American Journal of Otolaryngology–Head and Neck Medicine and Surgery 42 (2021) 102911

Contents lists available at ScienceDirect

American Journal of Otolaryngology–Head and Neck Medicine and Surgery

journal homepage: www.elsevier.com/locate/amjoto

Is there a correlation between viral load and olfactory & taste dysfunction in COVID-19 patients?☆

Avani Jain[a,*], A.K. Pandey[b], Jasleen Kaur[a], Lakshit Kumar[a], Mitasha Singh[c], Suman Das[b], Sandeep Purohit[d]

[a] Department of ENT, ESIC Medical College and Hospital, Faridabad, India
[b] Department of Physiology, ESIC Medical College and Hospital, Faridabad, India
[c] Department of Community Medicine, ESIC Medical College and Hospital, Faridabad, India
[d] Rajiv Gandhi Cancer Institute and Research Centre, New Delhi, India

ARTICLE INFO

Keywords:
COVID-19
Cycle threshold values
Gustatory
Olfactory
Viral load

ABSTRACT

Objectives: To evaluate the correlation between cycle threshold (Ct) value and occurrence of olfactory and taste dysfunction in COVID-19 patients.
Methods: This comparative study included COVID-19 patients diagnosed by reverse transcription- polymerase chain reaction (RT-PCR) based test at our hospital with mild to moderate disease. The demographic details and detailed clinical history of the patient, including history of loss of smell and taste was taken at the time of presentation. The patients were divided into 2 groups, group A: COVID-19 patients with OTD; group B: COVID-19 patients without OTD. 100 contiguous patients were recruited in each group. The COVID-19 test by RT-PCR was done and Ct value of the 3 genes: E (Envelope encoding) gene, N (Nucleocapsid encoding) gene, and RdRp (RNA-dependent RNA polymerase) gene, was used for data analysis. The Ct values of each of the three genes were compared between groups A and B.
Results: Group A and B did not differ significantly in terms of basic demographics. The differences in the Ct values of the 3 genes E gene, N gene and RdRp gene, of group A and B were found to be statistically significant ($p = 0.005$, $p = 0.001$ and $p = 0.002$, respectively).
Conclusion: The patients with OTD had a lower Ct value at diagnosis, and hence, a higher viral load than those without OTD. The evaluation of Ct value and viral load in COVID-19 patients may help in further reducing the transmission of the virus in the community.

1. Introduction

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) causes COVID-19, which is an ongoing global pandemic. Its diagnosis is confirmed by detection of viral nucleic acid by RT-PCR, in the upper respiratory samples via nasopharyngeal or oropharyngeal swabs or sputum [1]. Real time reverse transcription polymerase chain reaction (PCR) yields cycle threshold value (Ct value), which is defined as the number of amplification cycles required to reach a threshold for detection of the viral nucleic acid. Ct value is inversely proportional to the amount of target nucleic acid in the sample, i.e., lower the Ct value, greater the amount of target nucleic acid in the sample [2]. The association between Ct values and disease severity of COVID-19 is still controversial.

The common symptoms of COVID-19 include fever, cough, dyspnoea, sore throat, headache, myalgia, rhinorrhoea, diarrhoea [3,4]. Many professional organizations have now recognized olfactory and taste dysfunction (OTD) as symptoms of COVID-19 and included them in their diagnostic guidelines. There is a varying prevalence of olfactory and taste dysfunction in COVID-19 patients, with a higher prevalence reported in the European population as compared to the Asian population [5]. However, there is limited data comparing the Ct values with chemosensory loss. Here, we compared the Ct values, which reflect viral load, of COVID-19 patients with and without OTD.

☆ Conflicts of interest and source of funding (for all the authors) - none declared.

* Corresponding author at: Department of ENT, ESIC Medical College and Hospital, Faridabad, India.
E-mail address: avanijain87@hotmail.com (A. Jain).

https://doi.org/10.1016/j.amjoto.2021.102911
Received 15 December 2020;
Available online 12 January 2021

2. Methods

2.1. Study design and setting

This comparative study was carried out at ESIC Medical College and Hospital, Faridabad. It was approved by the Institutional Ethics Committee. Informed consent was taken prior to inclusion in the study.

2.2. Patient population

The patients who tested positive for COVID-19 infection by reverse transcription- polymerase chain reaction (RT-PCR) based test at our hospital with mild to moderate disease and willingness to participate in the study were included. The exclusion criteria were: children (<18 years of age), psychiatric or neurological disorders, history of previous surgery or radiation in the oral and nasal cavities, chronic rhinosinusitis, pre-existing smell and taste disturbances, assisted ventilation. The demographic details and detailed clinical history of the patient was taken at the time of presentation. Detailed history regarding loss of smell and taste was taken during ENT consultation or telephonically. The patients were divided into 2 groups, group A: COVID-19 patients with OTD; group B: COVID-19 patients without OTD. 100 contiguous patients were recruited in each group. All the patients were followed up till RT-PCR negative report.

