Clin. Lab. 2021;67:XXX-XXX

ORIGINAL ARTICLE

Environmental Assessment of SARS-CoV-2 for Internal Quality Management in a Clinical Laboratory

Yingping Wu [*], Zhiqiang Zhu [*], Wei Xu, Wenzi Bi, Jianyong Wu, Jintao He, Lili Niu, Xiaoping Xia

[*] *Yingping Wu and Zhiqiang Zhu are joint first authors*
Department of Clinical Laboratory, Fourth Affiliated Hospital, School of Medicine, Zhejiang University, Jinhua, China

SUMMARY

Background: **As an emerging infectious disease, coronavirus disease 2019 (COVID-19) exhibits occult infection, which might cause difficulties in controlling disease spread. The possibility of aerosol transmission in a relatively closed environment contributes to the high infectivity of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in hospitals. This study presents an environmental surveillance system for SARS-CoV-2 that is suitable for a clinical laboratory and may also lead to further assessment of infection prevention programs in different departments in hospitals.**
Methods: **The study was performed in a SARS-CoV-2 RNA laboratory involved in the diagnosis of COVID-19 in China. Reverse transcription-polymerase chain reaction (RT-PCR) assays were used to detect viral pathogens. Standard operating procedures (SOPs) for monitoring infectious pathogens were developed in this study.**
Results: **In total, more than 180 air and surface samples were tested for SARS-CoV-2 to determine whether the virus was present at the airborne and particle level. The employed molecular method effectively identified environmental contamination.**
Conclusions**: Our study suggests that regular environmental surveillance is critical in a clinical PCR laboratory. The presented strategy could also be used for monitoring and surveillance in negative pressure wards and clinics in hospitals to prevent hospital-acquired infections.**
(Clin. Lab. 2021;67:xx-xx. DOI: 10.7754/Clin.Lab.2021.200926)

Correspondence:
Yingping Wu
Department of Clinical Laboratory
Fourth Affiliated Hospital
School of Medicine
Zhejiang University
Jinhua
Zhejiang Province
China
Phone: +86 13858118992
Email: yingpingwu@zju.edu.cn

KEY WORDS

SARS-CoV-2, laboratory-associated infections, environmental surveillance, environmental contamination

INTRODUCTION

Among the hazards to healthcare personnel, hospital-acquired infections (HAIs), in particular laboratory-associated infections, may occur if inadequate protective measures are used [1]. Controlling air quality is crucial to reduce the dissemination of aerosolized biological particles, while appropriate hygiene and disinfection procedures remove respiratory pathogens from surfaces in hospitals [2]. A wide range of respiratory viruses are known to infect humans; these viruses not only cause recurrent epidemics of respiratory tract infections in humans that can lead to severe disease but also mutate to

Manuscript accepted March 9, 2021

be more efficiently transmitted among humans [3]. Human coronaviruses (HCoVs) have resulted in recent respiratory disease outbreaks; the responsible pathogens include severe acute respiratory syndrome coronavirus (SARS-CoV), Middle East respiratory syndrome coronavirus (MERS-CoV) and severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) [4,5]. Recently, SARS-CoV-2 rapidly spread worldwide; in Wuhan, China, 1,716 medical staff members have become infected, and 5 have died in early period of 2020 [6]. Based on the New Coronavirus Pneumonia Diagnosis and Treatment Plan guidelines (6th edition) in China, the main transmission route of SARS-CoV-2 are via respiratory droplets and direct contact. Recently, it has become clear that it is necessary to pay attention to aerosol transmission and contact spread of cold storage chain system in China, which has been found to be relatively common in hospitals [7]. Therefore, the management of HAIs should involve multiple departments that are at risk in hospitals.

As SARS-CoV-2 RNA testing of clinical specimens has increased, biosafety risks in clinical laboratories have also emerged. The clinical polymerase chain reaction (PCR) laboratory is part of the Clinical Laboratory Department, and the daily duty of the clinical PCR laboratory is to detect the DNA/RNA of infectious pathogens, including bacteria and viruses. Therefore, there is an urgent need for evaluation of the risk of transmission of environmental pathogens in a biosafety level 2 (BSL-2) laboratory. Improved biosafety of indoor air is one goal of internal quality control (IQC) in clinical laboratories [8].

Currently, there are no simple validated technologies that can be used to collect and identify viruses in the air rapidly and easily. At present, the most common techniques for collecting pathogens from the air are the airflow and liquid models [9,10]. These systems are complex, and their efficiency has not been thoroughly evaluated. It would be desirable to have a method that not only collects and identifies pathogens, but also eliminates them [11,12]. To evaluate the risk of SARS-CoV-2 spreading from clinical specimens in a clinical laboratory, we developed an operational method of performing regular monitoring for SARS-CoV-2 RNA by sampling the environment and the air, which are key components of IQC in our laboratory. To the best of our knowledge, this is the first report of a method of identifying SARS-CoV-2 contamination in clinical laboratories.

