SALIVA IN DIAGNOSIS OF HUMAN CORONA VIRUS- A REVIEW

Abstract

Saliva is a reservoir of various biological marker and provides superiority over other biological fluids. Rapid and accurate diagnosis of SARS CoV-2 is essential to control the ongoing covid-19 pandemic. Nasopharyngeal, oropharyngeal swab, bronchoalveolar lavage, sputum, urine, and blood are frequently tested sample but all these techniques are invasive and uncomfortable to the infected person. Therefore, the potential use of salivary sample can be taken into consideration as an alternative tool as it has various advantages over the current traditional methods. Salivary diagnostics also can be utilized as chair side tests for various diseases in the near future through conscientious testing.

Keywords- SARS CoV-2, covid-19,salivary biomarkers,diagnosis of viral infection

INTRODUCTION

The emergence of coronavirus in December 2019, has caused a large global outbreak and is a major public health issue. [1] Coronaviruses are enveloped, single stranded RNA viruses with high rates of mutation and recombination having the spike surface glycoprotein (S), small envelope protein (E), matrix protein (M), and nucleocapsid protein (N). [2] SARS-CoV-2, as it is commonly known, has been found to spread more rapidly than MERS-CoV with most common symptom being fever and cough.[3] Bilateral lung involvement with ground glass opacity is the most common finding of coronavirus disease (covid-19) from computed tomography images of the chest. [4] Spread by human to human transmission or direct contact and infection has estimated to have mean incubation period of 6.4 days and basic reproduction number of 2.24-3.58. [4] Efforts to control SARS-CoV-2 depend on the accurate and rapid diagnostic testing. These tests must be sensitive to mild and asymptomatic infections to promote effective self isolation and reduce transmission within high risk groups, consistent to reliably monitor disease and aid clinical decisions.[5] Nasopharyngeal or throat swabs are commonly used sampling method for viral load monitoring. Direct inspection of the patient's posterior pharynx and tonsils is recommended during throat swab collection. It can induce coughing and sneezing which generates aerosol and is a potential health hazard for health-care workers. Moreover, collection of nasopharyngeal specimens is a relatively invasive and uncomfortable procedure even can induce bleeding. A patient's reluctance to provide a sample can account for the scarcity of timepoints in viral load studies of respiratory virus infections. Previous studies have shown high concordance between nasopharyngeal aspirate and saliva as specimens for laboratory diagnosis of respiratory viruses. [3] The healthcare providers, doctors, nurses, and paramedic staff will be safe from the transmission of disease while using saliva as specimen. Hence, this method of sampling is advantageous compared with the use of nasopharyngeal aspirates.[6]

ROLE OF SALIVA

Saliva is an exocrine secretion of major and minor salivary gland comprising of approximately 99% water, variety of electrolytes, proteins represented by enzymes, immunoglobulins and other antimicrobial factor. [7] With the emerging latest technologies saliva has been studied thoroughly as a potential diagnostic apparatus to become a alteration for other biological fluids such as serum or urine in disease diagnosis. They have disclosed large numbers of medically important salivary biomarkers for various disease conditions including cancer, autoimmune, viral, bacterial, cardiovascular, and metabolic diseases.[8] Any alteration in the composition and quantity of saliva can help in the detection of various diseases.[7]

POSSIBLE DIRECT INVASIONS INTO ORAL TISSUES

SARSCoV-2 can be presented in saliva through three different routes.[7,9,10]

