

Investigating the Presence of SARS-CoV-2 on the Surfaces, Fomites, and in Indoor Air of a Referral COVID-19 Hospital, Shiraz, Iran

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Abstract

Background: Coronavirus disease (COVID-19) is an immensely transmissible viral infectious disease caused by Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2). This study aimed to assess the presence of SARS-CoV-2 in the indoor air, on the surfaces, and on the fomites of a COVID-19 referral hospital in Shiraz, Iran.

Methods: In this cross-sectional study, indoor air sampling was conducted utilizing a standard midge impinger containing 15 ml of viral transfer medium (VTM) equipped with a sampling pump with a flow rate of 10 L min⁻¹ for 60 minutes. Surfaces and fomites were sampled using sterile polyester swabs. The real-time reverse transcription-polymerase chain reaction (rRT-PCR) was utilized to detect SARS-CoV-2.

Results: The RNA of SARS-CoV-2 was detected in about 41.2% indoor air and 32% swab samples. Four out of the six (66.7%) indoor air samples up to a distance of 2 meters from the patient's bed in intensive care units (ICU-1, ICU-3), accident and emergency (A&E-2), and negative pressure rooms were positive for SARS-CoV-2 RNA. All air samples within 2 to 5 meters of the patient's bed were negative.

Conclusion: This study's results did not support the airborne SARS-CoV-2 transmission; However, it showed contamination of surfaces and fomites in the studied hospital's wards.

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Introduction

COVID-19 is an immensely contagious respiratory illness caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), first officially reported in late 2019 in Wuhan, Hubei Province, China.¹⁻³ It quickly spread to numerous countries worldwide and has been declared a global pandemic by the World Health Organization (WHO).⁴ Since the global COVID-19 pandemic, several variations in SARS-CoV-2 have occurred globally. The emerging variations appear to be spreading rapidly, leading to more cases of COVID-19.⁵

The first COVID-19 case in Iran was officially reported on February 19, 2020. During the first ten days of the COVID-19 epidemic in Iran, the reproductive number (R_0) was estimated to be 4.70 (95% CI: 4.23-5.23).⁶

When someone with respiratory tract infections breathes, talks, shouts, sneezes, or coughs, large and tiny infectious droplets are released into the environment. Like other bioaerosols, these virus-containing respiratory droplets of various sizes could be suspended in the atmosphere for minutes to hours due to complex motions and thermodynamic transformations.^{7,8}

Respiratory virus transmission occurs mainly in three ways. The first and most common form is direct physical contact with the infected person or touching contaminated surfaces/fomites. Second, transmission through virus-containing respiratory droplets of different sizes occurs in the vicinity of the infected individual. Third, suspended droplets or particles are transmitted through air.⁹

At the onset of the COVID-19 outbreak, knowledge of the modes of transmission was based on the same family's virus literature, such as the Middle East Respiratory Syndrome Coronavirus (MERS-CoV) and Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV-1). MERS is a viral respiratory disease found for the first time in Saudi Arabia in 2012. Another outbreak was observed in 2015 in the Republic of Korea.¹⁰ Following the outbreak of MERS in South Korea, several studies were conducted to assess the survival and stability of MERS-CoV in the air and on the surfaces in healthcare facilities. In a study by bin et al., the RNA of MERS-CoV was isolated for up to five days from surfaces after the patients' latest positive polymerase chain reaction (PCR) test of respiratory specimens.

Moreover, viral RNA has been isolated in several surface samples from anterooms, medical tools, ventilation systems, bedsheets, bedrails, intravascular fluid hangers, and X-ray imaging equipment. Kim et al. observed the uninjured MERS-CoV particles of viral cultures of the indoor air and surface samples using the electron microscope imaging technique.¹¹ ¹² Nonetheless, according to the WHO guidelines, MERS is mainly transmitted through close contact between Healthcare workers.^{13, 14}

Studies on the COVID-19 transmission routes have indicated that it could be transmitted from person to person via expelled respiratory droplets larger than 5 µm in diameter. Close contact (usually less than 1 m) with an infected individual could be another way of COVID-19 transmission. Furthermore, the transmission may occur indirectly through contact with contaminated surfaces/fomites.^{15, 16}

Airborne transmission in the case of viruses, especially the novel SARS-CoV-2, refers to the infection caused by exposure to tiny respiratory droplets or fine particles containing the virus, which can remain suspended in the air for extended periods and prolonged distances, usually more than two meters.¹⁷ With a smaller size, the aerosols and tiny droplets can disperse more in the atmosphere, making it possible for SARS-CoV-2 to transmit beyond 2 m into the air.¹⁸ Several studies have explained the airborne transmission route of Norwalk-like virus and SARS-CoV-1 in school children, hospitals, health care facilities, and airplanes.¹⁹⁻²² The result of a study conducted on a pig farm revealed that the H1N1 virus could be contagious to animals via airborne routes.²³

Studies conducted in Hong Kong and Canada at health care facilities and aircraft had shown the SARS-CoV-1 airborne route transmission. The similarities between SARS-CoV-1 and SARS-CoV-2 and evidence-based on virus transmission, in general, support the hypothesis of COVID-19 airborne transmission.²²

Current investigations have isolated SARS-CoV-2 RNA from aerosols in Wuhan hospitals and outdoor air in Italy.^{24, 25} Moreover, Viable SARS-CoV-2 was detached from air samples at a distance of 2 to 4.8 m from patients.²⁶ Nonetheless, in a Sean Wei Xiang Ong et al., did not detect SARS-CoV-2 in the air of patients' rooms. In a journal article, Y. H. Li et al. reported a similar finding.²⁷ Besides, in a study by Faridi et al., all collected indoor air samples at a distance of 2 to 5 m from the patients' beds were negative for SARS-CoV-2 RNA.^{28, 29}

To date, airborne transmission is not entirely understood, and air transmission of COVID-19 is controversial.³⁰ However, the Centers for Disease Control and Prevention (CDC) and the WHO guidelines suggest that COVID-19 airborne transmission can happen under particular circumstances and or with aerosol-generating procedures. According to the CDC, particular conditions include indoor spaces with improper ventilation and exposure to an infected individual for more than 30 minutes.^{9, 31, 32}

Hospitals play a vital role in societies to keep communities and healthcare workers healthy.³³ HCWs accounted for 21% of cases in previous SARS and MERS epidemics. Moreover, in Hong Kong, Singapore, and Canada, over half of all mortality due to SARS infection had occurred among HCWs.³⁴ Hence, HCWs have the highest risk of exposure to SARS-CoV-2.³⁵ In China, approximately 3,000 HCWs were infected with SARS-CoV-2, and 22 died due to COVID-19.³⁶ It has been estimated that 10 to 20% of all COVID-19 infection cases occur among the front-line HCWs, which means maintaining their safety is critical.³⁷ Intensive care units (ICUs) are the most common source of infection due to the high presence of viruses in the environment and infected medical equipment. Therefore, the HCWs treating ICU patients are at higher risk of becoming infected with COVID-19.³⁸ Hence, investigating virus transmission patterns in healthcare facilities would be crucial to protecting the HCWs from contracting COVID-19. The present study aimed to investigate the possibility of airborne transmission of COVID-19 and the presence of SARS-CoV-2 on the surfaces/fomites in a COVID-19 referral hospital in Shiraz, Iran, from March to April, 2021.

