$).[34] However, no significant difference in wound size was found between treatments on days 3 to 7. In a similar study assessing the impact of ozonated olive oil on wound size in palatal wounds, wound size on days 5, 7, 14, 21 and 28 was consistently smaller in wounds treated with the ozonated olive oil versus pure olive oil ($P < 0.05$).[32] The remaining three studies examined the impact of ozonated water or oil on skin ulcers.[29,31,33] A 2015 study found that ulcers treated with ozonated oil had smaller wound sizes on days 2, 4, and 6 than wounds treated with pure sesame oil or distilled water ($P < 0.05$).[29] A 2017 study found ozonated oil plus a-bisabolol spray applied to chronic venous leg ulcers produced smaller wound size at days 7, 14, and 30 compared to control.[33] Lastly, a 2019 study examined 136 patients with diabetic foot ulcers who received either vacuum-assisted closure (VAC) plus ozonated water or VAC only.[31] The results demonstrated a significant reduction in wound size for the VAC plus ozonated water group at weeks 1, 2, and 3 compared to VAC only ($P < 0.05$).

3.4 | Impact of ozonated oils/water on time to complete wound healing

3.4.1 | Animal studies

Five studies investigated the effect of ozonated oils on the time to complete wound healing in animals.[19,20,35-37] (Table 3) Four of the five studies found a faster time to complete wound healing[20,35-37] and one study[19] found no difference across treatment conditions. A within-subject trial on wounds in rats found that wounds treated with ozonated sesame, nigella sativa or hypericum perforatum oils were fully healed on day 18 whereas wounds with no

TABLE 3 Results of studies evaluating time to wound healing

Author	Time to wound healing
Animal studies	
de Araujo 2017[19]	Ozonated andiroba oil = ozonated saline solution = pure andiroba oil = saline solution (*ns*, range 30-32 days). All groups had significant wound healing by day 21***
di Filippo 2020[20]	Ozonated sunflower oil (25 days) < pure oil (27 days) < saline (30 days)
Canpolat 2021[35]	Ozone (18 days) < control (19 days)
Roshan 2018[36]	Ozonated oils, linseed oil, positive control < negative control*; ozonated oils < positive control* (group mean range 8-15 days)
Ye 2016[37]	Ozone (21 days) < chlorhexidine (26 days) < control (38 days)*
Human studies	
Hu 2019[31]	Ozone + VAC (13 days) < VAC (26 days)***
Patel 2011[32]	Ozone group healed by 28 days; control group still had wounds present at 28 days
Solovastru 2015[33]	At 30 days: 25% of patients in the ozone + a-bisabolol group completely healed versus 0% in control patients
Elshenawie 2013[34]	Complete healing of wound at week 5: ozone (60%) versus control (0%).
Kim 2020[38]	Ozone (19 days) < tap water (27 days) ***
Menendez 2002[39]	After 6 weeks of treatment: ozone = ketoconazole (*ns*)

Note: *$P \leq 0.05$; **$P \leq 0.01$; ***$P \leq 0.001$; days reported are group means where applicable.

Abbreviation: ns, no significant difference.