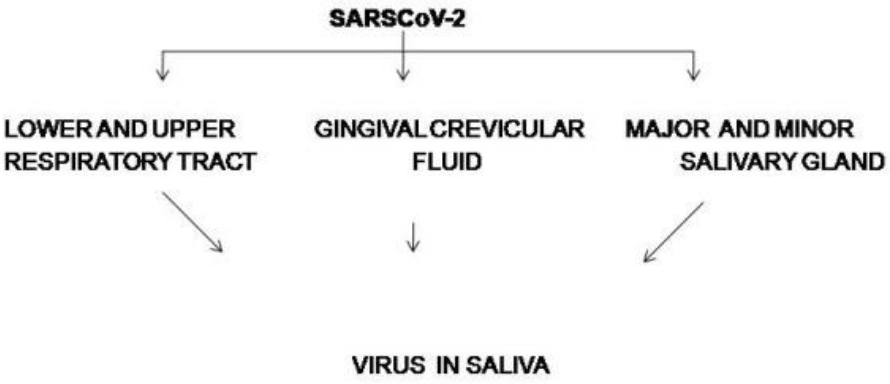

Figure 1: Routes of invasion of SARSCoV-2 in saliva

INTERACTION BETWEEN HOST CELL IN ORAL CAVITY AND VIRUS

The entry of coronavirus into the host cell is a multi-step process using multiple distinct domains in the spike protein that facilitates attachment of the virus to the surface of the cell, engagement of the receptor, processing of proteases and membrane fusion. The SARS-CoV-2 uses ACE2 as the receptor for viral entry and transmembrane protease serine 2 (TMPRSS2) for priming of the spike protein [2,11]

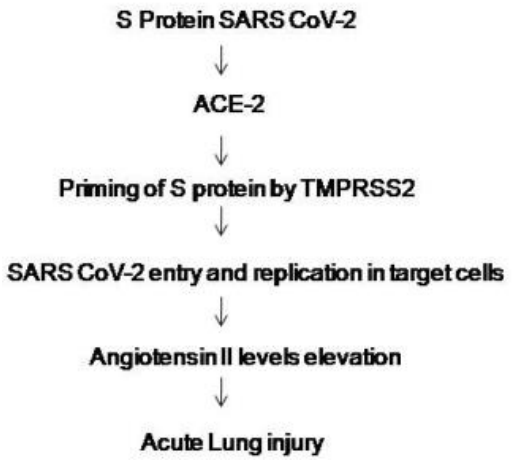

Figure 2: Relation of SARSCoV-2 with ACE receptor [12]

Expression of the enzyme furin on tongue has been implicated in virus infection by cleaving viral envelope glycoproteins and enhancing infection with host cells. [1] Hyposalivation can disrupt the physical barrier of the oral and airway mucosal surfaces. This enhances the viral colonization and adhesion. The decrease may also produce disturbances in the secretion of antimicrobial peptides and proteins.Considering the existence of various proteins with established antiviral characteristics in saliva may potentially obstruct virus replication especially SARS-CoV-2.[9]

SALIVA FOR DETECTIONS OF SARS CoV-2

RUCDR Infinite Biologic researchers at Rutgers University have successfully demonstrated saliva as a sample source for COVID-19 detection in compared to nasopharyngeal or oropharyngeal swabs.SARS-CoV-2 invades the epithelial cell of rhesus macaques in salivary gland ducts. However, it should be noted that saliva specimen also contains secretion from the nasopharynx and lungs via the action of cilia. [7] Viral load peaks can be detected early in salivary specimens at the onset of infection.[1] Studies have shown that some viral strain can be detected in saliva as long as 29 days after infection enhancing the disease detection.[10,13] Dental/oral and other health professionals must always be diligent in protecting against the spread of infectious disease. Small droplets with a diameter of less than or equal to 60 μm can cause short range transmission for individuals with distance less than one meter. In a desirable environment, small droplets are likely to fade away into droplet nuclei with a diameter of less than 10 μm. It then become capable of long-range aerosol transmission. For a susceptible host can enter the mouth, eyes, or be inhaled directly into the lungs thereby causing infection.[7,9]

Antibodies against Cov in saliva and its potential in diagnosis

Previous studies have shown the production of SARS-CoV-specific secretory immunoglobulin A (sIgA) in the saliva of animal models.[14] Immune responses, including the production of SARS-CoV-specific serum immunoglobulin G (IgG) and secretory immunoglobulin A (sIgA), were determined in animal mucosal secretions and tissues.[15] Saliva-based antibody tests are there to detect several viruses and the presence of immunoglobulin can help by rapid diagnosis of covid-19.