Methods

Area of Study

This cross-sectional study was accomplished in

July 2020 at Ali Asghar Hospital in Shiraz County, Iran, considered the referral hospital for hospitalization of COVID-19 patients by Shiraz University of Medical Sciences.

Shiraz, the capital of Fars province, is placed at the latitude of 29°36' N and the longitude of 52°32' E (northwest of Fars Province, south of Iran) with an average elevation of 1500 m above sea level and an area of 240 km² (Figure 1). Shiraz is the fifth-most-populous city in Iran, with almost two million populations.^{39, 40}

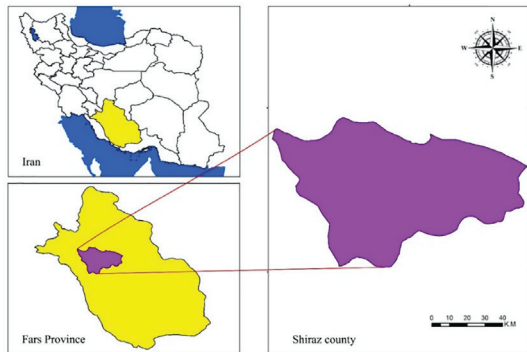


Figure 1: Location of the province of Fars and the county of Shiraz.

Ali Asghar Hospital has several wards, including triage, two accident and emergency (A&E) wards, five inpatients wards, three ICUs, one negative pressure room, a Computerized Tomography scan (CT scan), a laboratory, and an administrative department.

Environmental Sampling Indoor Air

Previous studies reported that the impinger technique is a flexible and successful method of collecting the SARS-CoV-1 and SARS-CoV-2 from the air. In this technique, airborne viruses are sucked into liquid or buffer and could be used for culture or molecular analysis.^{29, 41-43}

In the impinger technique, the sampling liquid evaporation limits the sampling time. The maximum efficiency is established in the first 60 minutes of sampling.⁴⁴ The U.S. Environmental Protection Agency (US EPA) recommends an air sample size of 750 to 6,000 liters for sampling the human-associated coronavirus.⁴⁵

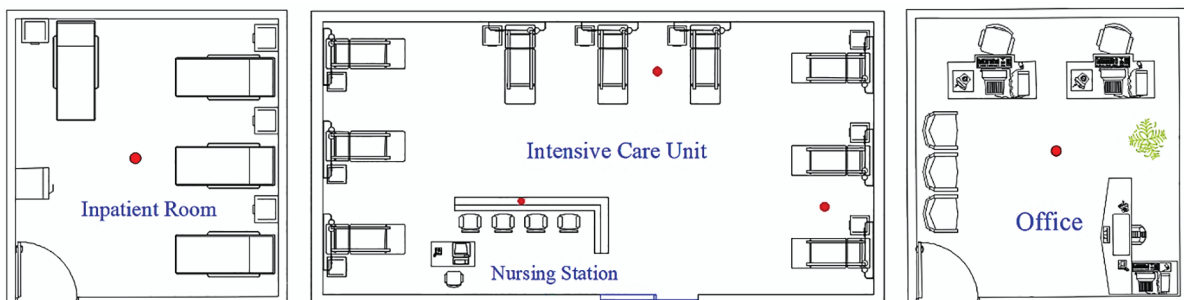


Figure 2: Schematic of air sampling locations in different hospital wards (red points are sampling locations).

This study collected 17 indoor air samples using a standard midjet impinger and a sampling pump (Legend Legacy, SKC Inc.) with a flow rate of 10 L min⁻¹. The indoor air sampling was performed for 60 minutes. Before each sampling collection, the impinger and the connecting tube were disinfected using ethyl alcohol 70%. Besides, the impinger was contained 15 ml of viral transfer medium (VTM). The VTM was contained Dulbecco's Modified Eagle Medium (DMEM), streptomycin (100 µg/ml), penicillin (100 IU/ml), and isoamyl alcohol (1%); as an anti-foaming agent.²⁹ Indoor air relative humidity (RH) and temperature were recorded using HD 110 KIMO during sampling (Sauermaann Group).

Figure 2 shows the schematic of air sampling locations in multiple wards of the studied hospital. Air sampling equipment was set up in different wards at an altitude of 1.5 m above the ground level as follows:

- At a distance of 2 m from the patient's bed in the ICU-1, ICU-2, ICU-3, the negative pressure room, the A&E-1, and A&E-2 wards.
- On the nursing station's counter (2 to 5 m from the patient's bed).
- In the admission and discharge office, inpatient's room, CT scan, laundry, office room in the administrative department (2 m from the personnel desk), and temporary waste storage area (TWS).

Surface and Fomites

Twenty-two samples were collected from surfaces and fomites using sterile polyester-tipped swabs (Dacron). Each collection vial contained 3 ml of the VTM. The surfaces were sampled before the daily cleaning and disinfection program. According to the WHO-recommended protocol, the surface sampling procedure was performed in 25 cm² for swab sampling. For the surface sampling, the wet swab with the VTM with appropriate pressure was rotated on the surfaces, and the swab was moved at least in two different directions.⁴⁶

SARS-CoV-2 Virus Detection Test

The real-time reverse transcription-polymerase chain reaction (rRT-PCR) is currently the golden standard and most reliable SARS-CoV-2 detection

method, which relies on recognizing unique ribonucleic acid (RNA) sequences in the SARS-CoV-2.^{47,48}

All the SARS-CoV-2 samples were immediately shipped to the laboratory under cold conditions (4°C) for further analysis. Collected samples were initially placed in a 50 ml Falcon containing phosphate-buffered saline (PBS) solution and stored in a refrigerator at 4°C. Samples were centrifuged at 13000 rpm. The SRAS-CoV-2 genome was then isolated using a virus RNA extraction kit (PCR cloning kit, SINACLON Co.-Iran) based on the manufacturer's instructions. Finally, the isolated RNA was tested using one-step rRT-PCR for the SRAS-CoV-2 genome.⁴⁹

The cycle threshold (Ct) indicates the number

of cycles required to amplify the viral RNA to a detectable level in the RT-PCR test. Besides, the Ct value is a semi-quantitative measure to determine the viral loads.⁴⁹ In the present study, the Ct cut-off value for positive RT-PCR of SARS-CoV-2 was 35.

