



Detection of Coronavirus Disease 2019 (COVID-19) Virus on the Surface of Hospital Settings by Quantitative Real-Time Polymerase Chain Reaction

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Abstract

Background & Objectives: Coronavirus disease 2019 (COVID-19) has had a profound impact on human health, with over 704,753,890 confirmed cases and 7,010,681 deaths reported to date. This study aimed to evaluate the contamination rate of COVID-19 on high-risk surfaces at Valiasr Hospital in Fasa, Iran, using molecular testing.

Materials & Methods: A total of 142 surface swabs, immersed in viral transport medium (VTM), were collected and transported to the Virology Reference Laboratory at the Cohort Center of Fasa University of Medical Sciences for COVID-19 virus testing. The presence of the virus was assessed using the real-time polymerase chain reaction (RT-qPCR) technique (QIAquant 96 5plex), following the manufacturer's protocol (Qiagen, MD, USA).

Results: Among the 142 samples obtained from surfaces and equipment in the COVID-19 ward, two samples tested positive for COVID-19. Similarly, two samples from the coronavirus isolation ward were found to be positive using RT-qPCR. The positive samples were collected from a patient's bed and the interior bed of an insulated room. No COVID-19 contamination was detected on hospital surface samples outside these areas.

Conclusion: This study identified a low rate of COVID-19 contamination on hospital surfaces and equipment in Fasa city. The findings suggest that the hospital environment could serve as a potential source of COVID-19 transmission, particularly among healthcare providers, visitors, and patients.

Keywords: Coronavirus Disease 2019, Molecular diagnosis, Contamination, Hospital environment

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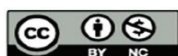
Introduction

Hospital infections are among the most significant causes of death worldwide, yet they can be effectively controlled through the

appropriate application of disinfectants and sanitizers (1, 2). Sterilization, disinfection, and decontamination constitute the main pillars of any infection control program. Cleaning remains the most critical measure for maintaining health and controlling the spread of diseases. The transmission of microorganisms can occur through direct contamination, vectors,

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individuals, equipment, animals, or animal products (3). This transmission is mitigated through various methods, including the use of disinfectants, sanitizers, and cleaners. The mechanisms of action, applications, and efficacy of different disinfectants vary significantly.

Given the importance of saving human lives during medical surgeries or other healthcare services, the use of disinfectants for critically ill patients is indispensable. Furthermore, disinfectants are essential for cleaning hospital environments and equipment (4, 5).

Pandemics of severe acute respiratory syndrome coronavirus (SARS-CoV) have profoundly impacted global healthcare and economies. Coronaviruses are a large family of viruses that can cause a broad range of diseases, from mild conditions, such as the common cold, to more severe illnesses, including Middle East respiratory syndrome (MERS) and severe acute respiratory syndrome (SARS) (6).

In 2019, a novel coronavirus causing coronavirus disease 2019 (COVID-19) was first identified in Wuhan, China. This new type of coronavirus, which had not previously been observed in humans, has since infected more than 704,753,890 individuals and caused 7,010,681 deaths globally to date. The number of COVID-19 infections continues to rise worldwide (7, 8).

Identifying COVID-19 transmission routes plays a crucial role in controlling the disease, although these routes have not been fully elucidated. COVID-19 is primarily transmitted between individuals through close physical contact and respiratory droplets (9). Even in modern, well-equipped hospitals in developed countries, the COVID-19 virus is likely present due to the admission of patients with diverse medical conditions and the existence of undetected sources of infection in such settings (10). Close contact in crowded environments, such as households, healthcare facilities, high-traffic areas, and residential zones, increases the

likelihood of COVID-19 transmission (11, 12).

