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Original Article

Microbiology Section

Saliva as a Viable and Simple Alternative to Nasopharyngeal and Oropharyngeal Swabs for COVID-19 Real-Time Reverse Transcriptase Polymerase Chain Reaction

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ABSTRACT

Introduction: Attributable to the difficulties in specimen collection, discomfort and symptoms caused on by Nasopharyngeal Swab (NPS) and Oropharyngeal Swab (OPS) collection, and significant risk to Healthcare Workers (HCW), evaluation of an alternative specimen for the diagnosis of Coronavirus Disease-2019 (COVID-19) is required. Saliva specimen could be an alternative specimen with many advantages over NPS and OPS, however little is known about how well it performs this purpose.

Aim: To assess the efficacy of saliva as a viable and simple alternative specimen to NPS and OPS for COVID-19 Real-Time reverse transcriptase Polymerase Chain Reaction (rRT-PCR).

Materials and Methods: The present cross-sectional study was conducted in the Department of Microbiology, SGT Medical College Hospital and Research Institute, Haryana, India, from July to December 2020. A total of 60 symptomatic and 20 asymptomatic COVID-19 patients were recruited for the study and specimen viz., saliva, NPS and OPS were collected at four different sampling points i.e., on day one, five, seven and 14 after confirmation of COVID-19 rRT-PCR test positivity. Data obtained from the study was analysed and expressed as median, frequency, interquartile range and Chi-square test was done for comparison of categorical variables.

Results: Majority of the patients in symptomatic hospitalised COVID-19 patients were males 49 (81.7%) and remaining were females 11 (18.3%) and in asymptomatic group 8 (40%) were males and 12 (60%) were females. Saliva was the most sensitive specimen (74.2%), followed by NPS, Naso Oropharyngeal Swab (NOPS) with 70.8% each and OPS (65.8%) for detection of Severe Acute Respiratory Syndrome Coronavirus-2 (SARS-CoV-2) in symptomatic patients at four different sampling points. Comparable findings were also observed in specimens obtained from asymptomatic individuals as well. In addition, the viral load was also highest in saliva sample, as measured by Cycle Threshold (Ct) value. Across all specimen types, high viral load (lower Ct values) was observed during the early period of infection. Majority of the study participants reported discomfort during NPS and OPS collection (90% and 85%, respectively), lacrimation, sneezing and gag reflex being the most commonly reported induced symptoms.

Conclusion: In the present study, saliva could be a viable and alternate specimen for COVID-19 diagnosis due to its ease in sample collection, specimen stability and reduced risk of transmission of infection due to droplets.

Keywords: Coronavirus disease-2019, Cycle threshold value, Diagnostic specimen

INTRODUCTION

In December 2019, Severe Acute Respiratory Syndrome Coronavirus-2 (SARS-CoV-2) emerged in China's Hubei Province, causing an outbreak of atypical viral pneumonia, Coronavirus Disease-2019 (COVID-19) [1,2]. Detection of SARS-CoV-2 in Nasopharyngeal Swab (NPS) and/or Oropharyngeal Swab (OPS) by Real-time reverse transcriptase Polymerase Chain Reaction (rRT-PCR) is still recommended for COVID-19 diagnosis by the World Health Organisation (WHO) [1,3,4]. However, collection of NPS and/or OPS specimens a minimally invasive procedure involving close contact with patients, thus posing a risk of disease transmission to the Healthcare Workers (HCWs) because of aerosol droplets generated during the procedure. In addition, patients also experience discomfort and induce symptoms such as gag reflex, cough or sneezing, which can also result in bleeding of the NPS or oropharyngeal tissue, especially in thrombocytopenic individuals [5-7].

