Received: 14 March 2021 | Accepted: 27 July 2021

DOI: 10.1111/coa.13844

ORIGINAL ARTICLE

WILEY

Virus load and incidence of olfactory, gustatory, respiratory, gastrointestinal disorders in COVID-19 patients: A retrospective cohort study

Mohammad Hosein Taziki Balajelini[1] | Abdolhalim Rajabi[2] | Masoud Mohammadi[3] | Hadi Razavi Nikoo[4] | Alijan Tabarraei[4] | Mohsen Mansouri[5] | Seyed Mehran Hosseini[6]

[1]Department of Otorhinolaryngology, Neuroscience Research Center, School of Medicine, Golestan University of Medical Sciences, Gorgan, Iran

[2]Department of Health Management and Social Development Research Center, Faculty of Health, Golestan University of Medical Sciences, Gorgan, Iran

[3]Department of Golestan Research Centre of Gastroenterology and Hepatology, Golestan University of Medical Sciences, Gorgan, Iran

[4]Department of Microbiology, School of Medicine, Infectious Diseases Research Center, Golestan University of Medical Sciences, Gorgan, Iran

[5]Department of Statistics and Information Technology Management, Golestan University of Medical Sciences, Gorgan, Iran

[6]Department of Physiology, School of Medicine, Neuroscience Research Center, Golestan University of Medical Sciences, Gorgan, Iran

Correspondence
Seyed Mehran Hosseini, Department of Physiology, School of Medicine, Neuroscience Research Center Golestan University of Medical Sciences, Gorgan, Golestan, Iran.
Email: hosseini@goums.ac.ir

Abdolhalim Rajabi, Faculty of Health, Golestan University of Medical Sciences, Gorgan, Iran.
Email: drrajabi.a@goums.ac.ir

Funding information
This study was supported to conduct research by Golestan University of Medical Sciences. No other funding was received for this study

Abstract

Objectives: This study investigated the relationship between viral load and the incidence of olfactory and gustatory dysfunction (OD and GD), the incidence of respiratory and gastrointestinal symptoms and the recovery of OD and GD in COVID-19 patients.

Design: A retrospective cohort study.

Setting and Participants: This study was conducted on 599 outpatients' cases in Golestan province between February and June 2020.

Main Outcome Measures: The incidence, severity (complete or partial) and recovery time of OD and GD and their associations with cycle threshold (CT) values of SARS-CoV-2 polymerase chain reaction were assessed.

Results: The mean age of patients was 38.27 ± 13.62 years. The incidence of general symptoms included myalgia 70.1%, headache 51.8%, fever 47.7% and dyspnoea 21.4%. 41.9% of patients had gastrointestinal symptoms, including abdominal pain 26.5%, diarrhoea 25.2%, nausea 20.5% and vomiting 12.9%. 12.2% of patients had comorbidity. The trimester recovery rates of OD and GD were 93.94% and 94.74% respectively. The mean recovery time of OD and GD was 14.56 ± 13.37 and 13.8 ± 3.77 days respectively. The mean CT value in all patients was 27.45 ± 4.55. There were significant associations between the mean of CT value with headache ($p = 0.04$), GD ($p = 0.002$) and OD ($p = 0.001$).

Conclusions: The finding of this study indicates a possible association between viral load with incidence of OD and GD in COVID-19 patient's cases and assures the recovery of OD/GD in these patients.

KEYWORDS
COVID-19, gustatory disorders, olfaction disorders, taste disorders, viral load

Abdolhalim Rajabi and Seyed Mehran Hosseini contributed equally to this work.

1 | INTRODUCTION

COVID-19 is caused by severe acute respiratory syndrome virus, SARS-CoV-2. Its cumulative incidence and its morbidity are still growing.[1] Gastrointestinal symptoms, such as diarrhoea, abdominal pain, nausea and vomiting, are also common manifestations in COVID-19.[2]

The symptoms of the disease are nonspecific in the early stages and are indistinguishable from the symptoms of the common cold. Even asymptomatic carriers of SARS-CoV-2 can spread the virus and are contagious.[3] Different factors were considered for COVID-19 screening. In summary, they include fever, dry cough, sputum, shortness of breath, sore throat and myalgia, a history of suspected contact with infected individuals, travel to an infected area. OD and GD also received special attention to be used for COVID-19 screening.[4] Following numerous reports of the high prevalence of OD and GD due to COVID-19 in European and American countries, the American Academy of Otolaryngology (Head and Neck Surgery) recommended adding anosmia, ageusia, and dysgeusia to screen for possible COVID-19 infection.[5,6] But, there are still many questions about pathophysiology and the association between OD and GD with the prognosis of COVID-19.