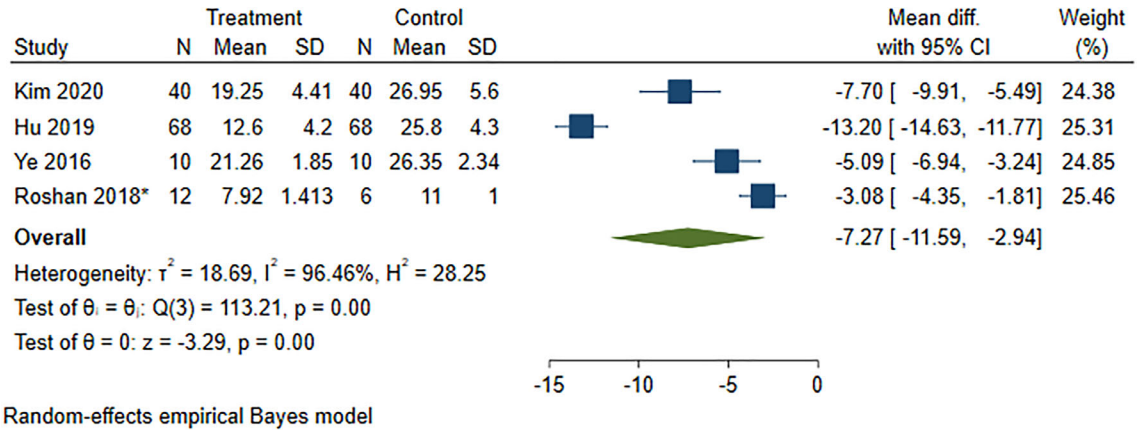

FIGURE 2 Forest plot describing meta-analysis of time-to-wound-healing

treatment were healed on day 19.[35] A 2018 study reported that wounds on rats treated with ozonated linseed oil were completely healed in 9 days, wounds treated with ozonated sunflower oil were treated in 10 days, and wounds treated with pure sunflower oil, pure linseed oil, Mebo ointment, or receiving no treatment were healed in 14 days.[36] A third study on rats conducted in 2016 demonstrated that wounds treated with ozonated water had a significantly shorter mean healing time of 21.26 days than wounds treated with chlorhexidine (26.35 days) or control (38.24 days) ($P < 0.05$).[37] Two studies on horses found inconsistent results.[19,20] A study of eight horses, each with eight wounds, found wounds treated with ozonated sunflower oil reached complete healing faster than wounds treated with pure oil or saline.[20] A similar study of five horses, each with four wounds, found time to complete healing in the wounds treated with ozonated andiroba oil or pure andiroba oil were healed on day 30, wounds treated with ozonated saline were healed on day 32, and wounds in the control group were healed on day 31.[19]

3.4.2 | Human studies

Six studies on patients with skin ulcers (n = 3), post-hemorrhoidectomy wounds (n = 1), palatal wounds (n = 1), or tinea pedis wounds (n = 1) found greater complete healing of wounds that received treatment with ozonated oils or water.[31-34,38,39] (Table 3) In a study of 29 patients with chronic venous leg ulcers, 25% of the patients in the ozone plus a-bisabolol group displayed complete healing at the end of treatment (day 30) as compared to 0% patients in the control group.[33] In a study of oral wounds, on the final study day (day 28), the wounds in the ozonated group were completely healed, whereas the control group still had wounds of a mean size of 2.05mm^2.[32] In patients post-hemorrhoidectomy, wounds treated with ozonated water sitz

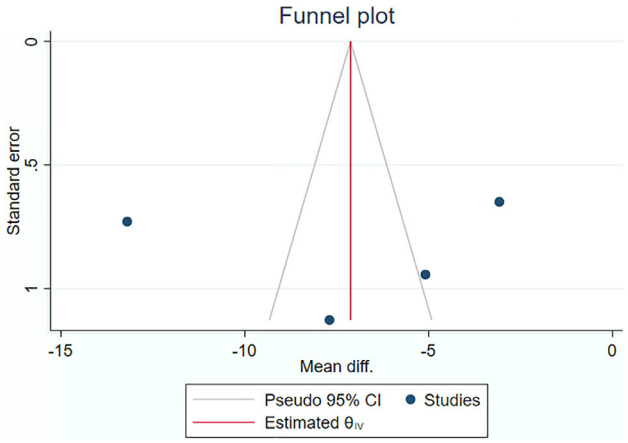

FIGURE 3 Funnel plot evaluating publication bias of studies included in meta-analysis

baths were completely healed in a mean of 2.75 weeks compared to 3.85 weeks for wounds treated with sitz baths using ordinary water ($P < 0.001$).[38] In a study of diabetic foot ulcers treated with VAC and ozonated water, wounds were healed after an average of 12.6 days with ozone + VAC treatment compared to 25.8 days in the VAC only group ($P < 0.001$).[31] In another study of diabetic foot ulcers, 30 patients were treated with either ozonated olive oil or control dressings for five weeks.[34] At the end of treatment, 60% of ulcers treated with ozonated olive oil were completely healed as compared to 0% of ulcers treated with control wound dressings. Moreover, 40% of ulcers were partially healed at the end of treatment and 66.7% of ulcers treated with control wound dressings were partially healed. In contrast to the findings described above, a study on patients with foot wounds from tinea pedis found that more patients in the ketoconazole cream 2% group had complete healing as compared to the ozonated oil group (81 versus 75, respectively) although this difference was not statistically significant.[39]

3.5 | Meta-analysis of time to complete wound healing studies

Four studies reported time to wound healing in human patients with sufficient data to be included in a meta-analysis. The study by Roshan et al included two experimental groups, ozonated linseed and ozonated sunflower oils, that produced statistically indifferent results within their study. Therefore, these two groups were mathematically combined (averaged means, pooled standard deviations) as a single treatment group in the meta-analysis. The control groups used in the analysis were the standard of care identified by each study.