Salivary droplets are considered a prime source of human-to-human transmission of SARS-CoV-2 when social distancing (>2 m) is not maintained [8]. Thus, saliva could be a promising alternative which is safe, non invasive and easy to collect specimen without requiring personal protective equipment for the diagnosis of COVID-19. Furthermore, saliva study can help explain the pathogenesis since epithelial oral cavity cells revealed abundant expression of the angiotensin-converting enzyme-2 receptor, which is essential for SARS-CoV-2 entry into cells [9]. Various comparative studies have reported that in 84.6% to >90% of the patients who had positive rRT-PCR for SARS-CoV-2 in NPS, the virus was also detected in saliva [7,10-13]. Despite the fact that saliva offers a number of advantages over NPS and OPS, there is little information on conclusive evidence about its diagnostic accuracy/performances. Thus, the present study aimed to evaluate saliva as an alternative specimen to NPS and/or OPS in COVID-19 diagnosis in a tertiary care hospital in north India. Additionally, the discomfort and the symptoms that were induced while collecting NPS or OPS specimens were evaluated.

MATERIALS AND METHODS

The cross-sectional study was conducted in the Department of Microbiology, SGT Medical College Hospital and Research Institute, Haryana, India, from July to December 2020. The study protocol was reviewed and approved by the Institutional Ethics Committee of SGT University (SGTU/FMHS/EC/2020/7). A total of 60 symptomatic hospitalised COVID-19 patients (group 1) and 20 asymptomatic COVID-19 patients (group 2) were recruited using convenience sampling.

Inclusion criteria: Symptomatic hospitalised COVID-19 patients aged >18 years, positive for SARS-CoV-2 by rRT-PCR by NPS

and OPS specimens admitted to the COVID-19 care ward of SGT hospital during the study period were recruited in group 1 after obtaining written informed consent. Based on their severity of illness, they were further divided into mild, moderate, severe and critically ill patients [14]. To compare if there is any difference in SARS-CoV-2 detection rate in various specimens from asymptomatic patients compared to symptomatic patients, asymptomatic HCWs from the SGT hospital sent on quarantine after one week of duty in COVID-19 specialised wards, as per our institutional protocol being followed then, and were thereafter COVID-19 positive by rRT-PCR test were included as group 2.

Exclusion criteria: Those participants whose follow-up specimens could be obtained or those who were not willing to participate in the study and refused to provide written informed consent were excluded from the study.

Study Procedure

After recruiting the study participants, a detailed proforma was filled out, which included information on demographics, clinical data including co-morbid conditions, level of discomfort and induced symptoms experienced by the study participants during NPS and/or OPS specimen collection. The levels of discomfort experienced by study participants were rated using an arbitrary rating scale (1-4), with 1 indicating no discomfort, 2-moderate (induced symptoms such as sneezing, gag reflex, lacrimation), 3-considerable (induced symptoms such as vomit, rhinorrhoea, headache) and 4-indicating extreme discomfort (bleeding/epistaxis) [15,16].

Specimen collection and processing: Participants in the study were recruited after receiving the COVID-19 rRT-PCR test positive report released by the molecular laboratory of SGT hospital. Thus, within 12 hours following the test report, a saliva specimen was taken from each study participant, in addition to NPS and OPS (day one sample or first sampling point). After receiving the rRT-PCR COVID-19 positive report, follow-up samples were taken on days five (second sampling point), seven (third sampling point), and 14 (fourth sampling point).

Nasopharyngeal swab and OPS specimens were collected as per Indian Council of Medical Research (ICMR) guidelines by trained technicians using sterile nylon flocked swabs and placed immediately into a tube containing 3.5 mL of Viral Transport Medium (VTM) [17]. For the collection of saliva specimen, patients were instructed to pool saliva in their mouth for a few minutes before collection and gently spit saliva into a sterile universal container. Neat saliva specimens were immediately transported to the laboratory in a cool box where an approximate 1:1 ratio of VTM was immediately added [7]. The specimens were properly labelled and packed in triple layer packing and transported to the laboratory on ice in a cold box and later stored at -20°C for further processing.