Methods of collection of saliva

There are many saliva collection devices available in the market for safe and sterile collection without compromising the quality and quantity. Self collection of saliva can also be done and usually early morning saliva sample is preferred before tooth brushing. Saliva specimen can be added to the viral transport medium.[16] Commercially available saliva sampling devices and their company names can be accessed by all researchers, healthcare providers, doctors, microbiologists, and virologists for the handling of samples. Different saliva collection devices used in the sampling of contagious infectious diseases are Salivette® (Sarstedt); Quantisal® (Immunalysis); SCS® (Greiner-BioOne), VersiSAL®, and SuperSAL by Oasis Diagnostics® Corporation.[6]
The potential benefits of salivary diagnostic tests are economical, easier to apply than serum sampling with no requirement for specialized healthcare workers. Numerous samples are simple to obtain, collection and monitoring can be done at home with good storage than serum sampling. Saliva does not clot and can be handled more efficiently than blood with lesser agitation during the diagnostic process . Thereby, salivary diagnostic testing can offer a cost-effective and convenient mechanism for early-diagnosis of Covid-19.[7,9]

Diagnostic Kits and efficiency

Several viral infections can be diagnosed depending on the type of salivary biomarkers, such as viral DNA and RNA, antigens and antibodies. A quantitative real-time reverse transcription–polymerase chain reaction (RT-PCR) assay and fractionation experiment can detect the load of SARS-CoV.[17] Nucliec acid extraction method, ELISPOT,POC technology can also be used to detect salivary biomarkers.[7,14,18]

Study done by **E. Pasomsub et al.(2020)**investigated the diagnosis of COVID-19 and found the sensitivity and specificity of the saliva sample RT-PCR were 84.2%, and 98.9%, respectively. [19] **Hiba Hamid et al.(2017)** concluded that POC technology using saliva can rapidly detect and effective in identifying and isolating potential carriers and contacts.[18] **Kelvin Kai-Wang** evaluated that posterior oropharyngeal saliva sample contains the highest saliva load which can account for fast spread of the infection.[3] **Anne L. Wyllie et al.(2020)** described that saliva has greater detection sensitivity and consistency throughout the course of infection of covid-19.[5]

Conclusion

Saliva acts as the promising diagnostic tool in a large setting of individuals requiring screening. This minimizes the load of collection of sample and nosocomial transmission of covid-19 to the health workers. The possibility of the salivary glands as a reservoir, harboring latent infection, which may reactivate later, should also be considered and this warrants further research.

REFERENCES

1. Xu R, Cui B, Duan X, Zhang P, Zhou X, Yuan Q. Saliva: potential diagnostic value and transmission of 2019-nCoV. Int J Oral Sci. 2020 Apr 17;12(1):11.

2. Vinayachandran D, Saravanakarthikeyan B. Salivary diagnostics in COVID-19: Future research implications. J Dent Sci [Internet]. 2020 Apr 23; Available from: http://dx.doi.org/10.1016/j.jds.2020.04.006

3. To KK-W, Tsang OT-Y, Leung W-S, Tam AR, Wu T-C, Lung DC, et al. Temporal profiles of viral load in posterior oropharyngeal saliva samples and serum antibody responses during infection by SARS-CoV-2: an observational cohort study. Lancet Infect Dis [Internet]. 2020; Available from: https://www.sciencedirect.com/science/article/pii/S1473309920301961

4. Lai C-C, Shih T-P, Ko W-C, Tang H-J, Hsueh P-R. Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) and coronavirus disease-2019 (COVID-19): The epidemic and the challenges. Int J Antimicrob Agents. 2020 Mar;55(3):105924.

