USE OF OXYGEN THERAPIES IN WOUND HEALING

FOCUS ON TOPICAL AND HYPERBARIC OXYGEN TREATMENT

JWC journal of wound care

A JOINT DOCUMENT

WoundsAustralia

EWMA European Wound Management Association

4. Topical oxygen therapies

Despite almost 50 years of clinical use, the subject of TOT for non-healing wounds remains controversial.[38–42] TOT can be defined as the administration of oxygen applied topically over injured tissue by either continuous delivery or pressurised systems. The availability to the wound tissue of topically applied higher pO_2 reverses localised hypoxia.[43] This causes both the direct killing of anaerobic bacteria and an enhancement of leukocyte function to address all other pathogens.[44,45] Once the inflammatory cascade subsides, the high availability of oxygen molecules in the wound tissue helps to upregulate angiogenic growth factors like vascular endothelial growth factor (VEGF) and fibroblast growth factor-2 (FGF-2).[45] This results in the prolific structured growth of new blood vessels and the stimulation of collagen synthesis by enhancing fibroblast activity.[46–48] These factors combined result in better wound bed granulation, strong collagen tissue formation, and wound closure.[46,47,49]

Background

The first report of TOT was published in 1969[41] wherein this therapy was called 'topical hyperbaric oxygen'. However, the term 'hyperbaric' as used in that paper was misleading and incorrect as currently used. Using specially constructed topical chambers on 52 patients with wounds of varying aetiologies, pure humidified oxygen was delivered under a constant pressure of 22mmHg; oxygen was applied continuously for 4–12 hours a day. Although uncontrolled by current standards, success was noted in the majority of cases with only six reported failures with an average healing time of three weeks in those treated with pressurised oxygen. It was found that wounds subjected to O_2 therapy at ambient pressures improved, but more slowly than those under pressure.[41] In the first RCT of topical 'hyperbaric' oxygen (THO) treatment, a total of only 28 patients were allocated to THO (n=12) and control (n=16) groups. All patients were admitted to the hospital for debridement, local dressings, intravenous antibiotics, and bedrest. The intervention group received THO in only four daily 90 minute sessions using a leg chamber providing humidified 100% oxygen under cycled pressures between 0 and 30mmHg. During the 14-day study period both groups experienced progressive reductions in the size of their DFUs. Not surprisingly, there were no significant differences in wound area reduction between the two groups. The obvious (and fatal) flaws in this study were the small numbers of patients treated and the very limited time period under study. There was simply insufficient power to detect any differences in treatments should any exist at only two weeks. The standard time frames that are currently employed for such DFU wound healing studies are 12-week treatment periods. Nonetheless, this study is often quoted as 'evidence' that THO is ineffective in promoting healing of foot ulcers.[50] In the following years there were inconsistent results in case series and reviews suggesting the putative benefits of administering oxygen topically to chronic wounds.[45,47,51–54]

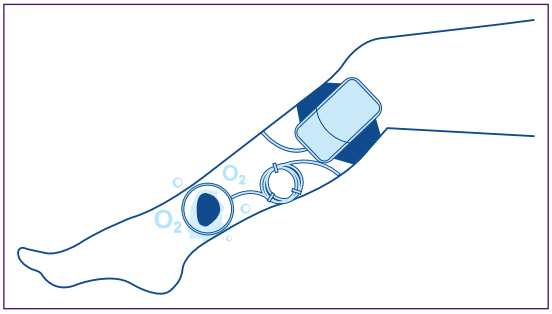
A subsequent non-randomised study sought to evaluate the healing benefits of both HBO and topical oxygen (TO) in a group of 57 patients with

a variety of chronic wounds.[45] Using standardised protocols for both therapies, healing outcomes were assessed at 14 weeks. Although they found no statistically significant change in wound volume reduction in the HBO group after this treatment period, the 25 wounds subjected to TOT showed a significant 57% reduction after 14 weeks of treatment (4 days each week). Additionally, wound edge tissue biopsies were taken to assess VEGF gene expression at baseline and at treatment end. Comparing VEGF expression at the final time point to the baseline measurement, those wounds treated with TO achieved a significant induction of VEGF expression, higher in those wounds where wound healing/volume reduction occurred. The overall difference in VEGF gene expression for HBO treated patients was not found to be statistically significant, although there was indeed an increase noted for most patients.[45] This study provides further evidence that treatment with topical oxygen can have a beneficial effect towards the healing of chronic wounds

