Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Oxidation and "Unconventional" Approaches to Infection

Robert Jay Rowen, Private Medical Practice, Santa Rosa, CA, United States

Introduction

> A paradigm shift in the treatment of infectious diseases is necessary to prevent antibiotics becoming obsolete, and where appropriate, alternatives to antibiotics ought to be considered.
>
> (Carson et al., 2006).

Within a generation after antibiotic introduction, bacterial infection morbidity/mortality sharply fell. However, within a few more human generations, rapid genetic sharing of resistance genes has returned bacterial induced deaths to front and center, therefore the world again faces a crisis. Dire calls have been issued for the development of new drugs. Nevertheless, just as fast as medicines are developed, pathogens rise to resistance (see fascinating but frightening video of *Escherichia coli* developing resistance over just 11 days to 1000× inhibitory concentration at outset, Science News, 2016; Baym et al., 2016).

The advent of modern medical technology has accelerated the natural evolution of microorganisms into self-serving and protective communities termed "biofilms," notoriously resistant to conventional treatment (Bjarnsholt, 2013). Therefore, there is urgent need to resurrect old but useful therapies, which were actually curing infection prior to the drug era, which therapies went lost or forgotten. Actually, medicine, due to the successful results of pharmaceutical approaches to treat infectious diseases, has paid scant attention to harnessing the body's own innate defenses to overcome pathogens, whether viral, bacterial, fungal, or other.

Immune cells respond to invaders with an oxidative or respiratory burst (Nguyen et al., 2017; Roos et al., 1988). Macrophages and neutrophils engage, engulf, and destroy pathogens. When encountering an invader, white cells may consume up to 30× the amount of oxygen as at rest. This "burst" induces synthesis of powerful oxidants, actual germicides, enabling lysosomal dissolution of the pathogen. Such oxidants include, but are not limited to, superoxide, hydrogen peroxide, nitric oxide, hydroxyl radical, and peroxynitrate (Prolo et al., 2014). Other reactive species are sodium hypochlorite (bleach), singlet oxygen, and ozone, which is the most powerful of the oxidants, recently discovered to be endogenously generated in humans from singlet oxygen catalyzed by antibodies (Babior et al., 2003).

Might it be possible to heal infection by augmenting these natural defenses? The medical literature cited in this chapter strongly suggests this. Animal life has coexisted on the planet with bacteria for hundreds of millions of years. If bacteria could become resistant to endogenously generated germicides, we would not be here.

Ozone therapy

Ozone (O_3 or triatomic oxygen)

More than 180 years ago, in 1840, triatomic oxygen was discovered by the electrolysis of oxygen creating an odorous gas that its discoverer, Christian Schonbein, called "ozone." Ozone was quickly discovered to destroy bacteria and within a few decades, rapidly gained fame in purifying water. Nikola Tesla patented the first American ozone generator in 1896. However, ozone's utility was hampered at those times by lack of ozone-resistant materials, as the molecule could oxidize naturally occurring hydrocarbons turning them into carbon dioxide and water. The advent of resistant plastics rehabilitated ozone therapy into healing fields by German scientists. In 1958 pediatrician Robert Mayer (1983) of Florida published "Using Ozone as a Chemotherapeutic Agent in the Treatment of Diseases," having learned of the therapy from a German prisoner in the Second World War. More recently, many of its mechanisms of action have been elucidated.

Generation of ozone

Ozone, a metastable pale blue gas, must be generated onsite and used quickly. Its half-life at room temperature is about 30 min, before dismutating back to O_2. O_3 resonates between four ionic structures, highly soluble in water. O_3 is generated from a high voltage corona arc discharge, similar to lightning. Medical "ozone" is actually a mixture of O_2 and O_3, 95–99% and 1–5%, respectively, or 5–70 μg ozone/cc. "Ozone" can be collected in a syringe for parenteral administration, or conducted into a bottle or bag to treat extracorporeal blood, or bubbled through water/saline to make an ozonated solution, or administered to body cavities (Elvis and Ekta, 2011; Shrisel and Suryakant, 2017; Bocci et al., 2012). The advantages and limits of each approach are reported in a subsequent paragraph. However, The U.S. Food and Drug Administration 21 CFR H (801) declares ozone to be a toxic gas with "no known useful medical applications" (FDA, 2020). This "statement" remains in the federal regulations despite thousands of scientific publications, including positive biochemical/metabolic effects in vivo (including humans), highlighting the benefits of ozone therapy. Indeed, ozone gas, when inhaled directly, can damage lungs; therefore, the system used in antimicrobial therapy must not permit leaks which could lead to inhalation. The lungs are singularly highly ozone sensitive.

Ozone antibacterial/antiviral properties

Ozone virtually instantly destroys bacteria, about 100 times faster than chlorine. Unlike chlorine, it leaves no toxic residue. Furthermore, it quickly cuts through and destroys biofilms (Bialoszewski et al., 2011) that are virtually impossible to treat conventionally. Ozone can also be used to control tuberculosis in diabetic patients.

Multiple reports demonstrate the need for viral envelope glycoproteins to be reduced (R-S-H rather than oxidized R-S-S-R) to enter cells, including Ebola (Chandran et al., 2008; Lee and Saphire, 2009) and coronavirus (Broer et al., 2006; Schoeman and Fielding, 2019; Lopez et al., 2008). CMV loses infectivity when oxidized, but 65% is restorable if chemically rereduced (Mirazimi et al., 1999). Many viruses including cytomegalovirus, HIV, Norwalk, bacteriophage MS2, hepatitis A, and poliovirus are nearly instantly inactivated by ozone (Wolf, 2019; Kekez and Sattar, 1997; Herbold et al., 1989; Emerson et al., 1982; Katzenelson et al., 1979; Roy et al., 1981b; Roy et al., 1981a). HIV cell entry is dependent on reduced thiol groups (Ryser et al., 1994; Markovic et al., 2004).

In vivo, ozone instantly reacts with blood components, extinguishing the delivered ozone, leaving only useful O_2 as a residue. The temporary oxidative stress generates reactive oxygen species (peroxides, aldehydes, etc.), which chemically are more electrophilic than sulfur, hence oxidizing the fragile viral thiol groups (ROOH +2 RSH → R-SSR + H2O). As an example, coronavirus spike proteins are rich in sulfhydryl groups and tryptophan (Broer et al., 2006), both very susceptible to oxidative damage (Sharma and Graham, 2010). Therefore, virions are inactivated while retaining their immunogenicity, thus speeding up infection resolution and enhancing natural resistance. Ozone may also damage the lipid component of enveloped virions, a desired effect since viruses cannot self-repair the damaged structures. Lorizate and Kräusslich (2011) discussed that agents that attack the lipid envelope could be useful as antivirals. However, they lamented that conventional agents lack specificity and are "thus unacceptably toxic." Ozone therapy does not so suffer.

Ozone biochemical and immune modulating effects

O_3 is a most powerful oxidant. Two research teams (Italy and Cuba) have independently found major biochemical modulations induced by ozone (Bocci, 2011; Menéndez and Weiser, 2016). Among these effects are: (1) increased red blood cell 2,3 diglycerophosphate (greater hemoglobin oxygen release), (2) improved rheological properties of red blood cells and endothelial nitric oxide production, (3) greater arterial/venous partial O_2 pressure difference, indicating greater mitochondrial consumption of oxygen (energy production), (4) immune system modulation and reduction of inflammation, (5) improvement of antioxidant status, antioxidant enzymes (including superoxide dismutase), and glutathione in cells.

All these effects are achieved by ozone virtually instantly reacting with unsaturated (electron rich) fatty acids and generating ozonides, which act as downstream cell signaling redox molecules and other shorter lived ROS (reactive oxygen species: peroxides, aldehydes, etc.). These affect and modulate redox status without altering blood cell membranes (Bocci, 2011; Menéndez and Weiser,

2016). The authors reported that damage to red cell membranes required higher concentrations of ozone (over 80 μg/cc) than clinically used (15–70 μg/cc). Significant hemolysis has not been reported with any form of ozone therapy.

