RESEARCH ARTICLE



The detection of SARS-CoV-2 in outpatient clinics and public facilities during the COVID-19 pandemic

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Hossein M. Elbadawy, Department of Pharmacology and Toxicology, College of Pharmacy, Taibah University, Madinah, Saudi Arabia. Email: hmbadawy@taibahu.edu.sa Abstract

The transmission of the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) can occur through an airborne route, in addition to contaminated surfaces and objects. In hospitals, it has been confirmed by several studies that SARS-CoV-2 can contaminate surfaces and medical equipment especially in hospitals dedicated to coronavirus disease 2019 (COVID-19) patients. The aim of this study was to detect the contamination of hands, objects, and surfaces in isolation rooms and also in outpatients' clinics in hospitals and polyclinics. Environmental contamination of public high-touch surfaces in public facilities was also investigated during an active COVID-19 pandemic. Random swabs were also taken from public shops, pharmacies, bakeries, groceries, banknotes, and automated teller machines (ATMs). Samples were analyzed for SARS-CoV-2 positivity using real-time polymerase chain reaction. In the COVID-19 regional reference hospital, only 3 out of 20 samples were positive for SARS-CoV-2 RNA. Hand swabs from SARS-CoV-2-positive patients in isolation rooms were occasionally positive for viral RNA. In outpatients' clinics, door handles were the most contaminated surfaces. Dental chairs, sinks, keyboards, ophthalmoscopes, and laboratory equipment were also contaminated. Although no positive swabs were found in shops and public facilities, random ATM swabs returned a positive result for SARS-CoV-2. Although there is no longer a focus on COVID-19 wards and isolation hospitals, more attention is required to decontaminate frequently touched surfaces in health-care facilities used by patients not diagnosed with COVID-19. Additionally, high-touch public surfaces such as ATMs require further disinfection procedures to limit the transmission of the infection.

KEYWORDS

ATM, COVID-19, outpatient, SARS-CoV-2, surfaces

1 | INTRODUCTION

The severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection, also known as the new coronavirus disease 2019 (COVID-19), was declared as a worldwide pandemic in 2020. COVID-19 was first reported in December 2019, in Wuhan city, China, and spread progressively throughout China and then to the whole world.¹ The world is waiting for science to find answers and solutions to the devastating global problem of COVID-19. The current count of SARS-CoV-2 infection is escalating. Although most patients have mild symptoms and a good prognosis after infection, some patients developed severe respiratory distress and died. The SARS-CoV-2 infection can be divided into three stages: viremia, pneumonia, and then the recovery phase or death. Learning about the mechanism by which the SARS-CoV-2 infection is spreading is essential for the prevention of the infection and to reduce mortalities.

COVID-19's high capacity for human-to-human transmission is alarming. During the course of the COVID-19 disease, no antiviral drug has yet shown any clear cut efficiency, however, one antiviral drug, namely, Remdesivir, has shown some promise.^{2,3} Antiviral drugs, despite limited efficiency, can be used in all stages of the disease. However, it is extremely important to take proper measurements to contain the spread of the virus and prevent infection. Understanding the routes by which the virus spreads is the main factor in breaking up the chain of transmission and maintaining the infection levels within the capacity of the health-care system.

The spread of the virus to the surrounding environment still has many mysteries. It was initially thought that the size of the virus will not allow it to spread via airborne transmission, and therefore, the World Health Organization (WHO) recommendations were not to wear a face mask unless a person is infected or looking after an infected person. However, this information changed recently, due to a better understanding of how the virus is spreading. Symptomatic patients are, possibly, contaminating surfaces around them, which can spread the infection if touched by healthy individuals,⁴ sometimes even after the recovery of a patient.⁵ The gap in knowledge is evident, as reported by the WHO.⁶ Under health-care settings, infection prevention and control is even more important. It has been reported that a large percentage of health-care professionals have been infected due to exposure to SARS-CoV-2 during work.^{7,8} Due to the lack of knowledge about the pathogen, the frontline health-care workers were most affected.⁷ Additionally, the period of exposure to such a threat is prolonged for health-care professionals who spend long hours at work. For sure, personal protecting equipment can help, however, this differs from one hospital to another and from one country to other countries around the world. Taking swabs from different surfaces in hospitals in Singapore showed that the most contaminated surfaces were the air exhaust vent and the floor, followed by bed rails and locker handles, then electric switches, seats and chairs, and toilets.⁹ Additionally, contamination was highest during the first week of illness.⁹ Another investigation showed that the air was clean in three tested rooms, where SARS-CoV-2 patients were staying, however, air vents and surfaces were mostly positive

