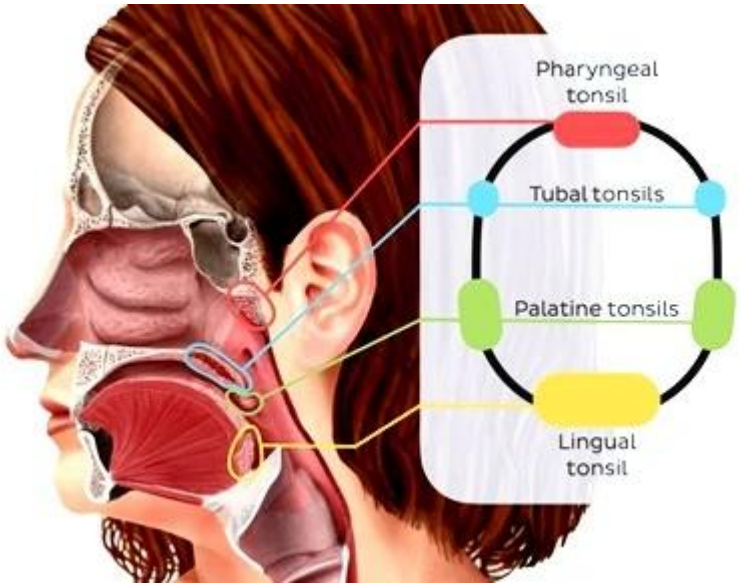
Epipharyngeal lymphoid tissue, located at the posterior part of the nasal cavity, and palatine tonsils are the main lymphatic organs of Waldeyer's ring, the innate immune system of the upper respiratory tract. To assess the epipharynx as a potential reservoir for severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the Japanese researchers in this study analyzed gene expression patterns in epipharyngeal tissue collected from patients with long COVID. They also investigated the efficacy of epipharyngeal abrasive therapy (EAT) in terms of local inflammation and gene expression.



Waldeyer's ring

Chronic epipharyngitis, which progresses from acute epipharyngitis caused by viral infection, is characterized by local symptoms such as chronic cough, sore throat, and pharyngeal discomfort. Epipharyngeal abrasive therapy (EAT) is a treatment method for chronic epipharyngitis developed in Japan. EAT was shown to alleviate local abnormalities in the epipharynx, improve symptoms, and downregulate the mRNA expression of inflammatory cytokines, including interleukin-6 (IL-6) and tumor necrosis factor-alpha (TNFα) in cases of chronic epipharyngitis unrelated to SARS-CoV-2 infection.

Epipharyngeal epithelium highly expresses angiotensin-converting enzyme 2 (ACE2) and transmembrane protease serine 2 (TMPRSS2), utilized by SARS-CoV-2 for host cell entry. The epipharyngeal region is significantly involved during acute infection. As chronic epipharyngitis has been found in the majority of long COVID patients, it is plausible that the residual effects in this region contribute to persistent symptoms. The same research team demonstrated in their previous publication the excellent therapeutic potential of EAT in the elimination of SARS-CoV-2 antigens from the epipharyngeal tissue of patients diagnosed with long COVID.

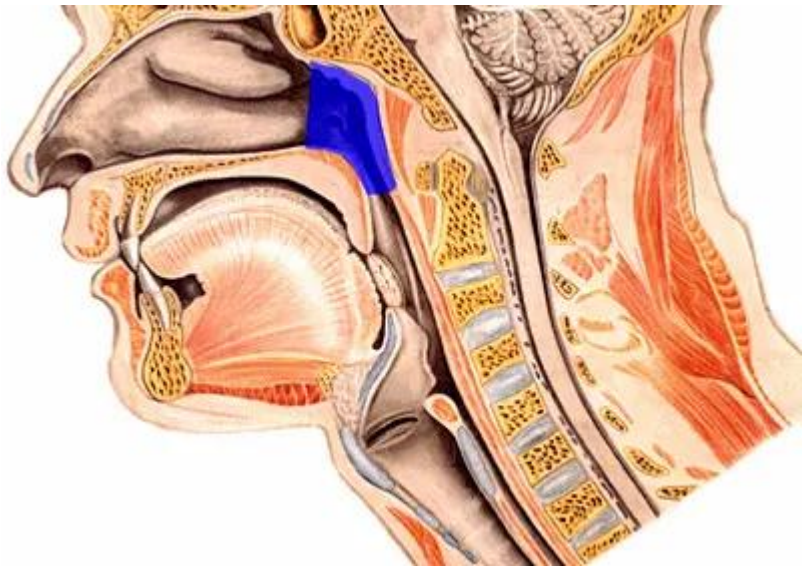
Interestingly, a recent study reported different lineages of SARS-CoV-2 in both epithelial and lymphomononuclear cells of the adenoids and palatine tonsils in one-quarter of children who underwent adenotonsillectomy. This suggests that hypertrophic adenoids and palatine tonsils in children may serve as reservoirs for SARS-CoV-2, even in the absence of COVID-19 symptoms.
https://discovermednews.com/hypertrophic-tonsils-as-sarscov2-reservoir-in-children/

About the study

The authors evaluated three patients with persistent post-COVID symptoms after infection with SARS-CoV-2 during the Omicron period. All three patients with persistent post-COVID symptoms underwent EAT. Patient 1 was a 45-year-old woman with continued dizziness for 2 months after COVID-19, patient 2 was a 21-year-old man who suffered from a persistent cough for 4 months after COVID-19, and patient 3 was a 24-year-old man with persistent severe fatigue that prevented him from attending college for 6 months after COVID-19. Patients 2 and 3 received the COVID-19 vaccine before the COVID-19 infection. The study also included three control patients who developed chronic epipharyngitis before December 2019, when the first cases of COVID-19 were reported.