Conventional thought is that oxidative stress is negative for human health. However, unlike pathological oxidative stress, the controlled oxidative processes of ozone actually induce antioxidant enzyme production. In particular, ozone activates the Nrf2 (nuclear factor erythroid) protein, which activates many protective enzyme pathways (Galiè et al., 2019). In activating the Nrf2 pathway in human multiple sclerosis patients, ozone induces higher levels of antioxidant enzymes, and lower pro-inflammatory peptides TNFα and IL-1β (Delgado-Roche et al., 2017). Ozone also induces heme oxygenase (Pecorelli et al., 2013).

It has been established that the ozone's hydrogen peroxide-mediated natural induction of cytokines by immune cells is safer and more effective than exogenous administration of a single cytokine. Bocci (2011) called ozone the "ideal cytokine inducer" and described ozone-treated erythrocytes as "super gifted," in reference to their enhanced ability to deliver oxygen (their specific function).

Cuban research has demonstrated valuable properties of ozone therapy in infection.

Preconditioning with ozone (ozone treatment before inflicted infection) improved survival of rats to subsequent lethal peritoneal injection of fecal material up to 62.5%. Such preconditioning also rivaled steroids in reducing tumor necrosis factor alpha release in endotoxic shock (Zamora et al., 2005). When antibiotics were added to ozone preconditioning, survival increased to 93% (Bette et al., 2006). Ozone therapy induces prostacyclin production, improving the prostacyclin/thromboxane ratio (Schulz et al., 2012), which would have a positive effect on circulation and reduction in inflammation.

Ozone safety

The largest safety study was conducted in 1980 by Jacobs (1982) for the German Medical Society for Ozone Therapy. She surveyed 2815 known European ozone therapists with a 25% response rate. The report tallied 384,775 patients and 5,579,238 ozone treatments. The risk of adverse incidents was only 0.7 per 100,000 treatments, often related to improper administration. Pediatrician Robert Mayer (1983) reported on 3000 administrations (including intrathecal ozone gas for meningitis), most in children. Mayer found ozone "without any side effects or toxicity noted." Robins (2018) reported performing over 300,000 ozone treatments utilizing direct intravenous gas (DIV) with no lasting ill effects other than vein irritation with this particular method. Rowen (2016) conducted and presented a survey of those he has trained in a delivery method called "hyperbaric ozone." No significant adverse problems reported in 30,000+ observed treatments. Ozone therapy appears to be one of the safest, if not the safest, treatment modalities ever known.

Delivery methods, advantages, and disadvantages

Ozone must be generated on site as its half-life at room temperature is about 30 min. Inhalation must be avoided/minimized. The lungs have very limited capacity to handle such powerful oxidative stress, considering that even breathing 100% O_2 for prolonged periods can be harmful to pulmonary tissues. However, although ozone gas cannot be inhaled, it can be safely bubbled or nebulized through oils for inhalation. This method was used in America's early ozone days, often to reduce tuberculosis organisms in lungs (Eberhart, 1913).

Nevertheless, medical reports suggest that ozone administered to any other body tissue is safe. The most common sites of delivery are to blood, joints, soft tissues, body cavities, subcutaneous, and intramuscular (Bocci, 2011; Menéndez and Weiser, 2016). Delivery is via direct gas injection, gas insufflation, drinking ozonated water, topical, and autohemotherapy (patient's blood treated with ozone outside the body and returned). Local treatments are applied for local conditions.

Ozone gas can be administered into body cavities and spaces such as bladder and joints. For bladder, the volume ranges from 10 cc to 60 cc based on condition, size of patient, and comfort. Concentration of gas ranges between 10 and 40 μg/cc (Bayrak et al., 2014). Joint therapy is universally well accepted to be effective for degenerative and inflammatory joint conditions (Trevisani and Udell, 2015). Typical treatment is preloading the joint (or soft tissue) with a small amount of local anesthetic (preservative and epinephrine free), followed by ozone gas at a concentration of 15–40 μg/cc. A usual knee treatment delivers 10–20 cc. Because ozone is germicidal and antibiotic resistance is not an issue, any type of infection can be treated.

Rectal administration is very inexpensive to administer at home, leaving little or no medical waste. Menendez and Weiser (2016) led Cuban studies indicated that this mode induces similar biochemical effects in the body as blood therapies. This mode improved COVID-19 disease reducing inflammatory markers of fibrinogen, D-dimer, urea, ferritin, LDH, IL-6, and CRP (Fernández-Cuadros et al., 2020).

The dosage depends on the body size of patient, 50–400 cc (volume increased as tolerated) beginning with concentration of 20 μg/cc and increasing to 40–45 μg/cc, as tolerated. The treatment can be performed a few times a week to even 3–4 times daily depending on need. It is generally applied by filling a plastic IV bag with ozone gas, attaching its outlet tube to a reusable catheter, which is placed in the rectum, and the bag contents slowly rolled in. The gas induces a significant bowel evacuation within minutes in most people. However, clinical results are not as rapid or effective, per treatment, as auto-hemotherapy (extracorporeal treatment of blood, which is then returned).

Systemic therapies are used for systemic conditions. There are no published reports of problems in combining methods. The most common systemic delivery method worldwide is direct venous gas (DIV) application. Intravenous oxygen gas therapy has been employed in Europe for generations and has been found to modulate the immune system through generation of 15-Lox-1 by

eosinophils (Chaitidis et al., 2004), and improve prostacyclin production (Stichtenoth et al., 2001). Intravenous oxygen gas therapy has been reported to induce strong immunological effects and improve blood rheology and oxygenation (Schmidt, 2002). DIV ozone is very inexpensive to deliver, requiring (aside from the ozone generator) only a syringe and fine butterfly needle, with minimum medical waste. Hence, DIV has great advantage in the third world where the method is most used. However, the presence of ozone, a powerful oxidant, in the gas mixture, can be irritating to veins and repeated use can lead to vein sclerosis. Dosage generally begins with 20 ccs of gas at concentration of 30–40 μg/cc with increase in volume as tolerated to 100 cc or more. DIV training is important before beginning. Some ozone users in western countries have opposed DIV fearing gas embolism after intravenous administration (Madrid, 2020). However, the gas is not air, which contains about 80% nitrogen, but 100% metabolically active oxygen (O_2/O_3 mixture), which is rapidly consumed.

The most popular delivery in the Western world is major autohemotherapy, which has two modes. The simplest is performed by withdrawing 50–200 cc heparinized blood into a plastic bag under gravity. An equal amount by volume of ozone gas, 20–70 μg/cc, is added and mixed with the blood by gentle rocking. The treated blood is then returned to the patient by gravity (total time about 2 h). Inert glass bottles are preferable to plastic.

A superior method is performed by using a glass evacuated bottle to withdraw up to 200 cc of blood. An equal amount of ozone gas is pumped into the bottle under up to two atmospheres of pressure (2 ATA), mixed, and then rapidly returned to the patient by the pressure. Although there is need for a more sophisticated ozone generator, this method has the advantage of more thorough admixing of gas and blood, and the delivery of actual solubilized oxygen gas in the returned blood, which, as mentioned above, has additional beneficial effects. Hyperbaric ozone therapy is far quicker (10–15 min/treatment) than gravity method. Furthermore, the hyperbaric ozone method (HBO_3) has the additional advantage of permitting additional "passes" of 200 cc of blood up to 10 passes or more, as pioneered by Johann Lahodny, MD of Austria (2017), who called this method "Ozone High Dose Therapy" (OHT), known in America as "10 pass." Very rapid clinical improvement in infected patients has been reported (American Academy, 2017). The disadvantage of both autohemotherapy methods is medical waste.

Ozone minor autohemotherapy is administered by withdrawing up to 10 cc of blood in a syringe prefilled with an equal amount of ozone gas at a desired concentration. The entire mixture is vigorously shaken and quickly returned intramuscular (IM). Bocci (2011) considered IM ozone as an immune stimulant and modulator. Ozone gas can be given directly IM, concentration up to 20 μg/cc with little discomfort. Generally, up to 20 cc of gas is injected. Likewise, the gas can be given to local skin infections, including warts, by subcutaneous route (Bocci, 2011).