2.3. Sample collection procedure

Nasopharyngeal swab for RT-PCR was taken for these patients at the time of presentation. The nasopharyngeal swab was transported in viral transport medium (VTM) in a triple-layered packaging in an ice-box from the sample collection site to the institutes molecular biology lab.

2.4. Laboratory procedure

RNA was extracted from 150 μL of VTM using viral RNA isolation kit, and eluted in 50 μL of nuclease-free water and used as a template for quantification of SARS-CoV-2 viral RNA levels by reverse transcription polymerase chain reaction (RT-PCR). The COVID-19 test by RT-PCR was done, with 3 gene detection: E (Envelope encoding) gene, N (Nucleocapsid encoding) gene, and RdRp (RNA-dependent RNA polymerase) gene. Cycle threshold (Ct) value of each gene was used for data analysis. The Ct values of each of the three genes were compared between groups A and B. Out of the Ct values of the 3 genes in each patient, the minimum Ct value was noted. The average minimum Ct values of groups A and B were compared for any statistical significance.

2.5. Statistical analysis

The data collected was entered in Microsoft excel software and analysed using Epi info version 7. The data was presented as proportions and mean. The test of significance applied to test the difference between categorical variable in two groups (Group A and B) was chi square and for continuous variable was student 't' test. The level of significance was set at 5%.

3. Results

Group A (COVID-19 patients with OTD) and group B (COVID-19 patients without OTD) included 100 patients each. Group A comprised 58 men and 42 women, whereas group B comprised 62 men and 38 women; and there was no statistically significant difference between the two groups ($p = 0.564$). The mean age in group A was 35.23 ± 11.99 years, and in group B was 35.32 ± 12.92 years; this difference was not statistically significant ($p = 0.96$). Therefore, group A and B did not differ significantly in terms of basic demographics. The symptoms of the patients in group A were: malaise (71%), sore throat (46%), cough (40%), fever (74%), and nasal discharge (26%). The symptoms of the patients in group B were: malaise (53%), sore throat (23%), cough (23%), fever (29%), and nasal discharge (7%).

The average Ct values of the 3 genes E gene, N gene and RdRp gene, of group A and B are mentioned in Table 1. The differences in the Ct values, of all the three genes, between the 2 groups was found to be statistically significant. The average minimum Ct values in group A and B were 23.78 ± 5.83 and 27.63 ± 4.83, respectively; this difference was also statistically significant ($p < 0.001$).

4. Discussion

Sudden onset loss of smell and taste are now well documented symptoms of COVID-19 and have now been incorporated in various diagnostic guidelines. Its prevalence varies between populations. According to a review and meta-analysis by von Bartheld et al. [5], the prevalence of olfactory dysfunction in the East Asian and European populations was found to be 22.4% and 48.4%, respectively; the prevalence of gustatory dysfunction in the East Asian and European populations was found to be 16.2% and 50.3%, respectively; the prevalence of olfactory and/or gustatory dysfunction in the East Asian and European populations was found to be 23.4% and 54.7%, respectively. A meta-analysis of 10 large cohort studies of olfactory dysfunction and nine large cohort studies of gustatory dysfunction, showed a prevalence of 52.5% and 43.93% of olfactory and gustatory dysfunction, respectively [6]. However, the pathophysiology mechanism of olfactory and taste dysfunction in COVID-19 is still unknown. The most likely mechanism is that SARS-CoV-2 targets the angiotensin-converting enzyme 2 (ACE2) receptors found on the sustentacular and basal cells of the nasal epithelium, including the olfactory epithelium. It is also postulated that the virus invades the central nervous system through the olfactory bulb [7].

The diagnosis of COVID-19 requires detection of SARS-CoV2 RNA by RT-PCR on respiratory samples. Higher viral load has been detected in the nasopharynx than oropharynx [8]. Therefore, in this study, nasopharyngeal swab was taken of all the patients. Quantitative RT-PCR provides real time quantification by first transcribing SARS-CoV-2 RNA into DNA by reverse transcriptase. Then quantitative PCR is performed where in a fluorescence signal increases proportionally to the amount of amplified nucleic acid. This yields a cycle threshold (Ct) value that is inversely proportional to the amount of target virus in the sample. Therefore, Ct value may indicate the viral replication activity level and viral load [[9]]. This may be helpful in isolating the patients with a higher viral load to reduce the transmission of the virus. The correlation of COVID-19 symptoms and viral load remains controversial. Some studies indicate that a high viral load might be a risk factor for severe disease [10,11]. Whereas, other studies did not observe any difference in the viral load between asymptomatic and symptomatic patients [12]. These differences may be due to the variation in the study design, the type and timing of respiratory sampling, the method of taking the respiratory sample. We collected only nasopharyngeal swabs of patients at the time of consultation. Only patients with mild to moderate disease were included in our study.