for the detection of the virus, showing that airflow systems can be shedding viral particles onto surfaces.⁴ Importantly, data on the spread of viruses on surfaces are not redundant. One study, involved only three patients, while another study in Korea showed results from hospital rooms occupied with 13 patients only.¹⁰ Additionally, very limited information is available regarding the transmission of the virus on surfaces in outpatients' clinics or in public high-touch surfaces. The virus was detected on patients' hands in one recent study¹¹; however, the debate is ongoing regarding the major route of transmission, whether it can be transmitted more through touching contaminated surfaces or by airborne droplets. Thus, more studies are required to confirm or oppose these findings and to highlight the precautions which should be taken to better control the spread of the infection. Information regarding the spread of the virus on surfaces and objects is essential for controlling the outbreak and is, therefore, the main target of this study.

2 | MATERIALS AND METHODS

Specific real-time reverse-transcriptase-polymerase chain reaction (RT-PCR) was used to detect the presence of SARS-CoV-2 on swabs taken from the hands of patients and from surfaces and objects. Sample collection was carried out by trained professionals from the infection control unit at the involved health-care facilities, taking all necessary precautions. Ethical approval was obtained for carrying out this study from the Institutional Review Board of the General Directorate of Health Affairs in Madinah (No. H-03-M-084).

2.1 | RT-PCR protocol

Collected swabs were processed for viral RNA extraction using the Viral Nucleic Acid Extraction Kit II (Geneaid Biotechnology). Briefly, 400 µl of VB buffer and 200 µl of phosphate-buffered saline swab-elutes were added to each tube and then incubated for 10 min at 35°C. The mixture was centrifuged at 16,000g for 2 min and the supernatant was transferred to a new 1.5 ml microcentrifuge tube. A volume of 400 µl of washing buffer (AD buffer) was added to each tube and mixed gently for 10 s. Afterwards, 200 µl of absolute ethanol was then added to the mixture before transferring the entire contents to a extraction column. The columns were centrifuged at 16,000g for 2 min to discard the flow-through and the column was then transferred to a new 2 ml collection tube. The column was then washed twice using 400 and 600 µl of W1 buffer. Finally, 50 µl of preheated elution buffer was added to the extraction columns and left standing for 3 min at room temperature to allow the elution buffer to be completely absorbed. RNA was then eluted from the extraction columns by centrifuging the columns at 16,000g for 30 s. RNA concentration and purity for extracted samples were assessed by analyzing 1 µl of the elute using a NanoDrop 1000 UV-VIS Spectrophotometer (Thermo Fisher Scientific).

For real-time PCR analysis, the center for disease control and prevention quantitative PCR (qPCR) Probe Assay was used.¹² The qRT-PCR reaction was prepared by adding 1.5 μ l of primer/probe mix (100pM pH8) to 8.5 μ l of Nuclease-free water (VWR International), and 5 μ l of the Oasig Lyophilised OneStep RT-qPCR Master Mix (PrimerDesign Ltd.). Thermal cycling was performed on the ABI-7500 Fast 96-well Real-Time PCR System (Applied Biosystems) with an initial reverse-transcription step of 10 min at 55°C step, followed by an enzyme activation step at 95°C for 2 min, then followed by 45 cycles of denaturation at 95°C for 10 s, then annealing and data acquisition at 60°C for 10 s. CT and cut-off analysis to determine the positive and negative samples were performed using the internal software of the ABI-7500 system (software version 2.0.4).

2.2 | Hand swabs

For taking hand swabs, patients who are SARS-CoV-2 positive within 2 days and who agreed to participate in the study were recruited after taking signed consent. For ER patients presenting in the emergency room (ER) as suspected COVID-19 cases, swabs of hands were taken, and the patients' records were followed up later to select those with PCR positive results and include their hand swab results in this study. A questionnaire was distributed to eight patients who agreed to participate and signed informed consent. The questionnaire was composed of questions about age, sex, days of home isolation before admission to hospital, hand wash hygiene, and adhering to home isolation rules.

2.3 | Hospitals and public swabs

Swabs were taken from COVID-19 isolation hospital and from outpatients' hospitals and polyclinics. High-touch surfaces were swabbed, and samples were sent for real-time PCR analysis following the protocol above. The investigation also included cell phones, automated teller machine (ATM) machines keypads, banknotes, and public shops, shopping carts, pharmacies, and bakeries.