The presence of the SARS-CoV-2 genome was assessed using a commercially available SARS-CoV-2 Test Kit (Pishtaz Teb Zaman Co.-Iran). According to the manufacturer's instructions, this kit is designed to detect SARS-CoV-2 using a one-step rRT-PCR method. The SARS-CoV-2 Test Kit is a molecular in vitro diagnostic technique that uses TaqMan probe-based technology for qualitatively detecting SARS-CoV-2. The virus detection targets are the nucleocapsid (N) genes and RNA-dependent RNA polymerase

Table 1: Results of indoor air sampling in different wards of the hospital

Samples	Hospital Wards	No. of patients or staff/status	SARS-CoV-2 presence or absence	Ventilation system	Temperature (°C)	Relative humidity (%)	Cycle threshold-ORF1ab gene	Cycle threshold-N gene
1	Intensive Care Unit -1	10 (Oxygen mask: 6, Intubated: 4)	+	Mechanical	23.4	30.6	30	31
2	Intensive Care Unit -2	10 (Oxygen mask: 5, Intubated: 5)	-	Mechanical	23.0	31.0	-	-
3	Intensive Care Unit -3	7 (Oxygen mask: 5, Intubated: 2)	+	Mechanical	24	31.0	33	33
4	Negative pressure room	2 / Intubated	+	Mechanical	23.1	19.0	33	34
5	Accident and emergency ward -1	15 (Oxygen mask: 2, Masked:13)	-	Mechanical/ Natural	22.8	33.0	-	-
6	Accident and emergency ward -2	15 (Oxygen mask: 1, Masked: 14)	+	Mechanical/ Natural	25.4	30.8	32	33
7	Accident and emergency ward -1 (Nursing station)	4 / Masked	-	Mechanical	21.8	27.6	-	-
8	Accident and emergency ward -2 (Nursing station)	4 / Masked	-	Mechanical	23.0	27.5	-	-
9	Surgery ward (Nursing station)	3 / Masked	-	Mechanical	24.4	32.0	-	-
10	Men's Internal Ward	4 / Masked	-	Mechanical/ Natural	21.0	27.6	-	-
11	Men's Internal Ward (Nursing station)	3 / masked	-	Mechanical/ Natural	22.5	29.0	-	-
12	Woman's Internal Ward	4 / masked	-	Mechanical/ Natural	23.0	33.7	-	-
13	Computerized Tomography scan	1 / masked	-	Mechanical	25.0	19.8	-	-
14	Admission and discharge office	2 / masked	+	Natural	19.0	21.0	33	34
15	Office in the administrative department	3 / masked	+	Natural	23.2	26.2	30	31
16	Landry	2 / masked	-	Mechanical	29.0	50.6	-	-
17	Temporary waste storage	1 / masked	+	Mechanical	27.6	41.7	31	31

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2); Severe acute respiratory syndrome coronavirus 2 (ORF1ab gene); Severe acute respiratory syndrome coronavirus 2 (N gene)

(RdRp). In addition, Ribonuclease P (RNase P) was used as an internal control.⁵⁰

Results

Indoor Air Sampling

Table 1 shows the presence or absence of SARS-CoV-2 RNA in indoor air samples of different wards of the studied hospital. The RNA of SARS-CoV-2 was found in 41.2% of indoor air samples. Typically, after screening and triage, patients with COVID-19 symptoms were admitted to healthcare settings, implying that the admission and discharge office staff were in close contact with COVID-19 patients. In this study, one indoor air sample collected from the admission and discharge office showed the presence of the SARS-CoV-2 RNA. Additionally, indoor air samples collected at a distance of 2 m from the patients' beds in ICU-1, ICU-3, and A&E-2 were positive. All collected indoor air samples from the nursing station, over 2 to 5 m from the patient's bed, were negative. The air sample taken from a crowded office in the administrative department was positive, possibly due to the occasional presence of asymptomatic HCWs or the presence of asymptomatic staff and inadequate ventilation.⁵¹ Besides, indoor air samples were positive for TWS. The SARS-CoV-2 RNA at a distance of 2 m from the patients' beds has been detected in the indoor air of the negative pressure room, probably due to the inadequate air exchange provided by the ventilation system.⁵² WHO recommended the minimum hourly average ventilation rates of 160 L/s/patient, 60 L/s/patient, and 2.5 L/s/m³ for airborne precaution rooms, general wards, outpatient departments, and corridors, respectively.⁵³ The minimum and maximum RH in different wards were 19% and 50.6%, respectively.

Surfaces and Fomites Sampling

Table 2 shows the surfaces and fomites sampling results. The results showed that 32% of the surfaces and fomites samples were positive. Also, 33% of samples taken from the medical ventilator and the patient's bed surface in the ICU wards were positive. Besides, all samples collected on the surfaces of the exhaust fans in ICUs and A&Es were negative SARS-CoV-2 RNA. 88.9% of the samples from main exhaust filters were positive in a study carried out by Karolina Nissen et al. in the wards at Uppsala University Hospital, Sweden. Hence, they suggested that there could be a chance for airborne transmission of SARS-CoV-2.⁵⁴ Surfaces that were more frequently touched, such as doorknobs in the bathroom of ward-1, were also positive. Samples from the food trolley were positive, suggesting that the virus infection may be transmitted from the wards to the hospital's kitchen. The surface sample collected from the protective counter screen in the admission and discharge office was positive for the SARS-CoV-2 RNA. Therefore, particular consideration should be given to the disinfection of surfaces in the admission and discharge offices.

Discussion

Indoor Air Sampling

Prior studies suggested that airborne transmission could be considered a critical transmission mode for COVID-19 in healthcare settings.⁵⁴ Similar findings were reported in a study conducted at the Nebraska University Hospital in the United States.⁵⁵ In a study performed at a hospital in Milan, Italy, all air samples in the ICU and the corridor were positive for SARS-CoV-2 RNA presence.⁵⁶ Experimental and direct visualization studies have reported that daily voice

Table 2: Results of the presence of SARS-CoV-2 RNA in surface and fomites samples of different hospital wards

Samples	Surfaces and formats	Presence or absence of SARS-CoV-2	Cycle threshold-threshold-ORF1ab gene	Cycle threshold-N gene
1	Medical ventilator (Intensive Care Unit -1)	+	30	31
2	Patient's bed (Intensive Care Unit -1)	+	31	31
3	Medical ventilator (Intensive Care Unit -2, Intensive Care Unit -3)	-	-	-
4	Patient's bed (Intensive Care Unit -2, Intensive Care Unit -3)	-	-	-
5	Exhaust fan (Intensive Care Unit -1, Intensive Care Unit -2, Intensive Care Unit -3)	-	-	-
6	The surface of the counter of the nursing station (Triage)	+	33	33
7	Patient's bed (Ward-1)	-	-	-
8	Office desk (Ward-1)	-	-	-
9	Exhaust fan (Ward-1 and Ward-2)	-	-	-
10	Doorknobs (Bathroom/Ward-1)	+	33	34
11	Hospital bedsheet (Landry)	-	-	-
12	Computerized Tomography scan patient's bed	-	-	-
13	Admission and discharge office (Protective Counter Screen)	+	32	34
14	Hospital food trolley surfaces	+	31	32
15	Temporary waste storage	-	-	-
16	Medical waste autoclaves	-	-	-

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2); Severe acute respiratory syndrome coronavirus 2 (ORF1ab gene); Severe acute respiratory syndrome coronavirus 2 (N gene)

communication can produce droplets of various sizes suspended in the air for 10 minutes or more.^{57, 58} Another experimental model showed healthy people could produce aerosols by coughing and speech.⁵¹ The authors suggested that the droplets could transmit COVID-19 in indoor environments.

