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Activation of the trigeminal system as a likely target of SARS-CoV-2 may contribute to anosmia in COVID-19

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Abstract

Clinical publications show consistently that headache is a common symptom in the coronavirus disease of 2019 (COVID-19). Several studies specifically investigated headache symptomatology and associated features in patients with COVID-19. The headache is frequently debilitating with manifold characters including migraine-like characteristics. Studies suggested that COVID-19 patients with headache vs. those without headache are more likely to have anosmia. We present a pathophysiological hypothesis which may explain this phenomenon, discuss current hypotheses about how the coronavirus SARS-CoV-2 enters the central nervous system and suggest that activation of the trigeminal nerve may contribute to both headache and anosmia in COVID-19.

Headache is a frequently described initial symptom of COVID-19 (1). The headache is mostly bilateral though it frequently has a migraine-like phenotype and lasts longer than two days in most cases (2,3). Remarkably, it shows a clear association with anosmia (4,5), however the pathophysiological basis for this association is as yet unclear. Meinhardt and colleagues (6) presented postmortem findings in patients with COVID-19 and discussed how SARS-CoV-2 enters the central nervous system (CNS) and causes multiple neurological symptoms including headache and loss of smell and taste. They showed the presence of SARS-CoV-2 ribonucleic acid (RNA) and protein mainly in the nasal mucosa and certain brain areas. They suggested a transmucosal entry via regional nervous structures, possibly followed by transport along the olfactory tract of the CNS. The olfactory neuroepithelium shows a high expression of ACE2 receptors used by SARS-CoV-2 to enter the cells, which has been suggested to cause the early olfactory dysfunctions (7). Another nervous structure, the trigeminal nerve, has been suggested by Caronna et al. (5) and Bolay et al. (8) to be as likely to serve as a point of entry into the

brain and if so, could not only explain the loss of taste and smell but also the headache associated with an infection of SARS-CoV-2 (Figure 1).

These hypotheses give rise to two principal questions. The first question is: how does the virus penetrate the brain and how it is transported? There are two possible routes for the direct passage of peptides from the nose to the brain, namely an intraneuronal pathway using the axonal transport, and an extraneural pathway through intercellular clefts in the olfactory epithelium directly connected with the subarachnoid space (9).

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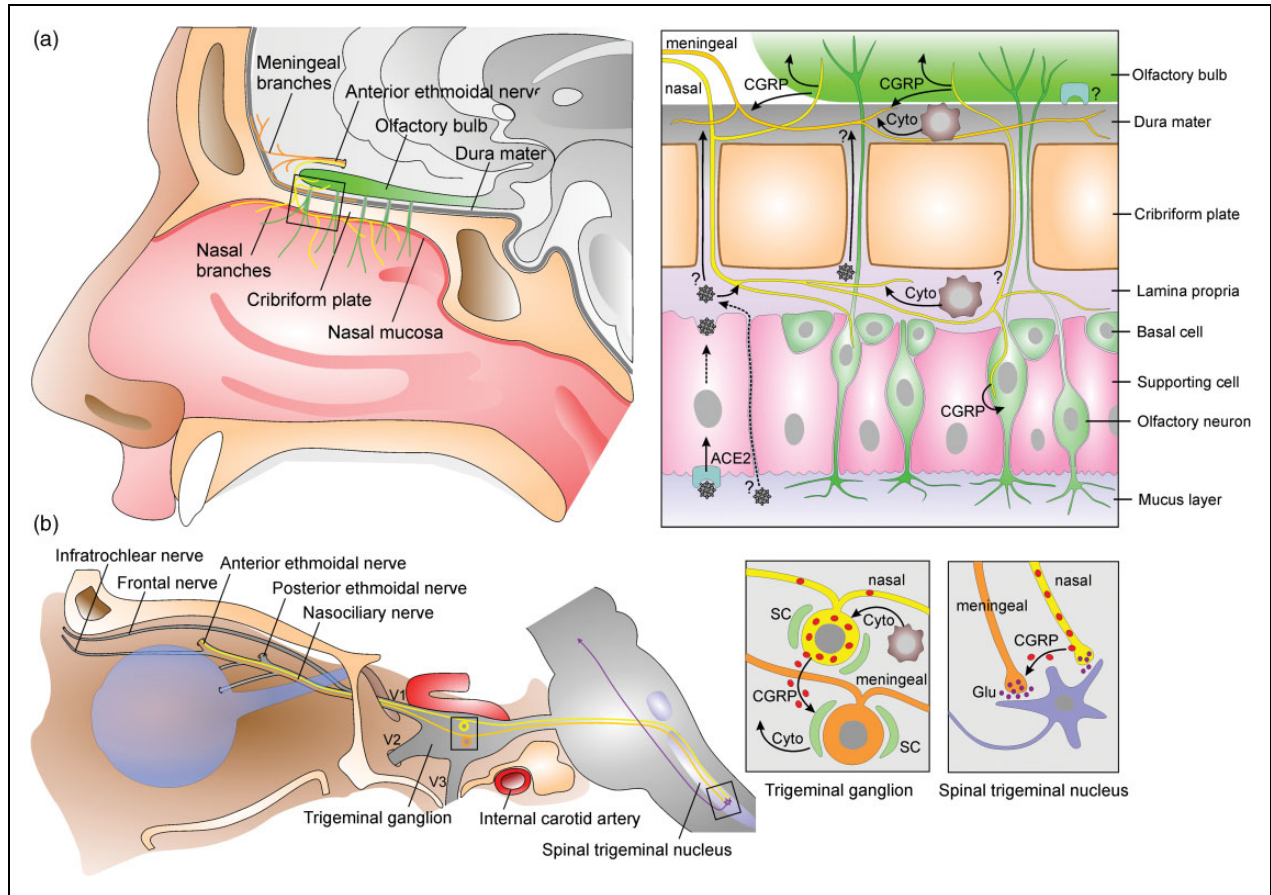Email: a.may@uke.uni-hamburg.de

