The Mechanism of Hyperbaric Oxygen Therapy in the Treatment of Chronic Wounds and Diabetic Foot Ulcers

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ABSTRACT

Non-healing wounds are a growing public health concern, and more than $25 billion per year in the US are spent caring for patients with chronic wounds. Many of these patients are referred to specialized wound centers, where hyperbaric oxygen therapy (HBOT) has become a mainstay in healing wounds, especially diabetic foot ulcers (DFU). However, it is costly, with a typical course of therapy running into the tens of thousands of dollars. Presently, as many as 30–40% of DFU patients with Wagner's Grade 3 and 4 ulcers treated with HBOT fail to heal by 24 weeks. Unfortunately, the patient will have already received lengthy therapy (30–60 daily treatments over 6–10 week time period) before having the wound deemed non-responsive. Currently, practitioners employ a combination of clinical markers, diagnostic testing and a four-week preliminary healing response, but this approach is inaccurate and delays definitive identification of HBOT responder and non-responder phenotypes.

KEYWORDS: hyperbaric oxygen therapy, diabetic foot ulcer, chronic wounds, molecular mechanism

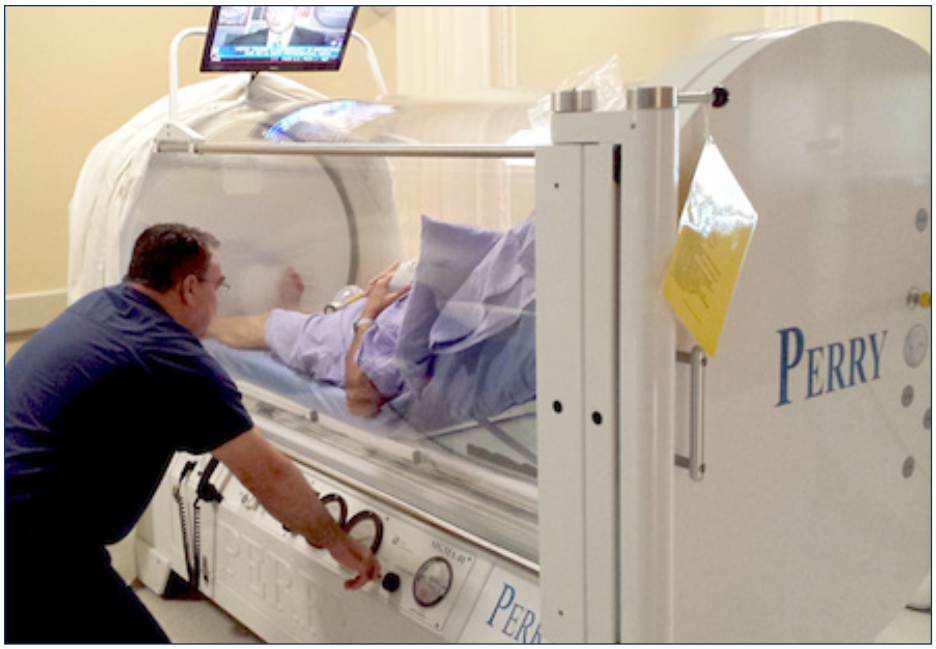
INTRODUCTION

Hyperbaric Oxygen Therapy (HBOT) is a treatment proposed for a myriad of ischemic conditions.[1-8] In the early 1960s, physicians began to consider its use for treating chronic wounds.[4,6] A patient prescribed the therapy is sealed within a large chamber (**Figure 1**) filled with 100% oxygen pressurized at 2.0 to 2.5 atmospheres absolute (ATA).[5] For comparison, this chamber pressure is equivalent to being about 45 feet underwater. The typical therapy session lasts for 1 to 2 hours and may be repeated for 30-40 treatments.[2,5]