Ozone treatment of human acute bacterial infections

Bacteria are virtually instantly destroyed by ozone gas exposure. However, except by local gas injection into a focal infection, all the effects of ozone therapy will be indirect and secondary to its profound modulation of biochemical processes and induction of downstream ozonide metabolites (Bocci, 2011; Menéndez and Weiser, 2016). While we lack desirable infection "double blinded" studies, many reports demonstrate that ozone can be an effective alternative or adjunctive therapy to current conventional drug practices. Local ozone injection and HBO_3 together with an oral antibiotic treatment were reported as the first medical treatment to successfully eradicate a prosthetic joint infection without surgery or parenteral antibiotics (Rowen, 2018a). Single treatment of HBO_3 alone eradicated a tick bite cellulitis in a man previously cleared of Lyme disease years before by HBO_3 without antibiotics (Rowen, 2018b). Used in combination with antibiotics, ozone was reported to prevent resistance to drugs in *Mycobacterium tuberculosis* in humans (Belianin, 1997; Belianin and Shmelev, 1998). Ozone therapy, in conjunction with removal of infected teeth, remitted a highly aggressive case of dermatomyositis (Rowen, 2018c). Topical ozone therapy induced a highly significant reduction in erythrocyte sedimentation rate in osteomyelitis patients compared to control group, though clinical outcomes were similar in both groups (Naderi Nabi et al. (2018). Topical application of ozonized oil has been used successfully against *Staphylococcus aureus*, including MRSA (Song et al., 2018).

Ozone therapy in human viral disease

Medicine has serious challenges in addressing viral diseases. Nevertheless, viruses have particular vulnerabilities to oxidation, as referred in a previous section. Rowen speculated that ozone therapy may be the ideal treatment for controlling viruses, summarizing viral inactivation by oxidation (Rowen, 2019).

Virions can be inactivated by ozone/ozonides while retaining their immunogenicity, quickening infection resolution and enhancing natural resistance. Outstanding clinical effectiveness of ozone therapy has been reported in meetings of ozone doctors sharing clinical work worldwide and in clinical case series of Ebola (Rowen et al., 2016) and coronavirus (Wu et al., 2020; Franzini et al., 2020).

Ozone in pediatric infections

Pediatrician Robert Mayer (1983) reported results from 1946 to 1964 of significant research on 2870 children. In summary, for nonbacterial infectious diarrhea, 1932 resolved multiple days of diarrhea, most in just one day, with one rectal insufflation of gas. He also reported data on intrathecal ozone for 5 viral (mumps, rubeola, St. Louis encephalitis) and two bacterial (*Francisella tularensis* and *Pseudomonas aeruginosa*) meningitis cases. All patients recovered except one; however, the low numbers prevent any statistical consideration. However, the case concerning *P. aeruginosa* is worth mentioning. This was a two-month premature infant, who, on day 5, developed overt meningitis, with spinal fluid WBC count of 13,600. *P. aeruginosa* was detected after culture. The child

received numerous antibiotics, and by day 13, spinal fluid WBC dropped to 450 with still a positive culture. The drugs were discontinued. Mayer provided intrathecal ozone at 89 μg/cc by first removing spinal fluid equal to the volume of gas to be injected. Twelve administrations were performed (intraspinal and intracisternal). The child fully recovered having normal milestones through a year's observations.

Topical ozone

Ozone has been used for decades in several methods of delivery: bathing a wound with the gas in bags, topical application of ozonized water or saline, and ozonized oil. Unsaturated bonds of fatty acids can trap O_3 holding it with stability for months/years. Ozonized vegetable oil has been found to destroy microbes with particular efficacy against *Mycobacterium*, presumably because of its susceptible waxy (fatty) vulnerable coat, and MRSA (Sechi, 2001). The mechanism is a germicidal effect. Lavage by ozonated saline solution proved the most effective irrigating solution (even over antibiotic solution) to prevent bacterial abscess formation in experimental peritonitis rat model (Ozmen et al., 1993). Many oils are available commercially. Saline or aqueous solutions must be made fresh due to dismutation of the dissolved raw gas.

Topical (by bag) and parenteral ozone application was added to a failed antibiotic and debridement regimen for seriously advanced wound infection, with 4/5 of tibia exposed, and pus extruding from knee joint with full resolution (Shah et al., 2011).

Ozone applications have been used successfully as a disinfectant in dentistry for many years. These include ozonized water, ozonized oils, and local injections. These have proven to be antimicrobial, immunostimulating, analgesic, and with no toxic effects on the local tissue environment. This results in a promising approach for "green dentistry" (Nagarakanti and Athuluru, 2011; Mobeen, 2017).

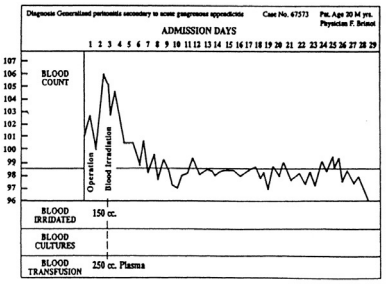
Ultraviolet blood irradiation (UBI)

Historical background

Termed "The Cure That Time Forgot" (Rowen, 1996), UBI has been used continuously in America and overseas for many generations. Ultraviolet energy is a known germicide, used topically at the turn of the last century to cure skin infections. The 1904 Nobel Prize in Physiology or Medicine was awarded to Niels Finsen for his work on UV treatment of various skin diseases. His success rate was 98% in thousands of cases, mostly cutaneous tuberculosis known as lupus vulgaris. Walter Ude reported a series of 100 cases of erysipelas (a cutaneous infection caused by *Streptococcus pyogenes*) in the 1920s, with high cure rates using irradiation of the skin with UV light (Ude, 1929).

American physicist Emmett Knott assembled the known information, and developed a device to irradiate blood extracorporeally. Experimenting on dogs, he settled on a dose of 3.5 cc blood per kg weight and specific exposure times (10 s per aliquot of blood) using a water-cooled boiling mercury bulb emitting 200 watts of energy. The key to the Knott's method was a quartz glass chamber, which induced turbulence to assure maximum exposure to the treated blood. Otherwise, the first few exposed cell layers would absorb all the UV energy. A woman with a septic abortion received the first human treatment. She survived and went on to have two children. Following this case, and availability of his machine, dozens of articles appeared in American literature detailing extremely rapid and stunning resolutions of otherwise (at that time) untreatable bacterial and viral infections (Miley and Christensen, 1948; Barrett, 1940). Most cases resolved in 1–2 sessions. No toxicity was reported in thousands of treatments.

Typical sepsis fever curves and clinical outcomes from UBI therapy, no antibiotics (Rebbeck and Miley, 1941):



Male of 20, who after operation was comatose, in shock, and apparently moribund, with a fulminating toxemia due to generalized peritonitis secondary to a ruptured appendix. Within 24 hours of ultraviolet blood-irradiation therapy detoxification was pronounced and the downhill course of the patient reversed. An eventful convalescence ensued.

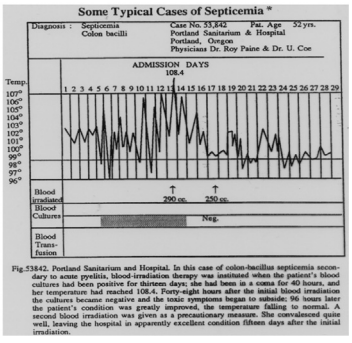


Fig. 53842. Portland Sanitarium and Hospital. In this case of colon-bacillus septicemia secondary to acute pyelitis, blood-irradiation therapy was instituted when the patient's blood cultures had been positive for thirteen days; she had been in a coma for 40 hours, and her temperature had reached 108.4. Forty-eight hours after the initial blood irradiation the cultures became negative and the toxic symptoms began to subside; 96 hours later the patient's condition was greatly improved, the temperature falling to normal. A second blood irradiation was given as a precautionary measure. She convalesced quite well, leaving the hospital in apparently excellent condition fifteen days after the initial irradiation.