Detection of COVID-19 is performed by Reverse Transcription-Polymerase Chain Reaction (RT-PCR) test.[7] This test shows the nucleic acid of the virus in the saliva–nasal secretions of the patient.[8] In this test, the cycle threshold value (CT) means the number of amplification cycles required to reach the detection threshold of virus nucleic acids. The value of CT is inversely related to the load of the virus in the sample and indirectly indicates the level of virus replication.[9] Sampling for RT-PCR test can be obtained from nasopharynx, oropharynx and lower respiratory tract. Some studies reported that SARS-CoV-2 virus loads in the nasal samples were more than the pharyngeal samples.[10] Viral loads have also been reported to typically increase during the first week after the onset of symptoms and remain high for subsequent weeks.[11] It has also been reported that the amount of virus in samples collected from the lower respiratory tract is very high, but due to specific conditions for this type of sampling, its application as routine practice in all health centres is limited. Thus, it has not been approved by FDA.[12]

There are few studies on virus-related factors that may affect the outcome, and therefore the American Infectious Diseases Association has emphasised quantitative testing as a prognostic factor for the diagnosis of SARS-CoV-2.[13] In this regard, the CT value of the RT-PCR test has been considered as an indicator of virus load in determining the prognosis and outcome of patients and the occurrence of some disorders.

Due to the high prevalence of COVID-19 and a large percentage of patients with OD and GD and the importance of determining the predictors of some complications and outcome of patients, this study was performed to determine the relationship between CT value of RT-PCR test and OD and GD in patients with COVID-19 in Golestan province in Iran.

Key points

- A retrospective analysis of 599 COVID-19 patients was performed to determine the incidence of olfactory and gustatory dysfunction and their recovery.
- The association of OD, GD, recovery rate and recovery time with cycle threshold (CT) values of SARS-CoV-2 RT PCR was assessed.
- The mean of CT value in patients with complete OD was significantly higher than in patients with partial OD. Also, the mean of CT in patients with or without GD showed a significant difference.
- There was no significant relation between a recovery time of OD and GD with the mean of CT value.
- There was a strong correlation between the recovery time of OD and GD.

2 | METHODS

A cohort study was conducted between February and June 2020 in Golestan province on the southern shore of the Caspian Sea in northern Iran. The strengthening of the reporting of observational studies in epidemiology (STROBE) statement was used for reporting the study. Patients who were suspected of having COVID-19 based on clinical signs were evaluated by a family physician and then the COVID-19 disease was confirmed by RT-PCR test. Nasopharyngeal samples were taken from patients with suspicious symptoms of COVID-19 in the first visit to the family physician. The sampling of patients was performed by trained experts. For each patient, a nasopharyngeal sample was taken through the nose and a nasopharyngeal sample was taken orally. Samples were transferred to the Department of Virology of the School of Medicine in Golestan University of Medical Sciences. RT-PCR was performed by ABI Step One Plus, and CT values were extracted. Demographic characteristics of patients including age, sex, time of infection, as well as respiratory symptoms, and gastrointestinal symptoms including fever, headache, myalgia, cough and diarrhoea, vomiting, abdominal pain and nausea were collected from patients' electronic records. The status of OD and GD of outpatients was followed and completed via a telephone call because of logistic limitations of the pandemic within a short period (at most 1–2 weeks). For more reassurance, an otolaryngologist examined patients in person who were unable to accurately describe their condition or for ruling out other aetiologies. Patients were called weekly to record changes or improve their OD or GD. Patients who recovered completely were recorded as the outcome, and patients who had partial recovery were followed up until complete recovery.

The severity of OD and GD was categorised into two types, complete and partial. Patients were classified according to age and three geographical locations in the province, East, Centre and West.