The collected specimens, namely NPS, OPS, NPS with OPS and saliva specimens were vortexed properly and then 200 µL of each specimen was subjected to nucleic acid, Ribonucleic Acid (RNA) extraction using a viral nucleic acid extraction kit as per manufacturer's instructions (TRUPCR Viral RNA Extraction Kit, 3B Blackbio Biotech India Ltd.,). After RNA extraction, rRT-PCR was performed by a multiplex rRT-PCR test using primers targeting Envelope gene (E-gene) and RNA dependent RNA Polymerase/Nucleocapsid genes (RdRp/N-genes) of SARS-CoV-2 (TRUPCR SARS-CoV-2 RT qPCR kit, 3B Blackbio Biotech India Ltd.,) in a 7500 fast real-time PCR system (Applied Biosystems). The test was performed in 25 µL reactions in a 0.1 mL PCR tube containing 10 µL master mix, 0.35 µL enzyme and 4.65 µL primer probe and 10 µL extracted RNA. The thermal cycling condition was: Complementary Deoxyribonucleic Acid (cDNA) synthesis at 50°C for 15 minutes, initial denaturation at 95°C for five minutes, followed by 38 cycles of amplification with denaturation at 95°C for five seconds, annealing at 60°C for 40 seconds, and extension at 72°C for 15 seconds. Positive and negative controls were included in each PCR run.

STATISTICAL ANALYSIS

Data on age and gender of the patients, number of days of sampling from the onset of symptoms, SARS-CoV-2 detection rate, Cycle Threshold (Ct) value and the level of discomfort and induced symptoms during collection of NPS and OPS were translated to a Microsoft (MS) excel spreadsheet, and manually assessed in terms of median, frequency and Interquartile range (IQR). For comparing categorical variables, Chi-square test was used, and a p-value <0.05 was considered as statistically significant.

RESULTS

Majority of the patients in group 1 i.e., symptomatic hospitalised COVID-19 patients were males 49 (81.7%) and remaining were females 11 (18.3%), with male:female ratio as 1:0.2, with a median age of 31 years (IQR: 27-41 years). Among them, 8 (13.3%) had serious illnesses requiring Intensive Care Unit (ICU) stay during hospital stay, while the rest of the study participants had mild to moderate illnesses (categories as described earlier) and were discharged within 14 days. None of the study participants' conditions deteriorated throughout the study. Five individuals with serious illnesses requiring ICU care were over the age of sixty, with two of them suffering from diabetes and one from chronic obstructive pulmonary disease. The median number of days from symptom onset was two days at the time of the initial sampling (IQR 3-2 days) [Table/Fig-1].

Characteristics		Group 1, (n=60) n (%)	Group 2, (n=20) n (%)
Age (years)	18-29	10 (16.7)	5 (25)
	30-49	37 (61.6)	13 (65)
	50-64	9 (15)	2 (10)
	≥65	4 (6.7)	0
Gender	Male	49 (81.7)	8 (40)
	Female	11 (18.3)	12 (60)
Severity of illness	Mild	19 (31.7)	-
	Moderate	33 (55)	-
	Severe	8 (13.3)	-
	Critically ill	0	-
Co-morbidities	Hypertension	6 (10)	0
	Diabetes	4 (6.7)	0
	Chronic obstructive pulmonary disease	3 (5)	0
	Cardiovascular disease	1 (1.7)	0
Sampling after onset of symptoms (days)	1	4 (6.7)	-
	2	29 (48.3)	-
	3	22 (36.7)	-
	4	5 (8.3)	-

[Table/Fig-1]: Demographic and clinical profile of study participants in group 1 (Symptomatic hospitalised patients) and group 2 (Asymptomatic healthcare workers).

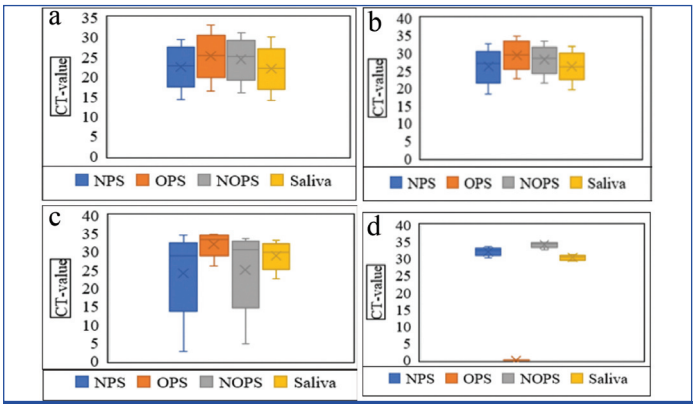
Of the 20 asymptomatic HCWs i.e., group 2 patients, the male:female ratio was 0.7:1, with a median age of 30 years (IQR 26-32 years) and none of them had co-morbid conditions. During the period of the present study, no asymptomatic participant's condition deteriorated [Table/Fig-1].