Tissue samples from all participants were collected by endoscopic epipharyngeal biopsy. Spatial gene expression analysis was performed using Visium HD technology, which precisely maps changes in gene expression in tissue and provides a comprehensive overview of the effects of SARS-CoV-2 on the tissue microenvironment at the cellular level. 16 distinct clusters were derived from seven samples: three samples from long COVID patients, two samples from long COVID patients after EAT, and two samples from COVID-19-uninfected individuals. Each cluster was categorized based on gene expression profiles. Clusters 6, 7,

9, 4, 12, and 13 were epithelial clusters, such as basal cells involved in regeneration and differentiation, while clusters 0, 1, 2, 3, 5, 8, 10, 11, 14, and 15 were non-epithelial clusters composed of immune cells, endothelial cells, fibroblasts, smooth muscle cells, dendritic cells, and stromal cells.



Epipharynx

Results

Analysis of signaling pathways

At the initial visit, *in situ* hybridization confirmed the presence of residual SARS-CoV-2 RNA in the epipharynx of all three patients with long COVID. In those patients, gene expression analysis revealed enriched pathways related to B cell activation and highly expressed Ighg3 and Ighm genes in plasma cells (Ighg3 encodes the Fc domain of IgG3 and Ighm encodes the Fc domain of IgM), implying persistent immune activation and antibody production. Several pathways, including positive regulation of the immune response and interferon signaling, were significantly enriched in the plasmacytoid dendritic cells (pDCs), suggesting that pDCs continue to recognize and respond to viral components. The activation of these pathways in pDCs, together with the heightened responses of B cells and plasma cells, indicates that a complex network of interactions contributes to prolonged inflammation and immune dysregulation in the upper pharynx of long COVID patients.

No significant pathways were detected in T cells. However, several other immune-related

pathways were enriched, including the regulation of the mitogen-activated protein kinase (MAPK) cascade and the binding of TNFs to their physiological receptors.

As SARS-CoV-2 infection leads to ciliary dysfunction, gene expression analysis also revealed a decrease in signaling pathways related to cilia organization and cilia movement in the epipharyngeal epithelium of the long COVID group. Histologic analysis confirmed structural damage to the cilia in the epipharynxes of patients with long COVID.

Effects of epipharyngeal abrasive therapy (EAT)

After administering EAT once a week for 3 months, the SARS-CoV-2 RNA expression disappeared in patients 1 and 2. In patient 3, a substantial reduction in viral RNA expression was observed, but a complete clearance was not achieved.

EAT improved the symptoms in all three patients with long COVID. The inflammation scores for these patients considerably decreased, and the Visual Analog Scale (VAS) scores showed alleviation of their primary complaints.

EAT significantly suppressed activated T-cell receptor (TCR)-signaling pathways and downregulated the expression of inflammatory cytokines IL-6 and TNF-α, which are closely linked to persistent inflammation and immune dysregulation in patients with long COVID. In addition, the long COVID post-EAT group had lower Ighm gene expression in the plasma cell cluster than the long COVID pre-EAT group.

In patient 2, the tissue surface covered by ciliated epithelium before EAT was not observed after treatment. A new cluster of squamous epithelial cells emerged, which confirmed histologic analysis. In the newly emerged cluster of squamous epithelial cells, spatial gene expression analysis showed significantly enriched pathways associated with squamous epithelium and high barrier function. According to the authors, the elimination of inflamed, dysfunctional ciliated epithelial cells and the induction of squamous metaplasia after EAT may contribute to restoring the barrier function of the epipharyngeal mucosa.

Conclusion

Spatial gene expression analysis showed the persistence of SARS-CoV-2-related inflammation in the epipharynx of patients with long COVID. EAT demonstrated great therapeutic potential by reducing excessive activation of TCR-related pathways, persistent inflammation, and overactivation of the humoral immune response in the epipharynx. This

contributed to clinical symptom improvement.

These results suggest a possible link between long COVID and chronic epipharyngitis and emphasize the therapeutic potential of EAT in treating the symptoms of long COVID. The authors concluded that larger sample sizes are required to validate these findings and elucidate the underlying mechanisms by which EAT manages long COVID symptoms.

This article was published in Scientific Reports.

Journal Reference

Nishi, K., Yoshimoto, S., Tanaka, T. *et al.* Spatial transcriptomics of the epipharynx in long COVID identifies SARS-CoV-2 signaling pathways and the therapeutic potential of epipharyngeal abrasive therapy. Sci Rep 2025; 15, 8618 (Open Access). https://doi.org/10.1038/s41598-025-92908-7