The transmission of microorganisms from environmental surfaces to patients occurs predominantly through physical contact. Environmental surfaces are more likely to become contaminated with the COVID-19 virus during healthcare procedures involving infected patients (13, 14). Hospital surfaces and healthcare environments are typically categorized as medical equipment and general household-like areas. Surfaces of medical equipment and facilities, including sphygmomanometers, stethoscopes, hemodialysis machines, imaging devices, bed rails, bedside tables, mechanical ventilators, toilets, suction devices, and mattresses, are frequently contaminated with infectious agents. These contaminated surfaces facilitate the transmission of these agents, thereby contributing to the occurrence and spread of diseases (15). Therefore, surfaces in COVID-19 wards must be thoroughly cleaned and disinfected to prevent further virus transmission.

Several studies have evaluated the stability of the COVID-19 virus on various surfaces. Findings from these studies indicate that the virus remains viable and capable of transmission on different surfaces for varying durations (16). For instance, the COVID-19 virus may survive up to one day on fabric and wood, two days on glass, four days on steel and plastic, and seven days on the outer layer of surgical masks (16).

Accordingly, Hu et al. (2020) conducted a study in Nanjing, China, to evaluate the contamination of contact surfaces by asymptomatic individuals, identifying contaminated surfaces as a significant factor in disease transmission (17). Faridi et al. (2020) reported that all air samples collected at a distance of 2–5 meters from the beds of COVID-19 patients tested negative (18). Similarly, Ahmad and Ali's studies found SARS-CoV on surfaces and objects in patient rooms and toilets, highlighting that the virus could remain on various surfaces for several days (19, 20).



Rahmani Samani et al.'s findings at Shahrekord Hajar Hospital, Iran, indicated that 46.15% of surface samples and 26.66% of air samples tested positive for SARS-CoV-2 RNA. These contaminations were confined to areas in close contact with COVID-19 patients (21). Other studies have reported varying prevalence rates of the virus, including 8% in Thailand (22), 11.7% in hospital rooms in Shanghai, China (23), 83% in European hospital air samples (24), 6.6% (45/682) in hospital rooms and salivary viral loads, 41.2% in indoor air, and 32% in swab samples in Shiraz, Iran (25), and 74.7% (239/320) on personal protective equipment in Shanghai, China (26).

The results of Wu et al.'s study showed that a significant proportion of touchable hospital surfaces were highly contaminated, emphasizing that hospital environments are a critical pathway for disease transmission. These findings underscore the necessity of strictly implementing environmental health principles to mitigate the spread of disease (3).

While numerous studies have been conducted in this field, no report on the current situation in the studied area is available. Considering the extensive impact of the coronavirus, which has affected a substantial portion of the world and particularly Iran, continuous and systematic monitoring should remain a priority for researchers and public health officials.

Given the still-unresolved aspects of COVID-19 transmission patterns, coupled with recommendations from studies conducted in other countries for further investigation, this study was undertaken. The primary objective was to identify potential risk factors and evaluate the extent of environmental contamination by COVID-19 patients. The study aimed to inform health protocols, optimize the frequency of disinfection at critical points, and, where necessary, improve healthcare management and infection control guidelines. Specifically, this study sought to examine the presence of

the COVID-19 virus on high-risk surfaces at Valiasr Hospital in Fasa, Iran, using real-time quantitative PCR (RT-qPCR).

Materials and Methods

Sampling area

This cross-sectional descriptive and analytical study was conducted in April 2020 at Fasa Valiasr Hospital, affiliated with Fasa University of Medical Sciences. Fasa is a city and the capital of Fasa County, Fars Province, Iran, located at an altitude of 1,150 m above sea level. With a population of approximately 205,187, Fasa is one of the most populous cities in the Fars province. Valiasr Hospital, which has a capacity of 251 beds, includes several medical wards, such as the emergency ward (e.g., supervised emergency, hospitalized emergency, and pediatric emergency), operating rooms, anesthesia, CCU, post-CCU, general gynecological surgery, internal medicine, obstetrics and gynecology, pediatrics, men's surgery, internal surgery, heart surgery, intensive care units (ICU), and neonatal intensive care units (NICU). The hospital also houses special care wards and various paraclinical units, including the Valiasr clinic, laboratory, radiology, sonography, CT scan, mammography, endoscopy, ECG, exercise testing, echocardiography, angiography, and pharmacy.

