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Study presence of COVID-19 (SARS-CoV-2) in the sweat of patients infected with Covid-19

Hadis Fathizadeh, Sepehr Taghizadeh, Rohollah Safari, Saeid Shabestari Khiabani, Bayaz Babak, Fatemeh Hamzavi, Khudaverdi Ganbarov, Silvano Esposito, Elham Zeinalzadeh, Sounkalo Dao, Sükran Köse, Hossein Samadi Kafil

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ABSTRACT

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) disease, which started in Wuhan, Chin, has now become a public health challenge in most countries around the world. Proper preventive measures are necessary to prevent the spread of the virus to help control the pandemic. Because, SARS-CoV-2 is new, its transmission route has not been fully understood. In this study, we aimed to investigate the presence of SARS-CoV-2 in the sweat secretion of COVID-19 patients. Sweat specimens of 25 COVID-19 patients were collected and tested for SARS-CoV-2 RNA by Real-time Polymerase Chain Reaction (RT-PCR) method. After RNA extraction and cDNA amplification, all samples were examined for the presence of ORF-1ab and N genes related to COVID-19. Results annotated by Realtime PCR machines software based on Dynamic algorithm. The results of this study showed the absence of SARS-CoV-2 in the sweat samples taken from the foreheads of infected people. Therefore, it can be concluded that the sweat of patients with COVID-19 cannot transmit SARS-CoV-2. However they can be easily contaminated with other body liquids.

1. Introduction

On December 31, 2019, a cluster of atypical pneumonia cases appeared in China [1]. Rapidly, investigations identified a novel family B betacoronavirus, now officially named as SARS-CoV-2 by the International Committee on Taxonomy of Viruses (ICTV), and introduced as the etiological factor responsible for this infection [2,3]. Acute fever, cough, myalgia, dyspnea, and radiological evidence of bilateral pneumonia with ground-glass opacity of lung have been observed in most patients with coronavirus disease 2019 (COVID-19), or SARS-CoV-2 infection [4,5]. Although, mildly symptomatic or asymptomatic cases have also been reported [6–8]. According to the findings of epidemiologic and genetic research, COVID-19 disease is transmitted by sustained human-to-human spread. It is now believed that transmission between individuals occurs mainly through respiratory droplets and contact [9]. On the other hand, with the discovery of SARS-CoV-2 in the feces of patients in the United States and China, it has been announced that there may be a risk of transmission of the virus through the feces of patients [10,11]. However, our knowledge of other possible ways of transmitting the virus, including vertical transmission (from infected mother to fetus), transmission through sperm and sweat, is not yet complete. In a study of 212 people infected with SARS-CoV-2, 114 reported “profuse sweating” and 102 of them reported “night sweats”. This indicates a high volume of substance for infection, if the sweat of these people has infectious SARS-CoV-2 [12]. On other hand, SARS-CoV-2 has similarities with acute respiratory coronavirus syndrome (SARS-CoV) and Middle
First, the patients were diagnosed based on clinical symptoms and contact history with further confirmation by positive results of qualitative real-time PCR. Taqman RT-PCR was performed using the MIC PCR thermocycler (Applied Biomolecular system, Australia) at the following cycling conditions: 1 cycle at 50 °C for 20 min, 1 cycle at 95 °C for 1 min and 45 cycles at 95 °C for 15 s, 60 °C for 35 s then 72.0 °C for 15 s. For analysis of the final results dynamic algorithm was used by cycling analysis in green and yellow and red fluorescence and cut off for fluorescence was 5%. Cut off for detection of virus according to the company report was above 200 copy number per ml.

3. Results

The 25 patients consisted of 15 males and 10 females. Their median age was 35 years (range 24–48 years). All of them had clinical features compatible with high viral load such as fatigue, fever, and low blood oxygenation levels and also positive result of real-time PCR of nasopharyngeal sample (19.2 < Ct ≤ 32) (Fig. 1).

Out of twenty-five patients, twenty of them had CRP positive and blood changes such as lymphocyte depletion were observed in 18 of them.

