Editorial: Nicotine and SARS-CoV-2: COVID-19 may be a disease of the nicotinic cholinergic system

Konstantinos Farsalinos

PII: S2214-7500(20)30292-4
DOI: https://doi.org/10.1016/j.toxrep.2020.04.012
Reference: TOXREP 884

To appear in: *Toxicology Reports*

Raymond Niaura

PII: S2214-7500(20)30292-4
DOI: https://doi.org/10.1016/j.toxrep.2020.04.012
Reference: TOXREP 884

To appear in: *Toxicology Reports*

Jacques Le Houezec

PII: S2214-7500(20)30292-4
DOI: https://doi.org/10.1016/j.toxrep.2020.04.012
Reference: TOXREP 884

To appear in: *Toxicology Reports*

Anastasia Barbouni

PII: S2214-7500(20)30292-4
DOI: https://doi.org/10.1016/j.toxrep.2020.04.012
Reference: TOXREP 884

To appear in: *Toxicology Reports*

Aristidis Tsatsakis

PII: S2214-7500(20)30292-4
DOI: https://doi.org/10.1016/j.toxrep.2020.04.012
Reference: TOXREP 884

To appear in: *Toxicology Reports*

Dimitrios Kouretas

PII: S2214-7500(20)30292-4
DOI: https://doi.org/10.1016/j.toxrep.2020.04.012
Reference: TOXREP 884

To appear in: *Toxicology Reports*

Apostolos Vantarakis

PII: S2214-7500(20)30292-4
DOI: https://doi.org/10.1016/j.toxrep.2020.04.012
Reference: TOXREP 884

To appear in: *Toxicology Reports*

Konstantinos Poulas

PII: S2214-7500(20)30292-4

DOI: https://doi.org/10.1016/j.toxrep.2020.04.012

Reference: TOXREP 884

To appear in: *Toxicology Reports*

Editorial: Nicotine and SARS-CoV-2: COVID-19 may be a disease of the nicotinic cholinergic system

Konstantinos Farsalinos[1,2*]kfarsalinos@gmail.com, Raymond Niaura[3], Jacques Le Houezec[1], Anastasia Barbouni[2], Aristidis Tsatsakis[4], Dimitrios Kouretas[5], Apostolos Vantarakis[6], Konstantinos Poulas[1]

[1] Laboratory of Mol. Biology and Immunology, Department of Pharmacy, University of Patras, Panepistimiopolis, 26500, Greece

[2] School of Public Health, Department of Public and Community Health, University of West Attica, 11521, Greece

[3] Departments of Social and Behavioral Science and Epidemiology, College of Global Public Health, New York University, USA

[4] Department Medicine, University of Crete, Greece

[5] Department of Biochemistry and Biotechnology, University of Thessaly, Larisa, 41500, Greece

[6] School of Medicine, University of Patras, Panepistimiopolis, 26500, Greece

Corresponding author:

Konstantinos Farsalinos, Leoforos Alexandras 196A, Athens 11521, Greece.

Email: Tel nr: +306977454837

Word count: 3639 words

Abstract

Corona Virus Disease 2019 (COVID-19) is a disease caused by a novel coronavirus, SARS-CoV-2. On 23 March we presented evidence of a low prevalence of smoking among hospitalized patients with COVID-19 in China, and we were the first to establish the hypothesis that nicotine may be beneficial for COVID-19 patients and should be evaluated in clinical trials due to its anti-inflammatory properties. While in many cases the disease is mild, severe COVID-19 involves a hyper-inflammatory response, commonly called cytokine storm, characterized by the release of pro-inflammatory cytokines that can lead to Acute Respiratory Distress Syndrome and death. The cholinergic anti-inflammatory pathway is an important immune-regulating system mediated by nAChRs that can control inflammation, and function as an immunomodulator through bi-directional communication between the immune and nervous systems. The clinical manifestations of cytokine storm observed in COVID-19 patients could be linked to a dysfunction of the cholinergic anti-inflammatory pathway. At the same time, several patients experience neurological symptoms that could be explained by the invasion of the virus to the terminal area of afferent vagus fibers or the origin of the efferent vagus fibers, and further dysregulation of the inflammatory response. Anosmia has been experienced by several patients, a phenomenon that has been observed in patients with Parkinson's disease and is caused by impaired cholinergic transmission. Thromboembolic complications, activation of platelets and endothelial damage with increased vascular permeability indicate ineffective control by the nicotinic cholinergic system. Considering that most of the manifestations of COVID-19 are linked to impairment of the nAChRs, we make the hypothesis that COVID-19 may be a disease of the nicotinic cholinergic system. We present regions with four or five amino acids homology between the SARS-CoV-2 and several neurotoxin molecules that act as competitive antagonists

in nAChRs. We propose that nicotine could be used therapeutically and should be urgently evaluated in clinical trials.

Keywords. SARS-CoV-2; COVID-19; ACE2; inflammation; smoking; nicotine; hospitalization.

Introduction

As of 20 April, almost 1.7 million people globally have been diagnosed with Corona Virus Disease 2019 (COVID-19), a pandemic that has evolved from the emergence of a new coronavirus strain, acute respiratory syndrome coronavirus 2 (SARS-CoV-2), in China. More than 170,000 deaths have been reported, while there are certainly many more cases of milder disease that have not been diagnosed and officially confirmed due to limited testing capacity in most countries. The pandemic is a global emergency due to the rapid transmission of the disease and the potential to overwhelm the healthcare systems, and is expected to have considerable economic and health impacts. Possible contributing factors, their possible role in the relatively high infection, death rates between countries and origin have recently been studied (Goumenou et al. 2020a, Goumenou et al. 2020b). This new outbreak has been additionally evaluated for current knowledge on coronaviruses based on a short history to epidemiology, pathogenesis, clinical manifestation of the disease, as well as treatment and prevention strategies (Docea et al 2020). The search for potential protective and therapeutic antiviral strategies is of particular and urgent concern (Skalny et al., 2020).

While in most cases, especially in young people without any comorbidities, the disease is expected to be relatively mild, there is a substantial proportion of patients who develop complications and need intensive care-unit support and mechanical intubation. In one case series of 1099 patients in China (Guan et al., 2020), 6.1% of cases suffered from the primary composite end-point of admission to an intensive care unit, the use of mechanical ventilation, or death. Patients with severe disease typically present with dyspnea and hypoxemia shortly after disease initiation, and may quickly progress to respiratory failure, acute respiratory distress syndrome (ARDS) and multi-organ failure (Chen et al., 2020). Predictors of adverse outcomes include

elevated levels of inflammatory markers and pro-inflammatory cytokines. A study of 150 COVID-19 cases reported that elevated levels of C-reactive protein (CRP), ferritin and IL-6 were associated with death (Ruan et al., 2020). IL-6, an important pro-inflammatory cytokine, was elevated in fatal cases of COVID-19 in another study of 191 patients (Zhou et al., 2020). Another study of 452 patients reported that those with severe disease showed lymphocytopenia, neutrophilia, low levels of monocytes, eosinophils and basophils, and elevated levels of infection-related biomarkers and inflammatory cytokines (Qin et al., 2020). Pathological examination of a case in China revealed bilateral diffuse alveolar damage, desquamation of pneumocytes, hyaline membrane formation and interstitial mononuclear inflammatory infiltrates (Xu et al., 2020). Flow cytometry of peripheral blood revealed reduced levels of CD4+ and CD8+ T cells, which however were hyper-activated, and elevated concentration of pro-inflammatory CCR6+ Th17 in CD4+ T cells. Such findings are hallmarks of ARDS and resemble features observed in SARS and Middle Eastern Respiratory Syndrome (Ng et al., 2016; Ding et al., 2003). Systemic vasculitis was also observed (Xu et al., 2020). Therefore, it seems that immune dysregulation may be implicated in the pathophysiology of severe COVID-19.