Continuous delivery of non-pressurised oxygen

This category of devices apply topical continuous delivery of non-pressurised (normobaric) oxygen (CDO) through small cannulas or thin tubes to essentially occlusive wound dressings. Small portable battery-powered oxygen generators (extraction units) supply a continuous flow of pure oxygen to the wounds 24 hours a day.[3] The wound dressings are typically changed weekly and the oxygen generators are generally replaced after one to two weeks of continuous use.

The interim results of the RCT of the TransCuO$_2$



Continuous delivery of non-pressurised oxygen

CDO device showed that wound closure at 12 weeks was not significantly associated with treatment per the protocol, active 11 (52.3%), sham 8 (38.1%), [relative risk (RR) 1.38; 95% confidence interval (CI): 0.7, 2.7), p=0.54].[55] However, in the recently published results of the completed RCT a significantly higher proportion of people healed in the active arm compared with the sham arm (46% versus 22%, p=0.02). This relative effect became greater in more chronic wounds (42.5% versus 13.5%, p=0.006). Patients randomised to the active device also experienced

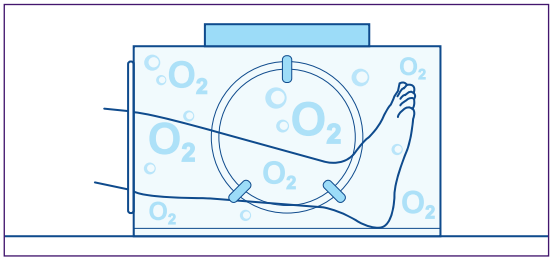
Table 3: Technologies available for distribution of topical oxygen in wound healing

Technologies available for distribution of topical oxygen in wound healing
Continuous delivery of non-pressurised oxygen (CDO)
Low constant pressure oxygen in a contained chamber
Higher cyclical pressure oxygen
Oxygen release through dressing or gel
Oxygen transfer
Application of oxygen species

significantly faster rates of closure relative to the sham (p<0.001). Unfortunately, this was only a per protocol analysis of the first 50 patients in each arm to complete the 12-week trial.[56]

Despite several small case studies indicating beneficial healing for chronic wounds,[57,58] results for the Epiflo device multicentre RCT have yet to be published in any journal. Nonetheless, information available on clinicaltrials.gov indicates that wound closure at 12 weeks was not statistically significantly associated with treatment per the protocol active 55.7%, sham 50.8% with 61 patients in each group.[59] A prior single centre randomised study of 17 DFU patients followed for four weeks indicated that the TO group achieved an average wound size reduction of 87% compared with 46% in the standard of care group (p<0.05).[60] While tissue and wound sample cellular and cytokine level changes were noted, these patients were not followed to complete healing and the sample size was too small to be widely generalisable.

The Natrox CDO device has been marketed for several years with posters and presentations indicating positive results in a variety of wounds. A small published case series on the treatment of venous leg ulcers (VLUs) indicated positive results towards healing and a reduction in pain scores during the treatment periods.[61] A recent small, single-centre, randomised non-placebo controlled trial of 20 patients with chronic DFUs compared this device with standard care alone over 8 weeks.[62] They found a significantly increased healing rate (wound area reduction) in those treated with the topical oxygen device compared with baseline at week 8 (p<0.001), but no such increased difference was noted in the control group (p<0.262). While all superficial ulcers healed in both groups, the TOT group seemed to show a more beneficial effect in more advanced ulcers. While published data is not yet available, a large RCT using this device is currently in progress to further determine its efficacy in healing chronic DFUs.



