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SARS-CoV-2-mediated encephalitis: Role of AT2R receptors in the blood-brain barrier

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ABSTRACT

At the end of 2019, there was an outbreak of a new Coronavirus 2019 (COVID-19 disease). Studies suggest that SARS-CoV-2 can cause infection in the central nervous system (CNS) and trigger neurological symptoms that include headache, nausea and vomiting, mental confusion and loss of smell or taste. These findings reveal that Coronaviruses have neurological tropism and neuroinvasive capacity. The spread of SARS-CoV-2 in the brain tissue possibly occurs through the systemic circulation as reported in patients affected by SARS-CoV. Evidence highlights similarity between the SARS-CoV genome and SARS-CoV-2 and that both interact with the angiotensin-converting enzyme type 2 (ACE2) located in the brain tissue of infected patients. Hence, the presence of ACE2 is likely in the CNS to mediate the entry of the SARS-CoV-2 virus into neural tissue. Our hypothesis suggests that SARS-CoV-2 can cause encephalitis through the production of inflammatory mediators and activation of immune system cells resulting from the interaction of the ACE2 receptor with the viral Spike protein that causes an increase in angiotensin II. This mechanism has the ability to activate immune system cells by exacerbating stimuli at the angiotensin 2 receptor (AT2R). Thus, it leads to a status of brain injury preceded by vascular damage and destruction of the blood-brain barrier, making it responsible for the installation of acute inflammation.

Introduction

At the end of 2019, an outbreak of a new Coronavirus 2019 (COVID-19) emerged in Wuhan in China, being called the causative agent of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) [1,2]. This virus is a *β-coronavirus* that belongs to the subfamily *Orthocoronavirinae* of the family *Coronaviridae* and order *Nividovirales* [2,3], with a positive single-stranded RNA genome approximately 26 to 32 kilobases in size [4], transmitted through respiratory, fecal-oral and body fluids [3]. According to the World Health Organization, more than 5 million people are infected in 216 countries and more than 331 thousand people died from COVID-19 [5]. Thus, this disease is considered as a global health threatdue to its the high rate of worldwide morbidity and mortality.

Recently, studies suggest that SARS-CoV-2 can cause infection in the central nervous system (CNS) and trigger neurological symptoms that include headache [6], nausea and vomiting [7], mental confusion and loss of smell or taste [8]. Corroborating these findings, other types of coronavirus caused neuroinfection [9,10]. In addition, autopsies of patients infected with SARS-CoV-2 reveal the presence of edema of the brain tissue and partial neuronal degeneration [11].

Evidence demonstrates a similarity of 79.5% of the SARS-CoV genome with SARS-CoV-2 [8] and also that both interact with ACE2 located in the brain tissue of infected patients. The presence of ACE2 in the CNS is likely to mediate the entry of the SARS-CoV-2 virus into the neural tissue and cause the destruction of the blood-brain barrier and subsequently neuroinfection [12]. One study showed that SARS-CoV particles expressed in the brain were almost exclusively in neurons [7].

Two cases of viral encephalitis caused by SARS-CoV-2 have been reported due to the presence of this virus RNA in the cerebrospinal fluid, suggesting the neuroinvasive potential of the virus [13,14]. The expression of the ACE2 receptor is the primary means of entry of SARS-CoV-2 intro brain tissue. Therefore, SARS-CoV-2 encephalitis can be expected to be a predominantly white matter disease [15].

Coronaviruses have been shown to have neurological tropism and neuroinvasive capacity [16,17] and can promote inflammation and demyelination in the CNS. After infection, this virus can reach the whole brain and cerebrospinal fluid [16]. The expression ACE2 in