Nonetheless, SARS-CoV-2 was not detected in the air of patients' rooms in a study by Sean Wei Xiang Ong.²⁸ Besides, Faridi et al. (2020) examined the indoor air of patients' rooms where patients with severe and critical symptoms of COVID-19 were hospitalized. The study showed that all collected indoor air samples at the distance of 2 to 5 m from the patient's bed were negative in terms of SARS-CoV-2,²⁹ which is consistent with our research findings.

The airborne transmission of SARS-CoV-2 can be influenced by different parameters such as RH and temperature. At RH below 40%, the probability of airborne transmission of SARS-CoV-2 is higher than the humidity of 90% or greater. The persistence of the SARS-CoV-2 at low temperatures (4°C) is significant, whereas it can no longer be detected after 5 minutes at temperatures around 70°C.^{59, 60} Studies in China reported a positive association between temperature and the spread of SARS-CoV-2 in Hubei, Hunan, and Anhui provinces, while a negative association was found in Zhejiang and Shandong provinces.⁶¹ On the other hand, some studies performed in China and the United States did not find significant correlations between ambient temperature and cumulative incidence of COVID-19.⁶²⁻⁶⁴ Findings of a study carried out by Zonglin He. et al. demonstrated that the mean temperature was more correlated with confirmed COVID-19 cases than RH.⁶⁵ In a study by Jingyuan Wang et al. (2021), the authors indicated a statistically significant negative relationship between R_0 , temperature, and RH in China and the United States of America.⁶⁶

Many studies on the airborne transmission of SARS-CoV-2 have used the RT-PCR test for diagnosis. Nevertheless, the RT-PCR test detects only SARS-CoV-2 RNA, not the viable or infectious form of the virus. This technique, therefore, makes it impossible to express the infectious ability of the virus.^{32, 67}

Surfaces and Fomites Sampling

Razzini, K. et al. (2020) demonstrated that 24.3% of the swab samples collected from the contaminated spaces in the ward of a hospital in Milan, Italy, were positive for the SARS-CoV-2 genome.⁵⁶ Likewise, 34.1% of samples collected from frequently touched surfaces during patient incubation were positive for SARS-CoV-2 RNA in a study by Xiaowen Hu. et al. in Qingdao, China. In that study, the collected samples from bathrooms and bedrooms were positive, with a rate of 46.7% and 50.0%, respectively. Furthermore, 60%, 40%, 40%, 33.3%, and 16.7% of the samples

were positive for cotton, ceramics, metal, wood, and plastics, respectively. On the other hand, SARS-CoV-2 has not been detected in living room.⁶⁸ Based on a study finding at the Imam Khomeini Hospital in Ardabil, Iran, 18% of frequently touched surfaces were positive for SARS-CoV-2 RNA.⁶⁹ In another study at a Chinese hospital, on-site SARS-CoV-2 contamination analysis was performed using the extraction-free loop-mediate isothermal amplification (LAMP) detection method. The results showed that the surface contamination rate was more than 70% in the isolation wards.⁷⁰

Based on the CDC and WHO guidelines, wearing a mask is the most effective action to prevent COVID-19 from spreading.⁷¹ In the current research, contamination of inside and outside surfaces of various face masks used by HCWs and patients in rooms affected by COVID-19 has been assessed. Based on the results, RNA of SARS-CoV-2 has not been detected.⁷¹

In the indirect COVID-19 transmission, the material of surfaces and fomites and the persistence of SARS-CoV-2 play an essential role.⁷² Several studies have reported temperature and humidity as the key factors affecting coronavirus survival on surfaces. Prior experience with SARS-CoV-1 indicated that the infection disappeared as the temperature increased in Summer.^{61, 63, 73} In addition, parameters such as temperature, humidity, and air quality may stimulate or decrease the rate of transmission and viability of viruses in the atmosphere or on surfaces.⁶² Shane Riddell et al. showed that rising temperature while maintaining extreme humidity decreases SARS-CoV-2 survival on surfaces.^{74, 75} In a journal article, Jennifer Biryukov et al. demonstrated that at 24°C, the SARS-CoV-2 half-life was 6.3 to 18.6 hours, depending on RH; however, when the temperature was raised to 35°C, the half-life was decreased to 1.0 to 8.9 hour.⁷⁶

In the case of SARS-CoV-2 detection, the RT-PCR technique is generally applied to recognize the virus's presence. Still, this method only detects the RNA of the virus and cannot detect the viable forms. Therefore, the viability of SARS-CoV-2 on surfaces is not well understood. Chin et al. demonstrated that the SARS-CoV-2 survive on surgical masks and cloth at 22°C for 96 h and 24 h, respectively.⁷⁷ On the other hand, Van Doremalen et al. compared the viability of SARS-CoV-1 and SARS-CoV-2 on surfaces and aerosols under experimental conditions. Their findings showed that both viruses have similar stability in the environment.⁷⁸ Hence, increasing virus survival on surfaces/fomites can increase the possibility of COVID-19 transmission.

Although various studies showed the contamination of surfaces/ fomites with SARS-CoV-2, the role of surface contact in transmitting COVID-19 is not yet

clear. The findings of a study performed by Harvey et al. revealed that the risk of COVID-19 infection from contact with highly touched surfaces was fewer than 5-in-10,000, and quantitative microbial risk assessment showed the minor role of fomites in the public transmission of SARS-CoV-2.⁷⁹ The CDC estimates that the probability of SARS-CoV-2 infection by contacting surfaces is less than 1 in 10,000, and the possibility of COVID-19 transmission from fomites to a susceptible individual after an infected person has been in an indoor space is insignificant after three days.⁸⁰ Nevertheless, the WHO considers surface contamination one of the main transmission routes, while Scientific Brief update of July 9, 2020 conveyed that there is no evidence of direct transmission of COVID-19 through fomites.⁸¹

Conclusion

This study aimed to examine the potential presence of SARS-CoV-2 in indoor air, surfaces, and fomites at a reference hospital for COVID-19 in Iran.

Our findings showed that 33% of the indoor air samples collected within 2 m of the patient's bed were positive, indicating the risk of SARS-CoV-2 transmission from a COVID-19 patient to a healthy person at a distance of 2 m or less. Moreover, findings demonstrated that all collected indoor air samples over 2 to 5 m from the patient's bed were negative. Given that the office's air sample was positive, the free movement of HCWs in close contact with the patients must be restricted, and natural or mechanical ventilation should be provided. In addition, our results showed the contamination of surfaces and fomites in the studied hospital's wards. In the COVID-19 pandemic, there is still initial uncertainty regarding transmission routes. Hence, regular disinfection should be performed with particular attention to frequently touched surfaces in healthcare facilities.