SARS-CoV-2 detection frequency and viral load: The present study evaluated matched NPS, OPS, Naso Oropharyngeal Swab (NOPS) and saliva specimens from all the study participants in both group 1 and group 2 at four distinct sampling points to compare SARS-CoV-19 detection by RT-PCR for each specimen type. The Ct values of RdRp gene for the rRT-PCR positive specimens were also analysed to determine the viral load.

In group 1 patients, SARS-CoV-2 was detected in all specimens obtained on first sampling point, with the lowest Ct value in saliva {IQR: 4.8 (24.1-19.4)}, reflecting a higher concentration of viral RNA, followed by NPS {IQR: 4.9 (25.5-20.7)}, NOPS {IQR: 5 (27.2-22.2)} and OPS {IQR: 4.6 (27.9-23.3)} [Table/Fig-2,3]. Analysis of specimens collected on second and third sampling points revealed the highest detection frequency in saliva specimen (100% and 86.7%, respectively) and lowest detection in OPS (91.7% and 71.7%, respectively). Comparable to first sampling point results, the Ct value of positive saliva specimens was lower than other specimens. The Ct value of each specimen type increased gradually on consecutive sampling points, SARS-CoV-2 rRT-PCR was positive at a low frequency even on sampling point 4 (day 14) specimens, i.e., 6.7% of NP and NOPS specimens and 10% of saliva specimens with high Ct values. Across all specimen types, lower Ct values were observed during the early period of infection. The frequency of detection of SARS-CoV-2 was significantly higher in saliva specimen with a higher viral load (low Ct value) compared to other specimens, viz., NPS, OPS and NOPS (p-value <0.05). Saliva was the most sensitive specimen (74.2%), followed by NPS, NOPS (70.8% each) and OPS (65.8%) for detection of SARS-CoV-2 in the present study. The overall percent agreement of rRT-PCR result of saliva to NPS and NOPS was 232/240 (96.7%) while to OPS was 220/240 (91.7%). Despite similar results between NPS and NOPS, the Ct value shows that NPS has a substantially higher viral load than NOPS.

Sampling point		SARS-CoV-2 rRT-PCR positivity in various specimens			
		NPS	OPS	NOPS	Saliva
1	N (%)	60 (100)	60 (100)	60 (100)	60 (100)
	Ct value (IQR)	4.9 (25.5-20.7)	4.6 (27.9-23.3)	5 (27.2-22.2)	4.8 (24.1-19.4)
2	n (%)	58 (96.7)	55 (91.7)	58 (96.7)	60 (100)
	Ct value (IQR)	3 (31-28)	3.7 (31.9-28.2)	3.2 (30.2-27)	3 (28-25.1)
3	n (%)	48 (80)	43 (71.7)	48 (80)	52 (86.7)
	Ct value (IQR)	3.8 (32.5-28.8)	2.5 (34.1-31.7)	3 (33.5-30.6)	3.4 (31.2-27.8)
4	n (%)	4 (6.7)	0	4 (6.7)	6 (10)
	Ct value (IQR)	1.2 (32.8-31.6)	-	0.5 (34.5-34)	1.1 (30.9-29.8)
Total		170	158	170	178

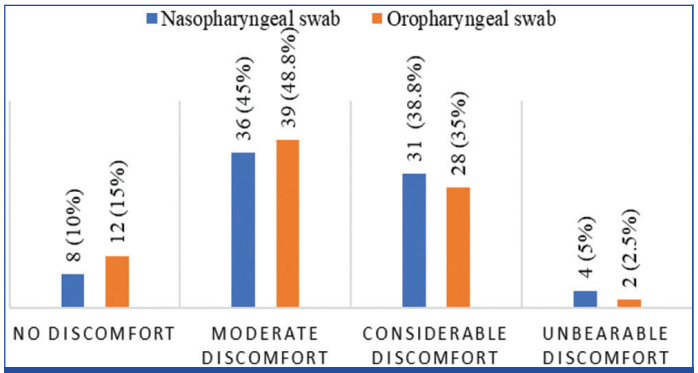
[Table/Fig-2]: Detection of SARS-CoV-2 by rRT-PCR in various clinical specimens collected from COVID-19 symptomatic admitted patients (n=60).
NPS vs Saliva, p-value=0.041; OPS vs Saliva p-value=0.046; NOPS vs Saliva; p-value=0.041