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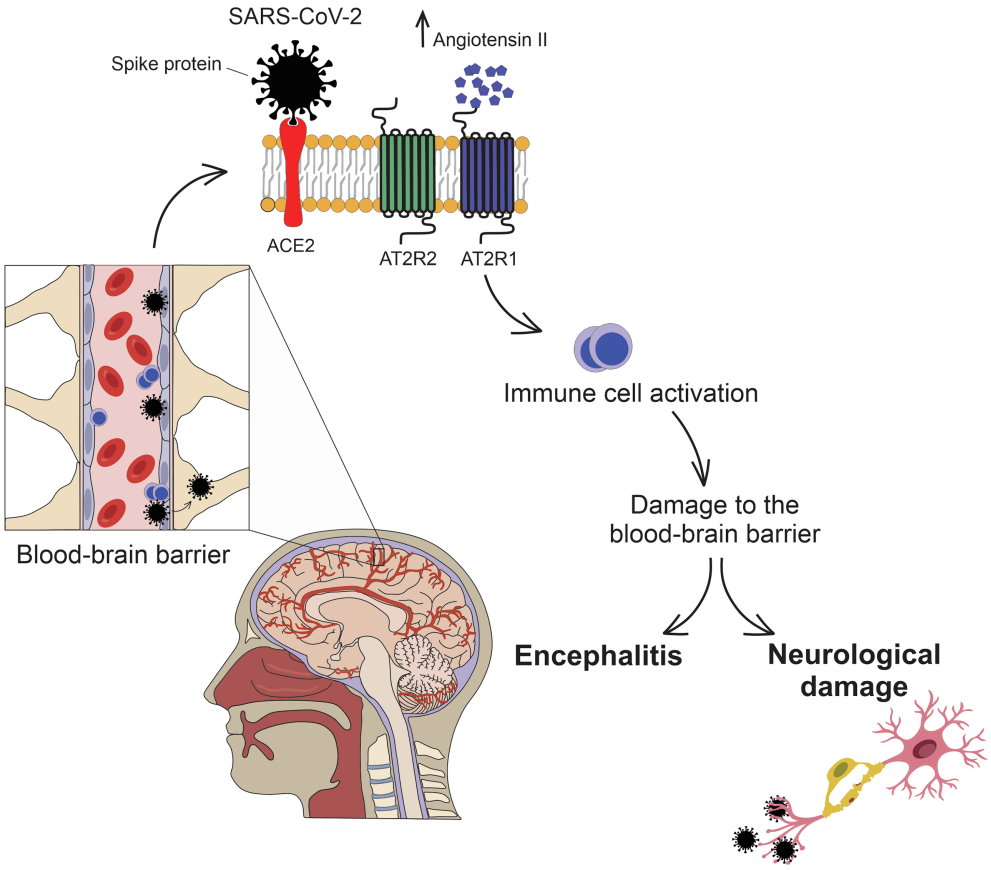

Fig.1. The interaction of the SARS-COV-2 viral spike protein with the ACE2 protein in the vascular endothelium causes an increase in the production of angiotensin II and exacerbation of stimuli in the AT2R1 receptor, leading to the activation of the immune response that causes vascular damage to the blood-brain barrier and encephalitis, even neurological damage.

neurons [18] and in the vascular endothelium [19] suggests the neurotropic potential of SARS-CoV-2 [7,20]. However, there is still no clear evidence of the pathophysiological mechanism of encephalitis caused by SARS-CoV-2.

The hypothesis

The spread of SARS-CoV-2 in brain tissue possibly occurs through systemic circulation as reported in patients affected by SARS-CoV [21]. Thus, our hypothesis is based on the fact that SARS-COV-2 triggers encephalitis through the interaction with the ACE2 protein in the capillary endothelium, causing an increase in the production of angiotensin II and consequently the activation of the immune response through exacerbation of stimuli at the AT2R receptor. That causes vascular damage, exposure and loss of blood-'brain barrier integrity by the release of cytokines and chemokines (Fig. 1).

Evaluation of the hypothesis

Encephalitis is characterized by inflammatory lesions in the brain parenchyma caused by pathogens, which include neuronal damage and damage to nerve tissues. Common symptoms include headache, fever, vomiting, convulsions and disturbances of consciousness [22]. Viral infections in the nervous system can cause serious structural and functional damage, including encephalitis, toxic encephalopathy, demyelinating lesions that develop after viral infections [8].

In viral encephalitis, the primary infection of the epithelium of the mucosa of the nose occurs by inhalation. Therefore, the virus infects neurons by hematogenous dissemination, spreading rapidly through the brain, affecting the thalamus and hippocampus [23]. SARS-CoV-2 encephalopathy is believed to occur by means of a similar mechanism. The frequent and persistent clinical manifestations of anosmia / dysgeusia is present in patients with COVID-19, and neurological symptoms are also reported in severe cases, which may be secondary to severe respiratory failure [24].

This possible propagation mechanism is marked by the viral presence in areas of the brain stem [21], as this structure contains centers for regulating the respiratory rhythm. Breathing is centrally controlled by the regulation of several neural groups. Through the nucleus of the solitary fascicle, the CNS receives information from the chemoreceptors that detect changes in the concentrations of CO2 and O2. Changes in these components lead to an increase or decrease in respiratory effort [25,26].

Thus, the stem nuclei have connections with the respiratory system and the entry of SARS-CoV-2 in this structure can trigger death by altering these neuronal groups, justifying that respiratory discomfort is not just the result of inflammatory pulmonary structural damage, but also due to the damage caused by the virus in the brain's respiratory centers [26].