Cytokine storm

While for decades common thinking suggested that every immune response to antigenic invasion was always beneficial in averting potential damage, studies in the 1980s identified that immune cells produce proteins with pleiotropic properties, having the potential to be either beneficial or harmful (Tracey, 2007). The proteins, called cytokines, were found to cause clinical manifestations similar to sepsis such as hemodynamic instability, fever, and localized inflammation (Tracey et al., 1986; Dinarello et al., 1986). Cytokines are important in mediating both immune cell recruitment and complex intracellular signaling control mechanisms that

characterize inflammation and infection control. They are expressed by numerous cells, including macrophages, monocytes, B cells and T cells, promote differentiation of T-helper cells and stimulate CD4+ cells (Turner et al., 2014). While activation of the immune system is important in fighting pathogens, dysregulation of cytokine production may lead to uncontrolled effects that can ultimately be detrimental to health (Dinarello, 2007; Turner et al., 2014).

Cytokine storm (also called macrophage activation syndrome) is a systemic inflammatory response that can be triggered by a variety of factors such as infections and drugs (Shimabukuro-Vornhagen et al., 2018). It represents a failure of the inflammatory response to return to homeostasis. The resulting unregulated immune activity can potentially lead to catastrophic tissue damage. The term first appeared in 1993 in an article relevant to graft-versus-host disease (Ferrara et al., 1993). Subsequently, cytokine storm was a phenomenon recognized in both viral and bacterial infections. It has been particularly studied in viral infections such as cytomegalovirus pneumonitis, influenza virus and SARS-CoV (Barry et al., 2000; Bermejo-Martin et al., 2009; Huang et al., 2005; Tisoncik et al., 2012). Bermejo-Martin et al. (2009) recruited both inpatients and outpatients during the first wave of the pandemic flu in 2009 (nvH1N1) and examined the effects of immune host responses to the evolution of mild or severe disease by measuring serum levels of several chemokines and cytokines. They found a dramatic increase of mediators that stimulate Th-1 responses and Th-17 responses (which are responsible for attacking intracellular pathogens and clearing pathogens during host defence reactions) among severe hospitalized patients compared to milder cases of nvH1N1 infection during the 2009 pandemic. The cytokine storm can result in acute lung injury and further progress to ARDS. This is characterized by local infiltration of inflammatory cells, increased vascular permeability and systemic spillover of inflammatory mediators that can cause systemic sepsis-

like symptoms (Tisoncik et al., 2012). While focus on cytokine storm detection relies mostly on measuring cytokines in the systemic circulation, it has been suggested that measuring systemic inflammatory mediators may underestimate the extent of the immunological cascade that takes place locally in deep tissues such as the respiratory tract (Tisoncik et al., 2012). Considering the above, controlling the inflammatory response may be an effective way of preventing collateral damage caused by the excessive activation of the immune system to clear pathogens.

Cholinergic anti-inflammatory pathway

Since the early 2000s, the cholinergic nervous system has been identified as an important pathway that modifies and controls the inflammatory response. Surgical dissection of the vagus nerve in mice led to enhanced TNF production and excessive response to endotoxin administration, while vagus nerve electrical stimulation inhibits the synthesis of TNF and prevents the acute inflammatory response (Borovikova et al., 2000; Tracey 2002; Blalock, 2002). Several animal experimental models inducing pro-inflammatory cytokines, such as sepsis, ischemia-reperfusion and pancreatitis have shown that vagus stimulation improves outcomes. This effect is mediated by the nicotinic acetylcholine receptor (nAChR) α7 subunit on macrophages (Wang et al., 2003). Mice deficient of the α7 subunit exhibited increased endotoxin-induced TNF production, and electrical vagus innervation failed to reduce serum TNF levels (Wang et al., 2003). B-lymphocytes also express α7 nAChRs. Macrophages appear to be very sensitive to acetylcholine, which suggests that any source of acetylcholine, even from non-neuronal sources such as epithelial and endothelial cells, could also modulate the activity of adjacent macrophages (Tracey, 2002). Besides TNF, other pro-inflammatory cytokines are inhibited by acetylcholine, such as high mobility group B1 (HMGB1), IL-1, and IL-6 (Ulloa, 2005).

Modulation of inflammatory and immune response by the central nervous system (CNS) through the vagus nerve is based on bi-directional communication between the immune and nervous systems. Afferent vagus nerve fibers, located in nucleus tractus solitarius, provide sensory input to the CNS about the inflammatory status that can result in the transmission of efferent signals, originating from the dorsal motor nucleus, to control the inflammatory response (Pavlov et al., 2003). Such a response is rapid and localized, unlike the diffusible anti-inflammatory network, which is slow, distributed, non-integrated and dependent on concentration gradients (Tracey, 2002).

Nicotine, nicotinic cholinergic system and COVID-19

Smoking is known to increase the risk for respiratory infection susceptibility and severity (Cohen et al., 1993; Millett et al., 2015). Considering that COVID-19 was declared by the World Health Organization as a pandemic, a substantial disease burden would be expected among the estimated 1.1 billion smokers, especially in countries with high smoking prevalence. Therefore, there were understandable concerns about this population subgroup (Berlin et al., 2020). Additionally, smoking-related disease conditions such as cardiovascular disease and COPD are also established risk factors for adverse outcomes in COVID-19 (Wu & McGoogan, 2020). China was the first country to be affected by the pandemic and has a high smoking prevalence. In 2018, the population smoking prevalence was 26.6% with a much higher prevalence in men (50.5%) than in women (2.1%) (World Health Organization, 2018). Therefore, a high smoking prevalence among patients with COVID-19 would be expected, even if smoking did not adversely affect disease susceptibility and severity.

On 23 March, a preliminary analysis by some members of our group examined data from 5 case series of hospitalized COVID-19 patients from China, and calculated a smoking prevalence of 10.2% (95% CI: 8.7–11.8%) while the estimated expected prevalence was 31.3% (95% CI: 8.7–11.8%) (Farsalinos et al., 2020a). The analysis was further expanded on 3 April by examining 13 Chinese studies and 5960 hospitalized COVID-19 patients, with a pooled smoking prevalence of 6.5% (95% CI: 4.9–8.2%) (Farsalinos et al., 2020b). On that date, we presented for the first time a hypothesis about the potential beneficial effects of nicotine, which was subsequently expanded (Farsalinos et al., 2020c). While there were limitations in the study analysis, mainly due to the inability to adjust for confounding factors, the findings of low smoking prevalence among hospitalized COVID-19 patients in China were consistent across all studies and in agreement with case series from USA (CDC, 2020; Richardson et al., 2020). The original hypothesis was based on the anti-inflammatory properties of nicotine through the cholinergic anti-inflammatory system, acknowledging that the disease appeared to involve a dysregulation of the immune response to viral invasion.