Oxygen delivery in a contained chamber

Low constant pressure oxygen in a contained chamber

The lower constant pressure devices include such devices as the O_2 Boot or OxyCare. In this approach oxygen is provided in a simple plastic chamber/boot that is placed around the extremity with the ulcer. Constant pressure is then maintained within the chamber up to 35mmHg. There are numerous studies that have been conducted on these types of devices over the last four decades that have ostensibly shown good clinical efficacy. However, the majority of these studies have consisted of case series or uncontrolled trials.[45] The one very poorly conducted RCT that used a similar device has been previously discussed.[50] Unfortunately, this study is often cited as evidence of the ineffectiveness of TO despite its being underpowered and of too short of a duration. This outcome is not surprising considering the fact that the therapy arm only received two treatments each week (four total treatments) with the O_2 therapy devices used.

Higher cyclical pressure oxygen

The Topical Wound Oxygen (TWO_2) system differs from other devices in that it applies a higher topical O_2 pressure between 5mmHg and 50mmHg, in a cyclical pressure waveform, combined with humidification. The benefit of this approach is that the higher pressure gradient results in O_2 molecules

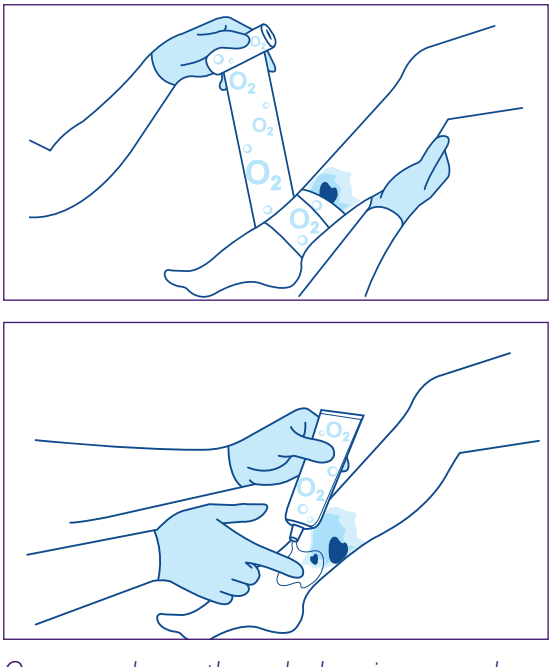
diffusing deeper into the hypoxic wound tissue and enhances multiple molecular and enzymatic functions.[46,63] The cyclical pressure applied with TWO_2 of between 5mmHg and 50mmHg creates sequential non-contact compression of the limb that helps to reduce peripheral oedema and stimulates wound site perfusion further.[48,64] Several prospective clinical studies have been conducted using this device on both VLUs and DFUs. One non-randomised parallel arm study of 83 patients was conducted on VLUs to measure the effect of TWO_2 compared with conventional compression dressings (CCD) on wound healing using the primary endpoint of the proportion of ulcers healed at 12 weeks.[48] At 12 weeks, 80% of TWO_2 managed ulcers were completely healed compared with 35% of the CCD-managed ulcers. Median time to full healing was 45 days in the TWO_2 arm and 182 days in CCD arm. Unfortunately, there was a good deal of selection bias pertaining to treatment allocation in this study. These same authors later conducted another comparative study that similarly investigated the efficacy of TWO_2 versus CCD in the management of refractory non-healing venous ulcers (RVUs) with a duration of at least two years.[64] This study was also non-randomised and allotment to treatment arm was primarily based on patient preference. A total of 132 patients were enrolled with 67 patients (mean age: 69 years) using TWO_2 and 65 patients (mean age: 68 years) with CCDs for 12 weeks or until full healing. At 12 weeks 76% of the TWO_2 managed ulcers had completely healed, compared with 46% of the CCD-managed ulcers with a median time to full healing of 57 days and 107 days, respectively. Interestingly, in those patients with meticillin-resistant *Staphylococcus aureus* (MRSA) colonised ulcers, MRSA elimination occurred in 46% of patients managed with TWO_2 and 0% of patients managed with CCD. Another prospective non-blinded, non-randomised study was conducted to examine the clinical efficacy of TWO_2 therapy in healing patients with severe DFUs referred to a community wound care clinic in Canada.[65] Patients were simply allocated to the TO if a unit was available or were otherwise treated with advanced moist wound therapy. At 12 weeks 82.4% of the ulcers in the TWO_2 therapy arm and 45.5% in the standard care arm (control) healed completely. Median time to complete healing was of 56 days in the TWO_2 therapy arm and 93 days in the control standard care arm. An ongoing study is currently enrolling subjects into a 220 patient multinational, multicentre, prospective, randomised, double blinded, placebo-controlled trial to evaluate the efficacy of TWO_2 in the treatment of chronic DFUs. The study's inclusion criterion allows for non-healing DFUs up to Stage 2D in the University of Texas Classification of Diabetic Foot Ulcers, defined as wounds penetrating to tendon or capsule with infection and ischaemia. It includes a two-week run-in period with best standard of care to flush out wounds that would heal with this alone and a 12-month follow-up to assess recurrence. With a standardised primary outcome of the incidence of complete wound closure at 12 weeks, this trial should not only address the need for TOT, but it should also make its results comparable with other advanced wound care therapies including systemic HBOT.[66]