5. Wyllie AL, Fournier J, Casanovas-Massana A, Campbell M, Tokuyama M, Vijayakumar P, et al. Saliva is more sensitive for SARS-CoV-2 detection in COVID-19 patients than nasopharyngeal swabs. Medrxiv [Internet]. 2020; Available from: https://www.medrxiv.org/content/10.1101/2020.04.16.20067835v1?fbclid=IwAR1sexAHAFxOuDiH7_7QKParCeIcotoL2DP8oMM27uKeV0bxE8d5IZYQXvM

6. Khurshid Z, Asiri FYI, Al Wadaani H. Human Saliva: Non-Invasive Fluid for Detecting Novel Coronavirus (2019-nCoV). Int J Environ Res Public Health [Internet]. 2020 Mar 26;17(7). Available from: http://dx.doi.org/10.3390/ijerph17072225

7. Santosh TS, Parmar R, Anand H, Srikanth K, Saritha M. A Review of Salivary Diagnostics and Its Potential Implication in Detection of Covid-19 [Internet]. Cureus. 2020. Available from: http://dx.doi.org/10.7759/cureus.7708

8. Pfaffe T, Cooper-White J, Beyerlein P, Kostner K, Punyadeera C. Diagnostic potential of saliva: current state and future applications. Clin Chem. 2011 May;57(5):675–87.

9. Baghizadeh Fini M. Oral saliva and COVID-19. Oral Oncol. 2020 May 27;108:104821.

10. Sabino-Silva R, Jardim ACG, Siqueira WL. Coronavirus COVID-19 impacts to dentistry and potential salivary diagnosis. Clin Oral Investig. 2020 Apr;24(4):1619–21.

11. Hoffmann M, Kleine-Weber H, Schroeder S, Krüger N, Herrler T, Erichsen S, et al. SARS-CoV-2 Cell Entry Depends on ACE2 and TMPRSS2 and Is Blocked by a Clinically Proven Protease Inhibitor. Cell. 2020 Apr 16;181(2):271–80.e8.

12. Kai H, Kai M. Interactions of coronaviruses with ACE2, angiotensin II, and RAS inhibitors—lessons from available evidence and insights into COVID-19. Hypertens Res [Internet]. 2020 Apr 27; Available from: https://doi.org/10.1038/s41440-020-0455-8

13. Díaz-Menéndez M, Crespillo-Andújar C. Zika Virus Infection: Risk of Spreading in Europe. Springer; 2017. 93 p.

14. Lu B, Huang Y, Huang L, Li B, Zheng Z, Chen Z, et al. Effect of mucosal and systemic immunization with virus-like particles of severe acute respiratory syndrome coronavirus in mice. Immunology. 2010

Jun;130(2):254–61.

15. Lu B, Huang Y, Huang L, Li B, Zheng Z, Chen Z, et al. Effect of mucosal and systemic immunization with virus-like particles of severe acute respiratory syndrome coronavirus in mice. Immunology. 2010 Jun 1;130(2):254–61.

16. To KKW, Tsang OTY, Leung WS, Tam AR. Temporal profiles of viral load in posterior oropharyngeal saliva samples and serum antibody responses during infection by SARS-CoV-2: an observational cohort …. Lancet Infect Dis [Internet]. 2020; Available from: https://www.sciencedirect.com/science/article/pii/S1473309920301961

17. Wang W-K, Chen S-Y, Liu I-J, Chen Y-C, Chen H-L, Yang C-F, et al. Detection of SARS-associated coronavirus in throat wash and saliva in early diagnosis. Emerg Infect Dis. 2004 Jul;10(7):1213–9.

18. Hamid H, Khurshid Z, Adanir N, Zafar MS, Zohaib S. COVID-19 Pandemic and Role of Human Saliva as a Testing Biofluid in Point-of-Care Technology. Eur J Dent [Internet]. 2020 Jun 3; Available from: http://dx.doi.org/10.1055/s-0040-1713020

19. Pasomsub E, Watcharananan SP, Boonyawat K, Janchompoo P, Wongtabtim G, Suksuwan W, et al. Saliva sample as a non-invasive specimen for the diagnosis of coronavirus disease-2019 (COVID-19): a cross-sectional study. Clin Microbiol Infect [Internet]. 2020; Available from: https://www.sciencedirect.com/science/article/pii/S1198743X20302780