In this study, we established a methodical strategy to effectively control viral contamination. In addition, we describe the importance of a clinical laboratory quality management program that involves monitoring air and surface contamination. It is clear that the COVID-19 pandemic will continue to spread worldwide via uncertain pathways. It is very important to focus on specific areas at high risk of contamination and implement effective decontamination measures. Finally, we provide several suggestions to improve quality control programs in clinical laboratories and make recommendations regarding the generalization of this SOP to other hospital wards to facilitate nosocomial infection management.

MATERIALS AND METHODS

Study design

In this study, all of the environmental samples were collected at the clinical laboratory of the Fourth Affiliated Hospital, Zhejiang University School of Medicine in Zhejiang Province of China, a hospital designated for the diagnosis and treatment of COVID-19 patients in February 2020. Air sampling was performed in 8 functional laboratories of the Clinical Laboratory Department: the clinical PCR laboratory, clinical biochemistry laboratory, clinical immunology laboratory, clinical microbiology laboratory, clinical hematology laboratory, body fluid laboratory, sample preparation area, and clinical fever laboratory. Sterile normal saline was used as a negative control. Most of these functional laboratories carry out sample testing services for patients with confirmed COVID-19. The Clinical Laboratory Department is a China National Accreditation Service for Conformity Assessment (CNAS)-certified laboratory, and all medical staff involved in SARS-CoV-2 detection were authorized and qualified professionals who obtained official permission to conduct these tests in China. The above functional laboratories were BSL-2 laboratories, with the exception of the PCR laboratory used for SARS-CoV-2 detection, which has a higher biosafety protection level of 3 (Table 1). Therefore, the experimental design for air and surface sampling was not identical in all laboratories.

Each laboratory was sampled 3 times per week to determine the contamination status after disinfection procedures. Therefore, a total of 72 samples and 2 other negative controls, were analyzed for airborne contaminant levels. Additionally, to reveal the patterns in which potential airborne contaminants settled on the surfaces in these laboratories, 120 surface samples were collected from 40 predetermined sampling locations at three different times.

Environmental sampling procedure

The sampling procedure was developed according to current Chinese hospital disinfection hygiene standards (GB 15982-2012), although a different dilution ratio was used. Air sampling plates with a 9 cm diameter that contained 2 mL of 0.9% sodium chloride solution were placed in the departments mentioned above for 24 hours. To sample object surfaces, sterile swabs moistened with saline were used to swab an approximately 10 cm x 10 cm area. Then, the swabs were placed individually into sterile tubes containing 2 mL of 0.9% sodium chloride solution. All collected samples were stored at -70°C until the downstream analysis was performed. Environmental surveillance was also performed three times each week after disinfection procedures.

Fluorescent RT-PCR assessment against SARS-CoV-2

RT-PCR was performed using a Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit (detection of 3 genes) (Liferiver, Shanghai, China), according to the manufacturer's instructions. Primers and probes used for multiplex nucleic acid amplification were designed to detect SARS-CoV-2 based on conserved regions in three genes (*RdRP*, *N*, and *E*). The one-step PCR protocol included one cycle at 45°C for 10 minutes and 95°C for 3 minutes, followed by 45 cycles at 95°C for 15 seconds and 58°C for 30 seconds, and single-point fluorescence detection at 58°C. The PCR system was operated according to the protocol in the manual. The crossing point (Cp) values for the relative quantification of gene expression were used to determine the viral load of SARS-CoV-2. As described in our previous report [13], samples containing the RdRP gene with Cp values ≤ 43.0 were considered positive for SARS-CoV-2 RNA. Samples containing the RdRP gene with Cp values of 0 or in which the *N* or *E* gene detected alone were considered negative for SARS-CoV-2 but positive for other coronaviruses.

Decontamination of the lab environment

The presence of SARS-CoV-2 in laboratories can cause widespread contamination of the air, working surfaces, and equipment and can affect the health of personnel. Based on the current Chinese standards for disinfection techniques in healthcare settings (WS/T 367-2012, China) and the characteristics of SARS-CoV-2, we used 75% ethanol, 1,000 mg/L hypochlorite solution, UV irradiation, HEPA filters, negative-pressure systems, and ionizing air and fresh air systems in different combinations to achieve decontamination. All environmental monitoring was carried out for three weeks.

