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High-dose Intravenous Vitamin C Treatment for COVID-19

by Adnan Erol, M.D.

(OMNS Mar 21, 2020) The evidence about COVID-19 pneumonia and well-established knowledge about related conditions suggests it is caused by the hyperactivation of immune effector cells. High-dose vitamin C may suppress these immune system effectors. As intravenous high-dose vitamin C treatment is known to be safe, this suggests that intravenous high-dose vitamin C may be the treatment of choice in the early stages of COVID-19.

Coronaviruses (CoVs) are large, enveloped, and positive sense RNA viruses that infect a broad range of vertebrates and cause disease of medical and veterinary significance. Human respiratory corona viruses have been known since the 1960s to circulate worldwide and to cause respiratory infection with rather mild symptoms, suggesting that they are well-adapted to the human host. However, zoonotic coronaviruses, such as severe acute respiratory syndrome (SARS) and Middle East respiratory syndrome coronavirus (MERS-CoV), can cause severe respiratory tract infection with high mortality [1].

Pulmonary pathology during severe coronavirus infection

The primary cell types found in the lower respiratory tract are alveolar epithelial cells and alveolar macrophages (AMs). AMs are susceptible to infections, but can also release a significant quantity of infectious virus particles. Pathological examinations of samples obtained from patients who died of SARS revealed diffuse alveolar damage, accompanied by prominent hyperplasia of pulmonary epithelial cells and presentation of activated alveolar and interstitial macrophages. Strikingly, these pulmonary manifestations were usually found after clearance of viremia (viruses in the blood) and in the absence of other opportunistic infections. Therefore, alveolar damage from local inflammatory responses may be due to an excessive host immune response [2].

In a murine model of SARS infection, fast and robust virus replication was accompanied by a delayed type I IFN (interferon) response. Accordingly, type I IFN expression was barely detectable in most cell types. Plasmacytoid dendritic cells are a notable exception. They utilize TLR7 (toll-like receptor-7) to sense viral nucleic acids and can induce robust expression of type I IFN following coronavirus infection. The extremely rapid replication of SARS-CoV together with the delayed onset of a type I IFN response caused extensive lung inflammation. This was accompanied by influx of inflammatory monocyte-macrophages, which are attracted by inflammatory signals. Furthermore, macrophages additionally produced high levels of inflammatory signals through stimulation of a type I IFN response, resulting in further macrophage influx in a pathological feedback loop. Altogether, massive accumulation of pathogenic inflammatory macrophages increased the severity of SARS. Moreover, type I IFN-

induced immune dysregulation enforced apoptosis of T cells, which would normally promote virus clearance, resulting in reduced numbers of virus-specific CD8 and CD4 T cells [1, 3].

Activation of effector immune cells

The rapid kinetics of SARS-CoV replication and the relative delay in type I IFN signaling may promote inflammatory M1 macrophage accumulation, suggesting that targeted antagonism of this pathway would improve outcomes in patients with severe coronavirus infections [2]. Notably, the 2019 novel coronavirus infection (COVID-19) behaves much like SARS-CoV; the virus responsible for COVID-19 has been named SARS-CoV-2. Typically, pneumonia from COVID-19 progresses rapidly with acute respiratory distress syndrome (ARDS) and septic shock, which are eventually followed by multiple organ failure due to a virus-induced cytokine storm in the body [4].

In response to infection, macrophages must react rapidly with a substantial pro-inflammatory burst to kill microorganisms and to recruit additional immune cells to the infection site. The inflammatory phenotype in macrophages is normally closely associated with a sharp increase in the rate of glycolysis. This causes activated macrophages and effector T lymphocytes to shift to a high glucose uptake, even under oxygen-rich conditions, which is known as the "Warburg effect", similar to cancer cells. The Warburg effect is associated with diverse cellular processes, such as angiogenesis, hypoxia, polarization of macrophages, and activation of T cells. This phenomenon is intimately linked to several disorders, including sepsis, autoimmune diseases and cancer [5].

Another interesting aspect of glycolysis induction in activated immune cells is the role of the glycolytic enzyme, glyceraldehyde-3-phosphate dehydrogenase (GAPDH). It has been shown that GAPDH binds to the mRNA coding for IFNγ, repressing its translation into protein. However, upon glycolysis activation, GAPDH dissociates from IFNγ mRNA, allowing to its translation into protein [6]. In addition, due to the glycolytic pathway stimulation in activated immune cells, their TCA (citric acid cycle) becomes disrupted. Therefore, an accumulation occurs for several metabolites, including succinate, which, in turn, may increase hypoxia-inducible factor-dependent activation of target genes, such as IL-1ß and the glucose transporter GLUT1 [7]. GLUT1 is required for the metabolic reprogramming, activation, and expansion of effector lymphocytes and M1 macrophages [7, 8].

Interaction between macrophages and alveolar epithelial type II (ATII) cells

Type I IFNs (type I interferons) produced by almost all cell types play a vital role in host defense against viral infection and cancer immunosurveillance. In response to viral products, pattern recognition receptors, such as retinoic-acid-inducible gene I (RIG-I)-like receptors (RLRs), send a signal to trigger type I IFN production in alveolar epithelial cells. Upon sensing cytosolic viral RNAs, these RLRs undergo conformational changes and oligomerization, to recruit a signaling adaptor called mitochondrial antiviral-signaling (MAVS) protein. Once activated, MAVS causes phosphorylation of IRF3, and subsequent transcription and expression of type I IFNs [9].