UBI was used to treat incomplete septic abortion, puerperal sepsis, acute pyelitis, peritonitis, sepsis, osteomyelitis, wound infections, and infectious thrombophlebitis. In 1947, George Miley and his team summarized 445 cases of acute pyogenic infection, including 151 consecutive cases (Miley and Christensen, 1947). Results showed a 100% recovery in 56 early cases, 98% recovery in 323 moderately advanced cases, and 45% in 66 apparently moribund patients (Table 1). Staphylococcal infections were also successfully treated, providing that sulfa drugs were not simultaneously used, since the sulfa nucleus absorbs UV photons and interferes with the UBI therapy (Miley, 1944).

UBI was reported highly effective in viral pneumonia, hepatitis, polio (Miley and Christensen, 1948), and even Clostridia caused botulism (Miley, 1946).

Table 1 Results in 445 cases of acute pyogenic infection given ultraviolet blood irradiation therapy at the Hahnemann hospital, Philadelphia, over a period of six and one-half years.

	No. of cases	*Recovered*	*Died*
Early			
Puerperal sepsis	3	3	
Incomplete septic abortion	12	12	
Acute furunculosis and carbunculosis	15	15	
Abscesses	2	2	
Acute Strep, hemolytic oropharyngitis	5	5	
Acute tracheobronchitis	12	12	
Acute wound infections	2	2	
Acute pansinusitis	1	1	
Acute pyelitis	1	1	
Acute otitis media	2	2	
Fever of unknown origin	1	1	
Moderately advanced			
Puerperal sepsis	14	14	
Incomplete septic abortion	57	57	
Acute furunculosis and carbunculosis	21	21	
Abscesses	13	13	
Acute Strep hermolytic oropharyngitis	5	5	
Endometritis and parametritis	17	17	
Acute wound infections	6	6	
Salpingitis	15	15	
Pentonitis	18	16	2
Osteomyelitis	26	25	1
Fever or unknown origin	13	13	
Lobar and bronchopneumonia	11	11	
Atypical (virus) pneumonia	10	10	
Preoperative	13	13	
Postoperative	44	41	3
Non-healing wounds	6	6	
Thrombophiebitis	34	34	
Apparently moribund			
Puerperal sepsis	4	3	1
Incomplete septic abortion	2	1	1
Generalized peritonitis	4	2	2
Abscesses Pelvic	6	5	1
Rectal	2	1	1
Scrotum	1	1	
Wound infections	3	2	1
Fever of unknown origin	2	1	1
Lobar and broncopneumonia	2	1	1
Tb. meningitis	3		3
Mesenteric thrombosis, diabetes mellitus	1		1
Septicemias:			
Staph. aurcus-albus with sulfa drugs	7		7
with sulfa drugs	9	9	
Str. hemolyticus	3	3	
Str. non-hemolyticus	2	1	1
Str. viridians–subacute bacterial endocarditis	13		13
Str. hemolyticus endocarditis	1		1
Str. non-hemolyticus endocarditis	1		1
Summary	**Early**	**Moderately Advanced**	**Apparently Moribund**
Number of cases	56	323	66
Number recovered	56	317	30
Percentage recovered	100%	98%	45%

Mechanism of action

UBI efficacy appeared to be consistent with a hormetic effect. Only about 5% of the blood volume was treated; therefore, the effect is not linked to blood sterilization. American physicians reported the following effects from UBI: toxin inactivation, bacteriostatic or bactericidal effect on bacteria, improvement of blood oxygen transport, leukocyte activation, immune stimulation, steroid hormone activation, vasodilation, reduced platelet aggregation, fibrinolysis, decreased blood viscosity, and improved microcirculation (Rowen, 1996). These effects singly and collectively would assist physiology in infection.

Still (2021) functioning Knott Hemo-irradiator, circa 1958

UBI in America largely faded with the advent of pharmaceutical antibiotics but continued in Germany and Russia with the development of less powerful but easily accessible devices (Douglass, 2003; Frick, 1986).

Germans found several metabolic effects, which mirror current findings in ozone therapy biochemistry. Seng (1988) reported the following effects from UBI therapy: (1) improves RBC rheology; (2) increases negative charge across RBC membrane; (3) reduces blood surface tension and platelet aggregation, while raising blood cells number; (4) increases the Pa-Pv O_2 difference and reduces blood pyruvate and lactate [suggesting improved mitochondrial activity]; (5) improves glucose tolerance; (6) reduces blood cholesterol, transaminases, and creatinine values; and (7) increases WBC phagocytosis and bactericidal blood capacity. A typical treatment today uses 60–200 cc of blood, passing it whole or diluted in saline through a quartz glass cuvette exposed to UV energy.

Analogies with ozone treatment

Ozone and UBI therapies have much in common. UV-C frequencies (253 nm) used in therapy are similar to the UV-C radiation that creates ozone naturally in the upper atmosphere. Conversely, ozone releases energy (a photon) when dismutating back to oxygen. Both quickly destroy pathogens and modulate immune function. Positive effects of both treatments can be achieved treating only 5–7% of blood. Better understanding the role of oxidation-related effects exerted by UBI in blood clearly links this treatment with ozone therapy. UBI has been shown to increase white blood cell production of hydrogen peroxide (Ivanov et al., 1989) (as does ozone). Almost identical to the findings of Bocci (2011) and Menendez and Weiser (2016) with ozone, UBI has been found to "highly increase" SOD and glutathione peroxidase while significantly decreasing signals of free radical damage (Dong et al., 2000). UBI may inactivate viral particles (since unlike host cells, viruses do not have repair mechanisms for genetic damage) meanwhile preserving immunogenicity. Furthermore, the cytokine storm that surrounds severe viral infections (Ebola, COVID-19) may be controlled by immune modulation that both these therapies provide (Hamblin, 2017). Ozone's huge advantage over UBI is that it can be locally administered to infected or compromised tissues improving local hypoxia in addition to its other stimulatory effects.

Intravenous hydrogen peroxide therapy

In the 1918 Spanish influenza epidemic, tens of millions died, predominantly of viral pneumonia. There was no treatment then, and little treatment still today for serious viral infections. British physician Oliver in India nearly halved the mortality of viral pneumonia with intravenous hydrogen peroxide. He emphasized that only the worst cases were so treated (Oliver and Murphy, 1920). In the 1980s Charles Farr, conducted informal studies utilizing a 0.03% H_2O_2 concentration in 250 cc 5% dextrose, infused over 1.5 h. The infusions were found to be safe and effectively shortened the duration of seasonal flu illnesses. He taught hundreds of physicians his methods and, to date, no adverse effects have been reported, except peripheral vein irritation (Farr, 1994). The method releases about 25 cc of oxygen gas intravenously. Nascent oxygen released by the infusion is postulated to create a pro-oxidant blood status, creating similar redox messenger molecules as ozone therapy, UBI, and high dose IV Vitamin C. The common protocol is twice weekly for 2–3 weeks, whether viral or bacterial disease.

However, since endothelial cells lack catalase, there is risk of local vein sclerosis at the administration site (as can high dose intravenous ascorbate), and repeated DIV ozone. As a result, H_2O_2 therapy has fallen out of favor among oxidation practitioners, replaced by ozone therapy and UBI. Nevertheless, based on Oliver's work, it may be a lifesaving treatment when another therapy is not possible or available. H_2O_2 can also be administered by a nebulizer (0.06% to 1% in water or saline solution) especially useful in case of pulmonary disease, including COVID-19 (Brownstein et al., 2020).

Hyperbaric oxygen therapy (HBOT)

HBOT is used as either a primary or adjunctive treatment in the management of infections such as gas gangrene, necrotizing fasciitis, diabetic foot infections, refractory osteomyelitis, neurosurgical infections, and fungal infections. HBOT acts as a bactericidal/bacteriostatic agent against anaerobic bacteria by increasing the formation of free oxygen radicals. HBOT restores the bacterial-killing capacity of leukocytes in hypoxic wounds by increasing tissue oxygen tensions (Cimşit et al., 2009).