As described in a previous study by our team [8], the standard procedure for laboratory decontamination that was employed once per day was as follows: (1) spraying a 75% ethyl alcohol solution into the air before cleaning the room; (2) wiping objects and equipment in the rooms with a hypochlorite solution to remove settled particles; (3) wiping equipment, including disassembled centrifuge rotors or PCR instruments, with a 75% ethyl alcohol solution; (4) increasing ventilation frequency; and (5) using in-room air cleaners, including portable or ceiling-mounted units with ionizing air, UV lights or ventilation. Separate sets of cleaning tools were used for each room and were not mixed. These steps were carried out twice each day after contamination was identified, and environmental samples were collected after decontamination.

Statistical analysis

Statistical analysis was performed using SPSS Statistics version 23 (IBM). A p-value of ≤ 0.05 was considered significant.

RESULTS

Biosafety protection measures in the clinical laboratory

The stability of SARS-CoV-2 is similar to that of SARS-CoV in aerosols and on surfaces; SARS-CoV-2 was found to exist in aerosols for hours and on surfaces for days [14]. SARS-CoV-2 is also highly stable at room temperature, at normal relative humidity, and on smooth surfaces [17]. During sampling in this study, the mean temperature and relative humidity were 20 - 23°C and 40 - 65%, respectively, and these conditions are suitable for the survival of SARS-CoV-2 (Table 1). Moreover, these relative humidity (70 - 80%) and temperature (12 - 15°C) conditions in laboratories promote bioaerosol survival and higher emission levels. However, SARS-CoV-2 is also susceptible to standard disinfection methods [15]. Our experience shows that it is essential to disinfect environmental surfaces, including sample storage refrigerators, and to record the humidity and temperature every day for effective monitoring. In laboratories, UV irradiation is effective at reducing the transmission of microorganisms by damaging their DNA and thereby rendering them noninfectious [16]. The ability of UV irradiation to kill pathogens depends on the relative humidity of the environment, and some studies have shown that the effectiveness of UV irradiation with regard to killing or inactivating microorganisms decreases when the relative humidity in a room exceeds 60%. UV lamps were mounted on the ceiling and placed on removable carts; these UV lamps are standard devices in clinical PCR laboratories in China. The use of UV light needs to be supported by the maintenance of an appropriate level of humidity. Furthermore, plasma air disinfectors are also widely used in our laboratories and are effective for disinfecting the air.

The transport of infectious droplets through the air can lead to an outbreak in an enclosed space. In the relatively closed environment of a laboratory, a ventilation system is essential.

Due to the high risk of infection associated with testing samples for the presence of SARS-CoV-2 RNA, the clinical PCR laboratories are the only areas with negative-pressure systems to increase the level of protection. Overall, these systems ensure greater protection and less risk of laboratory contamination. However, low biosafety levels may result in greater risk in the laboratory.

Sampling types and numbers show the diversity distribution in the clinical laboratory

It was noted that in a nosocomial SARS outbreak, an inoperative return air outlet could enhance the spread of aerosols in the same hospital ward [17]. When appropriate disinfection measures are taken, airborne contamination is preventable. Individual laboratories perform different tasks that are associated with various levels of risk of exposure; the level of personal protection required is adapted to that risk (Table 2). The ability of SARS-CoV and MERS-CoV to survive on dry surfaces

Table 1. Functional labs in the clinical laboratory.

Functional lab	Professional category	Approximate area size (m^2)	Existing biosafety facilities	Personal protective	Temperature and humidity values
PCR lab a. First Room: Reagent preparation area b. Second Room: Sample preparation area c. Third Room: Gene amplification area d. Fourth Room: Amplification product analysis area	nucleic acid detection for pathogens	80	negative-pressure system, ceiling UV light, removable UV car, biosafety cabinet, ventilation system	level-3	21 - 23°C, 40 - 50%
Clinical biochemistry lab	biochemistry tests	200	ventilation system	level-2	21 - 23°C, 40 - 50%
Clinical immunology lab	immunology tests	150	ventilation system	level-2	21 - 23°C, 40 - 50%
Clinical microbiology lab	microbial culture and identification (not including virus)	150	ventilation system, plasma air-sterilizing machine	level-2	21 - 23°C, 40 - 50%
Clinical hematology lab	complete blood count, coagulation function routine	100	ventilation system	level-2	21 - 23°C, 40 - 50%
Body fluid lab	routine urine test, stool-routine test, cell count and classification of sterile CSF, hydrothorax, and ascitic fluid	60	ventilation system, plasma air-sterilizing machine	level-2	21 - 23°C, 40 - 50%
Fever clinical lab	fever screening for hematology, body fluid, respiratory pathogen antigens screening	30	plasma air-sterilizing machine	level-2	20 - 22°C, 50 - 60%
Sample preparation area	classification and centrifugation of samples	60	ventilation system, plasma air-sterilizing machine, biosafety cabinet	level-2	21 - 23°C, 40 - 50%
Staff office	clean zone as negative control	100	N/A	N/A	15 - 20°C, 55 - 65%

Level-3, biosafety protective level 3 includes N95 respirators, 3M protective clothing, medical gloves, and protective goggles, medical positive pressure headgear. Level-2, biosafety protective level 2 includes N95 respirators, contagion gown, medical gloves, and protective goggles. m^2 - square meter.

has not been verified [18]. Therefore, we considered the importance of contaminated surfaces in laboratories and performed surface sampling for SARS-CoV-2 (Table 3).