Activated macrophages produce large amounts of lactate, which they readily export with carboxylate transporters [5]. Alveolar epithelial cells import the lactate, and use it as a substrate for mitochondrial oxidative energy (ATP) production [10]. In ATII cells, lactate inhibits the localization of MAVS into mitochondria, the RLR-MAVS association, and MAVS aggregation and downstream signaling activation. Lactate does this by binding to the TM domain of MAVS. Thus, macrophage released lactate may attenuate host innate immune response by decreasing type I IFN production for viral clearance [9].

Proposed mechanism of action of high-dose vitamin C in immune effector cells

Vitamin C is an essential anti-oxidant and enzymatic co-factor for physiological reactions, such as hormone production, collagen synthesis, and immune potentiation. Humans are unable to synthesize vitamin C; therefore, they must acquire vitamin C from dietary sources [11]. Vitamin C is transported across cellular membranes by the sodium vitamin C co-transporter (SVCT). In addition, vitamin C spontaneously oxidizes both intracellularly and extracellularly to its biologically inactive form, dehydroascorbate (DHA) [11, 12]. DHA is unstable at physiological pH and, unless it is reduced back to vitamin C by glutathione (GSH), it may irreversibly be hydrolyzed. Therefore, DHA is reduced to vitamin C after import at the expense of GSH, thioredoxin, and NADPH (reduced nicotinamide adenine dinucleotide phosphate). Consequently, production of reactive oxygen species (ROS) increases inside some activated immune cells (similar to cancer cells) due to the exhaustion of ROS scavenging systems involving redox couples, such as NADPH/NADP+ and GSH/GSSG (glutathione disulfide). Therefore, high-dose vitamin C may function as a pro-oxidant in a cell type-dependent manner [12].

Sepsis is characterized by systemic inflammation, increased oxidative stress, insulin resistance, and peripheral hypoxia. Remarkably, severe sepsis resulted in a ~43-fold increase in GAPDH expression [13]. GAPDH is a redox-sensitive enzyme that can become rate-limiting when glycolysis is upregulated due to the Warburg effect, as it is in both cancer cells [12] and activated immune cells. In addition to oxidizing and inhibiting GAPDH, the elevated ROS may also lead to the DNA damage and the activation of poly(ADP-ribose) polymerase (PARP). PARP activation leads to the consumption of NAD+ (nicotinamide adenine dinucleotide) following vitamin C treatment. Significantly, NAD+ is required for the enzymatic activity of GAPDH as a co-factor; therefore, the decrease in NAD+ further diminishes GAPDH enzymatic activity.

Altogether, high-dose vitamin C-induced inhibition of GAPDH decreases the generation of ATP and pyruvate that would otherwise induce an energetic crisis, ultimately leading to cell death [11, 12]. In other words, inhibition of GAPDH by vitamin C may in turn inhibit immune effector cells and their related immunosuppression. These results provide a mechanistic rationale for exploring the therapeutic use of vitamin C to prevent inflammatory hyperactivation in myeloid and lymphoid cells.

Intravenous high-dose vitamin C treatment for 2019-nCoV disease

The results of meta-analyses have been demonstrated that intravenous (IV) high-dose vitamin C treatment has significant benefits in the treatment of sepsis and septic shock. [14,15] Sepsis is a life-threatening organ dysfunction syndrome triggered by a systemic inflammatory reaction to

pathogenetic microorganisms and their products. ARDS, devastating and often lethal condition, is also common among patients with systemic inflammatory response, such as sepsis [16].

Rolipram, a typical phosphodiesterase-4 inhibitor, can inhibit TNFα production in activated macrophages and restrain acute inflammatory response. Rolipram was suggested as a novel drug treatment for sepsis and septic shock due to its potent immunosuppressive effects [17]. By analogy, the beneficial effects of intravenous high-dose vitamin C in sepsis and septic shock may also be due to its immunosuppressive effects.

While immune effector cells are dependent on glycolysis for their bioenergetic functions, lung epithelial cells use mitochondrial oxidative phosphorylation to produce ATP. Therefore, high-dose vitamin C treatment acts as a prooxidant for immune cells, but as an antioxidant for lung epithelial cells. Furthermore, vitamin C treatment may protect innate immunity of ATII through the inhibition of the lactate secretion, produced by the activated immune cells.

In connection with the prooxidant role of vitamin C, which requires pharmacological (millimolar) rather than physiological (micromolar) concentrations, reevaluating the high-dose infusion of vitamin C would be a timely choice for the COVID-19-related ARDS. Altogether, patients diagnosed with COVID-19 and hospitalized with the breathing difficulty and abnormal biomarkers would seem to be excellent candidates for a short period of high dose intravenous vitamin C treatment in the early period of the disease. However, a concern that may arise with high-dose vitamin C treatment is osmotic cell death of immune cells, (but not apoptosis) which might generate a local inflammation in alveolar medium. Therefore, IV glucocorticoid treatment should be added to attenuate the possible inflammatory complications of high-dose vitamin C treatment. A previously experienced and comparably well-tolerated treatment regimen for high-dose intravenous vitamin C could be the administration of 50 mg/ per kilogram body weight every 6 hours for 4 days [16] with a glucose restriction. In addition, hydrocortisone 50 mg IV every 6 hours for 7 days should be added to fight against therapy-induced inflammation. Vitamin C when used as a parenteral agent in high doses may act pleiotropically as a prooxidant to attenuate pro-inflammatory mediator expression, improving alveolar fluid clearance, and to act as an antioxidant to improve epithelial cell functions.

(Adnan Erol, MD, is a specialist in internal medicine at Erol Project Development House, Istanbul, Turkey. He is particularly interested in the functioning of the cell's energy metabolism and energy metabolism disorders. His email is eroladnan@protonmail.com)

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