Like ozone therapy, HBOT works synergistically with conventional antibiotics. One mechanism may be similar to ozone therapy. Breathing 100% oxygen at greater than atmospheric pressure (1 ATA) increases the formation of reactive oxygen species (ROS) (Korpinar and Uzun, 2019). This will create active signaling molecules from immune cells, while being directly active against pathogens. Considering its utility in several aforementioned serious infections, HBOT could be employed in any serious infection as an adjunctive therapy. As oxygen availability and utilization is the most important factor in biological processes, and infection can compromise blood flow, HBOT, by dissolving oxygen in body fluids, may overcome this local or systemic oxygen deprivation.

Typical use is daily at 2–3 ATA, for 1–2 h, until resolution of infection. Complications can include diving-like injury to the eardrum. Oxygen toxicity risk is low due to the relatively short lung exposure to high oxygen tension.

High-dose intravenous ascorbic acid

Ascorbic acid/ascorbate (commonly called "vitamin C") at low doses is an antioxidant acting as an electron donor. High-dose intravenous ascorbate acts as a prodrug for H_2O_2 generation (Chen et al., 2005), explaining observations over decades of its clinical utility in infection.

Septic patients have low circulating ascorbate levels (Wilson and Wu, 2012). Parenteral administration of ascorbate (10–50 g) raises plasma and tissue concentrations and may decrease morbidity.

Mechanism of action

In animal models of sepsis, intravenous ascorbate injection increases survival and protects several microvascular functions: capillary blood flow, microvascular permeability barrier, and arteriolar responsiveness to vasoconstrictors and vasodilators (Kuhn et al., 2018). The effects of parenteral ascorbate on microvascular function are both rapid and persistent. Ascorbate quickly accumulates in microvascular endothelial cells, scavenges reactive oxygen species, and acts through tetrahydrobiopterin to stimulate nitric oxide (NO) production by endothelial nitric oxide synthase, a normal and necessary process (Mortensen and Lykkesfeldt, 2014). However, sepsis can induce high levels of NO by other mechanisms, which can inhibit neutrophil migration and adhesion, and contribute to multiple organ dysfunction (Spiller et al., 2019). Ascorbate may attenuate the effects of excess NO in sepsis and offer organ protection (Oudemans-van Straaten et al., 2014).

Ascorbate may be a valuable adjuvant therapy in sepsis (Wilson, 2009). Ascorbate is important for immune cells, enhancing chemotaxis, ROS generation, and phagocytic destruction of microbes, being also required for apoptosis and clean-up of spent neutrophils. Deficiency results in impaired immunity. High doses (200 mg daily) reduce the severity and duration of the common cold (Carr and Maggini, 2017).

Infection raises metabolic demand for ascorbate. Healthy people may experience loose stools when exceeding 4–8 g orally daily. However, sick patients can tolerate intakes to 200 g daily without any GI distress, indicating far greater need and consumption tolerance of ascorbate when ill (Cathcart, 1981).

Clinical efficacy

Klenner (1949) was one of the first physicians to inject patients with high ascorbate doses. He reported in the 1950s his experiences with parenteral ascorbate, buffering ascorbic acid with sodium bicarbonate to neutralize pH. His schedule for home patients was "2,000 mg (injection) every six hours, supplemented by 1,000–2,000 mg orally every two hours." That amounted to 8000 mg IM, plus oral amounts (depending on sleep time) from 16,000 to 32,000 mg. Klenner reported stunning rapid and complete resolutions of viral diseases inclusive of 60 of 60 polio cases. Patients were often clinically well after 72 h.

Klenner reported on even higher doses ranging from 350 to 1200 mg per kg body weight to treat illness using parenteral ascorbate to cure measles, herpes simplex, viral encephalitis, and mononucleosis. He emphasized the need to maintain continuous high levels during the acute phase of the infection. Small children were given 2–3 g IM every two hours in the buttocks. Large doses, up to 60 g, could be added to 500 cc water together with other vitamins such as 500 mg thiamin HCl, 300 mg pyridoxine, 400 mg calcium pantothenate, 100 mg riboflavin, and 300 mg niacinamide, given once or twice daily.

Typical applications would be 25 g of ascorbate (buffered to neutral pH) into 500 ml of sterile water (18 cc diluent to each gram of ascorbate are necessary to ensure correct osmolality). He reported clearing viral pneumonia easily with parenteral treatments. Klenner also gave alkaline drinks to slow ascorbate renal excretion. With 350–400 mg/kg ascorbate every 2 h, he reported quick resolution of measles and chicken pox, sometimes requiring only a few treatments. Complications included vein irritation (IV route), subcutaneous thickening (local injections). Large oral doses could cause diarrhea, and genital or anal rash and itch (Smith, n.d.;

Klenner, 1971). To prevent these effects, Klenner often added small amounts of IM corticosteroids (desoxycorticosterone), 2.5 mg for children and 5 mg for adults. This combination therapy has been reconsidered in 2017 by Prof. Paul Marik who reported that intravenous vitamin C (1.5 g) with hydrocortisone (50 mg) and intravenous thiamine (200 mg) can reduce sepsis mortality from 40% to about 8% (Marik et al., 2016).

Conclusions

Generation of reactive oxygen species (ROS) may be promising for modulating the immune system and controlling infections, including those caused by drug-resistant organisms. Ozone therapy, Ultraviolet Blood Irradiation Therapy, intravenous ascorbate, H_2O_2, as well as Hyperbaric Oxygen Therapy are interesting therapies to be explored and exploited both as alternatives to, or in association with antimicrobials, providing that very uncommon side effects are kept in mind.

However, it should be emphasized that an important role in infection control is played by the innate immune defenses, and that the latter can be boosted not only with oxidants but also with correct nutrition. Vitamin A, zinc, iodine, and folate deficiencies, affecting 2 billion people in developing countries, impair the immune response, being a major contributor to severe infections and death (Katona and Katona-Apte, 2008). Micronutrient deficiencies (vitamins and minerals) are also common in the USA (Medical Economics, 2016; CDC.gov, 2012). Vitamins (A, B12, C, D, E, essential fatty acids) are important for ensuring the first defenses at the mucosal barrier level. Metals like zinc, iron, copper, and selenium all contribute to a correct catalytic/enzymatic activity ultimately helping immune functions. Besides these evident nutritional deficiencies, incorrect diet can alter immune defenses. As an example, the ingestion of simple carbohydrates, such as monosaccharides, can decrease the phagocytic index (Sanchez et al., 1973). Also, heavy metals (present from contamination or unwanted agricultural residues) can impair the immune system and reduce host resistance to invading pathogens (Lehmann et al., 2011; Fenga et al., 2017). Simultaneously, some foods possess useful antibacterial properties, due to their polyphenolic or other compounds. Among these are manuka honey (Johnston et al., 2018) and olive polyphenols. The latter displays antiviral activity in vitro (Bisignano et al., 1999; Rounds et al., 2012) including HIV (Bedoya et al., 2016) and SARS-CoV-2 transmission (Ergoren et al., 2020). In these changing times when important challenges such as multiple drug resistance arise, exploring new strategies to counteract infection is an urgent issue. Unfortunately, only few quantitative studies on old methods or alternative plant molecules are available in the literature. This may be because in vivo studies are expensive; and the final result, although successful, is not patentable. However, the natural world is rich in molecules that can have activity against microbes. The time is ripe to extend detailed and quantitative investigation to the myriad of compounds that can be used alone or in synergy with antibiotics or oxidation therapies. These sometimes "unapproved" but potentially lifesaving therapies can be the key for treating conventionally untreatable patients.

References

American Academy of Ozonotherapy (2017) *Annual Meeting, Dallas Texas.*

Babior B, Takeuchi C, Ruedi J, et al. (2003) Investigating antibody-catalyzed ozone generation by human neutrophils. *Proceedings. National Academy of Sciences. United States of America* 100: 3031–3034.