Moreover, washing hands with soap and water for a minimum of 20 seconds or using alcohol-based hand sanitizer with 60 to 70% alcohol is another means to eliminate SARS-CoV-2 and help prevent the surface transmission of COVID-19. Based on the WHO's updated guidelines, personal protective equipment is highly recommended for all HCWs in healthcare settings. Furthermore, air recirculation should be avoided, and proper natural and artificial ventilation should be provided to reduce COVID-19 transmission in healthcare settings.

According to the WHO recommendations, keeping physical distance, wearing a mask, frequent handwashing, and providing adequate ventilation in enclosed spaces are the possible ways to help prevent the spread of COVID-19 in the community.

Nowadays, COVID-19 is a major global challenge. Although our knowledge of transmission routes of

COVID-19 has improved, the argument for airborne transmission of COVID-19 remains. Therefore, further research should address the possible presence of viable SARS-CoV-2 on tiny droplets and aerosols in healthcare facilities and public environments.

The limitation of this study was that the RT-PCR test only detects SARS-CoV-2 RNA and cannot detect the viable virus, and, in the RT-PCR test, false-negative results occur in a low viral load.

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Conflict of interest: None declared.

References

- Andersen KG, Rambaut A, Lipkin WI, Holmes EC, Garry RF. The proximal origin of SARS-CoV-2. *Nat Med.* 2020;26(4):450-2. doi: 10.1038/s41591-020-0820-9. PubMed PMID: 32284615; PubMed Central PMCID: PMC7095063.
- Hu B, Guo H, Zhou P, Shi ZL. Characteristics of SARS-CoV-2 and COVID-19. *Nat Rev Microbiol.* 2021;19(3):141-54. doi: 10.1038/s41579-020-00459-7. PubMed PMID: 33024307; PubMed Central PMCID: PMC7537588.
- Sanche S, Lin YT, Xu C, Romero-Severson E, Hengartner N, Ke R. High Contagiousness and Rapid Spread of Severe Acute Respiratory Syndrome Coronavirus 2. *Emerg Infect Dis.* 2020;26(7):1470-7. doi: 10.3201/eid2607.200282. PubMed PMID: 32255761; PubMed Central PMCID: PMC7323562.
- Shakoor A, Chen X, Farooq TH, Shahzad U, Ashraf F, Rehman A, et al. Fluctuations in environmental pollutants and air quality during the lockdown in the USA and China: two sides of COVID-19 pandemic. *Air Qual Atmos Health.* 2020;13(11):1-8. doi: 10.1007/s11869-020-00888-6. PubMed PMID: 32837622; PubMed Central PMCID: PMC7415015.
- Abdool Karim SS, de Oliveira T. New SARS-CoV-2 Variants - Clinical, Public Health, and Vaccine Implications. *N Engl J Med.* 2021;384(19):1866-8. doi: 10.1056/NEJMc2100362. PubMed PMID: 33761203; PubMed Central PMCID: PMC8008749.
- Azimi SS, Koohi F, Aghaali M, Nikbakht R, Mahdavi M, Mokhayeri Y, et al. Estimation of the basic reproduction number (R_0) of the COVID-19 epidemic in Iran. *Med J Islam Repub Iran.* 2020;34:95. doi: 10.34171/mjiri.34.95. PubMed PMID: 33315980; PubMed Central PMCID: PMC7722950.
- Moreno T, Pinto RM, Bosch A, Moreno N, Alastuey

- A, Minguillon MC, et al. Tracing surface and airborne SARS-CoV-2 RNA inside public buses and subway trains. *Environ Int.* 2021;147:106326. doi: 10.1016/j.envint.2020.106326. PubMed PMID: 33340987; PubMed Central PMCID: PMC7723781.
- 8 Naddafi K, Jabbari H, Hoseini M, Nabizade R, Rahbar M, Yunesian M. Investigation of indoor and outdoor air bacterial density in Tehran subway system. *J Environ Health Sci Eng.* 2011.
 - 9 The Lancet Respiratory M. COVID-19 transmission-up in the air. *Lancet Respir Med.* 2020;8(12):1159. doi: 10.1016/S2213-2600(20)30514-2. PubMed PMID: 33129420; PubMed Central PMCID: PMC7598535.
 - 10 Park JE, Jung S, Kim A, Park JE. MERS transmission and risk factors: a systematic review. *BMC Public Health.* 2018;18(1):574. doi: 10.1186/s12889-018-5484-8. PubMed PMID: 29716568; PubMed Central PMCID: PMC5930778.
 - 11 Bin SY, Heo JY, Song MS, Lee J, Kim EH, Park SJ, et al. Environmental Contamination and Viral Shedding in MERS Patients During MERS-CoV Outbreak in South Korea. *Clin Infect Dis.* 2016;62(6):755-60. doi: 10.1093/cid/ciw1020. PubMed PMID: 26679623; PubMed Central PMCID: PMC7108026.
 - 12 Kim SH, Chang SY, Sung M, Park JH, Bin Kim H, Lee H, et al. Extensive Viable Middle East Respiratory Syndrome (MERS) Coronavirus Contamination in Air and Surrounding Environment in MERS Isolation Wards. *Clin Infect Dis.* 2016;63(3):363-9. doi: 10.1093/cid/ciw239. PubMed PMID: 27090992; PubMed Central PMCID: PMC7108054.
 - 13 World Health Organization. Investigation of cases of human infection with Middle East respiratory syndrome coronavirus (MERS-CoV): interim guidance. World Health Organization; June 2018.
 - 14 World Health Organization. Middle East respiratory syndrome coronavirus (MERS-CoV) 11 March 2019 [Available from: [https://www.who.int/news-room/fact-sheets/detail/middle-east-respiratory-syndrome-coronavirus-\(mers-cov\)](https://www.who.int/news-room/fact-sheets/detail/middle-east-respiratory-syndrome-coronavirus-(mers-cov))].
 - 15 World Health Organization. Modes of transmission of virus causing COVID-19: implications for IPC precaution recommendations: scientific brief, 27 March 2020. World Health Organization; 2020.
 - 16 Setti L, Passarini F, De Gennaro G, Barbieri P, Perrone MG, Borelli M, et al. Airborne Transmission Route of COVID-19: Why 2 Meters/6 Feet of Inter-Personal Distance Could Not Be Enough. *Int J Environ Res Public Health.* 2020;17(8). doi: 10.3390/ijerph17082932. PubMed PMID: 32340347; PubMed Central PMCID: PMC7215485.
 - 17 Scientific Brief. SARS-CoV-2 and Potential Airborne Transmission. *cdc.org*. Updated Oct. 5, 2020
 - 18 Godri Pollitt KJ, Peccia J, Ko AI, Kaminski N, Dela Cruz CS, Nebert DW, et al. COVID-19 vulnerability: the potential impact of genetic susceptibility and airborne transmission. *Hum Genomics.* 2020;14(1):17. doi: 10.1186/s40246-020-00267-3. PubMed PMID: 32398162; PubMed Central PMCID: PMC7214856.
 - 19 Li Y, Huang X, Yu IT, Wong TW, Qian H. Role of air distribution in SARS transmission during the largest nosocomial outbreak in Hong Kong. *Indoor Air.* 2005;15(2):83-95. doi: 10.1111/j.1600-0668.2004.00317.x. PubMed PMID: 15737151.
 - 20 Booth TF, Kournikakis B, Bastien N, Ho J, Kobasa D, Stadnyk L, et al. Detection of airborne severe acute respiratory syndrome (SARS) coronavirus and environmental contamination in SARS outbreak units. *J Infect Dis.* 2005;191(9):1472-7. doi: 10.1086/429634. PubMed PMID: 15809906; PubMed Central PMCID: PMC7202477.
 - 21 Olsen SJ, Chang HL, Cheung TY, Tang AF, Fisk TL, Ooi SP, et al. Transmission of the severe acute respiratory syndrome on aircraft. *N Engl J Med.* 2003;349(25):2416-22. doi: 10.1056/NEJMoa031349. PubMed PMID: 14681507.
 - 22 Morawska L, Cao J. Airborne transmission of SARS-CoV-2: The world should face the reality. *Environ Int.* 2020;139:105730. PubMed PMID: 32294574; PubMed Central PMCID: PMC7151430.
 - 23 Zhang H, Li X, Ma R, Li X, Zhou Y, Dong H, et al. Airborne spread and infection of a novel swine-origin influenza A (H1N1) virus. *Virol J.* 2013;10(1):204. doi: 10.1186/1743-422X-10-204. PubMed PMID: 23800032; PubMed Central PMCID: PMC3700749.
 - 24 Setti L, Passarini F, De Gennaro G, Barbieri P, Perrone MG, Borelli M, et al. SARS-Cov-2RNA found on particulate matter of Bergamo in Northern Italy: First evidence. *Environ Res.* 2020;188:109754. doi: 10.1016/j.envres.2020.109754. PubMed PMID: 32526492; PubMed Central PMCID: PMC7260575.
 - 25 Liu Y, Ning Z, Chen Y, Guo M, Liu Y, Gali NK, et al. Aerodynamic analysis of SARS-CoV-2 in two Wuhan hospitals. *Nature.* 2020;582(7813):557-60. doi: 10.1038/s41586-020-2271-3. PubMed PMID: 32340022.
 - 26 Lednicky JA, Lauzardo M, Fan ZH, Jutla A, Tilly TB, Gangwar M, et al. Viable SARS-CoV-2 in the air of a hospital room with COVID-19 patients. *Int J Infect Dis.* 2020;100:476-82. doi: 10.1016/j.ijid.2020.09.025. PubMed PMID: 32949774; PubMed Central PMCID: PMC7493737.
 - 27 Li YH, Fan YZ, Jiang L, Wang HB. Aerosol and environmental surface monitoring for SARS-CoV-2 RNA in a designated hospital for severe COVID-19 patients. *Epidemiol Infect.* 2020;148:e154. doi: 10.1017/S0950268820001570. PubMed PMID: 32660668; PubMed Central PMCID: PMC7371847.
 - 28 Ong SWX, Tan YK, Chia PY, Lee TH, Ng OT, Wong MSY, et al. Air, Surface Environmental, and Personal Protective Equipment Contamination by Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) From a Symptomatic Patient. *JAMA.* 2020;323(16):1610-2. doi: 10.1001/jama.2020.3227. PubMed PMID: 32129805; PubMed Central PMCID: PMC7057172.
 - 29 Faridi S, Niazi S, Sadeghi K, Naddafi K, Yavarian J,