2.1 | Statistical analysis

Statistical analysis was performed using STATA, version 14.1 (Stata Corp; Stata Corp LLC). Data are presented as mean ±standard deviation (SD) and frequency. Concerning quantitative variables, after variance equality and normal distribution of the values were checked, we used the Mann–Whitney U-test to compare the mean of CT value in binary groups, and to compare the mean of CT values in the categorical variable, we used the Kruskal–Wallis test. The correlation between time of recovery of olfactory and gustatory dysfunctions and CT values was assessed using Spearman's rank correlation coefficient. Multivariable analysis was performed for recovery of OD and GD with Cox proportional hazards models, including CT variable. A p value of <0.05 was considered statistically significant.

3 | RESULTS

In this study, 599 RT-PCR confirmed COVID-19 cases were included. There were 313 (52.2%) females and 286 (47.74%) males. The mean age 38.27 ± 13.62 years and 50.75% of cases were in the age group of 20–40 years. The clinical characteristics of patients are shown in Table 1.

The mean CT value in COVID-19 patients was 27.45 ± 4.55. The study showed that the mean CT value of women was 27.34 ± 4.63, and that of men was 27.56 ± 4.46 ($p = 0.54$). The mean of CT value was slightly higher in older age 27.68±4.85 in the age group 60 and above *vs*. 27.35±5.17 in the age group under 20 ($p = 0.94$).

Also, there were no significant differences in the mean of CT value of patients from different ethnicity and regions (eastern, western and central provinces). The mean of CT was 28.13 ± 4.55 in patients with OD and 26.91±4.49 in patients without OD. The mean of CT value between the two groups was significantly different ($p = 0.001$).

The mean of CT in patients with complete OD was significantly higher than patients with partial OD ($p = 0.02$). Also, the mean of CT was 28.16±4.52 in patients with GD and 27.01±4.52 in patients without GD ($p = 0.002$). However, the difference in mean CT in patients with complete and partial GD was not significant ($p = 0.32$).

In patients, without OD or GD, the mean of CT value was less than cases that had at least one of them ($p = 0.001$). However, the mean of CT value of OD-recovered patients and OD-unrecovered patients did not show a significant difference ($p = 0.62$). Also, there was no significant difference between people who had partial recovery and those who had complete recovery ($p = 0.38$).

These comparisons were also carried out for the GD recovery, and the results were similar to the OD recovery. Also, the multivariable Cox-regression analysis model indicated that there was no association between the mean of CT value and GD recovery (Hazard Ration (HR): 0.99; 95% CI: 0.96–1.02, $p = 0.95$) and OD recovery (HR: 0.99; 95% CI: 0.97–1.02, $p = 0.62$). The association of other variables with the recovery time of OD and GD is presented in another research.[14]

The mean of CT value, in COVID-19 patients, did not show a significant difference in terms of having or not having symptoms of fever, myalgia and shortness of breath. However, the mean of CT value of patients with headache symptoms was significantly higher than that of patients without headache symptoms ($p = 0.04$).

The mean of CT value was not significantly different in patients with and without gastrointestinal disorders ($p = 0.48$). Similarly, in the study of each gastrointestinal symptom (abdominal pain, diarrhoea, nausea and vomiting), there was no significant difference in the mean CT value in terms of having or not having these disorders. Also, the comorbidities of diseases, such as diabetes and hypertension, did not have a significant effect on the mean CT for COVID-19 patients.

There was not a significant correlation between OD recovery time and CT and also between GD recovery time and CT. However, there was a strong, direct and significant correlation between the recovery time of OD and GD ($r = 0.97$, $p < 0.001$; Figure 1).

4 | DISCUSSION

In this study, 599 patients were included in the study, of which 52.2% were female and the mean age of patients was 38.27 ± 3.62 years and the maximum age distribution was in the age group of 20–40 years (50.75%). Comparison of age and sex distribution of our patients who were followed up on an outpatient basis was similar to studies by Zhang-Hu et al.[15] However, the mean age of patients in our study was lower. The lower age of our patients may be because these patients have been followed up on an outpatient basis; however, in other studies, patients were hospitalised, most of whom were older.[16]

The mean of CT value in males and females and between age groups had no difference. This finding was similar to others.[17] However, in both studies, the mean of CT value had no statistical difference concerning age groups.