It is obviously inappropriate to suggest that anyone should initiate smoking or to continue to smoke due to the well-established smoking-related morbidities and the large number of potentially toxic chemicals in cigarette smoke. Furthermore, it is unlikely that any other compound in tobacco cigarette smoke, besides nicotine, would be implicated to the potential benefits observed in smokers. Moreover, due to the adverse effects of smoking and the fact that many smokers would suffer from co-morbidities (such as cardiovascular disease, COPD etc.), it is expected that the potential benefits of nicotine would be blunted when observed in smokers.

Nicotine is a cholinergic agonist. Therefore, it is an important inhibitor of pro-inflammatory cytokines acting through the cholinergic anti-inflammatory pathway via α7-nAChRs. Nicotine

inhibits TNF, IL-1, IL-6 and HMGB1 while it does not inhibit anti-inflammatory cytokines such as IL-10 (Li et al., 2011; Ulloa, 2005). *In vivo* animal models found nicotine to be protective against lipopolysaccharide-induced ARDS by reducing leukocyte infiltration and pro-inflammatory mediators in bronchoalveolar lavage fluid (Ni et al., 2011; Mabley et al., 2011). Such effects are relevant to COVID-19 since cytokine storm appears to be the hallmark in severe cases (Mehta et al., 2020; Huang et al., 2020). Several pro-inflammatory cytokines, such as IL-1β, IL-2, IL-6, IL-17, IL-8, TNF and CCL2 are elevated in COVID-19 patients (McGonagle et al., 2020). Treatment with anti-IL-6 anti-TNF medications has been proposed and clinical trials are already underway (Feldmann et al., 2020; Georgiev et al., 2020). However, it may be more effective to inhibit several instead of selectively one cytokine, while some cytokine inhibitors are associated with elevated risk of opportunistic infections (Rutherford et al., 2018). Also, it is possible that measuring blood levels of inflammatory cytokines does not accurately reflect the extent of the immune imbalance that exists locally in the lungs. In any case, the cholinergic anti-inflammatory system provides better control and modulation of the cytokine response compared to blocking a single agent, and nicotine could effectively contribute to maintaining a balanced immune response against viral infection. Therefore, it is possible that the clinical manifestations of cytokine storm in COVID-19 patients are the result of dysfunction of the cholinergic anti-inflammatory pathway.

SARS-CoV-2 is known to use the angiotensin converting enzyme 2 (ACE2) as a receptor for cell entry (Zhang et al., 2020). ACE2 has well-established vasodilatory, anti-inflammatory and antioxidant properties. Studies on smoking and ACE2 are contradictory. Studies published before the COVID-19 pandemic reported that smoking and nicotine down-regulate ACE2 (Oakes et al., 2018; Yue et al., 2018). However, more recent studies suggest that they up-regulate ACE2

(Cai et al., 2020; Leung et al., 2020; Blake et al., 2020). There is currently no evidence to suggest that up-regulation of ACE2 is associated with increased COVID-19 susceptibility or severity. In fact, up-regulation of ACE2 appears to be protective against tissue damage caused by SARS-CoV-2. ACE2 has been found to protect mice from developing ARDS (Hung et al., 2016; Imai et al., 2008; Imai et al., 2005). Data from SARS experimental studies suggest that continuous SARS-CoV-2 infection and replication induces immediate down-regulation of ACE2 that may be implicated in organ damage and disease severity (Vaduganathan et al., 2020). Further support for the beneficial role of ACE2 comes from data that estrogens appear to up-regulate ACE2 while children and younger adults have higher ACE2 levels compared to older people (Christiani et al., 2020; Ciaglia et al., 2020). At the same time, women, children and young people have milder COVID-19 symptoms. If accurate and verified, the recently-observed ACE2 up-regulation in smokers is probably induced as a defence mechanism to counteract the effects of angiotensin II. There is probably a dynamic balance between ACE and ACE2, which is continuously changing, depending on stressors and stimuli. Thus, there is uncertainty on whether nicotine affects COVID-19 progression through the renin-angiotensin-aldosterone axis and there is no known interaction between ACE2 and nAChR receptors.

Importantly, ACE2 is expressed in several regions in the brain. The regions where vagal afferent fibers terminate and vagal efferent fibers originate exhibit ACE2 expression (Xia &Lazartigues, 2010; Dubay et al., 2006). Neuroinvasion is a common feature of coronaviruses (Desfroges et al., 2020). Anosmia and ageusia have been reported by COVID-19 patients (Giacomelli et al., 2020). SARS-CoV-2 may enter the CNS either through the blood stream or via the olfactory nerve across the cribriform plate (Manji et al., 2020; Baig et al., 2020). A case series of 214 patients reported that 36.4% had neurological manifestations (Mao et al., 2020). Thus, it is possible that

the virus might infect the terminal areas of vagus afferent fibers or the origin of vagus efferent fiber causing down-regulation of ACE2 and resulting in local inflammation that could disrupt the cholinergic anti-inflammatory pathway and dysregulate the inflammatory response. Nicotine could have protective properties against possible brain inflammation caused by SARS-CoV-2, mediated through α7-AChRs (Bencherif et al., 2011).

A noteworthy parameter relative to anosmia and ageusia observed among COVID-19 patients is that these are characteristic and prodromal non-motor manifestations of Parkinson's disease (Oppo et al., 2020; Haehner et al., 2011). While ageusia has not been extensively studied, olfactory disturbance is a very common feature, observed in up to 95% of Parkinson's disease patients (Haehner et al., 2011) and this symptom may appear several years before the onset of motor symptoms. There is no olfactory improvement with dopamine agonists (Doty et al., 1988; Müller et al., 2002). Unlike the general population where smoking is associated with impaired olfactory function, smokers with Parkinson's disease experience less decline in olfactory sense compared to non-smokers, suggesting a protective effect of smoking (Sharer et al., 2015). This is explained by the fact the phenomenon has been linked to impairment of cholinergic transmission (Bohnen et al., 2010) while nicotine improved the olfactory impairment in a mouse model of Parkinson's disease (Yang et al., 2019). The olfactory bulb has a rich network of nAChRs, but α7 nAChRs may also be expressed on the axon terminals of the olfactory receptor neurons (D'Souza & Vijayaraghavan, 2014). While this may suggest facilitated brain infection through anterograde transport along the olfactory nerve, it is possible that olfactory receptor neurons may act as first-line viral sensors and initiate a rapid immune response (Butowt & Bilinska, 2020). This would explain the mild symptoms in COVID-19 patients with olfactory loss.In any case,

anosmia may represent another sign of dysfunction of the nicotinic cholinergic system in COVID-19.

