Supplement

Repurposing Of Sodium Thiosulfate (STS) In The Treatment Of COVID-19 Pandemic Based On A Personal Historical Account Of Intravenous STS Use In The Treatment Of Calciphylaxis.

On 20 March 2020, the news of repurposing an older malaria drug, hydroxychloroquine in combination with azithromycin (commonly referred as a Z-pak), to treat patients with COVID-19 provided me with the concept to share the sodium thiosulfate (STS) story in relation to its use for treating calciphylaxis and possibly repurposing it for the treatment of COVID-19. Currently, there are other multiple approved medications that are also being considered for repurposing as follows: Tocilizumab - sold under the name Actemra, Kaletra/Aluvia HIV drugs, Hydroxychloroquine as previously mentioned and Remdesivir - a broad spectrum anti-viral medication (personal communications and originally provided for further clinical studies by FDA, which has now been approved for emergency use authorization use by FDA- May 1, 2020). Multiple protease inhibitors are being actively pursued since they are known to impair viral replication including SARS-CoV-2. There are matrix metalloproteinase inhibitors (protease inhibitors) that are already approved including tetracycline, minocycline and doxycycline [1]; however, clinical testing other than with azithromycin being promoted in combination with hydroxychloroquine have been currently tested [1, 2]. How do we know if one medication in this combination treatment is working individually or does this combination actually have a synergistic effect? Unfortunately, most in the media only discuss hydroxychloroquine individually in a pro or a con standpoint and present confusion to each side of its use. Certainly this concept is not completely clear at the present time; however, there are ongoing clinical trials. Currently, the drug Hydroxychloroquine (HCQ) has been previously approved by FDA for the treatment of uncomplicated malaria, chronic discoid lupus erythematosus and systemic lupus erythematosus in adults; and acute and chronic rheumatoid arthritis in adults and now the FDA

has approved an abbreviated new drug application to address ongoing shortages of the drug. Further, it is now thought that HCQ may prevent endothelial dysfunction that may involve the ecGCx [3, 4, 5]. Additionally, the role of repurposing previously approved medications are in constant considerations [2]. There are undoubtedly many more medications under consideration, which may be mentioned as time evolves during this pandemic crisis.

The repurposing of STS began with two papers on vascular ossification calcification that included calciphylaxis in 2004-2005 [6, 7] and an older paper (1985 by Yatzidis H) that discussed the use of STS in the treatment of recurrent calcium urolithiasis [8]. Subsequently, multiple papers regarding the use of STS in the treatment of calciphylaxis were published [7, 9, 10, 11, 12, 13]. Importantly, there have been many other papers dealing with the use of STS in the treatment of calciphylaxis over the years (205 papers in PubMed.gov with search terms for sodium thiosulfate and calciphylaxis from 2004-2020) and STS has now become a part of the global multimodal treatment strategy for calciphylaxis.

Repurposing or repositioning of medications to treat emerging diseases is not a new concept; however, I wish to share the story of utilizing sodium thiosulfate (STS) as a repurposed drug in the treatment of calciphylaxis and how, even now, this drug could possibly assist in the treatment of COVID-19 viral pandemic of 2019 [2].

Possible Mechanisms of Sodium Thiosulfate (STS) As A Potent Antioxidant

Instead of one unpaired electron as in most antioxidants, STS has two unpaired electrons that can may be readily donated to sequester reactive oxygen/nitrogen species (RONS). During this chemical reaction process of sequestering RONS, STS also is capable of generating glutathione (GSH) a natural occurring intracellular antioxidant and thus, increase the cellular antioxidant strength in a region actively being damaged (Figs. 1 and 2). Also, STS treatment may improve

the sulfation as a sulfate donor to glycosaminoglycans (GAGs) as in figure 11of the main text and signified with red dots on the GAGs side chains of proteoglycans and glycoproteins; however, this has not been currently proven [14].

Known Side Effects Of Sodium Thiosulfate (STS) used to treat calciphylaxis

Here is what we know since sodium thiosulfate has become widely used in the global treatment of calciphylaxis. Obviously, any known allergy to STS or a similar sulfur containing product would preclude its use. Additionally, STS may induce an acidosis and with prolonged use could induce osteoporosis at least in animal models [7, 9, 10, 11, 12, 13].

Sodium Thiosulfate Protective Of Detrimental Cytokine and Reactive Oxygen/Nitrogen Species (RONS) Storm and Vascular Collapse

Upon initial infection with COVID-19 there is a cauldron of redox chemical reactions with an accumulation of excessive RONS (the first line of defense against the COVID 19 virus produced by the body's protective inflammatory response to infection as in figures 1 and 4 in main text. The cells of the immune response include the 1st responder's neutrophils, eosinophils, mast cells and chronic persistent macrophages - with monocyte migration and monocyte to macrophage transformation at the infectious site of the nasopharyngeal bronchial lining epithelial cells, pneumocytes and capillary blood-gas barrier (BGB) endothelial cells.