Our data showed a lower viral load in cases with OD and GD. This result is similar to the other reports.[18] In that study, it was also reported that the more severe the OD or GD, the higher the CT value, that is, the lower the virus load. At first glance, it may have been thought that people with OD or GD had a higher number of viruses, but the results show the opposite. Anosmia in COVID-19 is known as the main symptom, but its mechanism or pathophysiology remains still unclear.[19] One possibility is that the SARS-CoV-2 virus attacks ACE2 receptors in the basal and sustentacular cells of the olfactory epithelium. Another possibility is the ability of the virus to invade the central nervous system through the olfactory bulb.[20]

Although it is difficult to explain why COVID-19 patients with a lower load of the virus develop OD or GD, the possible reasons for this can be summarised as follows:

TABLE 1 Characteristics, presenting symptoms of patients with SARS-CoV-2 infection and CT Values from SARS-CoV-2 Polymerase Chain Reaction Assays in included cases in the study

Variables	N (%)/Mean ± SD	CT Values From SARS-CoV–2 PCR Mean ± SD	*p*-value*
Total (N, %)	599 (100)	27.45 ± 4.55	
	Sex (N, %)		
Men	286 (47.75)	27.56 ± 4.46	0.54[a]
Women	313 (52.25)	27.34 ± 4.63	
Age (Mean ± SD)	38.27 ± 13.62		
Age group (N, %)			
<20	40 (6.68)	27.35 ± 5.17	0.94[b]
20–40	304 (50.75)	27.38 ± 4.48	
40–60	210 (35.06)	27.51 ± 4.50	
≥60	45 (7.51)	27.68 ± 4.85	
Ethnicity (N, %)			
Fars	123 (34.6)	27.21 ± 4.71	0.87
Sistani-Persian	32 (9)	27.13 ± 4.68	
Turkmen	200 (56.3)	27.48 ± 4.35	
Location			
West	69 (11.58)	27.31 ± 4.02	0.54
Centre	101 (16.95)	27.91 ± 4.59	
East	426 (71.48)	27.38 ± 4.59	
Olfactory dysfunction (OD)			
Yes	263 (43.9)	28.13 ± 4.55	**0.001**
No	336 (56.1)	26.91 ± 4.49	
The severity of olfactory dysfunction			
Partial	60 (10)	26.87 ± 4.93	
Complete	203 (33.9)	28.50 ± 4.38	**0.02**
None	336 (56.1)	26.91 ± 4.49	
Gustatory dysfunction (GD)			
Yes	228 (38.1)	28.16 ± 4.52	**0.002**
No	371 (61.9)	27.01 ± 4.52	
The severity of Gustatory dysfunction			
Partial	64 (10.7)	27.57 ± 5.09	0.32
Complete	164 (27.4)	28.39 ± 4.27	
None	371 (61.9)	27.01 ± 4.52	
OD and GD[c]			
Yes	263 (44.1)	28.13 ± 4.54	**0.001**
No	335 (55.9)	26.91 ± 4.49	
Recovery of OD			
Yes	248 (93.94)	28.15 ± 4.55	0.62
No	16 (6.06)	27.57 ± 4.63	
Type of recovery of OD			
Complete	213 (80.68)	28.30 ± 4.57	0.38
Partial	35 (13.26)	27.23 ± 4.39	
Non-recovery	16 (6.06)	27.57 ± 4.63	

(Continues)

TABLE 1 (Continued)