Because sampling should reflect the actual environmental state, it is preferable to sample after disinfection but not during working hours. The first sampling results showed that the amplification curves of the surface samples from the clinical biochemistry laboratory were slightly abnormal. Targeting these key sites of contamination would improve the disinfection of this laboratory. Fortunately, contamination on hard surfaces is more easily removed than contamination in the air (Figure 1). Although contamination was detected in the first and

Table 2. Aerosol sampling for SARS-CoV-2 in different locations.

Location (n = 24) [a]	Sampling site	PCR results (Cp values) [*]		
		1st	2nd	3rd
PCR lab (n = 10) [a]	operation desk (first room)	0	0	0
	biosafety cabinet (level B2) 1 (second room)	<u>40</u>	0	0
	biosafety cabinet (level B2) 2 (second room)	0	0	0
	nucleic acid extraction instrument (second room)	0	0	0
	operation desk 1 (second room)	0	0	0
	operation desk 2 (second room)	0	0	0
	buffer ward (second room)	0	0	0
	operation desk 1 (third room)	0	0	0
	operation desk 2 (third room)	0	0	0
	operation desk (fourth room)	0	0	0
Clinical microbiology lab (n = 2) [a]	biosafety cabinet (level B1)	<u>40</u>	0	0
	operation desk	0	0	0
Sample preparation area (n = 3) [a]	biosafety cabinet (level A2) 1	0	0	0
	biosafety cabinet (level A2) 2	0	0	0
	operation desk	0	0	0
Body fluid lab (n = 1) [a]	operation desk	0	0	0
Clinical fever lab (n = 4) [a]	operation desk	0	0	0
	biosafety cabinet (level A2)	0	<u>40</u>	0
	nasopharyngeal swab collection window 1	0	0	0
	nasopharyngeal swab collection window 2	0	0	0
Clinical immunology lab (n = 1) [a]	operation desk	0	0	0
Clinical biochemistry lab (n = 1) [a]	operation desk	0	0	0
Clinical hematology lab (n = 1) [a]	operation desk	0	0	0
Staff office (n = 1) [a]	office desk	0	0	0

[a] - Numbers of tests.
[*] - Cp values of 0 means a non-amplified result. Underlined numbers indicate positive amplification curves.

second sampling periods, no contamination with SARS-CoV-2 was detected in the third sampling period (Figure 1). Regular monitoring can allow early identification and timely disinfection. In this study, we monitored the short-term implementation of the decontamination procedures for three weeks after the disinfection protocols were established (Table 2, Table 3). Although an outbreak of COVID-19 has not occurred in the clinical laboratory, despite slight amplification observed in the first and second sampling periods, monitoring for viruses, such as hepatitis B virus (HBV) and influenza virus, by PCR has continued routinely once every three months for four years.

Viral load of SARS-CoV-2 in the surrounding environment identified by RT-PCR

In general, for indoor environments, a total bacterial count is generally specified in indoor air quality standards [19], the conventional method of bacterial culture is not suitable for viruses. In this study, PCR was used to detect the presence of SARS-CoV-2.

The decontamination procedures were repeated twice every day. Over this period, 192 samples were collected at three time points. The Cp value was used as an index of SARS-CoV-2 RNA contamination and showed that the contamination decreased over the two-week period (Figure 1). The Cp value is inversely proportional to the

Table 3. Surface sampling for SARS-CoV-2 in different locations.