Barrett H (1940) The irradiation of autotransfused blood by ultraviolet spectral energy: Results of therapy in 110 cases. *Medical Clinics of North America* 723–732. May.

Baym M, Lieberman T, Kelsic E, et al. (2016) Spatiotemporal microbial evolution on antibiotic landscapes. *Science* 353(9). https://doi.org/10.1126/science.aag0822.

Bayrak O, Erturhan S, Seckiner I, Erbagci A, Ustun A, and Karakok M (2014) Chemical cystitis developed in experimental animals model: Topical effect of intravesical ozone application to bladder. *Urology Annals* 6(2): 122–126. https://doi.org/10.4103/0974-7796.130553.

Bedoya L, Beltran M, Obregon-Calderon P, et al. (2016) Hydroxytyrosol: a new class of microbicide displaying broad anti-HIV-1 activity. *AIDS* 30(18): 2767–2776. https://doi.org/10.1097/QAD.0000000000001283. 27677167. PMC5106087.

Belianin I (1997) The action of blood ozonization on the course of progressive pulmonary tuberculosis combined with diabetes mellitus. *Terapevticheskiĭ Arkhiv* 69: 44–48.

Belianin I and Shmelev E (1998) Blood ozonation in the treatment of patients with progressive pulmonary tuberculosis concurrent with diabetes mellitus. *Problemy Tuberkuleza* (1): 30–33.

Bette M, Nusing R, Mutters R, Zamora Z, Menendez S, and Schulz S (2006) Efficiency of tazobactam/piperacillin in lethal peritonitis is enhanced after preconditioning of rats with O_3/O_2-pneumoperitoneum. *Shock* 25: 23–29.

Bialoszewski D, Pietruczuk-Padzik A, Kalicinska A, et al. (2011) Activity of ozonated water and ozone against *Staphylococcus aureus* and *Pseudomonas aeruginosa* biofilms. *Medical Science Monitor* 17: Br339-334.

Bisignano G, Tomaino A, Lo Cascio R, et al. (1999) On the in-vitro antimicrobial activity of oleuropein and hydroxytyrosol. *The Journal of Pharmacy and Pharmacology* 51(8): 971–974. https://doi.org/10.1211/0022357991773258. 10504039.

Bjarnsholt T (2013) The role of bacterial biofilms in chronic infections. *APMIS. Supplementum* (136): 1–51. https://doi.org/10.1111/apm.12099. May 23635385.

Bocci V (2011) *OZONE: A New Medical Drug*, 2nd edn, Springer.

Bocci V, Zanardi I, Borelli E, et al. (2012) Reliable and effective oxygen-ozone therapy at crossroads with ozonated saline infusion and rectal insufflation. *The Journal of Pharmacy and Pharmacology* 64(4): 482–489.

Broer R, Boson B, Spaan W, Cosset F, and Corver J (2006) Important role for the transmembrane domain of severe acute respiratory syndrome coronavirus spike protein during entry. *Journal of Virology* 80(3): 1302–1310.

Brownstein D, Ng R, Rowen R, et al. (2020) A novel approach to treating COVID-19 using nutritional and oxidative therapies. *Science, Public Health Policy, and The Law* 2: 4–22.

Carr A and Maggini S (2017) Vitamin C and immune function. *Nutrients* 9(11): 1211. https://doi.org/10.3390/nu9111211. 29099763. PMC5707683.

Carson C, Hammer K, and Riley T (2006) Melaleuca alternifolia (Tea Tree) oil: a review of antimicrobial and other medicinal properties. *Clinical Microbiology Reviews* 19(1): 50–62. https://doi.org/10.1128/CMR.19.1.50-62.2006.

Cathcart R (1981) Vitamin C, titrating to bowel tolerance, anascorbemia, and acute induced scurvy. *Medical Hypotheses* 7(11): 1359–1376.

CDC.gov (2012) Second National Report on Biochemical Indicators of Diet and Nutrition in the U.S. Population. Available at: https://www.cdc.gov/nutritionreport/pdf/Nutrition_Book_complete508_final.pdf. Accessed on 22-6-2012.

Chaitidis P, Kreutzer F, Gerth C, Janata P, and Kuhn H (2004) Impact of intravenous oxygen therapy on the expression of reticulocyte-type 15-lipoxygenase in human volunteers. *Prostaglandins, Leukotrienes, and Essential Fatty Acids* 71(5): 271–276.

Chandran K, Li Q, Fabozzi G, Sullivan N, and Cunningham J (2008) Cysteine cathepsins are host factors for filovirus infection. In: *Keystone Symposia: Cell Biology of Virus Entry, Replication and Pathogenesis*, p. 29. Victoria, BC, Canada: Keystone Symposia.

Chen Q, Epsey M, Krishna M, et al. (2005) Pharmacologic ascorbic acid concentrations selectively kill cancer cells: action as a pro-drug to deliver hydrogen peroxide to tissues. *Proceedings of the National Academy of Sciences of the United States of America* 102(38): 13604–13609.

Cimşit M, Uzun G, and Yildiz S (2009) Hyperbaric oxygen therapy as an anti-infective agent. *Expert Review of Anti-Infective Therapy* (8): 1015–1026. https://doi.org/10.1586/eri.09.76. 19803709.

Delgado-Roche L, Riera-Romo M, Mesta F, Hernández-Matos Y, Barrios J, Martínez-Sánchez G, and Al-Dalaien S (2017) Medical ozone promotes Nrf2 phosphorylation reducing oxidative stress and pro-inflammatory cytokines in multiple sclerosis patients. *European Journal of Pharmacology* 811: 148–154. https://doi.org/10.1016/j.ejphar.2017.06.017 Sep 15. Epub 2017 Jun 13 28623000.

Dong Y, Shou T, Zhou Y, Jiang S, and Hua X (2000) Ultraviolet blood irradiation and oxygenation affects free radicals and antioxidase after rabbit spinal cord injury. *Chinese Medical Journal* 113: 991–995.

Douglass W (2003) *Into The Light*. S.A.: Rhino Publishing.

Eberhart N (1913) *A Working Manual of High Frequency Currents*. 218–288. Available at: https://twotowers.com/documents/a_working_manual_of_high_frequency_currents.pdf. Accessed 22-6-21.

Elvis A and Ekta J (2011) Ozone therapy: A clinical review. *Journal of Natural Science, Biology and Medicine* 2(1): 66–70. https://doi.org/10.4103/0976-9668.82319.

Emerson M, Sproul O, and Buck C (1982) Ozone inactivation of cell-associated viruses. *Applied and Environmental Microbiology* 43: 603–608.

Ergoren M, Paolacci S, Manara E, et al. (2020) A pilot study on the preventative potential of alpha-cyclodextrin and hydroxytyrosol against SARS-CoV-2 transmission. *Acta Biomed* 91(13-S): e2020022. https://doi.org/10.23750/abm.v91i13-S.10817 Nov 9 33170176. PMC8023124.

Farr C (1994) *Annual Meetings of International Oxidation Medicine Association, Anchorage Alaska, May.*

FDA. CFR Title 21. (2020) Available at: https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/cfrsearch.cfm?fr=801.415. Accessed 22-06-2021.

Fenga C, Gangemi S, Di Salvatore V, et al. (2017) Immunological effects of occupational exposure to lead (Review). *Molecular Medicine Reports* 15: 3355–3360. https://doi.org/10.3892/mmr.2017.6381.

Fernández-Cuadros M, Albaladejo-Florín M, Álava-Rabasa S, et al. (2020) Effect of rectal ozone (O_3) in severe COVID-19 pneumonia: Preliminary results [published online ahead of print, Aug 3]. *SN Comprehensive Clinical Medicine* 1–9. https://doi.org/10.1007/s42399-020-00374-1.

Franzini M, Valdenassi L, Ricevuti G, et al. (2020) Oxygen-ozone (O_2-O_3) immunoceutical therapy for patients with COVID-19. Preliminary evidence reported. *International Immunopharmacology* 88: 106879. https://doi.org/10.1016/j.intimp.2020.106879.