Variables	N (%)/Mean ± SD	CT Values From SARS-CoV–2 PCR Mean ± SD	*p*-value*
Recovery of GD			
Yes	216 (94.74)	28.11 ± 4.49	0.95
No	12 (5.26)	28.19 ± 4.93	
Type of recovery of GD			
Complete	182 (79.82)	28.22 ± 4.52	0.70
Partial	34 (14.91)	27.52 ± 4.33	
Non-recovery	12 (5.26)	28.19 ± 4.93	
Fever			
Yes	286 (47.7)	27.70 ± 4.52	0.15
No	313 (52.3)	27.73 ± 4.62	
Myalgia			
Yes	420 (70.1)	27.32 ± 4.52	0.38
No	179 (29.9)	27.73 ± 4.62	
Headache			
Yes	310 (51.8)	27.80 ± 4.51	**0.04**
No	289 (48.2)	27.07 ± 4.57	
Shortness of breath			
Yes	128 (21.4)	27.47 ± 4.83	0.86
No	471 (78.6)	27.44 ± 4.48	
Abdominal pain			
Yes	159 (26.5)	27.34 ± 4.57	0.74
No	440 (73.5)	27.49 ± 4.55	
Diarrhoea			
Yes	151 (25.2)	27.04 ± 4.60	0.24
No	448 (74.8)	27.58 ± 4.53	
Nausea			
Yes	123 (20.5)	27.51 ± 4.57	0.76
No	476 (79.5)	27.43 ± 4.55	
Vomit			
Yes	77 (12.9)	27.56 ± 4.62	0.80
No	522 (87.1)	27.43 ± 4.54	
Gastrointestinal disorders (GI)			
Yes	251 (41.9)	27.27 ± 4.56	0.48
No	348 (58.1)	27.57 ± 4.55	
History of diabetes			
Yes	31 (5.2)	28.23 ± 5.66	0.21
No	568 (94.8)	27.40 ± 4.48	
History of hypertension			
Yes	57 (9.5)	27.92 ± 5.08	0.33
No	542 (90.5)	27.40 ± 4.49	
Comorbidity			
Yes	72 (12.02)	28.08 ± 5.01	0.21
No	527 (87.98)	27.36 ± 4.48	

(Continues)

TABLE 1 (Continued)

Variables	N (%)/Mean ± SD	CT Values From SARS-CoV–2 PCR Mean ± SD	*p*-value*
Pregnant			
Yes	11 (3.9)	29.47 ± 3.59	0.10
No	271 (96.1)	27.17 ± 4.64	

*p values less than 0.05 are Bolded.

[a]For binary variable used the Mann-Whitney Test.

[b]For categorical variable used the Kruskal-Wallis Test.

[c]Exact isolation of the two dysfunction is not possible in this study. The statistically significant differences are shown as Bold numbers in p-value column.

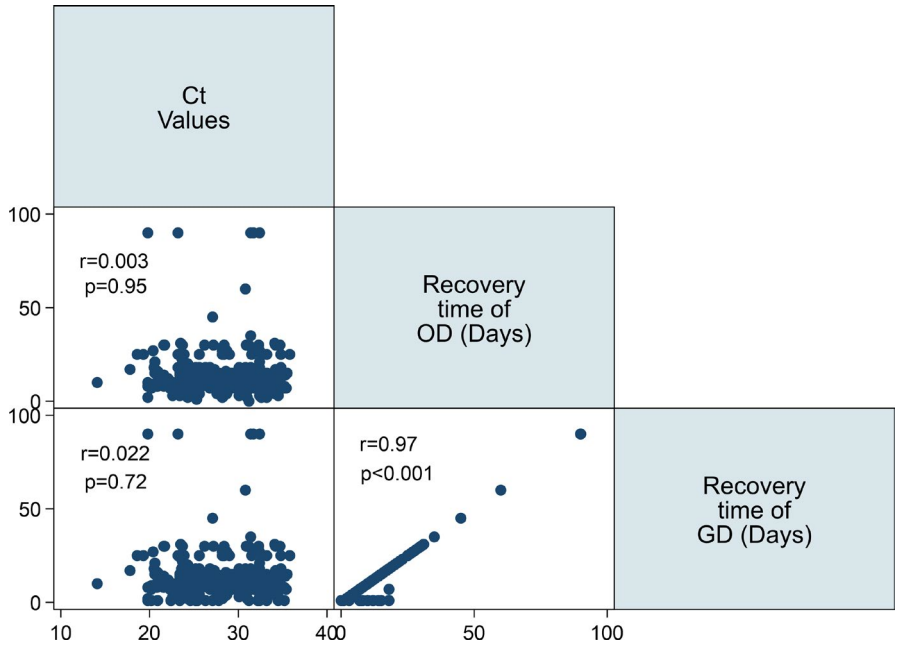

FIGURE 1 Matrix correlation between the recovery time of OD, GD and CT

The first reason may be due to an exaggerated utilisation of detergents in the initial phase of the pandemic. Therefore, it may be speculated that the olfactory epithelium damaged secondary to this chemical-induced inflammation and was more vulnerable to viral invasion. The second possible reason may be due to the presence of ACE as viral receptors in the olfactory epithelium. This inevitably increases the vulnerability of olfactory epithelium as one of the first exposed issues to SARS-CoV-2. Another reason may be related to the patient's anxiety due to the possibility of the sudden loss of sense of smell or taste, which can lead to early referral of patients and this anxiety has led to higher CT values.