Oxygen release through dressings or gels

Different kinds of products are available, either using the release of pure O_2 embedded in the dressing or releasing O_2 generated by a biochemical reaction in a hydrogel. In the O_2 containing dressings, pure O_2 is embedded, such as in vesicles, and released after the dressing is liquefied by the wound exudate. Continuous O_2 release dressings can be used as secondary dressing and release O_2 for up to six days. In order to optimise conditions for delivery at the wound, debridement and cleansing should be carried out at regular intervals before the dressings are applied.

In hydrogel dressings an increased concentration of dissolved O_2 is obtained via a chemical or

biochemical reaction. These occlusive dressings make use of the reactivity of 0.3% hydrogen peroxide, which is converted to water and dissolved O_2. This can diffuse via a permeable separator to the wound bed. In contrast, another product consists of two separate components must be applied together to activate the biochemical process. One component contains a hydrogel sheet containing glucose and a low-concentration gel matrix with less than 0.04% of iodide ions, and a second component sheet containing glucose oxidase. The glucose oxidase incorporated in the second gel sheet catalyses the oxidation of (beta)-D-glucose to D-gluconic acid and hydrogen peroxide in the presence of O_2. The hydrogen peroxide released as a result is thought to diffuse through the dressing and either oxidises iodide ions to free iodine and O_2 or, if it reaches the wound surface, is metabolised to water and O_2. Iodine has a beneficial antimicrobial effect within the gel and should help to prevent the proliferation of microorganisms at the wound–dressing interface, while the dissolved O_2 is believed to create beneficial effects within the wound.[3]



Oxygen release through dressings or gels

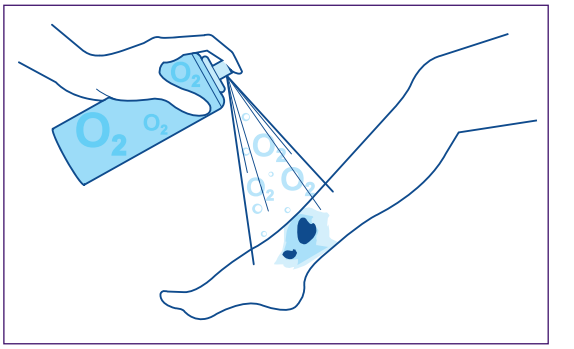
Several case study reports demonstrate improvements in the healing of different wound types.[67,68] As an example, in a non-controlled multicentre case series of 51 patients the dressing was tested over a six-week period in wounds with various aetiologies and a mean duration of 25.8 months. The results showed six wounds healed fully, 37 were judged to have improved, seven remained static and one deteriorated.[69] *In vitro* experiments have shown that such dressings are capable of significantly increasing O_2 levels in wounds.[70] Further evidence of its beneficial impacts on wound healing was generated by using these dressings on burn patients treating larger donor site wounds in comparison with standard care.[71] Moreover the oxygenating hydrogel dressings, which release O_2 and different levels of iodine into the wounds, were tested in different *in vitro* tests against various target organisms. It was shown that the dressings were significantly more effective against a broad spectrum of microorganisms including biofilm than controls.[72,73]