As these inflammatory cells attempt to clear the virus from these passageway epithelial cell's, they generate RONS with a staggering amount of unpaired reactive oxygen and nitrogen species of unpaired electrons and toxic cytokines, which over time creates a cytokine storm and RONS beget RONS and cytokines evoke more inflammatory invasion and thus create the cytokine storm not only at the epithelial cells but also the endothelial cells at the BGB. The healthy surrounding cells of the upper and lower pulmonary epithelial, pneumocytes and

capillary BGB endothelial cells become a source of ensuing collateral damage that originally was intended to result in deleting the invasion of viral envelope damage-associated molecular patterns (DAMPs) and pathogen-associated molecular pattern (PAMPs) recognized by these cells in a response to injury and wound healing mechanism to the invading COVID-19 virus and multiple virions in the pulmonary tissues.

In turn, this redox storm of RONS and cytokine storm leak into the systemic circulation and the protective lining of the endothelial cells endothelial glycocalyx (ecGCx) may become damaged and are attenuated and/or lost. This may result in a marked increased vascular permeability and loss of intravascular contents into the surrounding extravascular interstitial capillary space. Unfortunately, this may also result in increased capillary permeability and result in a 'capillary leak syndrome' [15, 16, 17], which may result in vascular collapse with decreased cardiac output and decreased perfusion of the brain with relative ischemia over time and result in death of the host (COVID-19 infected individuals) that have progressed to being on ventilator assisted respiration life support.

Indeed, the COVID-19 infection induced by this pulmonary infection with bilateral pneumonia's requiring ventilator treatment due to an acute respiratory distress syndrome (ARDS) is associated with increased pulmonary vascular permeability with increased lung weight and loss of aerated lung tissue; however, these patients actually may be dying of vascular collapse due to a loss of their local and systemic endothelial glycocalyx due to a cytokine and RONS storm.

STS treatments are provided by the intravenous route; however, all of these critically ill individuals have an established intravenous port to administer medication so this treatment

should not be viewed as invasive since these intravenous lines are already available. The antioxidant effect of STS might possibly restore some effective endothelial cell function and even re-establish the endothelial glycocalyx or assist in its restoration in addition to providing an increase in bioavailable nitic oxide. An increase in venous or arterial syndecans may be a key laboratory read out of endothelial glycocalyx loss and systemic vascular collapse [18]. Further, it has been found by deep-machine learning and artificial intelligence that increases in hemoglobin may be able to predict who will develop ARDS in COVID-19 patients in a small cohort from Wuhan, China [19]. Possibly, STS could bind the excessive iron cations involved in this process and be helpful due to its known chelation of cations. Additionally, patients on ventilators with COVID-19 certainly apply to these thoughts and concepts. The concept of increased pulmonary and possible systemic microvascular permeability is currently a possibility that is not being discussed a great deal at the moment but should be. Further, novel ideas for the prevention of this are of great importance during the COVID-19 pandemic. Medications that may help to eliminate the excessive RONS (redox storm) with excessive over reactive cytokines (cytokine storm) such as STS need to be considered as it relates to the capillary leak syndrome with vascular collapse due to viral sepsis of the COVID-19 disease.

Addendum

The obesity related cytokine-hormone Leptin may also be involved in multiple ways in COVID-19 since it is known to be elevated in some individuals with the metabolic syndrome with obesity, insulin and leptin resistance, hypertension, cardiovascular disease and T2DM. We need to think about leptin resistance and the possible lack of cellular leptin signaling (especially in the brain and inflammatory cells) due to leptin resistance especially since we know that part of the progression of COVID-19 is due to the viral, redox and cytokine storm with a dysfunctional

innate – adaptive immune system. Further, it is important to note that leptin is known to be closely tied to the proper function of the innate and adaptive immune systems and that both the macrophage and lymphocytes each have leptin receptors on their surface that may become dysfunctional in obesity and T2DM individuals with metabolic syndrome [20]. Even though leptin is often a perplexing and complicated hormone associated with obesity and excessive visceral white adipose tissue in glucose control, it also plays an important role in not only in glucose control but also thermogenesis and inflammation due to its central signaling of the hypothalamic nuclei that play an important role in obesity and T2DM. Also, it is important to note that leptin is a FDA approved drug (human recombinant leptin Myalept®) for the treatment of complications of leptin deficiency in patients with congenital or acquired generalized lipodystrophy. Since diet induced obesity and leptin deficient (ob/ob) mice leads to an impaired innate immune response to lipopolysaccharide infusion with an increased inflammatory sickness syndrome and prolonged behavioural response leptin should be at least be given some consideration in the near future [21].

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