Figure I Schematic representation illustrating the hypotheses about the movement of SARS-CoV-2 and its effects on nasal olfactory and trigeminal nerve fibers (A) as well as the interaction of trigeminal afferents in the trigeminal ganglion and the spinal trigeminal nucleus (B) leading to headache and anosmia. SARS-CoV-2 binding to ACE2 receptors expressed by supporting cells in the nasal mucosa causes inflammatory and immune reactions involving the release of cytokines (Cyto) from various immune cells in the mucosa and likely also in the dura mater. Possible alternative ways of SARS-CoV-2 penetration into the CNS are the foramina of the cribriform plate or the infection of trigeminal afferents. Cytokines and inflammatory mediators sensitize and activate nasal and meningeal trigeminal afferents, which may also be cross-sensitized by CGRP. However, massive CGRP release from trigeminal afferents can also contribute to attenuating the olfactory function in the nasal mucosa and the olfactory bulb. Cross-sensitization of trigeminal afferents may in addition occur in the trigeminal ganglion, where CGRP can induce cytokine production in satellite glial cells (SC), and in the spinal trigeminal nucleus, where CGRP contributes to synaptic transmission, partly by enhancing glutamate (Glu) release from other central trigeminal terminals.

Large molecules such as wheat germ agglutinin-horseradish peroxidase are known to hardly penetrate the blood-brain-barrier (BBB) but have been found in the CNS shortly after nasal application (10). Even large peptides such as melanocortin, vasopressin and insulin can be demonstrated in cerebrospinal fluid (CSF) within 30-80 minutes after intranasal application, probably bypassing the bloodstream (11). Triptans, which are the widely used treatment of choice for acute migraine and cluster headache attacks, do not or very sparsely penetrate the BBB, yet are effective through nasal application. More to the point, therapeutic concentrations in the CNS of a radiolabelled triptan (zolmitriptan), have been demonstrated shortly after nasal administration using positron emission tomography (PET) (12). An extracellular route via diffusion of SARS-CoV-2 particles through the cribriform plate between the nasal mucosa and the subarachnoidal space seems certainly possible and is supported by findings in mice showing that the CSF can leave the cranial cavity through the cribriform plate (13). On the other hand, there are several possibilities of neuronal transport once the virus gets into a neuron. At first glance, the fila olfactoria, the axons of the primary olfactory neurons, which enter the brain through the cribriform plate and synapse within the olfactory bulb, appear to be a target of such a transport. However, as mentioned above, the SARS-CoV-2 needs ACE2 receptors to

enter the cells but these receptors seem not to be expressed by olfactory sensory neurons but rather by sustentacular cells and basal cells in rodent, primate and human nasopharyngeal mucosa (14,15), although some colocalization of SARS-CoV spike protein immunoreactivity with neural cells in olfactory mucosa samples has been found by Meinhardt et al. (6). Another receptor that binds SARS-CoV-2, the neuropilin-1 receptor (NRP1), which is expressed in olfactory neurons, has recently been suggested as an alternative way to enter the neurons (16). For the trigeminal afferents innervating the nasopharyngeal mucosa no data are currently available regarding ACE2 expression, although this appears possible; at least ACE2 expression has been found in a subset of human dorsal root ganglion neurons (17,18). According to the findings of Meinhardt et al. (6), the density of virus RNA in the olfactory bulb is comparable to the presence of RNA in the trigeminal ganglion, where the cell bodies of the trigeminal afferents are located. However, in the mouse ACE2 expression has been found in superficial layers of the olfactory bulb but not in any neuronal cells (15).