Oxygen transfer

Haemoglobin as an O_2 carrier is another approach to topical wound treatment. Haemoglobin augments transport of O_2 by means of facilitated delivery.[74] The mode of action of this approach is based solely on the physical effect of facilitated delivery, and not on a pharmacological or metabolic effect. In wound treatment, the haemoglobin spray should be applied in addition to standard therapy. The spray can be used concomitantly with most existing treatment regimens.[3] In a pilot study the O_2 saturation of ulcer tissue was measured in five patients with chronic leg ulcers before application and 5 and 20 minutes after application using photoacoustic tomography. The average O_2 saturation showed

a significant increase up to 5mm depth from 56.4% before to 69% after 5 minutes and 78.8% after 20 minutes following a single application of haemoglobin spray. The authors conclude that the application of topical haemoglobin spray leads to an increase in O_2 saturation *in vivo* in patients with chronic leg ulcers.[75]

The authors of an RCT compared the application of the haemoglobin spray versus a sham product as add-on to best practice wound care over 13 weeks. In each treatment group there were 36 patients. In contrast with the control group, where no wound size reductions were observed, the patients treated with the complementary haemoglobin spray demonstrated a significant wound size reduction of 53%.[76] The clinical effects of a haemoglobin spray were also observed in a multicentre observational evaluation of 17 patients with 20 chronic DFUs. In 14 of the 18 wounds that completed the evaluation over a four-week period a mean reduction in wound size of 53.8% was observed. After 12 weeks 20% had healed, 53% were progressing towards healing, 20% increased in size and 7% were slow to heal.[77] In a case series of 11 patients with pressure ulcers (PUs) who were treated with haemoglobin spray for three months, nine wounds healed and two demonstrated reduced wound-size. From ten patients with pain at baseline, nine were pain-free by week 8. A rapid elimination of slough was observed in all patients.[78] In another set of recently collected data cohorts, sequential patients were recruited prospectively from patients with DFUs, chronic wounds (CWs), and sloughy wounds (SWs). The number of patients recruited to each cohort was 20, 50 and 100 respectively. As control group, data from clinical notes of an equal number of patients were collected retrospectively. These were selected sequentially by date in the notes without reported as matching to prospective cases. The DFU cohort was treated in a hospital setting and



Oxygen transfer

the CW/SW cohorts were treated in primary care. All three cohorts shared the inclusion criterion of a wound that failed to heal defined as a <40% reduction in area in the previous four weeks. In the DFU cohort the mean wound size reduction was greater in the haemoglobin spray group at week 4 (–63% versus –21%), week 16 (–91% versus –43%) and week 28 (–95% versus –63%). At week 28 follow-up, 15/20 patients in the haemoglobin spray cohort had complete healing compared with 8/20 in the control cohort. The CW cohort reported mean wound size reductions of –73% in the haemoglobin spray group compared with –12% in the control group at 4 weeks. The benefit persisted at 8 weeks (–87% versus –14%) and the final 26 week follow-up (–89% versus –75%). Altogether 45/50 patients had complete healing at the final 26-week follow-up compared with 19/50 in the control group. The SW cohort results were reported in a more limited fashion. At week 8 follow-up there was a mean wound size reduction of –93% in the haemoglobin spray group compared with –32% in the control group. At week six complete wound closure was observed for 65/100 patients in the haemoglobin spray group and 37/100 patients in the control group.[79,80]