A prominent feature of COVID-19 is coagulopathy that results in thromboembolic complications. Venous thromboembolism was reported in 25% of patients who were not under thromboprophylaxis, and was associated with higher mortality rate (Cui et al., 2020). Abnormal coagulation parameters were also associated with poor survival (Tang et al., 2020). Although venous thromboembolism is a well-known risk factor of any serious infection, additional mechanisms such as endothelial damage, increased vascular permeability and microvascular occlusion may be implicated in COVID-19 (Kollias et al., 2020). It is important to note that platelets express functional α7-AChRs (Schedel et al., 2011) while hematopoietic α7 nAChR deficiency increases inflammation and platelet activity (Kooijman et al., 2015). Recently, acetylcholine was found to be an endogenous inhibitor of platelet activation (Bennett et al., 2019). Therefore, dysfunction of the nicotinic cholinergic system could be implicated in the thrombotic and vascular complications of COVID-19.

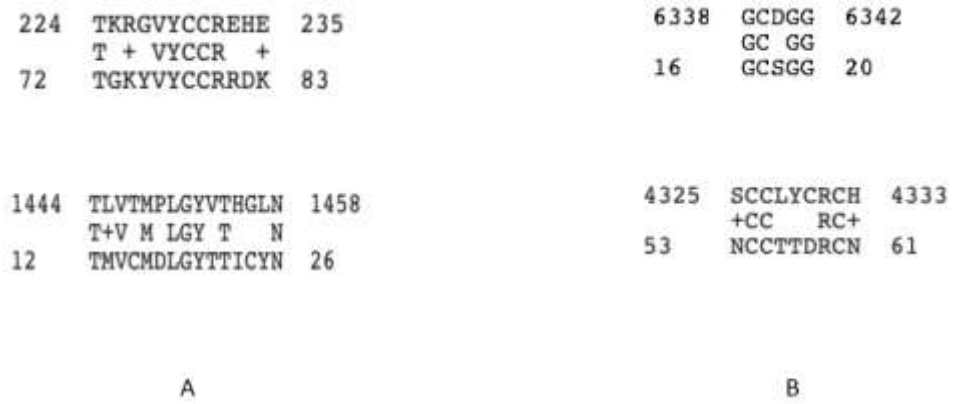
COVID-19 could be a disease of the nicotinic cholinergic system

The observation of a low prevalence of hospitalized COVID-19 patients in China led to the development of a hypothesis that nicotine could have protective effects by enhancing the cholinergic anti-inflammatory pathway (Farsalinos et al., 2020b). As more studies presented the clinical manifestations, laboratory findings and disease progression in COVID-19 patients, it became apparent that the nicotinic cholinergic system could explain most (if not all) of the disease characteristics. It would be unlikely for a single “defence system” to ameliorate all the

diverse and complex manifestations of COVID-19, unless that "defence mechanism" was the target of the viral host. Could that be possible?

SARS-CoV-2 appears to have originated from a bat coronavirus. Ji et al. (2020) carried out comprehensive sequence analysis in conjunction with relative synonymous codon usage bias and reported that the virus may have been a recombinant virus between the bat coronavirus and an unknown-origin coronavirus (Ji et al., 2020). One possible intermediate host could have been a snake coronavirus. Taking into consideration that snake venom toxins are competitive antagonists of acetylcholine on α7-nACh receptor with high affinity, we decided to explore the hypothesis that SARS-CoV-2 may have acquired sequences by any of the potential, and not defined yet, intermediates through genomic recombination. We compared the protein sequences between SARS-CoV-2 and snake venom neurotoxins. We were able to identify regions with four or five amino acids homology between the coronavirus and several neurotoxin molecules (e.g. SARS-CoV-2 compared with alpha Bungarotoxin, Fig 1A; SARS-CoV-2 and alpha-Cobratoxin, Fig 1B).

Figure 1. BLAST-P alignment of the SARS-CoV-2 protein against alpha Bungarotoxin (A) and alpha-Cobratoxin (B) indicating regions with high similarity.



Therefore, we hypothesize that these sequences on the SARS-CoV-2 proteins, being similar to the active sites of a neurotoxin, can result in binding to nAChRs and adversely affecting their function by preventing the action of acetylcholine.

Nicotine as a potential treatment for COVID-19

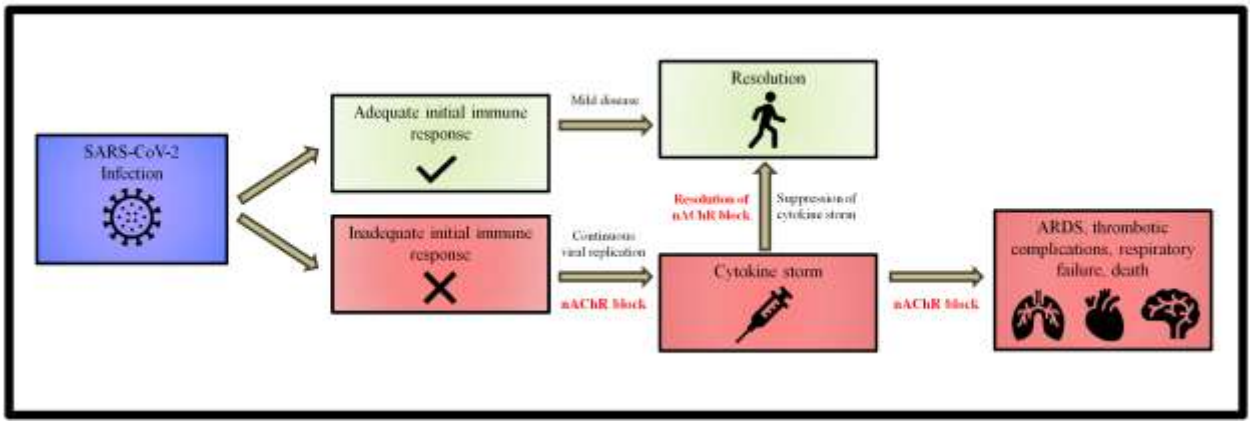
Nicotine could act as a competitive agonist for the nAChRs that could restore the compromised function of the nicotinic cholinergic system. This may be feasible through repurposing already approved (for other indications) pharmaceutical nicotine products such as nicotine patches for use by non-smokers, or even by using these products as already indicated (i.e. as smoking substitutes) among current smokers. These products are available over-the-counter in most countries. They have been administered therapeutically in non-smokers for neurological conditions and inflammatory bowel disease for larger periods than would be needed for COVID-19 (Pullan et al., 1994; Villafane et al., 2007; Newhouse et al., 2012). No abuse liability was observed in non-smokers despite being administerd for several weeks (Villafane et al., 2007; Newhouse et al., 2012). Besides gums and patches, nicotine can be administered though inhalation, with the use of a nebulizer or other aerosol systems, if necessary. Nicotine administration could be added on top of antiviral or other therapeutic options for COVID-19. By restoring and re-activating the cholinergic anti-inflammatory pathway, a more universal suppression of the cytokine storm could probably be achieved compared to administering inhibitors of a single cytokine. The potential need to provide pharmaceutical nicotine products to smokers and users of other nicotine products who experience abrupt withdrawal symptoms of nicotine when hospitalized for COVID-19 or aim to follow medical advice to quit smoking, should also be examined. Additionally, if the hypothesis about the beneficial effects of nicotine is valid, smokers who quit nicotine use when hospitalized will be deprived from these benefits.