The result of Real-time PCR test for sweat samples taken from the forehead of these patients was that all samples were negative for the presence of SARS-CoV-2 that has been shown in Fig. 2 and only two positive cases were observed (Ct: 32.4 and Ct:28.17).

4. Discussion

Findings of the present study indicate sweat of the patients are not infective and is safe. SARS-CoV-2 is known as the seventh and newest coronavirus, which started spreading in December 2019 from Wuhan, China, and quickly spread around the world, becoming the biggest global health concern [20]. Clinical signs of COVID-19 disease range from mild upper respiratory tract infection to a multisystemic disease with the inflammatory and thrombotic immunological response [21]. The main reason for the wide spread of this virus is its transmission power and contagion, and evidence shows that asymptomatic people can transmit the disease [22,23]. Development of practical and effective

Fig. 1. Average Cycle threshold (CT) of patients in nasopharyngeal samples in green fluorescence channels.
Fig. 2. Amplification curve of the Sar-Cov-2 identification by A) ORF1-ab (in Green channel), B) N gene (in Yellow channel) and C) Internal control (In Red Channel). These curves show accuracy and sensitivity of the identification of target genes. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)
strategies to protect high-risk individuals and prevent the spread of the virus is urgently needed. However, the pathogenicity of SARS-CoV2 infection and the pathways of virus transmission have not yet been clearly identified. In addition to proven routes of transmission of SARS-CoV-2, such as respiratory secretions and feces, the possibility of transmission through sweat is also considered. In this study, twenty-five infected patients, all of whom had clinical symptoms and high viral load in respiratory samples, were examined for the presence of SARS-CoV-2 in sweat samples. All of them were negative and only two positive cases were found. By asking patients who tested positive, we found that they had touched their foreheads during the test and that their hands may have been contaminated with oral secretions or mucusa, so the positive result was actually due to contamination of the sampling site with the secretions of other parts. Unfortunately, very little study has been done on the possible role of sweat in the transmission of any of the betacoronaviruses, especially with regard to other deadly viruses that can be transmitted through the sweat of an infected person [24]. In 2015, a report was presented that a Korean healthcare worker infected with the MERS-coronavirus during cardiopulmonary resuscitation (CPR) of a MERS patient. According to the report, during CPR, a large amount of fluid was splashed and the nurse becomes infected by touching the mask and wiping the sweat from her face. Although this is not certain, one of the possible ways that this person became infected was through mucosal exposure to sweat contaminated with MERS coronavirus [17]. The presence of the SARS-CoV-2 in eccrine glands indicates that sweat can be a source of transmission [25]. In a 2004 study of four SARS-CoV patients, Ding and his colleagues found SARS-CoV RNA and nucleo-protein in the sweat glands, kidney, and intestine by in situ hybridization and immunohistochemistry. Accordingly, there were speculations that SARS-CoV could be transmitted through sweat, feces and urine [16]. One of the ways of transmission by sweat is through skin contact with mucus, similar to what has been said about the Korean healthcare worker [17]. From a theoretical point of view, one way of transmission can be path skin-object-mucosa so that contaminated sweat may remain on objects, which in turn may cause infection if touched by non-infected people [26]. The receptor for SARS-CoV-2 to enter human cells is human angiotensin converting enzyme 2 (ACE2), and it is thought that organs with high levels of this receptor will be able to accept more viruses and therefore be more likely to become infected [27]. ACE2 is itself exist in the skin, eccrine glands and smooth muscles around the sebaceous glands as well as within these glands [28]. The fact that the number of ACE2 receptors in the skin can absorb the SARS-CoV-2 raises concerns about how the SARS-CoV-2 is transmitted. Therefore, it is possible that not only inhaling or touching the oral and respiratory secretions and mucosa alone does not cause infection, but also simpler routes such as touching the sweat-soaked objects of the infected person can cause the infection. However, the results of this study refute the hypotheses about the possibility of transmitting SARS-CoV-2 through sweat, but still more research in this case with more samples and sweat sampling of other parts of the body are recommended to ensure this result.