The forest plot (Figure 2) demonstrates that ozonated liquids significantly improved the time to wound healing by approximately one week, although considerable heterogeneity should be noted. While the funnel plot for publication bias (Figure 3) is difficult to interpret with only four studies, there is symmetry in the mean.

TABLE 4 Results of studies evaluating other clinical signs of wound healing

Author	Study findings
Elshenawie 2013[34]	Abnormal findings of wound healing at weeks 1, 3, and 5: ozone < control*
Sghaireen 2020[40]	Wound healing parameter scores (re-epithelization, haemostasis, and inflammation: ozone > control on day 1*; ozone = control on day 5 (*ns*)

Note: *$P < 0.05$.
Abbreviation: ns, no significant difference.

3.6 | Other clinical signs of wound healing

3.6.1 | Human studies

Two studies on human subjects reported clinical signs of wound healing by treatment type.[34,40] (Table 4)

	Random Sequence Generation	Allocation Concealment	Participant & Personnel Blinding	Outcome Assessment Blinding	Incomplete Outcome Data	Selective Reporting	Other Bias
Canpolat et al., 2021	L	-	L	-	L	L	L
de Araujo et al., 2017	L	-	L	L	L	L	-
Dharmavaram et al., 2015	-	-	L	-	L	L	L
di Filippo et al., 2020	-	-	H	L	L	L	L
Elshenawie et al., 2013	H	H	H	H	L	L	-
Filippi et al., 2001	L	-	L	L	L	L	-
Hu et al., 2019	-	-	H	L	L	L	L
Kim et al., 2020	L	-	-	-	L	L	-
Kim et al., 2009	-	-	-	H	L	L	-
Menendez et al., 2001	L	L	-	-	L	L	H
Pai et al., 2014	-	-	-	-	L	L	H
Patel et al., 2011	-	L	L	L	H	L	-
Rizaoglu et al., 2018	L	-	L	H	L	L	H
Roshan et al., 2018	-	-	-	-	L	L	-
Sanguanini et al., 2020	-	-	-	-	L	L	L
Sghaireen et al., 2020	L	-	L	-	L	L	L
Solovastru et al., 2015	-	-	L	-	L	L	-
Vahlepi et al., 2020	-	-	-	-	L	L	L
Valacchi et al., 2011	L	-	L	L	L	L	-
Xiao et al., 2017	-	-	-	-	L	L	-
Ye et al., 2016	-	-	L	L	L	L	-
Yuniati et al., 2021	L	-	-	-	L	H	H

FIGURE 4 Risk of bias, assessed as low (L), high (H), or unclear (–)

Evaluating the efficacy of ozonated olive oil to treat diabetic foot ulcers, significantly fewer abnormal findings of ulcer wound healing (ie, absence of healing epithelial edges, unchanged areas of wound, moist granulation tissue, clinical signs of wound infection/changes of colour or amount of wound exudates) were found in the ozonated oil group than the control group at week 1 ($P < 0.001$), week 3 ($P < 0.006$) and week 5 ($P < 0.001$).[34] In a study of oral wounds post dental implant surgery, compared to control wounds, wounds irrigated with ozonated water evidenced greater scores on clinical signs measuring re-epithelization, decreased haemostasis, reduced inflammation, and overall early wound healing at days 1 and 5 ($P < 0.05$).[40]

3.7 | Assessment of the risk of bias

Figure 4 summarises the results of the risk of bias assessment using the Cochrane Collaboration's Risk of Bias Tool.[18] Nine of the 22 studies reviewed had at least one bias category rated as having a high bias risk.[20,22,23,28,31,32,34,38,39] Three studies had high bias in blinding of participants and personnel, three had high risk in the blinding of outcome assessors, and four had other types of risk for high bias. The remaining 13 studies had a mixture of low risk and unclear risk ratings.[19,21,24-27,29,30,33,35-37,40]

4 | DISCUSSION

This systematic review aimed to examine the impact of ozonated oils/water on wound healing. Overall, the results demonstrated that ozonated liquids enhance wound healing in mammals. Specifically, benefits were observed in reduced time to complete wound healing, a greater decrease in wound size, and improved clinical signs of wound healing.

