Hyperbaric & Wound Care Associates

Exceptional Results

Enhanced Wound Healing by Binding Reactive Oxygen Species

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Introduction

Reactive oxygen species (ROS) are toxic waste products which produce oxidative stress during the inflammatory phase of wound healing. ROS result from the respiratory burst associated with revascularization, reperfusion and restructuring epithelial and connective tissues. The enzyme ribosomal S6 kinase (RSK) protects against excessive production of ROS. Overactivity of RSK from excessive production of ROS has been implicated in poor wound healing. Sterically hindered amines are known to bind ROS. A novel hydrophilic gel (Wound-Be-Gone®) formulated to deliver sterically hindered amines has been used in the care of patients with difficult to heal diabetic foot ulcers.

Methods

In the present study, we tested the hypothesis that the topical administration of a hydrophilic 2-hydroxyethyl methacrylate polymer containing a mixture of sterically hindered amines may function as an ROS antagonist, promote healing and inhibit scarring in cutaneous wounds. The study subjects were diabetic patients treated with a standard clinical regimen which included aggressive offloading, débridement, control of bioburden, optimization of diabetic control and vascular intervention as indicated. The patients were randomized to topical therapy of either hydrogel (control) or an ROS antagonist polymer (Wound-Be-Gone®, Wake Pharm). We present a series of the patients with diabetic foot ulcers that were randomized to the ROS antagonist polymer.

The Role of Sterically Hindered Amines in Scavenging Free Radicals

Sterically hindered amines (2,2,6,6-tetramethyl-substituted piperidines) are chemical compounds easily oxidized by electron transfer to parent cations in n-butyl chloride solution, by sulfate radical anions in aqueous solution, and by sensitized electron transfer to carbonyl triplets. In nonpolar surroundings, the radical cations of the tertiary piperidines have been directly observed by optical spectroscopy to exhibit absorption maxima below $\lambda = 300$ nm and around 550 nm. This occurs in the time span of nanoseconds. Subsequently, these sterically hindered amines deprotonate to a-alkylamine radicals which are also the first observable products of oxidation with sulfate radical anions in water. In the case of secondary piperidines, the amine radical cations deprotonate to aminyl radicals in times < 10 ns. The triplet-sensitized electron transfer to the benzophenone as well as cyclohexanone triplet results in amine-derived and ketyl-type radicals formed at a nearly diffusion-controlled rate, which suggests an electron- and subsequent proton-transfer mechanism. In the presence of oxygen, the amine-derived radicals are oxidized to nitroxyl radicals by different pathways for secondary and tertiary piperidines. For the reaction of the nitroxyl radicals with other radicals, rate constants are found to be quite similar (about 5×10^8 M-1 s-1) for several alkyl radicals and for the tert-butyloxyl radical and less than 10^5 M-1 s-1 for alkylperoxyl radicals. Because of the minor importance of radical reactions with the sterically hindered amines, the antioxidant effects of these compounds may be explainable by oxidation, primarily via cationic and subsequently radical intermediates, to nitroxyl radicals, which scavenge free radicals very efficiently.

The Role of Sterically Hindered Amines in Wound Healing

It is useful to think of ROS as toxic waste products which are the products of oxidative stress during the inflammatory phase of wound healing. ROS result from the respiratory burst associated with revascularization, reperfusion and restructuring epithelial and connective tissues. The enzyme ribosomal S6 kinase (RSK) is an enzyme which protects against excessive production of ROS. However, overactivity of RSK from excessive production of ROS has been implicated in poor wound healing and scarring. Because sterically hindered amines are known to react with ROS, producing ROS scavenging nitroxyl radicals as intermediates, the topical administration of sterically hindered amines sub serves ROS scavenging and healing in wounds. An ROS scavenging gel consisting of a hydrophilic 2-hydroxyethyl methacrylate polymer containing a sterically hindered amine has been synthesized (Wound-Be-Gone® (WBG), Wake Pharm) and has been shown to be effective at decreasing inflammation and promoting wound healing. The methacrylate polymer has the effect of absorbing excessive extracellular fluid containing substances which are known to stimulate pain receptors, thereby, producing immediate pain relief and optimizing the microenvironment for wound healing by coating the wound, protecting it from bacterial contamination and eliminating excessive accumulation of fluid containing algesic compounds, ROS and other substances which can inhibit wound healing.

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2. Patel RP, T Cornwell, and VM Darley-USMAR: The biochemistry of nitric oxide and peroxynitrite: implications for mitochondrial function. In: Understanding the process of ageing: The roles of mitochondria, free radicals, and antioxidants. (1999) Eds: E Cadenas and L Packer, Marcel Dekker, Inc. NY. Basel 39-40.

Case Study 1

CK is an 83 year old diabetic male who presented to the Wound Care Center with a 2 year history of a Right Foot D2 ulcer. The ulcer was due to trauma. Radiographs were negative for osteomyelitis although a healing fracture was noted. The patient's PAD was not amenable to revascularization. Offloading was continued. WBG was initiated at the time of the initial consult and the patient healed after 12 weeks.


2-18-09
Initial Consult, WBG Trial Initiated


3-18-09


4-22-09


5-28-09
Healed

Case Study 2

DB is a 43 year old male with diabetes and small vessel arterial insufficiency. He developed a necrotic ulcer on his Left Foot Great Toe, which required amputation. The surgical incision partially dehisced distally. The patient was treated with offloading and topical cadexomer iodine between 2-12-09 and 4-30-09 with little change in wound appearance. He was enrolled in the WBG trial on 4-30-09 and healed on 5-11-09.


2-12-09


3-24-09


4-30-09
WBG Trial Initiated


5-11-09
Healed

Case Study 3

LP is a 45 year old diabetic female who presented to the Wound Care Clinic with a Left Foot 4th webspace D5 ulceration. Initial management included evaluation for osteomyelitis (negative), vascular testing (ABI .85), offloading, and topical silver. On 5-6-09 the patient was enrolled in the WBG trial and healed rapidly.


2-6-09
Initial Consult


5-6-09
WBG Trial Initiated


6-2-09


7-29-09
Healed

Conclusions

Compared to controls, treatment using the ROS antagonist polymer (Wound-Be-Gone®) demonstrated decreased inflammation and improved healing trajectories. These findings suggest that the healing of cutaneous wounds may be improved by the topical administration of an ROS trapping agent. The mechanism of action appears to be related to decreased inflammatory mediators via oxygen free radial binding technology. Future studies should determine the physiological limitations of such ROS trapping agents.