CRediT authorship contribution statement

**Hadis Fathizadeh**: Writing - original draft, study design, Data collection, Data interpretation, manuscript preparation. **Sepehr Taghizadeh**: data interpretation, manuscript preparation. **Rohollah Safari**: Writing - original draft, Data collection, manuscript preparation. **Saeid Shabestari Khiani**: Writing - original draft, Data interpretation, manuscript preparation. **Bayaz Babak**: Writing - original draft, Data collection, manuscript preparation. **Fatemeh Hamzavi**: Writing - original draft, Data interpretation, manuscript preparation. **Silvano Esposito**: Writing - original draft, manuscript preparation. **Elham Zeinalzadeh**: Writing - original draft, Literature search, manuscript preparation. **Sounkalo Dao**: Writing - original draft, Formal analysis, Statistical analysis, manuscript preparation. **Sükrân Köse**: Writing - original draft, Formal analysis, Statistical analysis, manuscript preparation. Hossein Samadi Kaflí: Writing - original draft, Study design, Supervision, Funding acquisition, manuscript preparation.

Declaration of competing interest

None to declare.

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References

[1] M.A. Ozma, P. Maroufi, E. Khodadadi, S. Kose, I. Esposito, K. Ganbarov, et al., Clinical manifestation, diagnosis, prevention and control of SARS-CoV-2 (Covid-19) during the outbreak period, Infect Med 28 (2020) 153–165.
[2] N. Zhu, D. Zhang, W. Wang, X. Li, B. Yang, J. Song, et al., A novel coronavirus from patients with pneumonia in China, 2019. N. Engl. J. Med. 382 (2020) 727–733.
[3] H. Fathizadeh, P. Maroufi, M. Momen-Hezavei, S. Dao, S. Kose, K. Ganbarov, et al., Protection and disinfection policies against SARS-CoV-2 (COVID-19), Infect Med 28 (2020) 185–191.
[4] C. Huang, Y. Wang, X. Li, L. Ren, J. Zhao, Y. Hu, et al., Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China, Lancet 395 (2020) 497–506.
[5] E. Khodadadi, P. Maroufi, E. Khodadadi, I. Esposito, K. Ganbarov, S. Esposito, et al., Study of combining virtual screening and antiviral treatments of the SARS-CoV-2 (Covid-19), Microb. Pathog. (2020) 145.
[6] N. Chen, M. Zhou, X. Dong, J. Qu, F. Gong, Y. Han, et al., Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study, Lancet 395 (2020) 507–513.
[7] M. Wei, J. Yuan, Y. Liu, T. Fu, X. Yu, Z.J. Zhang, Novel coronavirus infection in hospitalized infants under 1 Year of age in China, J. Am. Med. Assoc. 323 (2020) 1313–1314.
[8] W. Arummanee, G.A.I. Ecoy, H.E.E. Khine, M. Duangkaew, E. Prompetchara, P. Chonvarachote, et al., Colicin N mediates apoptosis and suppresses integrin-modulated survival in human lung cancer cells, Molecules 25 (2020) 816.
[9] C. Del Rio, P.N. Malani, 2019 novel coronavirus important information for clinicians, J. Am. Med. Assoc. 323 (22) (2020) 1039–1040.
[10] M.L. Holshue, C. DeBolt, S. Lindquist, K.H. Lofy, J. Wiesman, H. Bruce, et al., First case of 2019 novel coronavirus in the United States, N. Engl. J. Med. 382 (2020) 929–936.
[11] P. Gholizadeh, R. Safari, P. Maroufi, E. Zeinalzadeh, P. Pagliano, K. Ganbarov, et al., Alteration of liver biomarkers in patients with SARS-CoV-2 (COVID-19), J. Inflammam. Res. (2020) 13.
[12] Y.L.Z. Zhang, B. Wang, J. Cang, Y. Ma, Gastrointestinal tract symptoms in coronavirus disease 2019: analysis of clinical symptoms in adult patients, medRxiv Preprint (2020).
[13] V.G. da Costa, M.L. Moreli, M.V. Saivish, The emergence of SARS, MERS and novel SARS-2 coronaviruses in the 21st century, Arch. Virol. 165 (2020) 1517–1526.
[14] K. Najafi, P. Maroufi, E. Khodadadi, E. Zeinalzadeh, K. Ganbarov, M. Arghazadeh, et al., SARS-CoV-2 receptor ACE2 and molecular pathway to enter target cells during infection, Rev. Med. Microbiol. 4 (2020) 1–12.
[15] R.E. Proper, Is sweat a possible route of transmission of SARS-CoV-2? Exp. Biol. Med. 245 (2020) 997–998.
[16] Y. Ding, L. He, Q. Zhang, Z. Huang, X. Che, J. Hou, et al., Organ distribution of severe acute respiratory syndrome (SARS) associated coronavirus (SARS-CoV) in SARS patients: implications for pathogens and virus transmission pathways, J. Pathol. 203 (2004) 622–630.
[17] H.S. Nam, M.Y. Yeon, J.W. Park, J.Y. Hong, J.W. Son, Healthcare worker infected with Middle East Respiratory Syndrome during cardiopulmonary resuscitation in Korea, Epidemiol. health 39 (2017), e2017052, 2015.
[18] S. Sanche, Y.T. Lin, C. Xu, E. Romero-Severson, N. Hengartner, R. Ke, High contagiousness and rapid spread of severe acute respiratory syndrome coronavirus 2, Emerg. Infect. Dis. 26 (2020) 1470–1477.
[19] P. Gholizadeh, M. Aghazadeh, M. Arghazadeh, H.S. Kafli, Suppressing the CRISPR/Cas adaptive immune system in bacterial infections, Eur. J. Clin. Microbiol. Infect. Dis. 11 (2017), 017-3036.
[20] Z. Deng, Y. Hu, P. Yang, P. Zheng, W. Peng, B. Ren, et al., Diagnosis and treatment of an acute severe pneumonia patient with COVID-19: case report, J. Med. Virol. (2020).
[21] S. Hoehl, H. Rabenaü, A. Berger, M. Kortenbusch, J. Ciniati, D. Bokjova, et al., Evidence of SARS-CoV-2 infection in returning travelers from Wuhan, China, N. Engl. J. Med. 382 (2020) 1278–1280.
[22] Q. Li, X. Guan, P. Wu, X. Wang, L. Zhou, Y. Tong, et al., Early transmission dynamics in Wuhan, China, of novel coronavirus-infected pneumonia, N. Engl. J. Med. 382 (2020) 1199–1207.