3 | RESULTS

3.1 | Hand hygiene of SARS-CoV-2 carriers

Hand swabs were taken from patients admitted to the intensive care unit (ICU) and patients in home isolation or in isolation rooms in hospitals. All 16 patients were PCR positive for SARS-CoV-2. Of these patients, 12.5% showed positive hand contamination with SARS-CoV-2 (Table 1), found only in patients in isolation with moderate to severe symptoms, however, under ICU settings, all hand swabs were negative. The hands of two patients at home isolation and another three patients presenting at visual triage in the hospital ER were all negative. Eight of these patients agreed to fill a brief questionnaire; six males and two females. The average age of the

TABLE 1Hand swabs

Location	Total swabs	Negative	Positive
ICU	3	3	0
Hospital isolation	8	6	2
Home isolation	2	2	0
ER reception	3	3	0

Abbreviations: ER, emergency room; ICU, intensive care unit.

participants was 62.75%, the youngest being 50 years old while the oldest was 80 years old. It was found that the two patients who did not observe regular hand hygiene were positive when hand swabs were taken from them. Although those patients were SARS-CoV-2 positive for 8.6 days before being hospitalized, during the stay at home only 25% (two patients) were strictly adhering to home isolation rules. The other six patients reported visiting mosques, shopping malls, and going to work while having symptoms.

3.2 | Detection of SARS-CoV-2 in COVID-19 wards and isolation rooms

To detect the presence of the SARS-CoV-2 virus in the COVID-19 reference hospital, 20 swabs were taken from different locations in the hospital and sent for real-time PCR analysis. Starting with wards dedicated for COVID-19 inpatient, several sites were swabbed (Table 2). Three out of 20 swabs were positive, including swabs taken from the pharmacy office (office, door, and chair), from the laboratory station where samples are being processed, and in the nursing station at the ICU unit. On the other hand, all swabs taken from isolation wards and other hospital facilities were negative (Table 2).

The contamination of surfaces and objects was investigated inside isolation rooms, at hospital or at home isolation. A total of 32 swabs were taken from banknotes, mobile phones, air condition filters, door handles, and sink faucets in isolation rooms in the hospital and at home, as detailed in Table 3. To investigate the contamination of surfaces and objects in self-isolated and inpatients rooms, swabs were taken from door handles and sink faucets of rooms occupied by SARS-CoV-2-positive patients at home or in hospital. The air condition filter in two home isolation patients who were positive within 2 days from taking the swabs was negative. Mobile phones and banknotes from patients at home or hospital isolation were also negative for SARS-CoV-2. Five random banknotes were taken from pharmacies, grocery shops, and fruit and vegetable shops were all negative for SARS-CoV-2 (Table 3).

3.3 | Contamination of public facilities with SARS-CoV-2

To study whether public high-touch surfaces can represent a possible route of transmission for SARS-CoV-2 infection, swabs

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Location	Swab site	Negative	Positive
Emergency room	Cardiac monitor CPR curtains	1 1	0 0
	CPR bed CPR monitor	1 1	0 0
Elevator	Office, chairs, bed and sink	1	0
Pharmacy office	Office and bed Counter and tables	0 1	1 0
Laboratory	Station Air flow cabinet	0 1	1 0
Human resources office	Counter, beds and offices	1	0
ICU	Infusion pump Ventilator	1	0
	Doctors room	1	0
	Intubation set	1	0
	Nurses station	0	1
Isolation wards	Medication trolly	1	0
	Nurses station	1	0
	HEPA filter surface (CPR)	1	0
	PC and keyboards	1	0
	Air condition filters	1	0

Abbreviations: CPR, cardiopulmonary resuscitation; HEPA, highefficiency particulate air; ICU, intensive care unit; PC, personal computer.

were taken from random ATM, elevator buttons, supermarket shopping carts, pharmacy doors, and counter and bakery doors. Elevators swabbed were located in hospitals and shopping malls. Although all swabs from elevators, supermarket shopping carts, pharmacies, and bakeries returned negative results; however, swabs taken from the ATM machine keypad showed 1 positive out of 10 swabbed machines. Each ATM was swabbed twice, one swab for the keypad and another for the touch screen. This may indicate that commonly touched surfaces by several individuals can be positive for SARS-CoV-2 and therefore requires regular decontamination (Table 4).

TABLE 4 High-touch surfaces in public facilities

Location	Total swabs	Swab site	Negative	Positive
ATM	10	Buttons and keypads	10	1
Elevator	3	Buttons pads	3	0
Supermarket	3	Shopping carts	3	0
Pharmacy	3	Counter and door handles	3	0
Bakery	3	Door handles	3	0

Abbreviation: ATM, automated teller machine.