In some studies, a significant relationship was observed between low CT value and an increase in LDH, and a decrease in lymphocytes, indicating a worse prognosis.[21,22] SARS-CoV-2-induced pneumonia has also been reported that lower CT values accompanied more tissue damage in the patient's lungs and a worse prognosis.[23] In the study of Zacharioudakis et al., which was performed on hospitalised patients with COVID-19 pneumonia, a change in CT value was reported in accordance with the general clinical condition of the patient. This means that with the improvement of the patient's general condition, the CT value also increased and this index was used to predict the patient's outcome.[24] In the study of Aquino-Jarquin et al, it was reported that CT value and clinical outcome were directly and nonlinearly related.[25] In the meta-analysis of Sonia et al., Low CT value was associated with increased disease severity and was reported to be consistent with the results of other studies.[26] The study by Huang et al. reported an increased risk of mortality with lower CT values.[27] In our study, only a significant relationship was found between headache and CT value. This means that patients with headaches had fewer viruses. This may be because in our study the patients were outpatients but in other studies, the patients were hospitalised.

Although many studies reported the relationship between CT value and prognosis and clinical severity, some other studies had not found such a relationship. In asymptomatic but infected individuals, the viral load was similar to that in symptomatic patients, and it was concluded that virus load alone was not a reliable indicator of predicting disease outcome. According to the results obtained on CT value in OD and GD, it seems that the exact pathophysiology of this phenomenon needs further study.

4.1 | Strength and limitation

For quantifying olfactory assessments in the COVID-19 studies, the most reported method was questionnaires, although interviewing or extracting clinical information from the patient's electronic health records were also used; however, these methods are subjective and imprecise.[21,28-31] There are several objective methods for the precise assessment of smell and taste. They include tests to determine the olfactory threshold and the type of aromatic substance. Although these two tests are accurate, they are completely non-objective and are recommended for relative verification of patients. The other test is the olfactory assay, which is completely objective but only used in specialised neurophysiological laboratories.[32,33] During the pandemic, measuring OD/GD with objective and non-objective methods was not always possible. This was one of the limitations of the present study. Self-assessed OD/GD has the potential of smell and taste confusion.[34-36] So, in this study, it is not possible to know how much of your patients had isolated smell, taste or both impaired.

The viral load of SARS-CoV-2 is known to vary during the course of infection.[10,37] However, in most studies, the CT values at the onset of symptoms have been evaluated.[26] A single CT value does not indicate the viral load during the course of the disease and provides only a point estimate of the viral load at the time of outpatient referral with clinical symptoms. Therefore, in the present study, CT values were measured in this way. This may be another limitation of the present study.

5 | CONCLUSIONS

The finding of this observational study indicates a possible association between lower viral load with some clinical manifestations, including olfactory dysfunction, severity of olfactory dysfunction, gustatory dysfunction and headache in COVID-19 outpatient cases and assures the recovery of OD/GD in these patients.

ACKNOWLEDGEMENTS

We would like to acknowledge the clinical laboratory experts who performed the SARS-CoV-2 laboratory testing.

CONFLICT OF INTEREST

None declared.

AUTHOR CONTRIBUTIONS

MHT, SMH and AT contributed to the study's concept and design. Study materials and their preparation, data collection provided by HRN, MM and MM. Data acquisition and analysis were performed by AR. The first draft of the manuscript was written by AR, SMH and MHT. However, the comments of all authors on the previous versions of the manuscript have been included in this manuscript. All authors have read and approved the manuscript.

ETHICAL STATEMENT

This study was approved by the Research Council of Golestan University of Medical Sciences with ethic code IR.GOUMS.REC.1399.031.