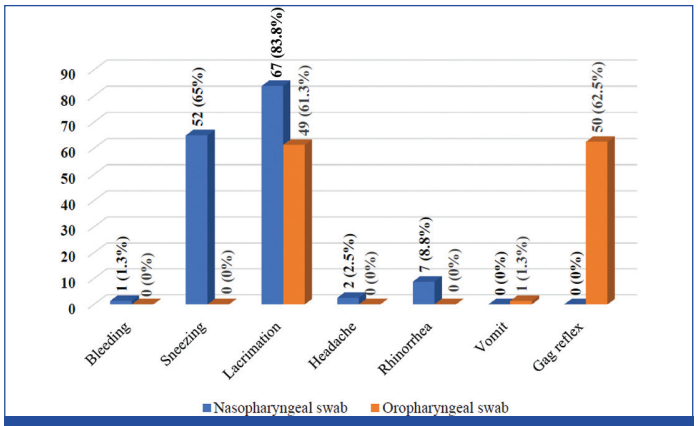
[Table/Fig-3]: Evaluation of SARS-CoV-2 viral load (as measured by Ct value) for various specimens collected on different sampling points. (a) First sampling point; (b) Second sampling point; (c) Third sampling point; (d) Fourth sampling point.

In group 2, the pattern of detection of SARS-CoV-2 in different specimens indicated comparable findings as those of group 1. Saliva, NPS and NOPS showed comparable observations on first and second sampling points, i.e., 20/20 (100%) and 11/20 (55%) SARS-CoV-2 detection rates, respectively, for the three specimen types. SARS-CoV-2 detection rates in the third sampling point were found to be 6/20 (30%) and 5/20 (25%), respectively for NPS and saliva specimens. Only two NPS specimens were found to be positive for SARS-CoV-2 at the fourth sampling point, but none of the saliva specimens did. The OPS was found to be the least sensitive specimen with SARS-CoV-2 detection rates of 17/20 (85%), 8/20 (40%), 1/20 (5%) and 0/20 (0%) in the first, second, third and fourth sampling points, respectively. Despite the similarity in SARS-CoV-2 detection rates between NPS and NOPS, NOPS had a higher Ct value than NPS, indicating lower viral loads. It is worth noting that the positive specimens from asymptomatic HCWs had Ct values that were significantly higher than the Ct values of the positive specimens obtained from symptomatic patients, indicating that the viral load in the previous group was lower (p-value <0.05).

Assessment of discomfort levels and symptoms during NPS and OPS sampling: Approximately half of participants in both the groups reported a moderate degree of discomfort during the collection of NPS and OPS, while a few reported experiencing unbearable discomforts [Table/Fig-4]. More individuals reported induced symptoms during NPS collection than during OPS collection. As revealed by the study participants, lacrimation (83.8%), sneezing (65%) and rhinorrhea (8.8%) were the most prevalently reported induced symptoms in the NPS collection whereas, gag reflex (62.5%), lacrimation (61.3%) and vomit (1.3%) were the most common ones in the OPS collection. Of the study participants, 65% of the individuals reported sneezing during NPS sampling while none reported sneezing during OPS sampling [Table/Fig-5]. Saliva collection is therefore favourable because no discomforts or induced symptoms were noted.



[Table/Fig-4]: Frequency (%) of varying degree of patient discomfort levels experienced during NPS and OPS sampling among both the groups of study participants (N=80).



[Table/Fig-5]: Frequency (%) of various symptoms experienced during NPS and OPS sampling among the study participants (N=80).

DISCUSSION

The most common validated specimen for respiratory viruses, including SARS-CoV-2, has been identified to be NPS [4]. The WHO and other regulatory authorities continue to recommend the identification of SARS-CoV-2 in respiratory specimens (i.e., NPS and/or OPS) using rRT-PCR as the gold standard method [1-4,18-20].