It is still unclear if viral load is associated with olfactory and taste dysfunction. We, therefore, studied the correlation between Ct value and OTD. We found that the Ct values of COVID-19 patients with OTD were significantly lower than the Ct values of the patients without OTD. This

Table 1
Comparison of average Ct values between group A and B.

	Average cycle threshold (Ct) value		
	E gene	N gene	RdRp gene
Group A	24.43 ± 6.70	25.91 ± 7.01	24.74 ± 5.54
Group B	27.39 ± 7.92	29.43 ± 7.72	27.50 ± 6.85
p value	0.005	0.001	0.002

result suggests that COVID-19 patients with OTD had a higher viral load than those without OTD. This association of OTD with a higher viral load may be helpful in preventing the transmission of the virus. Nakagawara et al. [13] similarly, demonstrated that fever and OTDs were significantly associated with a higher viral burden and longer time to negative RT-PCR. Biguenet et al. [14] found that anosmia was significantly associated with a lower viral load. However, they could not exclude that a delayed appearance of this symptom during the course of the disease has allowed time for the viral load to decrease.

This study has few limitations. Firstly, the symptoms of loss of smell and taste were self-reported and no objective or physiological test was done. However, the symptoms were prospectively collected and were not by recall basis. Secondly, this study did not include patients with severe disease, and only patients with mild to moderate disease were recruited in the study.

5. Conclusion

In conclusion, patients with OTD had a lower Ct value at diagnosis, and hence, a higher viral load. Loss of smell and taste are important symptoms in COVID-19 and need to be carefully assessed during the disease, even in patients with mild disease or asymptomatic patients. This may help in further reducing the transmission of the virus in the community.

References

[1] Wölfel R, Corman VM, Guggemos W, Seilmaier M, Zange S, Müller MA, et al. Virological assessment of hospitalized patients with COVID-2019. Nature 2020;581:465–9.

[2] Joynt GM, Wu WK. Understanding COVID-19: what does viral RNA load really mean? Lancet Infect Dis 2020;20:635–6.

[3] Young BE, Ong SWX, Kalimuddin S, Low JG, Tan SY, Loh J, et al. Epidemiologic features and clinical course of patients infected with SARS-CoV-2 in Singapore. JAMA 2020;323:1488–94.

[4] Wan S, Xiang Y, Fang W, Zheng Y, Li B, Hu Y, et al. Clinical features and treatment of COVID-19 patients in Northeast Chongqing. J Med Virol 2020;92:797–806.

[5] von Bartheld CS, Hagen MM, Butowt R. Prevalence of chemosensory dysfunction in COVID-19 patients: a systematic review and meta-analysis reveals significant ethnic differences. Preprint. medRxiv. 2020. https://doi.org/10.1101/2020.06.15.20132134. 2020.06.15.20132134. Published 2020 Jun 17.

[6] Tong JY, Wong A, Zhu D, Fastenberg JH, Tham T. The prevalence of olfactory and gustatory dysfunction in COVID-19 patients: a systematic review and meta-analysis. Otolaryngol Head Neck Surg 2020;163:3–11.

[7] Finsterer J, Stollberger C. Causes of hypogeusia/hyposmia in SARS-CoV2 infected patients. J Med Virol 2020. https://doi.org/10.1002/jmv.25903.

[8] Yoon JG, Yoon J, Song JY, et al. Clinical significance of a high SARS-CoV-2 viral load in the saliva. J Korean Med Sci 2020 May 25;35(20):e195. https://doi.org/10.3346/jkms.2020.35.e195.

[9] Tom MR, Mina MJ. To interpret the SARS-CoV-2 test, consider the cycle threshold value. Clin Infect Dis. 2020;71(16):2252–4.

[10] Liu Y, Yan LM, Wan L, Xiang TX, Le A, Liu JM, et al. Viral dynamics in mild and severe cases of COVID-19. Lancet Infect Dis 2020;20:656–7.

[11] Yu X, Sun S, Shi Y, Wang H, Zhao R, Sheng J. SARS-CoV-2 viral load in sputum correlates with risk of COVID-19 progression. Crit Care 2020;24:170.

[12] Zou L, Ruan F, Huang M, et al. SARS-CoV-2 viral load in upper respiratory specimens of infected patients. N Engl J Med 2020;382:1177–9.

[13] Nakagawara K, Masaki K, Uwamino Y, Kabata H, Uchida S, Uno S, et al. Acute onset olfactory/taste disorders are associated with a high viral burden in mild or asymptomatic SARS-CoV-2 infections. Int J Infect Dis 2020;99:19–22.

[14] Biguenet A, Bouiller K, Marty-Quinternet S, Brunel AS, Chirouze C, Lepiller Q. SARS-CoV-2 respiratory viral loads and association with clinical and biological features. J Med Virol Sep 5, 2020. https://doi.org/10.1002/jmv.26489 [Epub ahead of print. PMID: 32889755].