- Shamsipour M, et al. A field indoor air measurement of SARS-CoV-2 in the patient rooms of the largest hospital in Iran. *Sci Total Environ*. 2020;725:138401. doi: 10.1016/j.scitotenv.2020.138401. PubMed PMID: 32283308; PubMed Central PMCID: PMC7194859.
- 30 Tabatabaeizadeh SA. Airborne transmission of COVID-19 and the role of face mask to prevent it: a systematic review and meta-analysis. *Eur J Med Res*. 2021;26(1):1. doi: 10.1186/s40001-020-00475-6. PubMed PMID: 33388089; PubMed Central PMCID: PMC7776300.
- 31 Wilson NM, Norton A, Young FP, Collins DW. Airborne transmission of severe acute respiratory syndrome coronavirus-2 to healthcare workers: a narrative review. *Anaesthesia*. 2020;75(8):1086-95. doi: 10.1111/anae.15093. PubMed PMID: 32311771; PubMed Central PMCID: PMC7264768.
- 32 Ram K, Thakur RC, Singh DK, Kawamura K, Shimouchi A, Sekine Y, et al. Why airborne transmission hasn't been conclusive in case of COVID-19? An atmospheric science perspective. *Sci Total Environ*. 2021;773:145525. doi: 10.1016/j.scitotenv.2021.145525. PubMed PMID: 33940729; PubMed Central PMCID: PMC7984961.
- 33 Jaafari J, Dehghani MH, Hoseini M, Safari GH. Investigation of hospital solid waste management in Iran. *World Review of Science, Technology and Sustainable Development*. 2015;12(2):111-25. doi: 10.1504/WRSTSD.2015.073820.
- 34 Bahl P, Doolan C, de Silva C, Chughtai AA, Bourouiba L, MacIntyre CR. Airborne or droplet precautions for health workers treating COVID-19? *J Infect Dis*. 2020. doi: 10.1093/infdis/jiaa189. PubMed PMID: 32301491; PubMed Central PMCID: PMC7184471.
- 35 Adams JG, Walls RM. Supporting the Health Care Workforce During the COVID-19 Global Epidemic. *JAMA*. 2020;323(15):1439-40. doi: 10.1001/jama.2020.3972. PubMed PMID: 32163102.
- 36 Sim MR. The COVID-19 pandemic: major risks to healthcare and other workers on the front line. *Occup Environ Med*. 2020;77(5):281-2. doi: 10.1136/oemed-2020-106567. PubMed PMID: 32238444.
- 37 Nguyen LH, Drew DA, Graham MS, Joshi AD, Guo C-G, Ma W, et al. Risk of COVID-19 among front-line health-care workers and the general community: a prospective cohort study. *Lancet Glob Health*. 2020;5(9):e475-e83. doi: 10.1016/s2468-2667(20)30164-x. PubMed PMID: 32745512; PubMed Central PMCID: PMC7491202.
- 38 Pourdowlat G, Panahi P, Pooransari P, Ghorbani F. Prophylactic recommendation for healthcare workers in COVID-19 pandemic. *Front Med*. 2020;4(2s):e39-e.
- 39 Gharehchahi E, Mahvi AH, Amini H, Nabizadeh R, Akhlaghi AA, Shamsipour M, et al. Health impact assessment of air pollution in Shiraz, Iran: a two-part study. *J Environ Health Sci Eng*. 2013;11(1):11. doi: 10.1186/2052-336X-11-11. PubMed PMID: 24499576; PubMed Central PMCID: PMC3776287.
- 40 Shahsavani S, Hoseini M, Dehghani M, Fararouei M. Characterisation and potential source identification of polycyclic aromatic hydrocarbons in atmospheric particles (PM10) from urban and suburban residential areas in Shiraz, Iran. *Chemosphere*. 2017;183:557-64. doi: 10.1016/j.chemosphere.2017.05.101. PubMed PMID: 28570899.
- 41 Dart A, Thornburg J. Collection efficiencies of bioaerosol impingers for virus-containing aerosols. *Atmos Environ*. 2008;42(4):828-32. doi: 10.1016/j.atmosenv.2007.11.003.
- 42 Verreault D, Moineau S, Duchaine C. Methods for sampling of airborne viruses. *Microbiol Mol Biol Rev*. 2008;72(3):413-44. doi: 10.1128/MMBR.00002-08. PubMed PMID: 18772283; PubMed Central PMCID: PMC2546863.
- 43 Vosoughi M, Karami C, Dargahi A, Jeddi F, Jalali KM, Hadisi A, et al. Investigation of SARS-CoV-2 in hospital indoor air of COVID-19 patients' ward with impinger method. *Environ Sci Pollut Res Int*. 2021;28(36):50480-8. doi: 10.1007/s11356-021-14260-3. PubMed PMID: 33956316; PubMed Central PMCID: PMC8100364.
- 44 Lin X, Willeke K, Uleviccius V, Grinshpun SA. Effect of sampling time on the collection efficiency of all-glass impingers. *Am Ind Hyg*. 1997;58(7):480-8. doi: 10.1080/15428119791012577.
- 45 Campisano R, Hall K, Griggs J, Willison S, Reimer S, Mash H, et al. Selected analytical methods for environmental remediation and recovery (SAM) 2017. US Environmental Protection Agency, Washington, DC. 2017.
- 46 World Health Organization. Surface sampling of coronavirus disease (COVID-19): a practical "how to" protocol for health care and public health professionals. World Health Organization; 2020.
- 47 COVID LabCorp. RT-PCR test EUA Summary. Accelerated Emergency Use Authorization (EUA) Summary COVID-19 RT-PCR Test (Laboratory Corporation of America) Available online: www.fda.gov (accessed on 20 March 2020).
- 48 World Health Organization. Laboratory testing for coronavirus disease 2019 (COVID-19) in suspected human cases: interim guidance, 2 March 2020. World Health Organization; 2020.
- 49 Rao SN, Manissero D, Steele VR, Pareja J. A Systematic Review of the Clinical Utility of Cycle Threshold Values in the Context of COVID-19. *Infect Dis Ther*. 2020;9(3):573-86. doi: 10.1007/s40121-020-00324-3. PubMed PMID: 32725536; PubMed Central PMCID: PMC7386165.
- 50 Karami C, Normohammadi A, Dargahi A, Vosoughi M, Zandian H, Jeddi F, et al. Investigation of SARS-CoV-2 virus on nozzle surfaces of fuel supply stations in North West of Iran. *Sci Total Environ*. 2021;780:146641. doi: 10.1016/j.scitotenv.2021.146641. PubMed PMID: 34030290; PubMed Central PMCID: PMC7981268.
- 51 Somsen GA, van Rijn C, Kooij S, Bem RA, Bonn D. Small droplet aerosols in poorly ventilated spaces and SARS-CoV-2 transmission. *Lancet Respir Med*.