A total of 142 samples were collected from various wards, environmental surfaces, and equipment, including the COVID-19 ward, internal isolation ward, surgical ICU, CCU, emergency department, radiology unit, maternity ward, operating rooms, nursing stations, and ambulances used to transport COVID-19 patients. In these wards, samples were obtained from high-risk and sensitive areas, such as building surfaces (e.g., floors and walls), toilets, restrooms, and the COVID-19 and other wards. Additionally, samples were collected from medical equipment, including patient beds, oxygen devices, clothing and gowns of patients and personnel, endoscopes, CT scanners, and



other items in contact with COVID-19 patients.

Control samples were also tested, collected from areas and surfaces not exposed to COVID-19 patients, such as hospital parking lots and office departments. Sampling was performed on non-duplicated surfaces measuring 700 cm² (25×30 cm) in high-touch areas of critical wards where COVID-19 patients were hospitalized or transported. This involved placing flexible swabs, immersed in viral transport medium (VTM) (Easy Swab, Komed, SungNam, Korea), on high-touch surfaces to detect SARS-CoV-2 RNA (7, 18).

SARS-CoV-2 detection using RT-qPCR technique

All samples collected from surfaces and equipment were promptly referred to the COVID-19 diagnostic laboratory. RNA was extracted from the VTM medium using a QIAamp Viral RNA Mini Kit (QIAGEN, Hilden, Germany), following the manufacturer's instructions. The presence of the COVID-19 virus was analyzed using the RT-qPCR technique (QIAquant 96 5plex), in accordance with the manufacturer's protocol (Qiagen, MD, USA). A 20 µL aliquot of the extracted RNA was added to wells pre-filled with a reagent mix (30 µL). Each well was sealed, centrifuged at 2,000 rpm for 10 seconds, and placed into the QIAquant 96 5plex (Qiagen, MD, USA). Thermal cycling was performed at 50°C for 30 minutes for reverse transcription, followed by

95°C for 1 minute, and then 45 cycles of 95°C for 15 seconds and 60°C for 31 seconds. A cycle threshold (CT) value ≤40 was interpreted as positive (27).

Data analysis

After data collection, the information was analyzed using SPSS version 22 software. T-tests, analysis of variance, and chi-square tests were performed, with a significance level of 0.05 applied to all analyses.

Results

In this study, 142 swab samples were collected from environmental surfaces and equipment in various wards of Fasa Valiasr Hospital. The persistence and presence of the COVID-19 virus on these surfaces and equipment were examined. Table 1 presents the results of RT-qPCR testing for COVID-19 detection in different wards of Fasa Valiasr Hospital. Out of the 142 samples, four tested positive, including two samples from the COVID-19 ward and two from the internal isolation ward. Tables 2 and 3 summarize the results of RT-qPCR testing for COVID-19 on hospital equipment and environmental surfaces across different wards. The positive samples were obtained from a COVID-19 patient's bed and an isolation ward bed. The results revealed that all samples collected from building surfaces, tested using PCR, showed no contamination with COVID-19.

Table 1. Result of RT-PCR for COVID-19 detection in different wards of Fasa Valiasr Hospital

Hospital wards		No. of sample	No. of positive samples
1	COVID-19 ward	20	2
2	Emergency ward	8	0
3	Internal insulated ward	3	2
4	Internal ward	4	0
5	ICU surgical ward	9	0
6	CCU ward	5	0
7	Radiology ward	5	0
8	OB ward	5	0
9	COVID-19 Lab	6	0
10	surgery room	4	0