Frick G (1986) A Linke: Die Ultraviolet bestrahlung des Blutes, ihre Entwicklung und derzeitiger Stand. *Zschr Arztl Forth* 80.

Galiè M, Covi V, Tabaracci G, and Malatesta M (2019) The role of Nrf2 in the antioxidant cellular response to medical ozone exposure. *International Journal of Molecular Sciences* 20(16): 4009. https://doi.org/10.3390/ijms20164009. 31426459. PMC6720777.

Hamblin M (2017) Ultraviolet irradiation of blood: "The cure that time forgot"? *Advances in Experimental Medicine and Biology* 996: 295–309. https://doi.org/10.1007/978-3-319-56017-5_25. 29124710. PMC6122858.

Herbold K, Flehmig B, and Botzenhart K (1989) Comparison of ozone inactivation, in flowing water, of hepatitis A virus, poliovirus 1, and indicator organisms. *Applied and Environmental Microbiology* 55: 2949–2953.

Ivanov E, Kapshienko I, and Tril N (1989) Effect of the UV irradiation of autologous blood on the humoral link in the immune response of patients with chronic inflammatory processes. *Voprosy Kurortologii, Fizioterapii, i Lechebnoĭ Fizicheskoĭ Kultury* 1: 45–47.

Jacobs M (1982) Zwischenfalle und typische komplikationen in der ozone-sauerstoff-therapie. *OzoNachrichten* 1: 5.

Johnston M, McBride M, Dahiya D, Owusu-Apenten R, and Nigam P (2018) Antibacterial activity of Manuka honey and its components: An overview. *AIMS Microbiology* 4(4): 655–664. https://doi.org/10.3934/microbiol.2018.4.655.

Katona P and Katona-Apte J (2008) The Interaction between Nutrition and Infection. *Clinical Infectious Diseases* 46(10): 1582–1588. https://doi.org/10.1086/587658.

Katzenelson E, Koerner G, Biedermann N, Peleg M, and Shuval H (1979) Measurement of the inactivation kinetics of polio virus by ozone in a fast-flow mixer. *Applied and Environmental Microbiology* 37: 715–718.

Kekez M and Sattar S (1997) A new ozone-based method for virus inactivation: Preliminary study. *Physics in Medicine and Biology* 42: 2027–2039.

Klenner F (1949) *The treatment of poliomyelitis and other virus disease with vitamin C*. Southern Medicine and Surgery. July.

Klenner F (1971) Observations on the dose and administration of ascorbic acid when employed beyond the range of a vitamin in human pathology. *Journal of Applied Nutrition* 23(3 & 4) Winter. Available at: https://yost.com/health/klenner/klenner-1971.html.

Korpinar S and Uzun H (2019) The effects of hyperbaric oxygen at different pressures on oxidative stress and antioxidant status in rats. *Medicina* 55(5): 205. https://doi.org/10.3390/medicina55050205.

Kuhn S, Meissner K, Mayes L, and Bartels K (2018) Vitamin C in sepsis. *Current Opinion in Anaesthesiology* 31(1): 55–60. https://doi.org/10.1097/ACO.0000000000000549.

Lahodny J (2017) High dose ozone therapy. In: *World conference on ozone therapy in medicine, dentistry and veterinary – Ancona, Italy. September.*

Lee J and Saphire E (2009) Ebolavirus glycoprotein structure and mechanism of entry. *Future Virology* 4: 621–635.

Lehmann I, Sack U, and Lehmann J (2011) Metal ions affecting the immune system. *Metal Ions in Life Sciences* 8: 157–185. 21473381.

Lopez L, Riffle A, Pike S, Gardner D, and Hogue B (2008) Importance of conserved cysteine residues in the coronavirus envelope protein. *Journal of Virology* 82: 3000–3010.

Lorizate M and Kräusslich H (2011) Role of lipids in virus replication. *Cold Spring Harbor Perspectives in Biology* 3(10): a004820. https://doi.org/10.1101/cshperspect.a004820. 21628428. PMC3179339.

Madrid declaration on Ozone Therapy. (2020) Available at: https://isco3.org/madrid-declaration-on-ozone-therapy-3rd-edition-isco3/. Accessed 22-06-2021

Marik P, Khangoora V, Rivera R, Hooper M, and Catravas J (2016) Hydrocortisone, vitamin C, and thiamine for the treatment of severe sepsis and septic shock: A retrospective before-after study. *Chest* 151(6): 1229–1238. https://doi.org/10.1016/j.chest.2016.11.036. Epub 2016 Dec.

Markovic I, Stantchev T, Fields K, Tiffany L, Tomic M, et al. (2004) Thiol/disulfide exchange is a prerequisite for CXCR4-tropic HIV-1 envelope-mediated T-cell fusion during viral entry. *Blood* 103: 1586–1594.

Mayer R (1983) Experiences of a pediatrician using in the treatment of diseases in children and animals. In: *International Ozone Association World Congress, May, Wash. DC* Abstract available at: http://www.rexresearch.com/o/oz1-9.htm. Accessed 22-06-21 Full Mayer Monograph available at: https://ozonewithoutborders.ngo/wp-content/uploads/2021/06/Experiences-of-a-Pediatrician-Robert-Mayer.pdf. Accessed 24-06-2021.

Medical Economics (2016) The American Nutrient Gap: And How Vitamin and Mineral Supplements Can Help Fill It. Available at: https://www.medicaleconomics.com/view/american-nutrient-gap-and-how-vitamin-and-mineral-supplements-can-help-fill-it. Accessed 22-06-2021.

Menéndez S and Weiser M (2016) *Advances of Ozone Therapy in Medicine and Dentistry*. Santa Barbara, CA, USA: Ozone Therapies Group.

Miley G (1944) Efficacy of ultraviolet blood irradiation therapy and control of Staphylococcemias. *American Journal of Surgery* 64(3): 313–322.

Miley G (1946) Recovery from botulism coma following ultraviolet blood irradiation. *The Review of Gastroenterology* 13(1): 17–18.

Miley G and Christensen J (1947) Ultraviolet blood irradiation therapy: Further studies in acute infections. *American Journal of Surgery* 73(4): 486–493.

Miley G and Christensen J (1948) Ultraviolet blood irradiation therapy in acute virus and virus-like infections. *The Review of Gastroenterology* 15(4): 271–276.