In both animal and human trials, wounds treated with ozonated liquids were smaller than wounds treated with control or standard treatments at the study endpoints. In addition, all studies reported wound contracture occurring sooner with the application of ozonated liquids. Moreover, the findings of the meta-analysis indicated that the clinical use of ozonated liquids achieved complete wound healing faster than control. In sum, these findings support the potential clinical use of ozonated liquids to increase healing and wound closure.

Our literature review findings were generally consistent with and extended the results of the aforementioned, smaller systematic reviews that provided initial support for the benefits of ozone for wound healing. A 2018 review of nine studies evaluated the potential benefits and harms of diverse forms of ozone therapy (eg, gaseous, oils, water) as a treatment intervention for a heterogeneous population of chronic wounds in humans (eg, war wounds, burns, non-healing diabetic foot ulcers, venous ulcers, and arterial ulcers).[7] This review found a significant improvement in wound closure and results that favoured ozone application over standard treatments with respect to time to healing but could not derive conclusive evidence of ozone therapy being superior. A second 2018 published review including only three controlled trials evaluated topical ozone therapy's effectiveness as measured by healing rate, healing time reduction, and decreased bacterial load in venous leg ulcers.[8] The review demonstrated that ozone therapy is more effective than conventional therapy in each of these predetermined measures. Lastly, a 2015 systematic review of three RCTs evaluating the use of ozone in treating diabetic foot ulcers produced limited findings due to concerns about study bias.[9]

The present systematic review demonstrates favourable outcomes on wound healing in ozonated treatment groups and a favourable safety profile, consistent with other work.[40] Furthermore, this review focuses on topical formulations of ozone rather than gaseous forms, reducing the potential for pulmonary toxicity observed in some studies of inhaled ozone.[41] Therefore, based on the potential for positive outcomes with low risk attributed to topical ozone treatment for wound healing, further study is warranted to determine optimal ozone application in wound management.

An additional consideration for aqueous ozone may be in the area of wound irrigation. Wound irrigation utilising normal saline remains a standard component of wound care therapy.[41] The benefits of irrigation include removing surface material and decreasing the bacterial load to promote wound healing. However, there is currently no evidence supporting the addition of additives to irrigation fluid, such as iodine, chlorhexidine, hydrogen peroxide, or sodium hypochlorite.[42,43] Considering the potential benefits outlined in our systematic review, the authors hypothesize that aqueous ozone should be investigated as an adjuvant irrigation solution to existing wound therapy modalities.

This review has several limitations. Only four of the twenty-two studies were able to be quantitatively compared in the meta-analysis due to a lack of sufficient data for analysis. We also included human and animal experiments in this systematic review and the meta-analysis. While this design may contribute to heterogeneity, it was important to have a large enough sample size and incorporate all of the emerging data on topical ozone to represent the literature accurately. Additionally, our bias assessment found low overall risk with a few exceptions.

Finally, the results are limited by the selection of controls. In a presumed effort to reduce confounding variables, many studies compared ozonated and non-ozonated versions of the same solution, even when pure saline or olive oil may not have been appropriate treatments. Additional human trials are needed to compare standard wound healing regimens to topical ozone therapy head-to-head.

The findings from this review should encourage further study of topical ozonated solutions in wound care either as a primary treatment or as a complement to existing therapeutic modalities. Further, as these solutions are used in more extensive studies and gain clinical use worldwide, additional data can be collected to evaluate the overall impact of ozonated liquids on wound care.

5 | CONCLUSION

This systematic review finds that topical ozone solutions improve wound healing in mammals as measured by metrics such as time to complete healing, clinical signs of wound healing, and reduction in wound size. While additional study is needed to quantify the exact amount of improvement, ozonated liquids should be considered as part of an overall wound care strategy.

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CONFLICT OF INTEREST

Daniel J. Romary, Sarah A. Landsberger, K. Nicole Bradner, and Brian R. Leon were compensated through an unrestricted grant from 3Oe Scientific, Inc. BRL also serves on the medical advisory board of and holds equity in 3Oe Scientific, Inc.