Location (n = 40) [a]	Sampling site	PCR results (Cp values)		
		1st	2nd	3rd
PCR lab (n = 19) [a]	operation desk (first room)	0	0	0
	biosafety cabinet 1 (second room)	0	0	0
	biosafety cabinet 2 (second room)	0	0	0
	biosafety cabinet 3 (second room)	0	0	0
	nucleic acid extraction instrument inner 1 (second room)	0	0	0
	nucleic acid extraction instrument inner 2 (second room)	0	0	0
	nucleic acid extraction instrument outer (second room)	0	0	0
	regent refrigerator (second room)	0	0	0
	transfer window I inner (second room)	0	0	0
	transfer window I outer (second room)	0	0	0
	operation desk (second room)	0	0	0
	transfer window II (second room)	0	0	0
	inside door (second room)	0	<u>40</u>	0
	outside door (second room)	0	0	0
	buffer ward desk (second room)	0	0	0
	operation desk 1 (third room)	0	0	0
	operation desk 2 (third room)	0	0	0
	transfer window 1 (fourth room)	0	0	0
	transfer window 2 (fourth room)	0	0	0
Sample preparation area (n = 4) [a]	biosafety cabinet inner 1	0	0	0
	biosafety cabinet inner 2	0	0	0
	operation desk 1	0	0	0
	operation desk 2	0	0	0
Clinical microbiology lab (n = 3) [a]	biosafety cabinet 1	<u>40</u>	<u>40</u>	0
	biosafety cabinet 2	0	0	0
	operation desk	0	0	0
Clinical hematology lab (n = 3) [a]	operation desk 1	0	0	0
	operation desk 2	0	0	0
Body fluid lab (n = 2) [a]	operation desk 1	0	0	0
	operation desk 2	0	0	0
Fever clinical lab (n = 4) [a]	biosafety cabinet 1	0	0	0
	biosafety cabinet 2	0	0	0
	operation desk	0	0	0
	nasopharyngeal swab collection window	0	0	0
Clinical immunology lab (n = 2) [a]	operation desk 1	0	0	0
	operation desk 2	0	0	0
Clinical biochemistry lab (n = 2) [a]	operation desk 1	0	0	0
	operation desk 2	0	0	0
Clinical hematology lab (n = 1) [a]	operation desk	0	0	0
Staff office (n = 1) [a]	office desk	0	0	0

[a] - Numbers of tests. [*] - Cp values of 0 means a non-amplified result. Underlined numbers indicate positive amplification curves.

Table 4. Assessment of indoor environmental quality in isolation wards (n = 35).

Category	Sites	Sample type	RT-PCR results
Patient areas			
Negative pressure wards	**isolation ward of room no. 2**	**A/S**	**negative**
	buffer ward of isolation ward room no. 2	**A/S**	**negative**
	toilet room of isolation ward room no. 2	**S**	**negative**
	doorknob of isolation ward room no. 2	**S**	**negative**
	isolation ward of room no. 3	**A/S**	**negative**
	buffer ward of isolation ward room no. 3	**A/S**	**negative**
	toilet room of isolation ward room no. 3	**S**	**negative**
	doorknob of isolation ward room no. 3	**S**	**negative**
	wards of no. 13 - 15	**A/S**	**negative**
	wards of no. 16 - 17	**A/S**	**negative**
	wards of no. 16 - 17	**A/S**	**negative**
Semi-contaminated area			
	monitor	**S**	**negative**
	treatment vehicle	**S**	**negative**
	digital radiography machine	**S**	**negative**
	biohazard waste room	**S**	**negative**
	treatment room	**S**	**negative**
Medical staff areas			
	nurse station	**S**	**negative**
	buffer ward	**S**	**negative**

A - aerosol sampling, S - surface sampling, n - numbers of tests.

concentration of the target gene. A Cp value of 0 indicates non-amplification of the target gene. The results in Figure 1 show a low risk of SARS-CoV-2 exposure, even though six sites separately had weak amplification in the first and second environmental samplings. Based on the manual analysis, we considered the contamination to be due to other coronaviruses, not SARS-CoV-2. Air monitoring results showed weak positive amplification in the biosafety cabinet of the PCR laboratory (Figure 1), clinical microbiology laboratory, and clinical fever laboratory. Surface monitoring results showed slight contamination on the biosafety cabinet of the clinical microbiology laboratory, handles in the PCR laboratory, and the work bench of the biosafety cabinet in the clinical microbiology laboratory (twice). Moreover, daily work in the PCR and clinical microbiology laboratories involve the identification and culturing of microorganisms, and the pathogen load may be higher in these laboratories than in other laboratories because coronaviruses are ubiquitous.

After the disinfection measures were implemented, no amplification was observed in the samples from the third sampling. Clearly, these monitoring and decontamination measures were effective. Furthermore, the results of the indoor environmental assessment in isolation wards were negative in 35 key locations, which could be valuable for monitoring and preventing the occurrence of nosocomial infections (Table 4).