Studies on the new disease were not available at the outset of the pandemic because the aetiology was a novel respiratory virus, hence the previously widely accepted respiratory specimen i.e., NPS was used for diagnosis. However, collection of both NPS and OPS specimens faces several challenges as they are minimally invasive procedures causing discomfort and symptoms induced while collecting NPS or OPS specimens for the majority of individuals; for some, it is unbearably painful, which may reduce the willingness of individuals to undertake retesting when required [3]. Therefore, to avoid discomfort for the patients, many researchers have assessed saliva specimens as a non invasive specimen, in quest of additional alternative specimens for detection of SARS-CoV-2.

The present study assessed matched NPS, OPS, NOPS and saliva specimens for SARS-CoV-2 detection by rRT-PCR in 60 COVID-19 symptomatic patients and 20 COVID-19 asymptomatic HCWs. Saliva was found to be the most sensitive specimen to detect SARS-CoV-2 among the various specimens obtained from symptomatic patients processed at four distinct sampling points i.e., day one, five, seven, and 14. Saliva was then followed by NPS, NOPS, and OPS, with a statistical difference between them. Thus, saliva may be used as an equally effective alternative specimen for up to 14 days, which corresponds to the period of infectiousness for general population infection [20]. Similar to the present findings, other researchers have also reported that saliva could be an alternative specimen for COVID-19 screening and diagnosis [4,20,21]. A similar type of study conducted by Teo AKJ et al., on specimens collected from migrant workers with COVID-19 revealed that saliva was a sensitive and effective diagnostic specimen for the diagnosis of COVID-19. In addition, the study demonstrates that the probability of an rRT-PCR positive saliva test was higher than that of a NPS swab and self-administered nasal swab collected during the first and second weeks following the initial diagnosis, which is in accordance with the present study findings [21]. Another study by Beyene GT et al., also revealed that saliva was more sensitive than NPS, with 92.1% vs 52.6%, 77.4% vs 20.3% and 100% vs 50% on paired saliva NPS specimens collected at the time of admission (five to seven days), week two and week three after initial detection of SARS-CoV-2 RNA by RT-PCR using NPS specimens [20]. Bergevin MA et al., revealed that saliva and NOPS had comparable findings in a cohort of symptomatic patients presenting symptoms for <10 days, the sensitivity of NOPS was higher than that of saliva in patients with symptoms for more than 10 days (95.2% vs 71.4%) [22]. Thus, due to the numerous reports on saliva from various countries as reliable specimen, in the recent guidelines by the Centers for Disease Control and Prevention (CDC) for collection of specimens for COVID-19 diagnosis, saliva has been included as one of the upper respiratory specimens one of the specimens suitable for RT-PCR [19,23]. Furthermore, the United States (US) Food and Drug Administration (FDA) had authorised diagnostic test using self-collected saliva specimens for COVID-19 diagnosis [24].

In the present study, NPS specimens obtained from asymptomatic HCWs at four different sampling points were found to be more sensitive for detection of SARS-CoV-2 by rRT-PCR, i.e., 39/80 (48.8%) compared to matched saliva specimens i.e., 36/80 (45%). A study by Melo Costa M et al., also demonstrated less sensitivity of saliva specimens (82.9%) than NPS for detecting SARS-CoV-2 from specimens mostly taken from asymptomatic individuals [25]. Uddin MKM et al., also reported less sensitivity of saliva specimens (33%) compared with that of NPS to detect SARS-CoV-2 from asymptomatic patients [26]. OPS were the least sensitive specimen in the present analysis compared to other specimen types obtained from both symptomatic and asymptomatic study participants. Similar results were found in a study by Wang H et al., with OPS being less sensitive than NPS for the detection of SARS-CoV-2 [15]. Thus, saliva can be used as an effective alternate specimen for diagnosis of COVID-19 in early stage in both symptomatic as well as asymptomatic patients. To KK et al., also stated that the clinical management of patients with respiratory virus infections can be enhanced by additional saliva molecular testing, despite the fact that NPS continue to be the specimen of choice for the majority of patients due to their high viral contents [11]. Furthermore, saliva has the advantage in terms of sample stability; atleast 20 days at 4°C without VTM [27].