The second principal question is, why SARS-CoV-2 causes a loss of function (anosmia) in the olfactory system but a gain of function (headache) in the trigeminal system; and furthermore, are both these phenomena interconnected? Most of the trigeminal afferents innervating the nasal mucosa are nociceptors expressing transient receptor potential (TRP) cation channels of the type TRPV1, which may be activated by chemical compounds like capsaicin. Inflammatory responses caused by SARS-CoV-2 with a massive increase of cytokines in the nasal mucosa, particularly high TNF-α levels (19), may induce a cascade of nociceptive processes in the trigeminal system. TNF-α induces the expression of a variety of cytokines including IL-1β (20) as well as brain-derived neurotrophic factor (BDNF) (21) and TRPV1 receptor channels (22) in the trigeminal ganglion (TG). TNF-α and IL-1β stimulate expression and release of calcitonin gene-related peptide (CGRP) in TG neurons (23,24) and conversely, CGRP stimulates the expression of cytokines like IL-1β in TG satellite glial cells (25).

In rodents as well as humans, the afferent fibres from the nasal mucosa reach the anterior cranial fossa via the ethmoid nerve and travel in the dura mater to the trigeminal ganglion (26). Activation of these mucosal nerve endings following stimulation with capsaicin releases calcitonin gene-related peptide (CGRP) from the activated nociceptive afferents not only in the nasal mucosa but, importantly, also in the dura mater, leading to an increase in meningeal blood flow (27). If the mucosal trigeminal afferents passing through the dura mater are vigorously activated, the neuro-inflammatory cascades with increased neuropeptide release may spread to the dura mater involving the release of cytokines from macrophages (18) and secondarily activate meningeal nociceptors (28) to cause headache. Furthermore, the TG with multiple nociceptive interactions (29) and eventually the spinal trigeminal nucleus with converging afferent inputs from the nasal mucosa and the meninges (30) can be involved in the neuro-inflammatory cascades. Such a scenario may account for the acute headaches as well as the prolonged headaches accompanying the presumed cytokine storm in COVID-19 (5). The damage to the olfactory pathway caused by the virus or by the inflammatory process may cause anosmia on the one hand and stimulate the trigeminal system causing headache through the above interactions on the other hand. Lesions of the olfactory bulb following COVID-19 have been visualized by neuroimaging (31–33), substantiating structural changes that may underlie the anosmia. Inflammatory mediators produced and released through these lesions are likely to activate trigeminal afferents in the mucosa and the meninges surrounding the olfactory bulb.

However, the loss in olfactory function may not only be explained by the damage of the olfactory epithelium (34) but also by an involvement of the overactive trigeminal afferent system with an increased CGRP release, which in this context surprisingly has an inhibitory effect. The olfactory and trigeminal system are functionally connected and trigeminal activation is increased in patients with acquired anosmia (35). Part of this interaction may occur in the nasal mucosa, where it has experimentally been shown that CGRP released from activated trigeminal fibres inhibits the response of olfactory receptors to olfactory stimuli (36). In addition, from tracer experiments in rodents we know that trigeminal afferents innervating the nasal mucosa and travelling through the ethmoid nerve form collaterals innervating the olfactory bulb (37). There is experimental evidence that these trigeminal endings contribute to inhibitory effects on neurotransmission within the olfactory bulb, again by CGRP release (38). Thus, vigorous activation of trigeminal afferents injured by SARS-CoV-2 may contribute to both headache and concomitant anosmia.

In conclusion, the frequent association of headache and anosmia in COVID-19 may originate with inflammatory responses in the nasal mucosa but probably also requires the activation of meningeal nociceptors. SARS-CoV-2 may penetrate the cribriform plate to affect the meninges around the olfactory bulb. Although this is still a hypothesis that should be further investigated, CGRP released from activated trigeminal afferents may therefore contribute to suppress the olfactory functions in the nasal mucosa and the

olfactory bulb in those patients experiencing simultaneously headache and anosmia

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