DATA AVAILABILITY STATEMENT

Data sharing is not applicable to this article as no new data were created or analyzed in this study.

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APPENDIX: SEARCH STRATEGIES

Appendix: Search Strategies

Database: Ovid MEDLINE(R) and Epub Ahead of Print, In-Process, In-Data-Review & Other Non-Indexed Citations, Daily and Versions(R) <1946 to August 03, 2021>
Search Strategy:
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1 ozone/ and (water/ or exp oils/ or exp "plant oils"/ or exp therapeutics/ or therapy.fs. or 'therapeutic use'.fs.) 2544
2 ozone.mp. and (exp therapeutics/ or therapy.fs. or 'therapeutic use'.fs. or (oil* or water or aqueous or liquid or therap* or topical).ti,ab,oa,kw,kf.) 7952
3 (ozone adj3 (therap* or topical)).nm,ti,ab,oa,kw,kf. 822
4 ((ozone or ozonated or ozoni?ed) adj5 (oil* or water or aqueous or liquid)).ti,ab,oa,kw,kf. 1484
5 1 or 2 or 3 or 4 8148
6 exp "wounds and injuries"/ 941834
7 exp "wound healing"/ 132624
8 exp "wound infection"/ 48881
9 exp ulcer/ 14317
10 exp "skin ulcer"/ 47039
11 exp "foot ulcer"/ 11082
12 gangrene/ 8240
13 pressure ulcer/ 12782
14 exp "skin diseases, infectious"/ 120976
15 lacerations/ 3538
16 diabetic foot/ 9650
17 wound*.ti,ab,oa,kw,kf. 237036
18 ulcer*.ti,ab,oa,kw,kf. 221491
19 gangren*.ti,ab,oa,kw,kf. 18671
20 (diabet* adj3 (foot or feet)).ti,ab,oa,kw,kf. 10876
21 (((bed or pressure*) adj2 sore*) or (bedsore* or decubitus)).mp. 9248
22 ((skin or cutaneous or dermal) adj5 (diseas* or infectio* or abrasion* or avulsion* or bruise* or burn* or injur* or lesion* or scrape* or scratch*)).mp. 195721
23 laceration*.ti,ab,oa,kw,kf. 12714
24 or/6-23 1671561
25 ("clinical trial" or "clinical trial, phase i" or "clinical trial, phase ii" or clinical trial, phase iii or clinical trial, phase iv or controlled clinical trial or "multicenter study" or "randomized controlled trial").pt. or double-blind method/ or exp clinical trials as topic/ or controlled clinical trials as topic/ or exp randomized controlled trials/ or exp randomized controlled trials as topic/ or early termination of clinical trials as topic/ or multicenter studies as topic/ or ((randomi?ed adj10 trial*) or (controlled adj3 trial*) or (clinical adj2 trial*) or (treatment adj2 trial*) or ((single or doubl* or tripl* or treb*) and (blind* or mask*))).ti,ab,oa,kw,kf. or ("4 arm" or "four arm").ti,ab,oa,kw,kf. or (placebo or group* or "control group" or randomly or (random* adj2 alloc*)).ab. 5161367
26 5 and 24 and 25132
27 26 and english.la. 112

Database:Cochrane Library

Search Strategy:

ID Search Hits
#1 [mh ^ozone] AND ([mh ^water] OR [mh oils] OR [mh "plant oils"] OR [mh therapeutics] OR [mh /TH] or [mh /TU]) 189
#2 ozone:ti,ab,kw AND ([mh therapeutics] OR [mh /TH] OR [mh /TU]) 214
#3 (ozone NEAR/3 (therap* OR topical)):ti,ab,kw 295
#4 ((ozone OR ozonated OR ozoni?ed) NEAR/5 (oil* or water or aqueous or liquid)):ti,ab,kw 115
#5 #1 OR #2 OR #3 OR #4 468
#6 [mh "wounds and injuries"] 26922
#7 [mh "wound healing"] 6073
#8 [mh "wound infection"] 3698
#9 [mh ulcer] 1413
#10 [mh "skin ulcer"] 3000
#11 [mh "foot ulcer"] 1134
#12 [mh ^gangrene] 74
#13 [mh ^"pressure ulcer"] 775
#14 [mh "skin diseases, infectious"] 3452
#15 [mh ^lacerations] 217
#16 [mh ^"diabetic foot"] 1045
#17 wound*:ti,ab,kw 31932
#18 ulcer*:ti,ab,kw 28008
#19 gangren*:ti,ab,kw 770
#20 (diabet* NEAR/3 (foot OR feet)):ti,ab,kw 2933
#21 (((bed OR pressure*) NEAR/2 sore*) OR (bedsore* OR decubitus)):ti,ab,kw 1895
#22 ((skin OR cutaneous OR dermal) NEAR/5 (diseas* OR infectio* OR abrasion* OR avulsion* OR bruise* OR burn* OR injur* OR lesion* OR scrape* OR scratch*)):ti,ab,kw 13000
#23 (laceration*):ti,ab,kw 1302
#24 #6 OR #7 OR #8 OR #9 OR #10 OR #11 OR #12 OR #13 OR #14 OR #15 OR #16 OR #17 OR #18 OR #19 OR #21 OR #22 OR #23 92454
#25 ("clinical trial" or "clinical trial, phase i" or "clinical trial, phase ii" or clinical trial, phase iii or clinical trial, phase iv or controlled clinical trial or "multicenter study" or "randomized controlled trial"):pt or [mh ^"double-blind method"] OR [mh "clinical trials as topic"] OR [mh ^"controlled clinical trials as topic"] OR [mh "randomized controlled trials"] OR [mh "randomized controlled trials as topic"] OR [mh ^"early termination of clinical trials as topic"] OR [mh ^"multicenter studies as topic"] or ((randomi?ed NEAR/10 trial*) or (controlled adj3 trial*) or (clinical NEAR/2 trial*) or (treatment NEAR/2 trial*) or ((single or doubl* or tripl* or treb*) and (blind* or mask*))):ti,ab,kw or ("4 arm" or "four arm"):ti,ab,kw or (placebo or group* or "control group" or randomly or (random* NEAR/2 alloc*)):ab 1404833
#26 #5 and #24 and #25 52

Database:EMBASE(Elsevier)
Search Strategy:

No.	Query	Results	Date
#29.	#28 AND [english]/lim	296	4 Aug 2021
#28.	#5 AND #26 AND #27	317	4 Aug 2021
#27.	('double blind procedure'/de OR 'clinical trial (topic)'/de OR 'controlled clinical trial (topic)'/de OR 'randomized controlled trial'/exp OR 'randomized controlled trial (topic)'/exp OR 'early termination of clinical trial'/de OR multicenter) AND study AND topic OR ((randomi$e NEAR/10 trial*):ti,ab,kw) OR ((controlled NEAR/3 trial*):ti,ab,kw) OR ((clinical NEAR/2 trial*):ti,ab,kw) OR ((treatment NEAR/2 trial*):ti,ab,kw) OR ((single:ti,ab,kw OR doubl*:ti,ab,kw OR tripl*:ti,ab,kw OR treb*:ti,ab,kw) AND (blind*:ti,ab,kw OR mask*:ti,ab,kw)) OR '4 arm':ti,ab,kw OR 'four arm':ti,ab,kw OR placebo:ab OR group*:ab OR 'control group':ab OR randomly:ab OR ((random* NEAR/2 alloc*):ab)	6,421,062	4 Aug 2021
#26.	#6 OR #7 OR #8 OR #9 OR #10 OR #11 OR #12 OR #13 OR #14 OR #15 OR #16 OR #17 OR #18 OR #20 OR #21 OR #22 OR #23 OR #24 OR #25	3,127,876	4 Aug 2021
#25.	laceration*:ti,ab,kw	16,676	4 Aug 2021
#24.	((skin OR cutaneous OR dermal) NEAR/5 (diseas* OR infection* OR abrasion* OR avulsion* OR bruise* OR burn* OR injur* OR lesion* OR scrape* OR scratch*)):ti,ab,kw	185,590	4 Aug 2021
#23.	(((bed OR pressure*) NEAR/2 sore*):ti,ab,kw) OR bedsore*:ti,ab,kw OR decubitus:ti,ab,kw	13,078	4 Aug 2021
#22.	(diabet* NEAR/3 (foot OR feet)):ti,ab,kw	16,024	4 Aug 2021
#21.	gangrene*:ti,ab,kw	13,987	4 Aug 2021
#20.	ulcer*:ti,ab,kw	313,560	4 Aug 2021
#19.	wound*:ti,ab,kw	291,843	4 Aug 2021
#18.	'decubitus'/exp	23,961	4 Aug 2021
#17.	'laceration'/de	12,624	4 Aug 2021
#16.	'gangrene'/exp	18,200	4 Aug 2021
#15.	'diabetic foot'/de	17,502	4 Aug 2021
#14.	'skin infection'/exp	209,936	4 Aug 2021
#13.	'skin ulcer'/exp	81,183	4 Aug 2021
#12.	'ulcer'/exp	295,374	4 Aug 2021
#11.	'wound infection'/exp	50,052	4 Aug 2021
#10.	'wounds and injuries therapy'/de	482	4 Aug 2021
#9.	'wound healing'/exp	172,938	4 Aug 2021
#8.	'skin injury'/exp	117,605	4 Aug 2021
#7.	'injury'/exp	2,500,313	4 Aug 2021
#6.	'wound'/exp	305,281	4 Aug 2021
#5.	#1 OR #2 OR #3 OR #4	6,883	4 Aug 2021

