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# Potential mechanisms by which the oxygen-ozone ($O_2$-$O_3$) therapy could contribute to the treatment against the coronavirus COVID-19

**Luigi Valdenassi [1,2], Marianno Franzini [1,2], Giovanni Ricevuti [2], Luca Rinaldi[3], Umberto Tirelli [2,4]**

[1]Oxygen-Ozone Therapy Scientific Society (SIOOT), Gorle, Italy
[2]University of Pavia, Pavia, Italy
[3]Department of Advanced Surgical and Medical Sciences, University of Campania Luigi Vanvitelli, Naples, Italy.
[4]Tirelli Medical Group, Pordenone, Italy Clinic.

**Corresponding author**: Umberto Tirelli, MD, utirelli@cro.it

On 31 December, at the end of 2019, some cases of pneumonia of unknown etiology were notified to the World Health Organization (WHO) Country Office in China, regarding Wuhan, a city of Hubei province[1]. This unknown agent, a few weeks later, was identified as part of the coronavirus family and named 2019 novel coronavirus. Since then, the epidemic of 2019 novel coronavirus (currently renamed SARS-CoV-2) and causing the disease Covid-19 has expanded from Wuhan throughout China and is being exported to a growing number of countries, with an increasing number of other cases with different rate of transmission[2].

Biochemical and pharmacological characteristics of ozone provide reasons for considering this molecule useful in the treatment of several viral infections [1], specifically in the treatment of COVID-19.

Ozone is an oxidant which shows a paradoxical activity when in contact with organic molecules, thus causing a powerful antioxidant response[3].

In fact, reacting with target substrates in biological fluids (PUFA and GSH), leads to the creation of hydroperoxides and aldehydes. Among them, 4-HNE (4 hydroxyl - nonenal) is a key element in signal transduction, involved in upregulation of glutathione and also in enhancing the resistance to apoptosis resulting from pro-oxidant agents.

It causes a significant adaptive stress response, by stimulating anti-oxidizing and detoxifying enzymes expression.

The 4-HNE partially excreted by the liver and the kidneys, is mostly attached to Cys 34 albumin molecule, GSH and cysteine[4].

These molecules through circulation easily transfer 4-HNE into the cytoplasm of many cells.

Cells cytoplasm contains an inactive transcription factor called Nrf2, bounded to a larger inactive factor containing SH groups called Keap-1, rich in cysteine.

When attached to Cys 273 or Cys 288 of Keap-1, 4-HNE releases the key molecule Nrf 2 (Nuclear Factor Erythroid 2-Related Factor2). This leads to several anti-oxidizing enzymes expression: SOD, GPx, GST, CAT, HO-1, NQO-1, HSP and phase II drug metabolism enzymes[4].

HO-1 catalyzes the degradation of heme to carbon monoxide (CO), which modulates NF-KB determining a decreased pro-inflammatory cytokines expression and anti-inflammatory cytokines direct induction.

Ozone shows an anti-oxidizing and anti-inflammatory action, being NF-KB and Nrf2 transcription agents which modulate gene expression of pro-inflammatory and anti-inflammatory cytokines.

The accumulation of LOPs and 4-HNE during oxidative stress and in the presence of disease, generates a feedback mechanism which transmits signals and stimulates networks capable of stopping critical oxidation events, common to several conditions.

By reacting with PUFA and aldehydes, ozone generates hydroperoxides and particularly $H_2O_2$., it rapidly spreads through cells of the immune system. It also bioregulates signal transduction thus promoting immune responses, modulating interferon and interleukins through the activation of NF-KB, thus increasing the release of cytokines.

This process is part of an endogenic system. In 2002 Lerner and Wentworth have underlined the fact that the human body is able to produce ozone in order to protect itself from infectious agents. This happens by involving neutrophils and antibodies of the immune system which by producing ozone, use its oxidizing power in order to destroy bacteria and viruses present on cell walls[5-7].

Tanaka has shown how flu viruses can be inactivated by low concentrations of ozone in the environment and on smooth surfaces.

Other studies have shown that ozone can play a determining role against bacteria, viruses and fungi diseases[3-8].

Byron K. Murray and others have highlighted a decrease of viral infectivity after exposure to ozone. This causes lipidic peroxidation of virus capsid, thus interrupting its reproductive cycle, preventing the necessary contact between the virus and the receptor.

Other studies have shown how ozone can inactivate virus strains with or without envelope[8].

Some strains like HSV-1 (Herpes Simplex type 1 Mc Intyre) and VSV (Vesicular Stomatitis Virus Indiana) after being ozonized have shown a 6 LOG 10 reduction of infectious particles in 15 minutes.

VAC strains (Elstree strain) and H1 N1 (Influenza A), have shown a reduction up to 5 LOG 10 respectively in 40 and 30 minutes. These results show important changes in different virus strains morphology[8].

In addition to the anti-oxidizing, anti-inflammatory and anti-viral action we can consider the relevant activity of stimulation and immune response linked to NFAT, transcription factor linked to different cytokines (IL-2, IL-6, TNF-Alfa e IFN-Gamma) with its support to lymphocytes and macrophages, forming the first line of defense.

Another important characteristic of ozone therapy against COVID-19 infection is shown by the contrast ability toward severe hypoxemia, typical of this virus[17].

Tests carried out using NIRS spectroscopy, led to increased oxygenation (in the given case, cerebral) shown by an increase of oxygenated hemoglobin and constant values of the non-oxygenated one[18].

Ozone is a molecule which acts on different levels and in different physiopathological fields. Therefore we believe that it would be useful to propose this method as a support to the drug therapy currently in treatment against viral infections in general and particularly against COVID-19 and within a integrative medicine approach[10-11].

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