In France, the Addiction Prevention Network (RESPADD) officially recommends the use of nicotine replacement therapies for smokers when hospitalized for any illness (RESPADD, 2019). Clinical trials will dictate future approaches and the role of nicotine in COVID-19, while further experimental studies should examine the affinity of the virus to nAChRs.

Conclusions

In conclusion, we noticed that most of the clinical characteristics of severe COVID-19 could be explained by dysregulation of the cholinergic anti-inflammatory system. The observation that patients eventually develop cytokine storm which results in rapid clinical deterioration, led to the development of a hypothesis about the series of events associated with adverse outcomes in COVID-19 (Fig. 2).

Figure 2. Progression of COVID-19 after SARS-CoV-2 infection.



Once someone is infected with SARS-CoV-2, the immune system is mobilized. As the virus replicates, cell and viral debris or virions may interact with the nAChRs blocking the action of the cholinergic anti-inflammatory pathway. If the initial immune response is not enough to combat the viral invasion at an early stage, the extensive and prolonged replication of the virus

will eventually block a large part the cholinergic anti-inflammatory pathway seriously compromising its ability to control and regulate the immune response. The uncontrolled action of pro-inflammatory cytokines will result in the development of cytokine storm, with acute lung injury leading to ARDS, coagulation disturbances and multiorgan failure. Based on this hypothesis, COVID-19 appears to eventually become a disease of the nicotinic cholinergic system. Nicotine could maintain or restore the function of the cholinergic anti-inflammatory system and thus control the release and activity of pro-inflammatory cytokines. This could prevent or suppress the cytokine storm. This hypothesis needs to be examined in the laboratory and the clinical setting.

Ethics approval and consent to participate. Not applicable.

Consent for publication. Not applicable

Competing interests. The authors report no competing interests for the past 36 months.

Funding. No funding was provided for this study.

Acknowledgements. None

References

1. Guan WJ, Ni ZY, Hu Y, Liang WH, Ou CQ, He JX, Liu L, Shan H, Lei CL, Hui DSC, Du B, Li LJ, Zeng G, Yuen KY, Chen RC, Tang CL, Wang T, Chen PY, Xiang J, Li SY, Wang JL, Liang ZJ, Peng YX, Wei L, Liu Y, Hu YH, Peng P, Wang JM, Liu JY, Chen Z, Li G, Zheng ZJ, Qiu SQ, Luo J, Ye CJ, Zhu SY, Zhong NS; China Medical Treatment Expert Group for

Covid-19. Clinical Characteristics of Coronavirus Disease 2019 in China. N Engl J Med. 2020. doi: 10.1056/NEJMoa2002032.

2. Baig AM, Khaleeq A, Ali U, Syeda H. Evidence of the COVID-19 Virus Targeting the CNS: Tissue Distribution, Host-Virus Interaction, and Proposed Neurotropic Mechanisms. ACS Chem Neurosci. 2020 Apr 1;11(7):995-998. doi: 10.1021/acschemneuro.0c00122.

3. Barry SM, Johnson MA, Janossy G. Cytopathology or immunopathology? The puzzle of cytomegalovirus pneumonitis revisited. Bone Marrow Transplant. 2000 Sep; 26(6):591-7.

4. Bencherif M, Lippiello PM, Lucas R, Marrero MB. Alpha7 nicotinic receptors as novel therapeutic targets for inflammation-based diseases. Cell Mol Life Sci. 2011 Mar;68(6):931-49. doi: 10.1007/s00018-010-0525-1

5. Bennett JA, Ture SK, Schmidt RA, Mastrangelo MA, Cameron SJ, Terry LE, Yule DI, Morrell CN, Lowenstein CJ. Acetylcholine Inhibits Platelet Activation. J Pharmacol Exp Ther. 2019 May;369(2):182-187. doi: 10.1124/jpet.118.253583.

6. Berlin I, Thomas D, Le Faou AL, Cornuz J. COVID-19 and smoking. Nicotine Tob Res. 2020 Apr 3. pii: ntaa059. doi: 10.1093/ntr/ntaa059.

7. Bermejo-Martin JF1, Ortiz de Lejarazu R, Pumarola T, Rello J, Almansa R, Ramírez P, Martin-Loeches I, Varillas D, Gallegos MC, Serón C, Micheloud D, Gomez JM, Tenorio-Abreu A, Ramos MJ, Molina ML, Huidobro S, Sanchez E, Gordón M, Fernández V, Del Castillo A, Marcos MA, Villanueva B, López CJ, Rodríguez-Domínguez M, Galan JC, Cantón R, Lietor A, Rojo S, Eiros JM, Hinojosa C, Gonzalez I, Torner N, Banner D, Leon A, Cuesta P, Rowe T,

Kelvin DJ. Th1 and Th17 hypercytokinemia as early host response signature in severe pandemic influenza. Crit Care. 2009;13(6):R201. doi: 10.1186/cc8208..

8. Blake SJ, Barnsley K, Lu W, McAlinden KD, Eapen MS, Sohal SS. Smoking Upregulates Angiotensin-Converting Enzyme-2 Receptor: A Potential Adhesion Site for Novel Coronavirus SARS-CoV-2 (Covid-19).). J Clin Med 2020; 9(3):841. doi. 10.3390/jcm9030841.

9. Blalock JE. Harnessing a neural-immune circuit to control inflammation and shock. J Exp Med 2002; 195:F25–8.

10. Bohnen NI, Müller ML, Kotagal V, Koeppe RA, Kilbourn MA, Albin RL, Frey KA. Olfactory dysfunction, central cholinergic integrity and cognitive impairment in Parkinson's disease. Brain. 2010 Jun;133(Pt 6):1747-54. doi: 10.1093/brain/awq079.

11. Borovikova LV, Ivanova S, Zhang M, Yang H, Botchkina GI, Watkins LR, Wang H, Abumrad N, Eaton JW, Tracey KJ. Vagus nerve stimulation attenuates the systemic inflammatory response to endotoxin. Nature. 2000 May 25;405(6785):458-62.

12. Butowt R, Bilinska K. SARS-CoV-2: Olfaction, Brain Infection, and the Urgent Need for Clinical Samples Allowing Earlier Virus Detection. ACS Chem Neurosci. 2020 Apr 13. doi: 10.1021/acschemneuro.0c00172.

13. Cai G. Bulk and Single-Cell Transcriptomics Identify Tobacco-Use Disparity in Lung Gene Expression of ACE2, the Receptor of 2019-nCov. Preprints 2020, 2020020051 doi. 10.20944/preprints202002.0051.v3 (prepublication)

14. Centers for Disease Control and Prevention. Preliminary Estimates of the Prevalence of Selected Underlying Health Conditions Among Patients with Coronavirus Disease 2019 —

United States, February 12–March 28, 2020. MMWR Morb Mortal Wkly Rep 2020. doi: 10.15585/mmwr.mm6913e2.