DISCUSSION

Some studies have focused on environmental monitoring [20], including cold storage chain systems in China; therefore, monitoring is important for managing patients and controlling the infectious transmission of SARS-CoV-2 [21]. There is a risk of nosocomial infection in hospitals, with a 0.77% positive detection rate on the surfaces in the isolation ward housing intensive care patients and a 3.57% positive detection rate in the air. Clinical laboratories are high-risk departments in hospitals because specimens from patients with COVID-19 must be tested in a relatively closed area. The objective of this study was to identify environmental SARS-CoV-2 contamination in our clinical laboratory and to determine its concentration and distribution. Various factors,

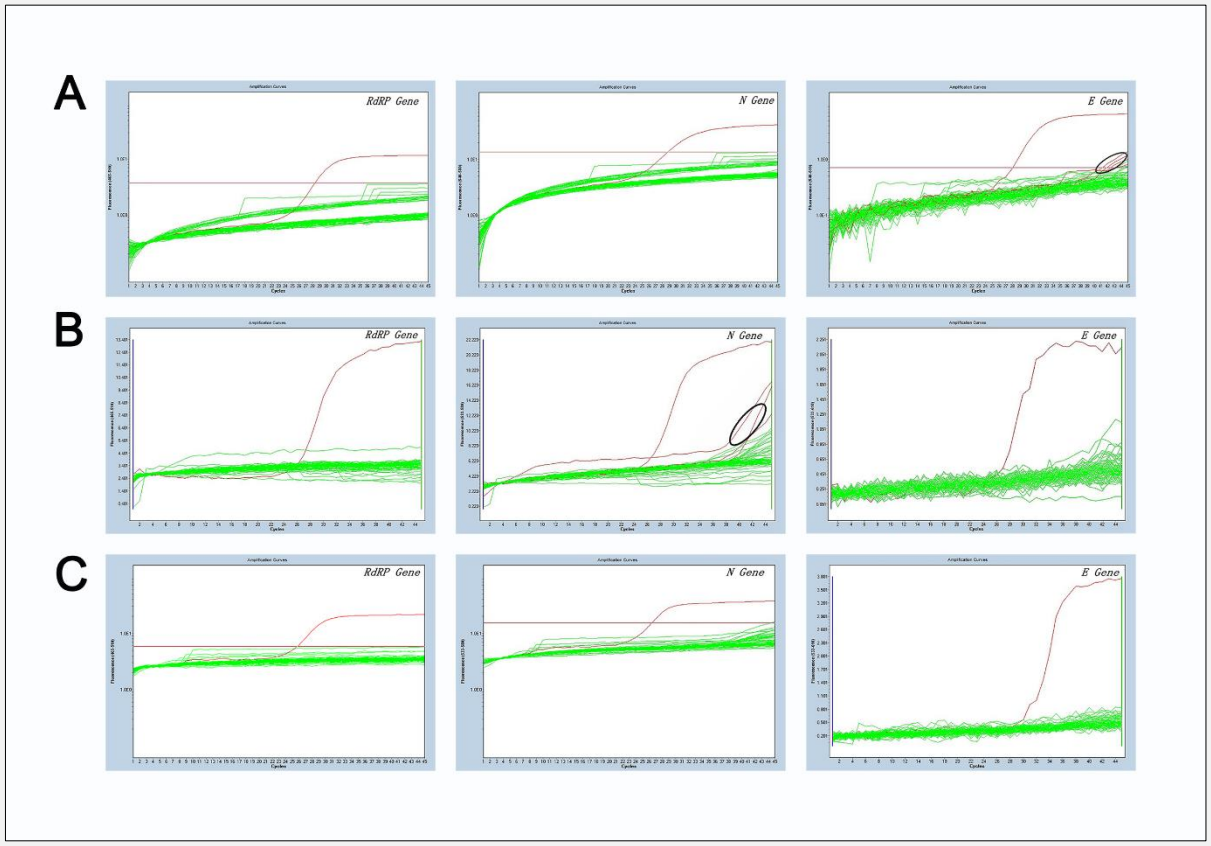

Figure 1. Real-time PCR assays were performed three times for air and surface monitoring in our clinical laboratory.

The first (A), second (B), and third (C) samplings were tested for the presence of three viral genes, namely, *RdRP*, *N*, and *E*. Amplification of the E gene was observed in the first samples at three sites, while amplification of the *N* gene was found in the second samples. No contamination with SARS-CoV-2 was detected in the third set of samples. Positive amplification is labeled with an oval. The Y-axis represents the number of cycles, and the X-axis represents the amount of fluorescence.

including the relative humidity, temperature, intensity of UV, ventilation frequency, and disinfectant use, have been verified to influence environmental contamination (Table 1). In this study, we have shown that tighter control of environmental contamination may reduce or prevent airborne and contact transmission. Furthermore, our strategy was implemented in high-risk and other areas of the hospital to monitor contamination in the air. The greatest fear when encountering any new or re-emerging pathogen is how and when it can be transmitted, including whether it can be transmitted through the air. Nosocomial cluster outbreaks are the predominant type of outbreak seen in the healthcare setting for most infectious diseases, including influenza and SARS [22, 23]. The practical importance and frequency of hospital-associated infections remain controversial, although the occurrence of infectious pathogens has increased sharply in recent years [24]. The methods of monitoring infection transmission routes are always improving; however, the surveillance and management of airborne pathogens remain difficult. The presence of organisms, especially viruses, in the airborne environment was more common than expected. Although many pathogens can be identified using conventional culture-based methods, the distribution of viruses in the hospital cannot be ascertained in the same manner [25]. In this paper, a simple strategy of environmental sampling in a clinical laboratory was used for the first time to identify contamination with SARS-CoV-2 in a real-world setting. It is important to note that positive molecular detection results may not necessarily reflect the presence of viable viruses.