Diabetic foot ulcers (DFU) occur in approximately 10% of diabetic patients and this may lead to serious complications, including amputation, in approximately 2%.[9] There are more than 23 million people in the United States with diabetes, and there is an estimated worldwide prevalence of 5%.[9] The continued care of DFUs is an expensive and time-consuming process and is exacerbated by a recurrence rate of almost 70% over a 5-year period.[9] Severe complications contribute to an annual mortality rate of 11% for those with a DFU and 22% for those with a lower extremity amputation[9]. Older patients are more likely to develop DFUs.[9] Management of DFUs begins with debridement, offloading, and infection control.[10] Debridement is the removal of necrotic tissue to expose viable tissue.[10] Offloading by wheelchair, cast, or crutches is very effective for compliant patients, with wound healing rates of 73–100%.[10] Infections at the DFU are common and are usually polymicrobial. Common pathogens found within the ulcer include Staphylococcus aureus, Group B streptococci, enterobacteriaceae, Pseudomonas aeruginosa, and enterococci.[10]

When DFUs do not heal despite adequate conservative management or progress to Wagner Grade 3 or 4, HBOT can be considered as an adjuvant therapy.[2,11] However, its efficacy is not universally accepted.[2,5,8,11-13] A 2013 study of patients with similar case presentations showed neither

Figure 1. A hyperbaric medicine provider attending to a patient receiving HBOT



improvement in wound healing nor a decrease in amputation following HBOT.[13] While this work brought significant doubt to the effectiveness of HBOT, a 2015 Cochrane report on HBOT for chronic wounds found that HBOT has strong clinical evidence for improved short-term healing (early wound healing response), limited clinical evidence for improved long-term healing (final stage wound healing), and limited evidence for decreasing the rate of lower limb amputation.[2,5,11] As cost-effective care becomes an increasing priority under the Patient Protection and Affordable Care Act of 2010, the expense of HBOT may no longer be justified without stronger evidence for consistent benefit. Indeed, each treatment costs between $200 and $1,250 and requires significant investment of time and compliance on the part of the patient.[2]

The subset of patients most likely to benefit from HBOT is still a matter of debate.[5,8,12] Studies failed to show an association between ankle-brachial index (systolic pressure in the ankle divided by systolic pressure in the arm) or toe blood pressure and healing of ulcers.[2,5] In-chamber wound-area transcutaneous oxygen pressure (TcP_{O2}) less than 200mmHg during HBOT has a 74% reliability of predicting non-healing; however, this is not a screening option available to centers without access to hyperbaric chambers.[14] Genetic assessment of patients may offer a new way of directing HBOT.[2] The authors of this paper are currently conducting a study to differentiate genetic expression profiles of responders and non-responders to HBOT, such that predictors for response may be identified.

The rest of this paper outlines the cellular and molecular mechanisms by which HBOT promotes wound healing.

HBOT increases oxygen delivery to tissues

At normal atmospheric conditions, nearly 100% of oxygen is transported by binding to hemoglobin, and only a small amount is dissolved in the plasma.[2,15] Oxygen delivery occurs when oxygen molecules leave the circulatory system and diffuse down their concentration gradient into cells. The concentration gradient is in turn determined by the partial pressures of oxygen in the capillaries and the tissue in immediate proximity.[15] Poorly perfused tissues create steeper gradients that induce greater oxygen delivery, but they also have a larger cumulative demand.[15] Patients suffering from microvasular diseases such as diabetes have fewer capillaries to provide oxygenation to the tissues.[2] HBOT combats this state of hypoxia by increasing the amount of oxygen dissolved in plasma as well as the partial pressure of oxygen in the tissue fluid.[5] This increases the cumulative amount of oxygen available to tissues, thereby meeting the increased oxygen demand of poorly perfused tissues.[2,3,5,8] Oxygen delivery to hypoxic tissues has been shown by modeling and clinical observation to be approximately 16-fold higher with HBOT.[16]

HBOT promotes angiogenesis, wound healing, and immune response through cell signaling

HBOT raises the partial pressure of oxygen in blood and subsequently in tissues, and this has been shown to have many downstream biological effects: angiogenesis, wound healing, and increased immune system response.[2,3,5,8,17] Various cytokines, gases and other macromolecules mediate these complex cellular responses. Angiogenesis is the process by which existing blood vessel networks expand to meet increased demand for blood and oxygen within tissues.[18] Angiogenesis can proceed by two main processes: endothelial cell migration, in which new vasculature forms as an extension of the existing network, and division of blood vessel lumen, in which the cross-sectional area of the existing capillary network increases.[18] Essential for these processes is having an adequate number of cells to create new blood vessels, and research has shown that circulating progenitor cells are recruited as a result of HBOT.[11] HBOT has a stimulatory effect on endothelial nitric oxide synthase (eNOS), which produces nitric oxide (NO), a signal necessary for the activation and recruitment of progenitor cells.[11,19,20] In patients with diabetes, eNOS is inhibited; however, HBOT can overwhelm the inhibitory effect of diabetes and induce NO synthesis, thereby promoting angiogenesis and accelerating wound healing.[11,21-23]