15. Chen N, Zhou M, Dong X, et al. Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study. Lancet. 2020;395(10223):507-513. doi:10.1016/S0140-6736(20)30211-7

16. Ciaglia E, Vecchione C, Puca AA. COVID-19 infection and the predictive ACE2 soluble levels: the favourable protection of children and women. Front. Pediatr 2020. doi: 10.3389/fped.2020.00206

17. Cohen S, Tyrrell DA, Russell MA, Jarvis MJ, Smith AP. Smoking, alcohol consumption, and susceptibility to the common cold. Am J Public Health. 1993 Sep;83(9):1277-83.

18. Cristiani L, Mancino E, Matera L, Nenna R, Pierangeli A, Scagnolari C, Midulla F. Will children reveal their secret? The coronavirus dilemma. Eur Respir J. 2020 Apr 2. pii: 2000749. doi: 10.1183/13993003.00749-2020.

19. Cui S, Chen S, Li X, Liu S, Wang F. Prevalence of venous thromboembolism in patients with severe novel coronavirus pneumonia. J Thromb Haemost. 2020. 10.1111/jth.14830.

20. D'Elia RV, Harrison K, Oyston PC, Lukaszewski RA, Clark GC. Targeting the "cytokine storm" for therapeutic benefit. Clin Vaccine Immunol. 2013 Mar;20(3):319-27. doi: 10.1128/CVI.00636-12.

21. D'Souza RD, Vijayaraghavan S. Paying attention to smell: cholinergic signaling in the olfactory bulb. Front Synaptic Neurosci. 2014 Sep 25;6:21. doi: 10.3389/fnsyn.2014.00021. eCollection 2014.

22. Desforges M., Le Coupanec A., Dubeau P. Human coronaviruses and other respiratory viruses: underestimated opportunistic pathogens of the central nervous system? Viruses. 2020;12:14

23. Dinarello CA, Cannon JG, Wolff SM, Bernheim HA, Beutler B, Cerami A, Figari IS, Palladino MA Jr, O'Connor JV. Tumor necrosis factor (cachectin) is an endogenous pyrogen and induces production of interleukin 1. J Exp Med. 1986 Jun 1;163(6):1433-50.

24. Dinarello CA. Historical insights into cytokines. Eur J Immunol. 2007 Nov;37 Suppl 1:S34-45.

25. Dinarello CA. Infection, fever, and exogenous and endogenous pyrogens: some concepts have changed. J Endotoxin Res. 2004;10(4):201-22.

26. Ding Y, Wang H, Shen H, Li Z, Geng J, Han H, Cai J, Li X, Kang W, Weng D, Lu Y, Wu D, He L, Yao K. The clinical pathology of severe acute respiratory syndrome (SARS): a report from China. J Pathol. 2003 Jul;200(3):282-9.

27. Docea, A. O., Tsatsakis, A., Albulescu, D., Cristea, O., Zlatian, O., Vinceti, M., Moschos, S. A., Tsoukalas, D., Goumenou, M., Drakoulis, N., Dumanov, J. M., Tutelyan, V. A., Onischenko, G. G., Aschner, M., Spandidos, D. A., Calina, D."A new threat from an old enemy: Re emergence of coronavirus (Review)". International Journal of Molecular Medicine 45.6 (2020): 1631-1643.

28. Doobay MF, Talman LS, Obr TD, Tian X, Davisson RL, Lazartigues E. Differential expression of neuronal ACE2 in transgenic mice with overexpression of the brain renin-angiotensin system. Am J Physiol Regul Integr Comp Physiol. 2007 Jan;292(1):R373-81.

29. Doty RL, Deems DA, Stellar S. Olfactory dysfunction in parkinsonism: a general deficit unrelated to neurologic signs, disease stage, or disease duration. Neurology. 1988 Aug;38(8):1237-44.

30. Farsalinos K, Barbouni A, Niaura R. Smoking, vaping and hospitalization for COVID-19. Qeios ID: Z69O8A.2. 2020a. doi. 10.32388/Z69O8A.2.

31. Farsalinos K, Barbouni A, Niaura R. Smoking, vaping and hospitalization for COVID-19. Qeios ID: Z69O8A.11. 2020b. doi. 10.32388/Z69O8A.11.

32. Farsalinos K, Barbouni A, Niaura R. Systematic review of the prevalence of current smoking among hospitalized COVID-19 patients in China: could nicotine be a therapeutic option? Intern Emerg Med 2020c (in press).

33. Feldmann M, Maini RN, Woody JN, Holgate ST, Winter G, Rowland M, Richards D, Hussell T. Trials of anti-tumour necrosis factor therapy for COVID-19 are urgently needed. Lancet 2020. doi. 10.1016/S0140-6736(20)30858-8.

34. Ferrara JL, Abhyankar S, Gilliland DG. Cytokine storm of graft-versus-host disease: a critical effector role for interleukin-1. Transplant Proc. 1993 Feb;25(1 Pt 2):1216-7.

35. Georgiev T. Coronavirus disease 2019 (COVID-19) and anti-rheumatic drugs. Rheumatol Int. 2020 Mar 30. doi: 10.1007/s00296-020-04570-z.

36. Giacomelli A, Pezzati L, Conti F, Bernacchia D, Siano M, Oreni L, Rusconi S, Gervasoni C, Ridolfo AL, Rizzardini G, Antinori S, Galli M. Self-reported olfactory and taste disorders in SARS-CoV-2 patients: a cross-sectional study. Clin Infect Dis. 2020 Mar 26. pii: ciaa330. doi: 10.1093/cid/ciaa330.

37. Goumenou, M., Sarigiannis, D., Tsatsakis, A., Anesti, O., Docea, A. O., Petrakis, D., Tsoukalas, D., Kostoff, R., Rakitskii, V., Spandidos, D. A., Aschner, M., Calina, D."COVID 19 in Northern Italy: An integrative overview of factors possibly influencing the sharp increase of the outbreak (Review)". Molecular Medicine Reports (2020a), 0.0 (1899): 0-0.

38. Goumenou, M., Spandidos, D. A., Tsatsakis, A."[Editorial] Possibility of transmission through dogs being a contributing factor to the extreme Covid 19 outbreak in North Italy". Molecular Medicine Reports 21.6 (2020b): 2293-2295.

39. Haehner A, Hummel T, Reichmann H. Olfactory loss in Parkinson's disease. Parkinsons Dis. 2011;2011:450939. doi: 10.4061/2011/450939.

40. Hofer S, Eisenbach C, Lukic IK, Schneider L, Bode K, Brueckmann M, Mautner S, Wente MN, Encke J, Werner J, Dalpke AH, Stremmel W, Nawroth PP, Martin E, Krammer PH, Bierhaus A, Weigand MA. Pharmacologic cholinesterase inhibition improves survival in experimental sepsis. Crit Care Med. 2008 Feb;36(2):404-8.

41. Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, Zhang L, Fan G, Xu J, Gu X, Cheng Z, Yu T, Xia J, Wei Y, Wu W, Xie X, Yin W, Li H, Liu M, Xiao Y, Gao H, Guo L, Xie J, Wang G, Jiang R, Gao Z, Jin Q, Wang J, Cao B. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet. 2020;395:497-506.

42. Huang KJ, Su IJ, Theron M, Wu YC, Lai SK, Liu CC, Lei HY. An interferon-gamma-related cytokine storm in SARS patients. J Med Virol. 2005 Feb; 75(2):185-94.