3.4 | Detection of SARS-CoV-2 virus in outpatient polyclinics

Previous studies investigated the contamination of COVID-19 hospital wards; however, the focus of this study was to inspect the outpatient health-care facilities. Of note, COVID-19 patients are not treated in these facilities as they have a designated reference center and an isolation hospital dedicated only for COVID-19 at this time of the pandemic. To study the contamination of surfaces in outpatient polyclinics, surfaces and instruments were swabbed and analyzed by real-time PCR for SARS-CoV2 positivity. Three swabs from the reception desk at three hospitals and polyclinics were negative. At each, a swab from the automated card payment station was taken, along with a swab from the front desk. Office, chairs, bed, and sink were negative for SARS-CoV-2 in one room used for minor surgeries. Offices, chairs, and beds were all negative in family medicine, general practice, internal medicine, and blood withdrawal rooms. However, door handles for general practice and family medicine clinics were positive. In dentistry clinics, swabs taken from five door handles at five different dentistry clinics returned two positive results, while five dental chairs and sink (one swab per clinic) also returned two positive results. Instruments such as ECG, X-ray, and ultrasound machines were all negative for SARS-CoV-2 contamination; however, equipment in the ophthalmology clinic returned a positive result out of three. Equipment included tonometers, autorefractors, slit-lamp microscopes, and phoropters. Instruments and touchpads in two laboratories returned one positive result, and also one out of three

TABLE 3Objects and surfaces in isolation rooms

Object	Total swabs	Swab site	Negative	Positive
Banknotes	10	ICU and isolation patients and public shops	10	0
Mobile phones	10	Isolation rooms in hospitals and home isolation	10	0
Air condition filters	2	Home isolation rooms	2	0
Door handles	5	Isolation rooms in hospitals and home isolation	5	0
Sink faucets	5	Isolation rooms in hospitals and home isolation	5	0

Abbreviation: ICU, intensive care unit.

swabs taken from the laboratory office was positive. Offices and beds in pediatric and psychology clinics were negative for SARS-CoV-2; however, one out of two swabs taken from door handles of pediatric clinics was positive. Finally, swabs taken from doors and sink faucets of bathrooms in hospitals and polyclinics were all negative. The results show that health-care facilities are an active site for infection via contaminated surfaces (Table 5).

4 | DISCUSSION

The contamination of surfaces and objects with SARS-CoV-2 has been proposed as one of the main routes of transmission of the infection.^{4,9,13} Several articles have confirmed the contamination of surfaces and objects with SARS-CoV-2 RNA in isolation wards and ICU units housing COVID-19 patients; however, other outpatients' health-care facilities were not thoroughly investigated. Patients using seeking medical care in the facilities are supposedly not infected, but the threat of asymptomatic carriers, spreading the infection in air and surfaces, is still a concern.

The results of this study have confirmed the presence of viral RNA at surfaces frequently touched by patients. Hands of SARS-CoV-2-positive patients can be contaminated with viral particles while touching a surface or an object, resulting in a

contaminated environment. However, until recently, hand swabs were not attempted. One study, which is still in press at the time of writing up this article, showed that the hands of COVID-19 patients can be positive for SARS-CoV-2 RNA.¹¹ This was further confirmed by our results, showing 12.5% of swabbed hands were positive for COVID-19 patients with moderate to severe symptoms, while no positive results were seen in ICU patients, showing that hand hygiene is better maintained in ICU because of medical supervision. Additionally, patients in ICU exhibit lower mobility and less activity in general, thus not using hands as often as for patients who are SARS-CoV-2 positive and maintained under self-isolation or in hospitals. High-touch surfaces can, for sure, be a source of infection. The possible areas where the public can contract the virus by frequent touching were investigated, and the SARS-CoV-2 RNA was detected in outpatient clinics and public facilities by taking random swabs. Starting with hospitals and polyclinics, not regularly receiving COVID-19 patients, several hospitals and health-care facilities were included in this study. A number of these hospitals were not designated to deal with SARS-CoV-2 patients, as they are usually referred to the COVID-19 reference hospital in the city. Yet, viral contamination of surfaces and equipment was evident. Specifically, door handles in hospitals and polyclinics were found to be contaminated with SARS-CoV-2. This further confirms the results shown previously in other countries. In Italy, for example, a study in 2020 showed that

Location	Total swabs	Swab site	Negative	Positive
Reception	3	Payment stations, desk	3	0
General practice	6	Office and bed Door handle	3 2	0 1
Family medicine	3	Office, examination bed Door handle	2 1	0 1
Surgery Room	1	Office, chairs, bed and sink	1	0
Laboratory	6	Chair for blood withdrawal Instruments, keyboards, touch screens and working benches	2 2	0 1
		Keyboard, mouse, phone and office	2	1
Ophthalmology	6	Equipment Door handle	2 3	1 0
Dental clinic	10	Dental chair, sink and office Door handle	5 5	2 2
Pediatric clinic	4	Bed and office Door handle	2 2	0 1
Internal Medicine	1	Office and bed	1	0
Emergency room	3	Counter, beds and offices	3	0
X-ray	1	Radiography machine	1	0
Psychiatry clinic	1	Door and office	1	0
Bathroom	3	Doors, sink faucets	3	0