Neurons do not normally express the major histocompatibility complex (MHC) antigen of any kind, but damaged or electrically silent neurons express the MHC class I antigen that makes neurons targets of CD8+ T lymphocytes [27]. If there is any deregulation in the neurons infected by the coronavirus, this process can occur to eliminate the compromised cell. Consequently, it can trigger an inflammatory process.

Thus, the virus hosts in brain cells and cerebrospinal fluid, triggering immune responses and inflammatory brain tissue, including meninges. Glial cells secrete chemokines and cytokines, and the infection triggers a vigorous response from the innate immune system until adaptive immunity is able to assist in the abolition of the active infection [16]. One study showed that a patient with SARS-CoV-2 infection and encephalitis had a severe increase in inflammatory cytokines in the CSF, such as Interleukin-8, TNF-α, a finding that demonstrates the inflammatory involvement in neurological repercussions related to COVID-19 [28]. Literature data [29] reported a relationship between

acute SARS-CoV-2 infection and aseptic encephalitis with focal neurological symptoms and signs.

At the beginning of the immune response, pattern recognition receptors, called toll-like receptors (TLRs), located in cells of the innate immune system, recognize and bind to conserved viral motifs, known as pathogen-associated molecular patterns. That triggers the dimerization of TLRs, which subsequently activate the signaling pathways that initiate the production of pro-inflammatory cytokines, such as interferons (IFNs), TNF-α and several interleukins [26].

Clinical and experimental studies have showed that the family of coronaviruses shows tropism for the CNS and that they are not always confined to the respiratory system. They can often invade the CNS affecting neurons and glial cells and cause various neurological pathologies [17,30]. Respiratory viruses that affect humans such as influenza virus, *orthopneumovirus* (respiratory syncytial virus), human *metapneumovirus* and coronavirus have been related to many various neurological manifestations in patients with severe respiratory disease [16].

Respiratory syncytial virus detected in cerebrospinal fluid can cause encephalitis, epileptic seizures [31], cerebellitis and ataxia [32]. The Hendra and Nepah viruses in the *Paramyxoviridae* family cause severe pneumonia and encephalitis [33]. The influenza virus can affect the CNS and induce neurological complications such as meningitis [34], encephalitis [35], myelitis [36] and Guillain-Barré syndrome [37], among others. SARS-CoV-2 infection is associated with cases of encephalopathy, encephalitis, necrotizing hemorrhagic encephalopathy, stroke, epileptic seizures, rhabdomyolysis and Guillain-Barre syndrome [30].

Experimental studies with animals and human coronaviruses such as OC43 and 229E have been shown to induce infection in oligodendrocytes and neuroglia [38,39]. The OC43 coronavirus has a neuroinvasive character, triggering demyelination and flaccid paralysis [40]. In the murine model, the coronavirus OC43 has selective tropism for neurons and uses the axonal transport system as a means of spreading the virus among neurons [17], and these mechanisms are also observed in cell cultures [41]. In addition, the RNA of this virus was detected in the CNS of mice suffering from encephalitis induced by this virus. Another coronavirus, the SARS-CoV has penetrated the CNS, allowing the spread of the infection to different regions of the brain [21]. It was also seen that SARS-CoV infection can cause neuronal death in mice knocked out by the human ACE2 receptor [21]. Corroborating these findings, patients with encephalitis have SARS-CoV-2 in the cerebrospinal fluid using genomic sequencing techniques [13,14].

The ACE2 receptor counter-regulates angiotensin II production by binding to the AT2R receptor. Angiotensin has the capacity to activate several cells of the immune system, such as macrophages [42] that induce the production of pro-inflammatory cytokines (Interleukin-6, TNF-α, INF- γ among others) [43]. A study of experimental viral encephalitis in mice identified cellular events that occur in the brain endothelium during the loss of blood-brain barrier integrity caused by INF-γ signaling that induces cell separation [44].

Based on the information that supports our hypothesis of the pathophysiology of SARS-CoV-2 in encephalitis, one can guess that the new coronavirus can act on the CNS and cause neurological symptoms by pathological mechanisms as proposed above. However, the infection and pathophysiology of encephalitis caused by SARS-CoV-2 remain unknown, and further studies are needed to legitimize the scientific mechanisms assumed in this hypothesis.

Authors' contributions

Antônio Kleiton de Sousa developed and wrote the hypothesis and elaborated the figures. Diva de Aguiar Magalhães formatted the article and helped to develop the hypothesis. Jairo dos Santos Ferreira and André Luiz dos Reis Barbosa helped in the writing, review and scientific contributions of the manuscript.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.mehy.2020.110213.

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