43. Hung YH, Hsieh WY, Hsieh JS, Liu FC, Tsai CH, Lu LC, Huang CY, Wu CL, Lin CS3. Alternative Roles of STAT3 and MAPK Signaling Pathways in the MMPs Activation and

Progression of Lung Injury Induced by Cigarette Smoke Exposure in ACE2 Knockout Mice. Int J Biol Sci 2016;12:454–465. doi:10.7150/ijbs.13379.

44. Imai Y, Kuba K, Penninger JM. The discovery of angiotensin-converting enzyme 2 and its role in acute lung injury in mice. Exp Physiol. 2008 May;93(5):543-8. doi: 10.1113/expphysiol.2007.040048.

45. Imai Y, Kuba K, Rao S, Huan Y, Guo F, Guan B, Yang P, Sarao R, Wada T, Leong-Poi H, Crackower MA, Fukamizu A, Hui CC, Hein L, Uhlig S, Slutsky AS, Jiang C, Penninger JM. Angiotensin-converting enzyme 2 protects from severe acute lung failure. Nature. 2005 Jul 7;436(7047):112-6.

46. Ji W, Wang W, Zhao X, Zai J, Li X. Cross-species transmission of the newly identified coronavirus 2019-nCoV. J Med Virol. 2020 Apr;92(4):433-440. doi: 10.1002/jmv.25682.

47. Kollias A, Kyriakoulis KG, Dimakakos E, Poulakou G, Stergiou GS, Syrigos K. Thromboembolic risk and anticoagulant therapy in COVID-19 patients: Emerging evidence and call for action. Br J Haematol. 2020 Apr 18. doi: 10.1111/bjh.16727.

48. Kooijman S, Meurs I, van der Stoep M, Habets KL, Lammers B, Berbée JF, Havekes LM, van Eck M, Romijn JA, Korporaal SJ, Rensen PC. Hematopoietic α7 nicotinic acetylcholine receptor deficiency increases inflammation and platelet activation status, but does not aggravate atherosclerosis. J Thromb Haemost. 2015 Jan;13(1):126-35. doi: 10.1111/jth.12765.

49. Leung JM, Yang CX, Tam A, Shaipanich T, Hackett TL, Singhera GK, Dorscheid DR, Sin DD. ACE-2 Expression in the Small Airway Epithelia of Smokers and COPD Patients:

Implications for COVID-19. medRxiv 2020.03.18.20038455. doi. 10.1101/2020.03.18.20038455. (prepublication).

50. Manji H, Carr AS, Brownlee WJ, Lunn MP. Neurology in the time of covid-19. BMJ 2020. doi. 0.1136/jnnp-2020-323414.

51. Mao L, Jin H, Wang M, Hu Y, Chen S, He Q, Chang J, Hong C, Zhou Y, Wang D, Miao X, Li Y, Hu B. Neurologic Manifestations of Hospitalized Patients With Coronavirus Disease 2019 in Wuhan, China. JAMA Neurol. 2020 Apr 10. doi: 10.1001/jamaneurol.2020.1127.

52. McGonagle D, Sharif K, O'Regan A, Bridgewood C. The Role of Cytokines including Interleukin-6 in COVID-19 induced Pneumonia and Macrophage Activation Syndrome-Like Disease. Autoimmun Rev. 2020 Apr 3:102537. doi: 10.1016/j.autrev.2020.102537.

53. Mehta P, McAuley DF, Brown M, Sanchez E, Tattersall RS, Manson JJ. HLH Across Speciality Collaboration, UK. COVID-19: consider cytokine storm syndromes and immunosuppression. Lancet. 2020 Mar 28;395(10229):1033-1034. doi: 10.1016/S0140-6736(20)30628-0.

54. Millett ER, De Stavola BL, Quint JK, Smeeth L, Thomas SL. Risk factors for hospital admission in the 28 days following a community-acquired pneumonia diagnosis in older adults, and their contribution to increasing hospitalisation rates over time: a cohort study. BMJ Open. 2015 Dec 1;5(12):e008737. doi: 10.1136/bmjopen-2015-008737.

55. Müller A, Müngersdorf M, Reichmann H, Strehle G, Hummel T. Olfactory function in Parkinsonian syndromes. J Clin Neurosci. 2002 Sep; 9(5):521-4.

56. Newhouse P, Kellar K, Aisen P, White H, Wesnes K, Coderre E, Pfaff A, Wilkins H, Howard D, Levin ED. Nicotine treatment of mild cognitive impairment: a 6-month double-blind pilot clinical trial. Neurology. 2012 Jan 10;78(2):91-101. doi: 10.1212/WNL.0b013e31823efcbb.

57. Ng DL, Al Hosani F, Keating MK, Gerber SI, Jones TL, Metcalfe MG, Tong S, Tao Y, Alami NN, Haynes LM, Mutei MA, Abdel-Wareth L, Uyeki TM, Swerdlow DL, Barakat M, Zaki SR. Clinicopathologic, Immunohistochemical, and Ultrastructural Findings of a Fatal Case of Middle East Respiratory Syndrome Coronavirus Infection in the United Arab Emirates, April 2014. Am J Pathol. 2016 Mar;186(3):652-8. doi: 10.1016/j.ajpath.2015.10.024.

58. Oakes JM, Fuchs RM, Gardner JD, Lazartigues E, Yue X. Nicotine and the renin-angiotensin system. Am J Physiol Regul Integr Comp Physiol. 2018;315(5):R895-R906. doi: 10.1152/ajpregu.00099.2018.

59. Oppo V, Melis M, Melis M, Tomassini Barbarossa I, Cossu G. "Smelling and Tasting" Parkinson's Disease: Using Senses to Improve the Knowledge of the Disease. Front Aging Neurosci. 2020 Feb 25;12:43. doi: 10.3389/fnagi.2020.00043. eCollection 2020.

60. Pavlov VA, Wang H, Czura CJ, Friedman SG, Tracey KJ. The cholinergic anti-inflammatory pathway: a missing link in neuroimmunomodulation. Mol Med. 2003 May-Aug;9(5-8):125-34. Review.

61. Pullan RD, Rhodes J, Ganesh S, Mani V, Morris JS, Williams GT, Newcombe RG, Russell MA, Feyerabend C, Thomas GA, Sawe U. Transdermal nicotine for active ulcerative colitis. N Engl J Med. 1994 Mar 24;330(12):811-5.

62. Qin C, Zhou L, Hu Z, Zhang S, Yang S, Tao Y, Xie C, Ma K, Shang K, Wang W, Tian DS. Dysregulation of immune response in patients with COVID-19 in Wuhan, China. Clin Infect Dis. 2020 Mar 12. pii: ciaa248. doi: 10.1093/cid/ciaa248.3

63. RESPADD. Prendre en charge les fumeurs dans les lieux de santé. Un livret d'aide à la pratique pour les professionnels. 2020. Available at: https://www.respadd.org/wp-content/uploads/2019/09/Livret-Prise-en-charge-LSST-09-2019-BAT3.pdf (accessed on April 18, 2020).