Air quality varies among different settings in hospitals. Healthcare settings are directly and inevitably affected by infectious aerosolized pathogens. The major factors affecting hospital-acquired infections are microbial

agents, patient susceptibility, and environmental factors [26]. Routine air monitoring is time-consuming, labor-intensive, and valid only for the moment and location of collection. It has been suggested that particle counting could be used [27]; however, its value in the prediction of airborne bacteria in various parts of hospital environments has rarely been investigated [28]. In a clinical laboratory, there is a high level of risk of generating aerosols when devices such as automated blood cell analyzers, coagulation analyzers, chemical detectors, and immunological detectors are used. Although the New Coronavirus Pneumonia Diagnosis and Treatment Plan (6th edition) and New Biosafety Guidelines (2nd edition) were released by the National Health Commission of the People's Republic of China, evaluation of the contribution to contamination of splashes and aerosols generated by some operational steps, including nucleic acid extraction, microbial smear generation, centrifugation and specimen lid opening, has not been performed. Therefore, assessing the distribution of SARS-CoV-2 is particularly important, due to the high level of sensitivity of the detection method; this is especially true for environmental surveillance, although few studies are available with regard to clinical laboratories in hospitals.

To date, there have been few reports of medical staff in clinical laboratories contracting COVID-19; however, the key risks still exist while handling clinical samples from COVID-19 patients during the pre-analytical and analytical phases [31]. We speculate that there are two reasons for this observation. First, clinical laboratories are BSL-2 facilities with ventilation purification systems designed to maintain a clean environment, and medical staff in laboratories have a strong awareness of biosafety protocols and typically use BSL-2 personal protective equipment, including an N95 respirator [29], protective suit, medical gloves and protective goggles. These methods may be effective for preventing COVID-19. In addition to sufficient personal protection, laboratory staff may employ additional protective protocols, including those pertaining to specimen handling. Second, it should be noted that PCR laboratories are always fully managed in major hospitals and are equipped with a negative-pressure system, ceiling UV lights, biological safety cabinets and separate ventilation systems. During the initial outbreak of COVID-19 in Wuhan, PCR laboratories could be rapidly upgraded to BSL-3 facilities. To assess the factors related to SARS-CoV-2 detection in BSL-2 facilities, various environmental-factors, including clinical specimen type, exposure risk, biosecurity equipment and disinfection frequency, were studied. Our results showed that no biosafety risks existed in our clinical laboratory, and our experience with environmental monitoring could be applied to other zones of the hospital, especially operating theatres and intensive care units (ICUs).

Surveillance is the key method used to control and manage infections in specialized units of hospitals [30]. In this study, we developed and implemented a simple detection system for monitoring and surveillance of the clinical laboratory environment, which can also be applied in other areas in the hospital; in addition, our results provide information on the current situation regarding specific airborne pathogens in the clinical laboratories of hospitals. Moreover, our results highlight that the risk of exposure to potentially harmful airborne pathogens is widespread in hospitals, which conflicts with the results of previous studies on conventional hospital infection surveillance systems. The possibility of aerosol transmission exists during prolonged exposure to high concentrations of aerosol in a relatively closed environment. Clinical laboratories have advanced molecular detection technologies for pathogens, and this surveillance system should become part of the routine procedures. Furthermore, PCR for specific highly pathogenic microbes can be considered the main index by which indoor air quality is evaluated by the hospital control manager (Table 4). Environmental surveillance systems for specific highly pathogenic microbes, including SARS-CoV, MERS-CoV, and SARS-CoV-2, are lacking in clinical laboratories, even in general hospitals in China.

Our study has some limitations. We monitored environmental contamination only in the clinical laboratory and isolation wards and not in other departments in the hospital. Additional trials should be carried out in negative-pressure wards or ICUs; however, departments that experience nosocomial infections often focus on preventing disease outbreaks rather than routine surveillance. We note that reports by Loh TP et al. [31,32] show that only 15% of laboratories have formulated decontamination steps. However, in the early stage of the COVID-19 outbreak, we systematically decontaminated the laboratory and monitored the environment to ensure the biosafety of our laboratory. Moreover, the detection of SARS-CoV-2 RNA merely indicates that some nucleic acids were present in the environment and did not necessarily indicate the presence of live, infectious viruses.