Wound healing is a normal process following injury that comprises four phases: hemostasis, inflammation, proliferation, and tissue remodeling.[24] Oxygen availability is critical in wound healing primarily for facilitating oxidative phosphorylation for normal cellular function.[24] However, during the initial phases of wound healing, the wound is hypoxic.[24] This leads to signaling for angiogenesis and other wound healing factors (hypoxia-inducible factors - HIF, platelet-derived growth factor - PDGF, transforming growth factor beta - TGF-B, vascular endothelial growth factor - VEGF, tumor necrosis factor alpha - TNF-α, and pre-pro-endothelin 1 - PPET-1), but conversely if the wound is chronically hypoxic there will be impaired healing.[11,24] This temporal difference in the effect of hypoxia is thought to be largely determined by HIF expression where early wound healing was improved with HBOT and HIF levels were decreased. However, HIF expression was elevated in hyperoxic conditions and lead to increased VEFG expression.[11] In addition to the aforementioned cytokines, SDF-1 has been shown to be a key determinant of wound healing and is activated by HBOT.[22] Lack of SDF-1 expression appears to partially explain why chronic hypoxic wounds (as in diabetes) do not heal.[22]

HBOT has been shown to decrease inflammation by inhibiting prostaglandin, IFN-γ, IL-1, and IL-6 formation.[25] This anti-inflammatory effect may improve general immune system function by decreasing immunosuppressive agents (prostaglandins, IL-1, IL10).[25] The immune system response is further augmented with HBOT by aiding the production of reactive oxygen species (ROS) by leukocytes.[2,11]

In addition to cytokine suppression, anti-inflammatory activity, and immune response, HBOT has effects on antioxidant production. [26]

HBOT and the antioxidant response pathway

Injury, infection, and chronic disease lead to stress response pathway activation.[27] Cells produce antioxidants in response to these stresses.[27] The main system that regulates antioxidant production is the Nrf2-Keap1 / cytoplasmic oxididative stress system.[27] Keap1 is a cytoplasmic chaperone protein that binds to Nrf2 – a transcription factor.[27] Without cellular stress, Nrf2 is ubiquinated and destroyed at a high rate.[27] With cellular stress, Nrf2 is no longer ubiquinated at a high rate and is able to translocate to the nucleus to activate antioxidant response elements (AREs) and over 200 antioxidant genes.[27-30] Gene expression analysis initially suggested that Nrf2 was increased universally following HBOT, suggesting that cytoprotection in endothelial cells by activation of antioxidant pathways was a key mechanism of HBOT.[28,29] More refined and longer time-scale expression analysis has revealed a more complex systemic response to HBOT.[30]. Nrf2 espression peaked at 4 hours after exposure to HBOT and was expressed at control levels at 24 hours following exposure.[29] Subsequent studies into the antioxidant pathways activated by HBOT reveal that diabetes activates Nrf2 expression likely because of systemic hyperglycemia and microvascular injury.[30] HBOT, although shown to increase Nrf2 expression within a few hours of exposure actually leads to a long-term decrease in Nrf2 expression when HBOT was continued in a clinically relevant exposure pattern in db/db mice.[30] This bi-phasic response is thought to indicate a short-term increase in cytoprotective antioxidant proteins that are stimulated by HBOT exposure, but eventually contribute to a long-term decrease in antioxidant production due to the cytoprotective effects of continued HBOT.[30]

CONCLUSION

For over 50 years HBOT has been regularly used for chronic wound care and yet the underlying mechanisms and clinical effectiveness are rightly still called into question. To provide direction to the field, more advanced analyses of the gene expression may prove to be useful. In addition to providing clarity to the usefulness of HBOT it serves the larger purpose of a more robust understanding of wound healing.

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