[23] Y. Bai, L. Yao, T. Wei, F. Tian, D.Y. Jin, L. Chen, et al., Presumed asymptomatic carrier transmission of COVID-19, J. Am. Med. Assoc. 323 (2020) 1406–1407.

[24] P. Vetter, W.A. Fischer 2nd, M. Schiβer, M. Jacobs, D.G. Bausch, L. Kaiser, Ebola virus shedding and transmission: review of current evidence, J. Infect. Dis. 214 (2016). S177–S84.

[25] C. Santonja, F. Heras, L. Núñez, L. Requena, COVID-19 chilblain-like lesion: immunohistochemical demonstration of SARS-CoV-2 spike protein in blood vessel endothelium and sweat gland epithelium in a polymerase chain reaction-negative patient, Br. J. Dermatol. (2020).

[26] N. Van Doremalen, T. Bushmaker, D.H. Morris, M.G. Holbrook, A. Gamble, B. N. Williamson, et al., Aerosol and surface stability of SARS-CoV-2 as compared with SARS-CoV-1, N. Engl. J. Med. 382 (2020) 1564–1567.

[27] M. Hoffmann, H. Kleine-Weber, S. Schroder, N. Krüger, T. Herrler, S. Erichsen, et al., SARS-CoV-2 cell entry depends on ACE2 and TMPRSS2 and is blocked by a clinically proven protease inhibitor, Cell (2020).

[28] I. Hamming, W. Timens, M. Bulthuis, A. Lely, Navis Gv, H. van Goor, Tissue distribution of ACE2 protein, the functional receptor for SARS coronavirus. A first step in understanding SARS pathogenesis, J. Pathol. 203 (2004) 631–637.