 TABLE 5
 Outpatients' health-care facilities

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door handles were frequently positive for SARS-CoV-2.^{13–15} Keyboards in hospitals were also contaminated with the virus and can be a source of infection as reported before,¹⁴ and as confirmed by the results presented in this study. Medical equipment and instruments were occasionally positive for SARS-CoV-2 RNA, in line with findings from previous reports, showing that two-third of medical equipment was contaminated with the virus.¹⁵

It was recently reported that the detection of the virus always declines after the first week of the disease time course.¹⁶ This can be the cause of not detecting viral contamination in patients in self-isolation or in ICU. Additionally, viral detection in rooms occupied by patients with severe pneumonia was reported to be small.¹⁰ Viral load was reportedly lower in patients with progressive pneumonia.¹⁷ For this, SARS-CoV-2, being undetectable in ICU, can be rationalized as due to lower viral load, regular disinfection, and limited movement in the room as patients are mostly on external oxygen support or mechanical ventilation.

Regarding dental clinics, it was shown that dental chairs and sinks in the dentistry clinic can be positive for SARS-CoV-2. Although viral contamination was not previously detected in dental clinics, it was shown that saliva is a source of SARS-CoV-2 infection.¹⁸ Dentistry was proposed as one of the major medical fields affected by the pandemic and strict measures should be taken to prevent infection.¹⁹ Regarding swabs from public areas where frequent touching is expected, the main finding was the detection of the viral RNA on ATMs. It was confirmed for the first time in this study that ATMs can be contaminated with SARS-CoV-2 and if not disinfected regularly, may represent a source of infection. ATMs can be considered a high-touch surface that may carry the virus for hours, due to the material of the keypad and the frequency with which is being touched by several individuals. Although it was not reported before that ATMs, specifically, are contaminated by SARS-CoV-2, a company called Phylagenlaunched a SARS-CoV-2 test kit for surfaces and while testing the kit on 75 frequently touched surfaces in Los Angeles, including supermarkets, petrol stations, escalators, public shops, gyms, and ATMs, they obtained 11 positive results of these swabs. However, the study did not show which surfaces tested positive. As the presence of viral RNA on random ATM swabs was shown in this study, enhanced cleaning and disinfection measures are required to prevent the spread of the infection via ATM. Routine cleaning of high-touch surfaces is not enough, as deep cleaning using effective antiviral disinfectants is required to be performed more frequently and thoroughly.

Collectively, it was concluded that outpatients' health-care facilities can be contaminated with SARS-CoV-2, it was evident that medical equipment not commonly used for COVID-19 patients are also contaminated. Additionally, dental clinics were more frequently contaminated rather than other clinics due to the invasive nature of the dental procedures performed. Regarding public facilities and high-touch surfaces, the virus was mainly detectable on keypads and touch screens of ATM money dispensers requiring further action to be taken to minimize the risk of COVID-19 transmission.

5 | CONCLUSION

The SARS-CoV-2 RNA was detected mainly in nonclean areas, while ICU units were mostly clean. Under health-care settings, door handles were the most contaminated objects, while medical equipment such as dental chairs and ophthalmoscopes were also positive for SARS-CoV-2 RNA. Hand swabs taken from patients in ICU were negative, while the viral RNA was detectable on the hands of patients in isolation rooms. Random samples from mobile phones and banknotes were negative; however, the ATM returned some positive results, showing its possible role in the spread of infection. The results showed the necessity to take more protective measures to prevent the spread of the infection through door handles and medical instruments. Additionally, it is confirmed that the ATM can be a source of infection.

CONFLICT OF INTERESTS

The author declares that there are no conflict of interests.

AUTHOR CONTRIBUTIONS

Study concept and design: Hossein M. Elbadawy and Amin K. Khattab. Analysis and interpretation of data: Meky Abouzied, Ameira S. Mahmoud, and Heba M. Eltahir. Drafting of the manuscript: Hossein M. Elbadawy, Ali Alalawi and Saleh Bahashwan. Experimental work: Bandar A. Suliman, Ziab Alahmadey, Fahad Dakilallah Aljohani, and Hamza Sundogji. Statistical analysis: Hossein M. Elbadawy. Critical revision of the manuscript was done by all authors.

PEER REVIEW

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DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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