Irrespective of the specimen types, authors observed that Ct values for SARS-CoV-2 rRT-PCR positive specimens were lower in those collected at the early period of infection, i.e., first sampling point, compared to specimens collected later in the period indicating higher viral load. Teo AKJ et al., also observed comparable results in saliva, NPS, and self-administered nasal swab specimens among migrant workers infected with COVID-19, whose onset of illness could be estimated [21]. Although, there was no discernible difference between the Ct values of saliva and NPS in the present study, it was observed that the viral load, as determined by Ct value, was highest in saliva, followed by NPS, NOPS and OPS. In a study from Singapore, viral load in paired saliva NPS was compared on specimens obtained on the day of hospital admission and it was found that saliva had a significantly higher viral load than NPS (p-value <0.001) [21]. Similar findings have been shown in a study by Wyllie AL et al., in which viral load in saliva was higher than NPS specimens [28]. Rao M et al., also showed that the Ct values for E and RdRp genes were significantly lower in saliva (median IQR 30.6 (27.5-32.8) and 31.2 (27.3-33.6) than those for NPS (median IQR 33.2 (30.0-35.1) and 33.7 (30.0-36.0), indicating high viral loads in saliva specimens [29]. Wang H et al., also reported a higher viral load in NPS than in OPS with significantly lower mean Ct values of NPS (37.8, 95% CI: 37.0-38.6) than those of OPS (39.4, 95% CI: 38.9-39.8) by 1.6 (95% CI 1.0-2.2, p-value <0.001), indicating that the SARS-CoV-2 load was significantly higher in NPS specimens [15]. In contrast to the present study, Justo AFO et al., demonstrated higher Ct values in saliva compared to NPS, though their sensitivity for detection of SARS-CoV-2 was same i.e., 83.3% [30]. These divergent outcomes among studies may be the result of various types of sampling/processing methods detection reagents used and the difference in study populations.

In the present study, authors assessed the patients' discomfort levels during NPS and OPS sampling and found that approximately half of the study participants experienced a moderate degree of discomfort during NPS and OPS sampling and six out of 80 individuals reported unbearable discomfort. As reported in this study, other studies have also reported discomfort and induced symptoms during NPS collection [15,20]. A study from China by Wang H et al., reported 37.9% and 41.7% of individuals reporting moderate degrees of discomfort during NPS and OPS sampling [15]. In contrast to the current study findings, Marra P et al., stated that the collection of NPS is largely a none to minimum discomfort inducing method and the variations in discomfort could be attributed to anatomical characteristics, highlighting the necessity for a patient specific and anatomy focused strategy [31]. With further assessment of the symptoms, authors found that lacrimation and sneezing were the most common symptoms encountered by study participants during NPS collection, whereas gag reflex and lacrimation were the most common symptoms during OPS collection. The findings of a study by Wang H et al., were in accordance with the present study findings, though the individuals who experienced vomiting during OPS collection were fewer in this study [15]. Although, NPS and OPS are recommended specimens for diagnosis of COVID-19, saliva may be used as an alternative specimen to avoid discomforts during specimen collection in certain scenarios, particularly in young children, individuals with nasal polyps or other anomalies, prior history of epistaxis or severe

discomfort during specimen collection and in instances when self-collection is requisite.

Limitation(s)

The primary limitations were the small sample size, from a single institute and disproportion between the two groups, which made it challenging for us to make appropriate inferences. Due to the small sample size, inferences about the effectiveness of saliva as a diagnostic specimen for COVID-19 in asymptomatic individuals may not be generalisable. In addition, follow-up sampling beyond 14 days after diagnosis of COVID-19 was not feasible as majority of the cases recovered and got discharged from the hospital.

CONCLUSION(S)

The present study findings suggest that saliva may be used accurately for the diagnosis of SARS-CoV-2 early after the beginning of symptoms, since it has the advantages of ease of sample collection, reduced transmission of infection to HCWs owing to droplet, and sample stability. Being non intrusive and causing no discomfort, it would be preferable specimen for paediatric age group as well. Thus, saliva could be recommended for diagnosing COVID-19 and monitoring viral load among individuals and also for mass surveillance. Saliva collection and processing methods need to be standardised and further studies need to be done to assess its effectiveness as a validated diagnostic specimen.

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