DATA AVAILABILITY STATEMENT

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

ORCID

Abdolhalim Rajabi https://orcid.org/0000-0002-0698-8166
Seyed Mehran Hosseini https://orcid.org/0000-0002-4783-7428

REFERENCES

1. Taylor AW, Price K, Gill TK, et al. Multimorbidity - not just an older person's issue. Results from an Australian biomedical study. *BMC Public Health*. 2010;10(1):718.
2. Zhou P, Yang X-L, Wang X-G, et al. A pneumonia outbreak associated with a new coronavirus of probable bat origin. *Nature*. 2020;579(7798):270-273.
3. Nishiura H, Kobayashi T, Miyama T, et al. Estimation of the asymptomatic ratio of novel coronavirus infections (COVID-19). *Int J Infect Dis*. 2020;94:154-155.
4. Hopkins C, Surda P, Kumar N. Presentation of new onset anosmia during the COVID-19 pandemic. *Rhinology J*. 2020;58(3):295-298.
5. Lechien JR, Chiesa-Estomba CM, De Siati DR, et al. Olfactory and gustatory dysfunctions as a clinical presentation of mild-to-moderate forms of the coronavirus disease (COVID-19): a multicenter European study. *Eur Arch Otorhinolaryngol*. 2020;277(8):2251-2261.
6. AlShakhs A, Almomen A, AlYaeesh I, et al. The association of smell and taste dysfunction with COVID19, and their functional impacts. *Indian J Otolaryngol Head Neck Surg*. 2021;1-6.
7. Tang Y-W, Schmitz JE, Persing DH, Stratton CW. Laboratory diagnosis of COVID-19: current issues and challenges. *J Clin Microbiol*. 2020;58(6):e00512.
8. Wölfel R, Corman VM, Guggemos W, et al. Virological assessment of hospitalized patients with COVID-2019. *Nature*. 2020;581(7809):465-469.
9. Joynt GM, Wu WK. Understanding COVID-19: what does viral RNA load really mean? *Lancet Infect Dis*. 2020;20(6):635-636.
10. 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(12):1177-1179.
11. Pan Y, Zhang D, Yang P, Poon LL, Wang Q. Viral load of SARS-CoV-2 in clinical samples. *Lancet Infect Dis*. 2020;20(4):411-412.
12. Huang Y, Chen S, Yang Z, et al. SARS-CoV-2 viral load in clinical samples of critically ill patients. *Am J Respir Crit Care Med*. 2020;201(11):1435-1438.
13. Lima PB, Santoro IL, Caetano LB, Cabral AL, Fernandes AL. Performance of a word labeled visual analog scale in determining the degree of dyspnea during exercise-induced bronchoconstriction in children and adolescents with asthma. *J Bras Pneumol*. 2010;36(5):532-538.
14. Balajelini MHT, Vakili MA, Rajabi A, Mohammadi M, Tabarraei A, Hosseini SM. Recovery of Olfactory and Gustatory Dysfunctions in COVID-19 Patients in Iran. A Prospective Cohort Study. 2020.
15. Zhang H, Liao Y-S, Gong J, Liu J, Xia X, Zhang H. Clinical characteristics of Coronavirus Disease (COVID-19) patients with gastrointestinal symptoms: a report of 164 cases. *Dig Liver Dis*. 2020;52(10):1076-1079.
16. Taziki Balajelini MH, Vakili MA, Saeidi M, Tabarraei A, Hosseini SM. Using anti-SARS-CoV-2 IgG and IgM antibodies to detect