Mirazimi A, Mousavi-Jazi M, Sundqvist V, and Svensson L (1999) Free thiol groups are essential for infectivity of human cytomegalovirus. *The Journal of General Virology* 80: 2861–2865.
Mobeen A (2017) Ozone therapy in oral diseases: A reply to antibiotic resistance. *World Journal of Pharmaceutical Research* 1476–1481.
Mortensen A and Lykkesfeldt J (2014) Does vitamin C enhance nitric oxide bioavailability in a tetrahydrobiopterin-dependent manner? In vitro, in vivo and clinical studies. *Nitric Oxide* 36: 51–57. https://doi.org/10.1016/j.niox.2013.12.001 Epub. 2013 Dec 9 24333161.
Naderi Nabi B, Sedighinejad A, Mirbolouk A, Farzi F, Haghighi M, et al. (2018) The effectiveness of ozone therapy in chronic osteomyelitis: A randomized controlled clinical trial. *Archives of Clinical Infectious Diseases* 13(2): e61320. https://doi.org/10.5812/archcid.61320.
Nagarakanti S and Athuluru D (2011) Ozone: A new revolution in dentistry. *Webmed Central Dentistry* 2(12): WMC002685. https://doi.org/10.9754/journal.wmc.2011.002685.
Nguyen G, Green E, and Mecsas J (2017) Neutrophils to the ROScue: Mechanisms of NADPH oxidase activation and bacterial resistance. *Frontiers in Cellular and Infection Microbiology* 7: 373. https://doi.org/10.3389/fcimb.2017.00373. Aug 25 28890882. PMC5574878.
Oliver T and Murphy D (1920) Influenzal pneumonia: The intravenous injection of hydrogen peroxide. *The Lancet* 195(5034): 432–433.
Oudemans-van Straaten H, Spoelstra-de Man A, and de Waard M (2014) Vitamin C revisited. *Critical Care* 18(4): 460. https://doi.org/10.1186/s13054-014-0460-x. 25185110. PMC4423646.
Ozmen V, Thomas W, Healy J, et al. (1993) Irrigation of the abdominal cavity in the treatment of experimentally induced microbial peritonitis: Efficacy of ozonated saline. *The American Surgeon* 59(5): 297–303. 8489098.
Pecorelli A, Bocci V, Acquaviva A, Belmonte G, et al. (2013) NRF2 activation is involved in ozonated human serum upregulation of HO-1 in endothelial cells. *Toxicology and Applied Pharmacology* 267: 30–40.
Prolo C, Alvarez M, and Radi R (2014) Peroxynitrite, a potent macrophage-derived oxidizing cytotoxin to combat invading pathogens. *BioFactors* 40(2): 215–225. https://doi.org/10.1002/biof.1150.
Rebbeck E and Miley G (1941) *Review of Gastroenterology*. January-February, 1943 11.
Robins H (2018) *Ozone Without Borders Annual Meeting, Costa Rica. April.*
Roos D, Lutter R, and Bolscher B (1988) The respiratory burst and the NADPH oxidase of phagocytic leukocytes. In: Sbarra AJ and Strauss RR (eds.) *The Respiratory Burst and Its Physiological Significance*. Boston, MA: Springerhttps://doi.org/10.1007/978-1-4684-5496-3_3.
Rounds L, Havens C, Feinstein Y, et al. (2012) Plant extracts, spices, and essential oils inactivate Escherichia coli O157:H7 and reduce formation of potentially carcinogenic heterocyclic amines in cooked beef patties. *Journal of Agricultural and Food Chemistry* 60(14): 3792–3799. https://doi.org/10.1021/jf204062p.
Rowen R (1996) Ultraviolet blood irradiation therapy (Photo-Oxidation) the cure that time forgot. *International Journal of Biosocial and Medical Research* 14(2): 115–132.
Rowen R (2016) Safety of hyperbaric ozone therapy. In: *Presented at American Academy of Ozonotherapy annual meeting. Dallas, Texas April.*
Rowen R (2018a) Ozone therapy in conjunction with oral antibiotics as a successful primary and sole treatment for chronic septic prosthetic joint: Review and case report. *Medical Gas Research* 8: 67–71.
Rowen R (2018b) Ozone therapy as a primary and sole treatment for acute bacterial infection: case report. *Medical Gas Research* 8(3): 121–124.
Rowen R (2018c) Remission of aggressive autoimmune disease (dermatomyositis) with removal of infective jaw pathology and ozone therapy: review and case report. *Autoimmunity Highlights* 9(1): 7.
Rowen R (2019) Ozone and oxidation therapies as a solution to the emerging crisis in infectious disease management: A review of current knowledge and experience. *Medical Gas Research* 9(4): 232–237.
Rowen R, Robins H, Carew K, et al. (2016) Rapid resolution of hemorrhagic fever (Ebola) in Sierra Leone with ozone therapy. *African Journal of Infectious Diseases* 10: 49–54.
Roy D, Chian E, and Englebrecht R (1981a) Kinetics of enteroviral inactivation by ozone. *Journal of the Environmental Engineering Division* 107(5).
Roy D, Wong P, Engelbrecht R, and Chian E (1981b) Mechanism of enteroviral inactivation by ozone. *Applied and Environmental Microbiology* 41: 718–723.
Ryser H, Levy E, Mandel R, and Disciullo G (1994) Inhibition of human immunodeficiency virus infection by agents that interfere with thiol-disulfide interchange upon virus-receptor interaction. *Proceedings. National Academy of Sciences. United States of America* 91: 4559–4563.
Sanchez A, Reeser J, Lau P, et al. (1973) Role of sugars in human neutrophilic phagocytosis. *The American Journal of Clinical Nutrition* 26(11): 1180–1184.
Schmidt H (2002) Regelsberger's intravenous oxygen therapy–an interpretation of results in practice from a biochemical and physiological point of view. *Forschende Komplementärmedizin und Klassische Naturheilkunde* 9(1): 7–18.
Schoeman D and Fielding B (2019) Coronavirus envelope protein: Current knowledge. *Virology Journal* 16: 69.
Schulz S, Ninke S, Watzer B, and Nusing R (2012) Ozone induces synthesis of systemic prostacyclin by cyclooxygenase-2 dependent mechanism in vivo. *Biochemical Pharmacology* 83(4): 506–513.
Science News (2016) Available at: https://www.youtube.com/watch?v=yybsSqcB7mE. Accessed 22-06-2021.
Sechi L (2001) Antibacterial activity of ozonized sunflower oil (Oleozon). *Journal of Applied Microbiology* 90: 279–284.
Seng G (1988) Hernatogenic Oxydationstherapie, *Therapeuticon Six*, June: 370-373.
Shah P, Shyam A, and Shah S (2011) Adjuvant combined ozone therapy for extensive wound over tibia. *Indian Journal of Orthopaedics* 45(4): 376–379.
Sharma V and Graham N (2010) Oxidation of Amino Acids, Peptides and Proteins by Ozone: A Review. *Ozone: Science & Engineering* 32(2): 81–90.
Shrisel H and Suryakant S (2017) Ozone therapy: An excellent treatment for various diseases. *International Journal of Pharmacy and Pharmaceutical Research* 10(3): 303–310.
Smith L (n.d.) Clinical Guide to the Use of Vitamin C. The Clinical Experiences of Frederick R. Klenner, M.D. abbreviated, summarized and annotated by Lendon H. Smith, M.D. Available at: http://vitaminc.co.nz/pdf/CLINICAL-GUIDE-TO-THE-USE-OF-VITAMIN-C-FREDERICK-KLENNER-MD.pdf. Accessed 22-06-2021
Song M, Zeng Q, Xiang Y, et al. (2018) The antibacterial effect of topical ozone on the treatment of MRSA skin infection. *Molecular Medicine Reports* 17(2): 2449–2455.
Spiller F, Formiga R, Coimbra J, et al. (2019) Targeting nitric oxide as a key modulator of sepsis, arthritis and pain. *Nitric Oxide* 89: 32–40. https://doi.org/10.1016/j.niox.2019.04.011. Epub Apr 30. 31051258.
Stichtenoth D, Kreutzer F, Gutzki F, Tsikas D, Nowak V, and Frölich J (2001) Effects of intravenous oxygen on prostacyclin and thromboxane formation in patients with peripheral occlusive arterial disease. *Prostaglandins, Leukotrienes, and Essential Fatty Acids* 65(4): 211–214.
Trevisani V and Udell J (2015) *American College of Rheumatology (ACR) Annual Meeting: Abstract 311*. Presented November 8.
Ude W (1929) Ultraviolet radiation therapy in erysipela. *Radiology* 13: 504.
Wilson J (2009) Mechanism of action of vitamin C in sepsis: Ascorbate modulates redox signaling in endothelium. *BioFactors* 35(1): 5–13. https://doi.org/10.1002/biof.7. 19319840. PMC2767105.
Wilson J and Wu F (2012) Vitamin C in sepsis. *Sub-Cellular Biochemistry* 56: 67–83. https://doi.org/10.1007/978-94-007-2199-9_5. 22116695.
Wolf W (2019) Inactivation of waterborne viruses by ozone: Kinetics and mechanisms. Available at: https://infoscience.epfl.ch/record/268519?ln=en.
Wu J, Tan CS, Yu H, et al. (2020) Recovery of four COVID-19 patients via ozonated autohemotherapy. *Innovation (N Y)* 1(3): 100060. https://doi.org/10.1016/j.xinn.2020.100060.
Zamora Z, Borrego A, López O, et al. (2005) Effects of ozone oxidative preconditioning on TNF-alpha release and antioxidant-prooxidant intracellular balance in mice during endotoxic shock. *Mediators of Inflammation* 1: 16–22.