Based on the published evidence and positive clinical outcomes regarding the efficacy of haemoglobin spray practical-oriented clinical algorithms have

been established for this kind of treatment both by the German-speaking D.A.CH.-(Germany, Austria, Switzerland) region[81] and in England.[82]

Application of oxygen species

Another therapeutic approach using topically applied O_2 in wound treatment is based on the fact that ROS can be used in antimicrobial treatment and perhaps as a signalling molecule that support wound healing processes.[79,80] ROS are effective in destroying a broad range of pathogens and also biofilms. Their mode of action is typically the physical destruction of the pathogen's cell-wall integrity and hence they are not linked to the problems of antibiotic resistance, which are related to a range of pharmacological effects. There is an increasing spectrum of products using ROS for antimicrobial and cleansing wound therapy available. A product containing hyperosmotic ionised seawater, ROS, triplet oxygen $^3O_2$ and a high pH-value is thought to reduce wound swelling, inflammation, microbial contamination and to stimulate cellular signalling transduction pathways. It is available as a rinsing solution and a wound gel. The antimicrobial effects are mediated primarily by the singlet O_2.

These effects are regulated by the basic pH value

Table 4. Types of topical oxygen devices and therapies currently available

TOT type	Medical devices	Treatment details				
	Company, Product			Treatment location	Moist wound environment	GRADE
Higher cyclical pressure oxygen	Aoti Inc., TWO_2	50mbar to 5mbar cycles;	Pressure low, >1bar Flow rate high Treatment time: 60–90 minutes Treatment frequency: 3–7 days	Open wound in chamber or bag	Possible	Grade 18, (RCT, controlled cohort studies, various case series) positive effect shown
Low constant pressure oxygen in a contained chamber	OxyCare GmbH, O_2TopiCare System	2-5 l/min;<50mbar;	Pressure: low, >>1bar Flow rate: high Treatment time: 60–90 minutes Treatment frequency: 3–7 days	Open wound in chamber or bag	Possible	
	GWR Medical, TO_2	2-5 l/min;<50mbar;	Pressure: low, >1bar Flow rate: high Treatment time: 60–90 minutes Treatment frequency: 3–7 days	Open wound in chamber or bag	Possible	
Continuous delivery of non-pressurised oxygen (CDO)	Ogenix Inc., EpiFLO	Continuous, slow flow of pure oxygen of 3 ml/hr for 15 days through a cannula to blanket the wound.	Pressure: low, <1bar Flow rate: low Treatment time: 24 hours Treatment frequency: 7 days	Occlusive wound dressing	yes	Grade 2C, (1 Interim report on RCT showed no advantage versus sham. Cohort studies, various case series) only weak evidence
	Inotec AMD Ltd., Natrox	Continuous, slow flow of pure oxygen of ~12ml/hour for several days via a thin flexible tube to the Oxygen Delivery System which is in direct contact with the wound surface	Pressure: low, <1bar Flow rate: low Treatment time: 24 hours Treatment frequency: 7 days	Occlusive wound dressing	yes	

which supports a high concentration of hydroxyl ions, which act as an antioxidant.

In a cohort study conducted in four wound clinics, the clinical efficacy of singlet O_2 solution was evaluated. In 73 patients with critically colonised and/or infected, malodorous wounds, covered with slough/fibrin, or wounds showing inflammation of the periwound skin were included. After 42 days 33% of the wounds in the study had healed, 57% had improved and 10% remained stagnant. All wounds had shown clinical signs and symptoms of critical colonisation and/or infection at day 0, at day 42 the infection was completely eradicated and inflammation was reduced in 60%.[83]