outpatient cases with olfactory and taste disorders suspected as mild form of COVID-19: a retrospective survey. *SN Compr Clin Med*. 2020;2(12):2554-2560.
17. Buchan BW, Hoff JS, Gmehlin CG, et al. Distribution of SARS-CoV-2 PCR cycle threshold values provide practical insight into overall and target-specific sensitivity among symptomatic patients. *Am J Clin Pathol*. 2020;154(4):479-485.
18. Cho RH, To ZW, Yeung ZW, et al. COVID-19 Viral Load in the Severity of and Recovery from Olfactory and Gustatory Dysfunction. *Laryngoscope*. 2020;130(11):2680-2685.
19. Potter MR, Chen JH, Lobban NS, Doty RL, editors. Olfactory dysfunction from acute upper respiratory infections: relationship to season of onset. *Int Forum Allergy Rhinol*. 2020;10(6):706-712.
20. Netland J, Meyerholz DK, Moore S, Cassell M, Perlman S. Severe acute respiratory syndrome coronavirus infection causes neuronal death in the absence of encephalitis in mice transgenic for human ACE2. *J Virol*. 2008;82(15):7264-7275.
21. Yan CH, Faraji F, Prajapati DP, Ostrander BT, DeConde AS, editors. Self-reported olfactory loss associates with outpatient clinical course in COVID-19. *Int Forum Allergy Rhinol*. 2020;10(7):821-831.
22. Kermali M, Khalsa RK, Pillai K, Ismail Z, Harky A. The role of biomarkers in diagnosis of COVID-19 – a systematic review. *Life Sci*. 2020;254:117788.
23. Kishaba T, Tamaki H, Shimaoka Y, Fukuyama H, Yamashiro S. Staging of acute exacerbation in patients with idiopathic pulmonary fibrosis. *Lung*. 2014;192(1):141-149.
24. Zacharioudakis IM, Zervou FN, Prasad PJ, et al. Association of SARS-CoV-2 genomic load trends with clinical status in COVID-19: A retrospective analysis from an academic hospital center in New York City. *PLoS One*. 2020;15(11):e0242399.
25. Aquino-Jarquin G. The Raw Cycle Threshold Values From Reverse-transcription Polymerase Chain Reaction Detection Are Not Viral Load Quantitation Units. *Clin Infect Dis*. 2021;72(8):1489–1490.
26. Rao SN, Manissero D, Steele VR, Pareja J. A narrative systematic review of the clinical utility of cycle threshold values in the context of COVID-19. *Infect Dis Ther*. 2020;1-14.
27. Huang J-T, Ran R-X, Lv Z-H, et al. Chronological Changes of Viral Shedding in Adult Inpatients With COVID-19 in Wuhan, China. *Clin Infect Dis*. 2020;71(16):2158-2166.
28. Mao L, Jin H, Wang M, et al. Neurological manifestations of hospitalized patients with Coronavirus Disease 2019 in Wuhan, China. *JAMA Neurol*. 2020;1(77):683–690.
29. Trubiano JA, Vogrin S, Kwong JC, Homes N. Alterations in smell or taste—classic coronavirus disease 2019? *Clin Infect Dis*. 2020;71(16):2307-2309.
30. Landis BN, Hummel T, Hugentobler M, Giger R, Lacroix J. Ratings of overall olfactory function. *Chem Senses*. 2003;28(8):691-694.
31. Philpott CM, Wolstenholme CR, Goodenough PC, Clark A, Murty GE. Comparison of subjective perception with objective measurement of olfaction. *Otolaryngol Head Neck Surg*. 2006;134(3):488-490.
32. Flint PW, Haughey BH, Robbins KT, et al. *Cummings otolaryngology-head and neck surgery e-book*, 6th ed. Elsevier Health Sciences; 2014.
33. Haxel B, Bertz-Duffy S, Fruth K, Letzel S, Mann W, Muttray A. Comparison of subjective olfaction ratings in patients with and without olfactory disorders. *J Laryngol Otol*. 2012;126(7):692.
34. Deems DA, Doty RL, Settle RG, et al. Smell and taste disorders, A study of 750 patients from the university of Pennsylvania smell and taste center. *Arch Otolaryngol Head Neck Surg*. 1991;117(5):519-528.
35. Liu DT, Besser G, Prem B, et al. Association between orthonasal olfaction and chemosensory perception in patients with smell loss. *Laryngoscope*. 2020;130(9):2213-2219.
36. Welge-Luessen A, Hummel T, Stojan T, Wolfensberger M. What is the correlation between ratings and measures of olfactory function in patients with olfactory loss? *Am J Rhinol*. 2005;19(6):567-571.
37. Zheng S, Fan J, Yu F, et al. Viral load dynamics and disease severity in patients infected with SARS-CoV-2 in Zhejiang province, China, January-March 2020: retrospective cohort study. *BMJ*. 2020;2020:369.

How to cite this article: Taziki Balajelini MH, Rajabi A, Mohammadi M, et al. Virus load and incidence of olfactory, gustatory, respiratory, gastrointestinal disorders in COVID-19 patients: A retrospective cohort study. *Clin Otolaryngol*. 2021;46:1331–1338. https://doi.org/10.1111/coa.13844