64. Richardson S, Hirsch JS, Narasimhan MN, Crawford JM, McGinn T, Davidson KW. The Northwell COVID-19 Research Consortium. Presenting Characteristics, Comorbidities, and Outcomes Among 5700 Patients Hospitalized With COVID-19 in the New York City Area. JAMA. 2020. Doi.10.1001/jama.2020.6775

65. Ruan Q, Yang K, Wang W, Jiang L, Song J. Clinical predictors of mortality due to COVID-19 based on an analysis of data of 150 patients from Wuhan, China. Intensive Care Med. 2020 Mar 3. doi: 10.1007/s00134-020-05991-x.

66. Rutherford AI, Subessinghe S, Hyrich KL, Galloway JB. Serious infection across biologic-treated patients with rheumatoid arthritis: results from the British Society for Rheumatology Biologics Register for Rheumatoid Arthritis. Ann Rheum Dis. 2018;77:905–910.

67. Schedel A, Thornton S, Schloss P, Klüter H, Bugert P. Human platelets express functional alpha7-nicotinic acetylcholine receptors. Arterioscler Thromb Vasc Biol. 2011 Apr;31(4):928-34. doi: 10.1161/ATVBAHA.110.218297

68. Shimabukuro-Vornhagen A, Gödel P, Subklewe M, Stemmler HJ, Schlößer HA, Schlaak M, Kochanek M, Böll B, von Bergwelt-Baildon MS. Cytokine release syndrome. J Immunother Cancer. 2018 Jun 15;6(1):56. doi: 10.1186/s40425-018-0343-9.

69. Skalny, A. V., Rink, L., Ajsuvakova, O. P., Aschner, M., Gritsenko, V. A., Alekseenko, S. ., Svistunov, A. A., Petrakis, D., Spandidos, D. A., Aaseth, J., Tsatsakis, A., Tinkov, A. A."Zinc and respiratory tract infections: Perspectives for COVID 19 (Review)". International Journal of Molecular Medicine (2020), 0.0 (1899): 0-0.

70. Tang N, Li D, Wang X, Sun Z. Abnormal Coagulation parameters are associated with poor prognosis in patients with novel coronavirus pneumonia. J Thromb Haemost. 2020;18:844-847.

71. Tisoncik JR, Korth MJ, Simmons CP, Farrar J, Martin TR, Katze MG. Into the eye of the cytokine storm. Microbiol Mol Biol Rev. 2012 Mar;76(1):16-32. doi: 10.1128/MMBR.05015-11.

72. Tracey KJ, Beutler B, Lowry SF, Merryweather J, Wolpe S, Milsark IW, Hariri RJ, Fahey TJ 3rd, Zentella A, Albert JD, et al. Shock and tissue injury induced by recombinant human cachectin. Science. 1986 Oct 24;234(4775):470-4.

73. Tracey KJ. Physiology and immunology of the cholinergic antiinflammatory pathway. J Clin Invest. 2007 Feb;117(2):289-96.

74. Tracey KJ. The inflammatory reflex. Nature 2002; 420:853–9.

75. Turner MD, Nedjai B, Hurst T, Pennington DJ. Cytokines and chemokines: At the crossroads of cell signalling and inflammatory disease. Biochim Biophys Acta. 2014 Nov;1843(11):2563-2582. doi: 10.1016/j.bbamcr.2014.05.014.

76. Ulloa L. The vagus nerve and the nicotinic anti-inflammatory pathway. Nat Rev Drug Discov. 2005 Aug;4(8):673-84.

77. Vaduganathan M, Vardeny O, Michel T, McMurray JJV, Pfeffer MA, Solomon SD. Renin-Angiotensin-Aldosterone System Inhibitors in Patients with Covid-19. N Engl J Med. 2020. doi: 10.1056/NEJMsr2005760.

78. Villafane G, Cesaro P, Rialland A, Baloul S, Azimi S, Bourdet C, Le Houezec J, Macquin-Mavier I, Maison P. Chronic high dose transdermal nicotine in Parkinson's disease: an open trial. Eur J Neurol. 2007 Dec;14(12):1313-6. Epub 2007 Oct 17.

79. Wang H, Yu M, Ochani M, Amella CA, Tanovic M, Susarla S, Li JH, Wang H, Yang H, Ulloa L, Al-Abed Y, Czura CJ, Tracey KJ. Nicotinic acetylcholine receptor alpha7 subunit is an essential regulator of inflammation. Nature. 2003 Jan 23;421(6921):384-8.

80. World Health Organization (WHO). Global Adult Tobacco Survey (GATS). Fact sheet China 2018. Available at: https://www.who.int/docs/default-source/wpro---documents/countries/china/2018-gats-china-factsheet-cn-en.pdf?sfvrsn=3f4e2da9_2 (accessed on March 30, 2020).

81. Wu Z, McGoogan JM. Characteristics of and Important Lessons From the Coronavirus Disease 2019 (COVID-19) Outbreak in China: Summary of a Report of 72 314 Cases From the Chinese Center for Disease Control and Prevention. JAMA. 2020 Feb 24. doi: 10.1001/jama.2020.2648.

82. Xia H, Lazartigues E. Angiotensin-converting enzyme 2: central regulator for cardiovascular function. Curr Hypertens Rep. 2010 Jun;12(3):170-5. doi: 10.1007/s11906-010-0105-7.

83. Xu Z, Shi L, Wang Y, Zhang J, Huang L, Zhang C, Liu S, Zhao P, Liu H, Zhu L, Tai Y, Bai C, Gao T, Song J, Xia P, Dong J, Zhao J, Wang FS. Pathological findings of COVID-19 associated with acute respiratory distress syndrome. Lancet Respir Med. 2020 Apr;8(4):420-422. doi: 10.1016/S2213-2600(20)30076-X.

84. Yang J, Lv DJ, Li LX, Wang YL, Qi D, Chen J, Mao CJ, Wang F, Liu Y, Hu LF, Liu CF. Nicotine improved the olfactory impairment in MPTP-induced mouse model of Parkinson's disease. Neurotoxicology. 2019 Jul;73:175-182. doi: 10.1016/j.neuro.2019.02.008.

85. Yue X, Basting TM, Flanagan TW, Xu J, Lobell TD, Gilpin NW, Gardner JD, Lazartigues E. Nicotine Downregulates the Compensatory Angiotensin-Converting Enzyme 2/Angiotensin Type 2 Receptor of the Renin–Angiotensin System. Ann Am Thorac Soc. 2018 Apr; 15(Suppl 2): S126–S127. doi: 10.1513/AnnalsATS.201706-464MG.

86. Zhang H, Penninger JM, Li Y, Zhong N, Slutsky AS. Angiotensin-converting enzyme 2 (ACE2) as a SARS-CoV-2 receptor: molecular mechanisms and potential therapeutic target. Intensive Care Med. 2020 Apr;46(4):586-590. doi: 10.1007/s00134-020-05985-9.

87. Zhou F, Yu T, Du R, Fan G, Liu Y, Liu Z, Xiang J, Wang Y, Song B, Gu X, Guan L, Wei Y, Li H, Wu X, Xu J, Tu S, Zhang Y, Chen H, Cao B. Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. Lancet. 2020. doi: 10.1016/S0140-6736(20)30566-3.