Table 4. Types of topical oxygen devices and therapies currently available

Oxygen release through dressing or gel	OxyBand Technologies Inc., OxyBand	Oxygen release for up to 5 days after contact with moisture within a simple occlusive wound dressing	Pressure: na Flow rate: na Treatment time: 24 hours Treatment frequency: 7 days	Occlusive wound dressing	yes	Grade 2B, (1RCT, cohort studies, various case series) only weak recommendation for oxyzyme by Nice due to lack of efficacy
	AcryMed/ Kimberly Clark, OxygeneSys Continuous	Use as a foam dressing, Oxygen release for up to 5 days when dressing is moistened	Pressure: na Flow rate: na Treatment time: 24 hours Treatment frequency: 7 days	Occlusive wound dressing	yes	
	AcryMed/ Kimberly Clark, OxygeneSys On Demand	Oxygen release for up to 5 days after contact with moisture within a simple occlusive wound dressing	Pressure: na Flow rate: na Treatment time: 24 hours Treatment frequency: 7 days	Occlusive wound dressing	yes	
	Crawford Healthcare Ltd, Oxyzyme	Use as a primary dressing, in early stage wound treatment. Oxygen release when both layers are attached to each other	Pressure: na Flow rate: na Treatment time: 24 hours Treatment frequency: 7 days	×	yes	
Oxygen transfer	SastoMed GmbH, Granulox	Liquid spray with 10% purified haemoglobin, applied as thin layer to the wound bed, and before wound is covered by a non-occlusive dressing, twice weekly up to once daily application depends on wound status	Pressure: na Flow rate: na Treatment time: 24 hours Treatment frequency: 7 days	×	yes	Grade 1B, (1RCT. 1 controlled open label study 3 controlled cohort studies, various case series) positive effect statistically shown, >50,000 treatments in more than 20 countries with no relevant side effects, clear positive benefit risk value

Other products contain super-oxidised solution or gel manufactured through the electrolysis of ultra-pure water and NaCl. The active ingredient as source of ROS is hypochlorous acid (HOCl), a major inorganic bactericidal compound of innate immunity.[84] HOCl has been shown to be effective against a broad range of microorganisms either as stabilised neutral or acidic HOCl-solutions.[85] These solutions are intended for use in the cleansing and debridement phase primarily to decrease the microbial load by eliminating pathogenic microorganisms.

In an RCT, a stabilised super-oxidised solutions at neutral to acidic pH was tested for the treatment of 40 patients with postsurgical lesions larger than 5cm^2 in DFUs. The outcome of the use of the SOS was compared with use of povidone iodine as a local medication. Patients were followed-up weekly for six months. The authors were able to demonstrate that the healing rates, time taken for cultures to become negative and duration of antibiotic therapy were significantly shorter in the group treated with super-oxidised solution.[86] The authors claim that the cost of the super-oxidised solutions is lower than standard treatment with a saving of 40% on the total expenditure, especially due to less antibiotic therapy and following surgical procedures. Results are in accordance with findings of other clinical trials performed. Recently, a safety, effectiveness and cost-effectiveness evaluation of stabilised super-oxidised solutions in comparison with povidone iodine (PVP-I) treatments was published.[87] The authors concluded that such solutions are a safe, effective and cost-effective irrigation and cleansing agents and can provide an economical alternative to the other available antimicrobial agents.

Conclusion

The clinical results achieved with these methods indicate possible benefits over standard care alone. As for many other products used in wound care management, the clinical evidence for the efficacy of topical oxygen-based treatment is not homogeneous and ranges from uncontrolled case reports to RCTs with some limitations. Although most of the published data does not meet the highest standards of evidence, it suggests that such adjunctive therapies are easy to handle, safe and may be potentially effective modalities for use in modern strategies of wound care in specific subpopulations. Interesting question about the concomitant action of TOT with other therapeutic procedures, including HBOT, vascular interventions or skin transplantation, still remains unanswered.

Recommendations

There is a limited but expanding evidence base for successful healing after treatment with TO products, especially in a subset of non-healing patients who failed to achieve an adequate healing response in standard treatment settings. Although the authors endorse the adjunctive administration of TO therapies for non-healing chronic wounds, more robust data from multi-centre prospective placebo-controlled trials affirming their clinical efficacy will be required before this promising therapy can be given a stronger recommendation.