Table 2. Result of RT-PCR for COVID-19 in Hospital Equipment

Equipment	No. of sample	RT-PCR for COVID-19 result
1 COVID-19 ambu bag	2	Negative
2 Ventilator	2	Negative
3 Stethoscope	2	Negative
4 Central suction	1	Negative
5 COVID-19 patient bed	2	Positive
6 CT scan machine	1	Negative
7 Bed CT scan	2	Negative
8 Radiology keyboard	1	Negative
9 surgery room monitor	1	Negative
10 COVID-19 patient ambulance suction	2	Negative
11 COVID-19 patient ambulance Bed	2	Negative
12 Interior insulated room bed	3	Positive
13 CCU monitor	1	Negative
14 COVID-19 ward personnel uniform	2	Negative
15 COVID-19 Lab staff uniforms	2	Negative
16 OB ward bed	2	Negative
17 CCU bed	2	Negative
18 surgery room bed	2	Negative
19 ICU patient clothing	2	Negative
20 Surgical ICU suction	1	Negative
21 Emergency box	2	Negative
22 COVID-19 patient room chair	2	Negative
23 COVID-19 section file chart	1	Negative
24 COVID-19 Food trolley table	2	Negative
25 DC Shock Emergency ward	1	Negative

Discussion

Various studies have proposed specific modes of transmission for this virus, including airborne transmission and contamination of surfaces. This virus can survive for hours in aerosols and may be transmitted indirectly through the contamination of hospital equipment surfaces (28, 29). In general, effective emergency management of health services requires a rapid assessment of the current epidemiological situation to implement preventive and control measures. However, few studies on the transmissibility of SARS-CoV-2 from contaminated surfaces and equipment have been conducted in Iran (30, 31). In the present study, all samples collected from building surfaces and medical equipment in different wards of Fasa Valiasr Hospital—one of the primary hospitalization centers for COVID-19 patients—were examined for the presence of

the SARS-CoV-2 virus. These surfaces included the walls and chairs of COVID-19 patient rooms, the COVID-19 ward, the telephone in the COVID-19 ward, nursing stations, surgical ICU walls, maternity ward phones, separating curtains in the COVID-19 ward, wardrobes in the COVID-19 ward, ventilators, CT scan room beds, Ambu bags, suction devices, surgical room monitors, COVID-19 patient ambulances, beds in internal isolation rooms, CCU beds, surgical room beds, and staff uniforms in areas vulnerable to coronavirus contamination. The results of this study showed that the hospital environment is likely to be contaminated with SARS-CoV-2 during the provision of healthcare services to patients.

Among the 142 samples obtained, four (5.79%) from the internal isolation wards and COVID-19 wards tested positive by PCR.



Table 3. Result of RT-PCR for COVID-19 in environmental surfaces in different wards

Location, surface		No. of sample	RT-PCR for COVID-19
1	COVID-19 room wall	1	Negative
2	COVID-19 room chair	1	Negative
3	COVID-19 health service	2	Negative
4	COVID-19 section phone	1	Negative
5	COVID-19 computer keyboard	1	Negative
6	COVID-19 Nursing Station	2	Negative
7	Emergency Nursing Station	2	Negative
8	Emergency ward phone	1	Negative
9	Internal health service	2	Negative
10	Wall surgery ICU	1	Negative
11	Surgical clothing ICU patient	1	Negative
12	Surgical ICU health service	1	Negative
13	Surgical ICU wardrobe	1	Negative
14	Surgery ICU Phone	1	Negative
15	CCU Nursing Station	2	Negative
16	ICU wardrobe	2	Negative
17	COVID-19 part separator curtain	1	Negative
18	Maternity ward phone	1	Negative
19	COVID-19 Lab Glasses	1	Negative
20	Knobs in COVID-19 Lab	1	Negative
21	Administration Area	2	Negative
22	Parking Lot	2	Negative

The COVID-19-positive samples were collected from patient beds in the COVID-19 ward and internal isolation rooms. These results suggest that inpatient wards for COVID-19 patients and internal isolation areas are particularly vulnerable to coronavirus contamination. It appears that the COVID-19 ward has been a potential hotspot for the spread of the virus, especially during the initial stages of the pandemic, likely due to overcrowding.