#4. ((ozone OR ozonated OR ozoni$ed) NEAR/5 (oil* OR water OR aqueous OR liquid)):ti,ab,kw 2,012 4 Aug 2021
#3. (ozone NEAR/3 (therap* OR topical)):ti,ab,kw 1,223 4 Aug 2021
#2. 'ozone therapy'/exp 1,167 4 Aug 2021
#1. 'ozone'/de AND ('water'/de OR 'oils'/exp OR 'vegetable oil'/exp OR 'therapy'/exp) 4,843 4 Aug 2021

WEB OF SCIENCE
Search Strategy:

(TS=(ozone AND (therap* OR topical)) OR TS=((ozone OR ozonated OR ozoni$ed) NEAR/5 (oil* OR water OR aqueous OR liquid))) AND (TS=wound* OR TS=ulcer* OR TS=gangren* OR TS=laceration* OR TS=(diabet* NEAR/3 (foot OR feet)) OR TS=(((bed OR pressure*) NEAR/3 sore*) OR bedsore OR decubitus) OR TS=((skin or cutaneous or dermal) NEAR/5 (diseas* or infectio* or abrasion* or avulsion* or bruise* or burn* or injur* or lesion* or scrape* or scratch*))) AND TS=((randomi?ed NEAR/10 trial*) OR (controlled NEAR/3 trial*) or (clinical NEAR/2 trial*) or (treatment NEAR/2 trial*) OR "multicenter study" OR “double-blind method" OR ((single or doubl* OR tripl* OR treb*) and (blind* OR mask*)) OR ("4 arm" OR "four arm") OR placebo OR group* OR "control group" OR randomly OR (random* NEAR/2 alloc*)) AND LA=(English)

Database: Scopus
Search Strategy:

ALL(ozone AND (therap* OR topical)) OR ((ozone OR ozonated OR ozoni?ed) W/5 (oil* OR water OR aqueous OR liquid))) AND ALL(wound* OR ulcer* OR gangren* OR laceration* OR (diabet* W/3 (foot OR feet)) OR ((bed OR pressure*) W/3 sore*) OR bedsore OR decubitus) OR ALL((skin or cutaneous or dermal) W/5 (diseas* or infectio* or abrasion* or avulsion* or bruise* or burn* or injur* or lesion* or scrape* or scratch*))) AND ALL((randomi?ed W/10 trial*) OR (controlled W/3 trial*) or (clinical W/2 trial*) or (treatment W/2 trial*) OR "multicenter study" OR "double-blind method" OR ((single or doubl* OR tripl* OR treb*) and (blind* OR mask*)) OR ("4 arm" OR "four arm") OR placebo OR group* OR "control group" OR randomly OR (random* W/2 alloc*)) AND LANGUAGE(english)

Database:Google Scholar
Results: 59
(intitle:ozone|ozonated|ozonised|ozonized) (intitle:wound|wounds|ulcer|ulcers|diabetic foot|skin infection|cutaneous|laceration|gangrene|decubitus|bed sore|skin disease|burn|burns|injury) (Randomized Controlled Trial|Clinical Trial)