- 2020;8(7):658-9. doi: 10.1016/S2213-2600(20)30245-9. PubMed PMID: 32473123; PubMed Central PMCID: PMC7255254.
- 52 Jayaweera M, Perera H, Gunawardana B, Manatunge J. Transmission of COVID-19 virus by droplets and aerosols: A critical review on the unresolved dichotomy. *Environ Res.* 2020;188:109819. doi: 10.1016/j.envres.2020.109819. PubMed PMID: 32569870; PubMed Central PMCID: PMC7293495.
- 53 Chartier Y, Pessoa-Silva C. Natural ventilation for infection control in health-care settings. 2009.
- 54 Nissen K, Krambrich J, Akaberi D, Hoffman T, Ling J, Lundkvist A, et al. Long-distance airborne dispersal of SARS-CoV-2 in COVID-19 wards. *Sci Rep.* 2020;10(1):19589. doi: 10.1038/s41598-020-76442-2. PubMed PMID: 33177563; PubMed Central PMCID: PMC7659316.
- 55 Santarpia JL, Rivera DN, Herrera V, Morwitzer MJ, Creager H, Santarpia GW, et al. Transmission potential of SARS-CoV-2 in viral shedding observed at the University of Nebraska Medical Center. *MedRxiv.* 2020. doi: 10.1101/2020.03.23.20039446.
- 56 Razzini K, Castrica M, Menchetti L, Maggi L, Negroni L, Orfeo NV, et al. SARS-CoV-2 RNA detection in the air and on surfaces in the COVID-19 ward of a hospital in Milan, Italy. *Sci Total Environ.* 2020;742:140540. doi: 10.1016/j.scitotenv.2020.140540. PubMed PMID: 32619843; PubMed Central PMCID: PMC7319646.
- 57 Asadi S, Bouvier N, Wexler AS, Ristenpart WD. The coronavirus pandemic and aerosols: Does COVID-19 transmit via expiratory particles? *Aerosol Sci Technol.* 2020;0(0):1-4. doi: 10.1080/02786826.2020.1749229. PubMed PMID: 32308568; PubMed Central PMCID: PMC7157964.
- 58 Stadnytskyi V, Bax CE, Bax A, Anfinrud P. The airborne lifetime of small speech droplets and their potential importance in SARS-CoV-2 transmission. *Proc Natl Acad Sci U S A.* 2020;117(22):11875-7. doi: 10.1073/pnas.2006874117. PubMed PMID: 32404416; PubMed Central PMCID: PMC7275719.
- 59 Delikhooon M, Guzman MI, Nabizadeh R, Norouzian Baghani A. Modes of Transmission of Severe Acute Respiratory Syndrome-Coronavirus-2 (SARS-CoV-2) and Factors Influencing on the Airborne Transmission: A Review. *Int J Environ Res Public Health.* 2021;18(2):395. doi: 10.3390/ijerph18020395. PubMed PMID: 33419142; PubMed Central PMCID: PMC7825517.
- 60 Ahlawat A, Wiedensohler A, Mishra SK. An Overview on the role of relative humidity in airborne transmission of SARS-CoV-2 in indoor environments. *Aerosol Air Qual Res.* 2020;20(9):1856-61. doi: 10.4209/aaqr.2020.06.0302
- 61 Shahzad F, Shahzad U, Fareed Z, Iqbal N, Hashmi SH, Ahmad F. Asymmetric nexus between temperature and COVID-19 in the top ten affected provinces of China: A current application of quantile-on-quantile approach. *Sci Total Environ.* 2020;736:139115. doi: 10.1016/j.scitotenv.2020.139115. PubMed PMID: 32470687; PubMed Central PMCID: PMC7194057.
- 62 Dogan B, Ben Jebli M, Shahzad K, Farooq TH, Shahzad U. Investigating the Effects of Meteorological Parameters on COVID-19: Case Study of New Jersey, United States. *Environ Res.* 2020;191:110148. doi: 10.1016/j.envres.2020.110148. PubMed PMID: 32877703; PubMed Central PMCID: PMC7456582.
- 63 Iqbal N, Fareed Z, Shahzad F, He X, Shahzad U, Lina M. The nexus between COVID-19, temperature and exchange rate in Wuhan city: New findings from partial and multiple wavelet coherence. *Sci Total Environ.* 2020;729:138916. doi: 10.1016/j.scitotenv.2020.138916. PubMed PMID: 32388129; PubMed Central PMCID: PMC7194511.
- 64 Yao Y, Pan J, Liu Z, Meng X, Wang W, Kan H, et al. No association of COVID-19 transmission with temperature or UV radiation in Chinese cities. *European Respiratory Journal.* 2020;55(5). doi: 10.1183/13993003.00517-2020. PubMed PMID: 32269084; PubMed Central PMCID: PMC7144256.
- 65 He Z, Chin Y, Yu S, Huang J, Zhang CJP, Zhu K, et al. The Influence of Average Temperature and Relative Humidity on New Cases of COVID-19: Time-Series Analysis. *JMIR Public Health Surveill.* 2021;7(1):e20495. doi: 10.2196/20495. PubMed PMID: 33232262; PubMed Central PMCID: PMC7836910.
- 66 Wang J, Tang K, Feng K, Lin X, Lv W, Chen K, et al. Impact of temperature and relative humidity on the transmission of COVID-19: a modelling study in China and the United States. *BMJ Open.* 2021;11(2):e043863. doi: 10.1136/bmjopen-2020-043863. PubMed PMID: 33597143; PubMed Central PMCID: PMC7893211.
- 67 Sepulcri C, Dentone C, Mikulska M, Bruzzone B, Lai A, Fenoglio D, et al. The longest persistence of viable SARS-CoV-2 with recurrence of viremia and relapsing symptomatic COVID-19 in an immunocompromised patient – a case study. *medRxiv.* 2021:2021.01.23.21249554. doi: 10.1101/2021.01.23.21249554.
- 68 Hu X, Ni W, Wang Z, Ma G, Pan B, Dong L, et al. The distribution of SARS-CoV-2 contamination on the environmental surfaces during incubation period of COVID-19 patients. *Ecotoxicol Environ Saf.* 2021;208:111438. doi: 10.1016/j.ecoenv.2020.111438. PubMed PMID: 33039873; PubMed Central PMCID: PMC7526608.
- 69 Dargahi A, Jeddi F, Vosoughi M, Karami C, Hadisi A, Ahamad Mokhtari S, et al. Investigation of SARS CoV-2 virus in environmental surface. *Environ Res.* 2021;195:110765. doi: 10.1016/j.envres.2021.110765. PubMed PMID: 33497684; PubMed Central PMCID: PMC7826054.
- 70 Wan B, Zhang X, Luo D, Zhang T, Chen X, Yao Y, et al. On-site analysis of COVID-19 on the surfaces in wards. *Sci Total Environ.* 2021;753:141758. doi: 10.1016/j.scitotenv.2020.141758. PubMed PMID: 32898806; PubMed Central PMCID: PMC7434306.
- 71 Dargahi A, Jeddi F, Ghobadi H, Vosoughi M, Karami C, Sarailoo M, et al. Evaluation of masks' internal