This finding aligns with previous studies on the persistence of the virus on dry surfaces and equipment. According to the literature, coronaviruses can survive for up to nine days on inanimate surfaces at room temperature, making incomplete or infrequent disinfection processes significant contributors to viral persistence and transmission. The presence of SARS-CoV-2 on surfaces is a major concern.

The results of this study emphasize that surfaces with high usage and congestion can

serve as reservoirs for the accumulation and spread of infectious agents. For instance, Wang et al. (2020) demonstrated that a proper and continuous disinfection process, along with COVID-19 prevention training for hospital staff, is crucial for effective disease management (13). Similarly, Faridi et al. (2020) reported that all samples collected from respiratory air at a distance of 2–5 meters from patients' beds tested negative for the COVID-19 virus (18). Additionally, Lee et al. (2020) found that 16% of samples were infected with the virus prior to disinfection (32).

In a related study, Guo et al. (2020) in Wuhan, China, reported widespread contamination, including on floors, computer mice, trash cans, and bed railings (19). Reported rates of contamination on hospital surfaces vary significantly: 8% in Thailand (22), 11.7% in hospital rooms in Shanghai, China (23), 83% in air samples from European hospitals (24), 6.6%



in hospital rooms and salivary viral loads in Shiraz, Iran, and 41.2% in indoor air with 32% in swab samples (25). Additionally, contamination rates as high as 74.7% (239/320) were observed on personal protective equipment in Shanghai, China (26).

As mentioned earlier, the percentage of positive COVID-19 samples in Ye et al.'s study was higher than in the present study. This discrepancy could be attributed to variations in sampling methods, ventilation system performance, and disinfection protocols across different study locations.

The findings of this study, together with other studies from various countries, indicate that the coronavirus exhibits high persistence in medical environments, particularly in inpatient wards for COVID-19 patients and specialized care units. The spread of the virus on surfaces and equipment in these settings is notably high. Interestingly, negative results were observed for surfaces related to treatment staff, such as nursing station telephones, chairs, computer keyboards, and staff uniforms. This suggests that health protocols are being effectively followed by the hospital staff.

The difference in the number of positive COVID-19 samples between the present study and other studies could also be due to differences in sampling locations. For example, some studies sampled patient masks and tables in front of patients, which were not sampled in the current study. Instead, this study identified patient beds in the COVID-19 ward and internal isolation rooms as the primary positive samples. The risk of contamination of surfaces and equipment with SARS-CoV-2 should be regularly emphasized to healthcare staff to prevent further spread of the virus. Providing standard guidelines and closely monitoring infection control practices should be integral to COVID-19 prevention strategies. This study faced certain limitations, including the relatively small number of samples, the short duration of the study, and restrictions on sampling.

Conclusion

The findings of this study indicate that the internal isolation wards and COVID-19 departments of the hospital may serve as potential sources of virus transmission, although the contamination rates observed herein were low. By completely segregating COVID-19 departments from other areas, healthcare services in non-COVID-19 departments can proceed as per pre-pandemic procedures. This study demonstrates that implementing robust disinfection processes for contaminated surfaces and equipment, along with infection prevention training, can effectively reduce the spread of the COVID-19 virus and interrupt its transmission cycle across hospital departments.

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Conflict of interest

The authors declare no conflicts of interest.

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Ethical Considerations

This article does not involve any studies on animals conducted by the authors.

Code of Ethics

The ethical code for this study is IR.FUMS.REC.1399.090.

Authors' Contribution

A.G. and N.A. conceptualized the study. A.G., N.A., Y.M., and M.D. conducted the study. A.G., N.A., Y.M., and A.G. drafted the manuscript. Z.M. and R.H. approved the study design. All authors reviewed and finalized the manuscript.



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