

ACE2 Receptor Gene Expression & SARS-CoV-2 Latency in Salivary Glands

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Introduction: COVID-19 disease is due to Severe Acute Respiratory Syndrome Coronavirus-2 (SARS-CoV-2). Viral entry involves binding of the angiotensin converting enzyme 2 (ACE2) receptor on host cell membrane followed by spike-protein cleavage by transmembrane serine protease 2 (TMPRSS2). Salivary glands express both entry factors, ACE2 and TMPRSS2, in ductal and acinar cells. Patients confirmed COVID-19 positive through nasopharyngeal PCR have shown active viral infections in saliva. We hypothesize salivary glands are sites of infectivity for SARS-CoV-2.

Objectives: This project investigates SARS-CoV-2 latency and infectivity in three ways: 1. determine relative ACE2 mRNA expression of salivary glands, 2. determine possible SARS-CoV-2 latency within the salivary glands, and 3. induce ACE2 gene expression following inflammatory cytokine treatment in human derived salivary stem/progenitor cells (hS/PCs).

Experimental Methods: Five salivary gland specimens from consenting patients scheduled for oral surgery who tested negative for SARS-CoV-2 via PCR were included in this study. RNA was isolated from frozen tissue and RT-qPCR was performed to determine transcript levels of ACE2 and SARS-CoV-2. hS/PCs were treated with inflammatory cytokines INF- γ , TNF- α and combination of INF- γ and TNF- α for 24 hours. RNA was then extracted and RT-qPCR was performed.

Results: Salivary gland samples had low levels of ACE2. One out of the five samples showed PCR positivity for SARS-CoV-2 using the N1 primers. Treatment of hS/PCs from a 50-year-old male (M50) with both IFN- γ alone and TNF- α alone had higher ACE2 mRNA levels compared to the no treatment control. The combination treatment showed no ACE2 gene expression.

Conclusion: Salivary glands serve as potential reservoirs for SARS-CoV-2 latency and affect viral transmission. They express low mRNA levels of ACE2. One of our samples tested positive for SARS-CoV-2, but requires further investigation. Inflammatory cytokine treatment of hS/PCs can induce ACE2 gene expression for future infectivity studies in salivary gland models.

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