- and external surfaces used by health care workers and patients in coronavirus-2 (SARS-CoV-2) wards. *Environ Res.* 2021;196:110948. doi: 10.1016/j.envres.2021.110948. PubMed PMID: 33684411; PubMed Central PMCID: PMC7935683.
- 72 Aydogdu MO, Altun E, Chung E, Ren G, Homer-Vanniasinkam S, Chen B, et al. Surface interactions and viability of coronaviruses. *J R Soc Interface.* 2021;18(174):20200798. doi: 10.1098/rsif.2020.0798. PubMed PMID: 33402019; PubMed Central PMCID: PMC7879773.
- 73 Shahzad K, Shahzad U, Iqbal N, Shahzad F, Fareed Z. Effects of climatological parameters on the outbreak spread of COVID-19 in highly affected regions of Spain. *Environ Sci Pollut Res Int.* 2020;27(31):39657-66. doi: 10.1007/s11356-020-10551-3. PubMed PMID: 32827296; PubMed Central PMCID: PMC7442890.
- 74 Riddell S, Goldie S, Hill A, Eagles D, Drew TW. The effect of temperature on persistence of SARS-CoV-2 on common surfaces. *Virology.* 2020;17(1):145. doi: 10.1186/s12985-020-01418-7. PubMed PMID: 33028356; PubMed Central PMCID: PMC7538848.
- 75 Fareed Z, Iqbal N, Shahzad F, Shah SGM, Zulfiqar B, Shahzad K, et al. Co-variance nexus between COVID-19 mortality, humidity, and air quality index in Wuhan, China: New insights from partial and multiple wavelet coherence. *Air Qual Atmos Health.* 2020;13:673-82. doi: 10.1007/s11869-020-00847-1. PubMed PMID: 32837610; PubMed Central PMCID: PMC7279636.
- 76 Biryukov J, Boydston J, Dunning R, Yeager J, Wood S, Reese A, et al. Increasing temperature and relative humidity accelerates inactivation of SARS-CoV-2 on surfaces. *mSphere.* 2020;5(4). doi: 10.1128/mSphere.00441-20. PubMed PMID: 32611701; PubMed Central PMCID: PMC7333574.
- 77 Chin A, Chu J, Perera M, Hui K, Yen H-L, Chan M, et al. Stability of SARS-CoV-2 in different environmental conditions. *medRxiv.* 2020.
- 78 van Doremalen N, Bushmaker T, Morris DH, Holbrook MG, Gamble A, Williamson BN, et al. Aerosol and Surface Stability of SARS-CoV-2 as Compared with SARS-CoV-1. *N Engl J Med.* 2020;382(16):1564-7. doi: 10.1056/NEJMc2004973. PubMed PMID: 32182409; PubMed Central PMCID: PMC7121658.
- 79 Harvey AP, Fuhrmeister ER, Cantrell ME, Pitol AK, Swarthout JM, Powers JE, et al. Longitudinal Monitoring of SARS-CoV-2 RNA on High-Touch Surfaces in a Community Setting. *Environ Sci Technol Lett.* 2021;8(2):168-75. doi: 10.1021/acs.estlett.0c00875. PubMed PMID: 34192125; PubMed Central PMCID: PMC7927285.
- 80 Centers for Disease Control Prevention. Science brief: SARS-CoV-2 and surface (fomite) transmission for indoor community environments. Updated Apr; 2021
- 81 World Health Organization. Transmission of SARS-CoV-2: implications for infection prevention precautions: scientific brief, 09 July 2020. World Health Organization; 2020.