CONCLUSION

To date, for basic control of the epidemic situation, we still carry out routine monitoring of the key departments of the hospital, such as the fever clinic, emergency department, respiratory department, and even the cold chain system of the staff canteen, so as to ensure the implementation of hospital infection monitoring. In conclusion, we suggest that this environmental surveillance system is suitable for routine implementation in a clinical laboratory after the emergence of specific pathogens. In addition, the use of environmental surveillance systems may also lead to the further assessment of infection prevention programs in different departments of hospitals, with the aim of providing a reliable method of managing hospital department and developing improved hospital pathogen surveillance systems.

Financial Support:
This work was supported by grants from Zhejiang Province Public Welfare Technology Application Project (No. LBY21H190001 to YP Wu), the Jinhua Science and Technology Bureau (and No. 2018-4-141 to YP Wu), and the Department of Education of Zhejiang Province (No. Y201839729 and Y202045827 to YP Wu). We are grateful to all of our colleagues in our laboratory for their support.

Acknowledgment:
We are grateful to all colleagues in our lab for their support.

Declaration of Interest:
None.

References:

1. Lippi G, Adeli K, Ferrari M, et al. Biosafety measures for preventing infection from COVID-19 in clinical laboratories: IFCC Taskforce Recommendations. Clin Chem Lab Med 2020;58(7): 1053-62 (PMID: 32396137).

2. Verde SC, Almeida SM, Matos J, et al. Microbiological assessment of indoor air quality at different hospital sites. Res Microbiol 2015;166(7):557-63 (PMID: 25869221).

3. Otter JA, Donskey C, Yezli S, Douthwaite S, Goldenberg SD, Weber DJ. Transmission of SARS and MERS Coronaviruses and Influenza Virus in Healthcare Settings: The Possible Role of Dry Surface Contamination. J Hosp Infect 2016;96:235-50 (PMID: 26 597631).

4. Menachery VD, Yount BL Jr, Debbink K, et al. A SARS-like cluster of circulating bat coronaviruses shows potential for human emergence. Nat Med 2015;21(12):1508-13 (PMID: 26552008).

5. Cui J, Li F, Shi ZL. Origin and evolution of pathogenic coronaviruses. Nat Rev Microbiol 2019;17(3):181-92 (PMID: 30531947).

6. Epidemiology Working Group for NCIP Epidemic Response, Chinese Center for Disease Control and Prevention. [The epidemiological characteristics of an outbreak of 2019 novel coronavirus diseases (COVID-19) in China]. Zhonghua Liu Xing Bing Xue Za Zhi 2020;41(2):145-51 (PMID: 32064853).

7. Liu Y, Ning Z, Chen Y, et al. Aerodynamic analysis of SARS-CoV-2 in two Wuhan hospitals. Nature 2020;582(7813):557-60 (PMID: 32340022).

8. Wu Y, Wu J, Zhang Z, Cheng C. DNA decontamination methods for internal quality management in clinical PCR laboratories. J Clin Lab Anal 2018;32(3):e22290 (PMID: 28665527).

9. Turgeon N, Toulouse MJ, Martel B, Moineau S, Duchaine C. Comparison of five bacteriophages as models for viral aerosol studies. Appl Environ Microbiol 2014;80:4242-50 (PMID: 24795 379).

10. Verani M, Bigazzi R, Carducci A. Viral contamination of aerosol and surfaces through toilet use in health care and other settings. Am J Infect Control 2014;42:758-62 (PMID: 24818773).

11. Hatagishi E, Okamoto M, Ohmiya S, et al. Establishment and clinical applications of a portable system for capturing influenza viruses released through coughing. PLoS One 2014;9(8):e103560 (PMID: 25083787).

12. Bergeron V, Chalfine A, Misset B, et al. Supplemental treatment of air in airborne infection isolation rooms using high-throughput in-room air decontamination units. Am J Infect Control 2011;39: 314-20 (PMID: 21095042).

13. Wu Y, Xu W, Zhu Z, Xia XP. Laboratory verification of an RT-PCR assay for SARS-CoV-2. J Clin Lab Anal 2020;34(10):e 23507 (PMID: 32754967).

14. Van Doremalen N, Bushmaker T, Morris DH, et al. Aerosol and Surface Stability of SARS-CoV-2as Compared with SARS-CoV-1. N Engl J Med 2020;382(16):1564-7 (PMID: 32182409).

15. Chin A, Chu J, Perera M, et al. Stability of SARS-CoV-2 in different environmental conditions. Lancet Microbe 2020;1(1):e10 (PMID: 32835322).

16. Medical Advisory Secretariat. Air Cleaning Technologies: An Evidence-Based Analysis. Ont Health Technol Assess Ser 2005;5 (17):1-52 (PMID: 23074468).

17. 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;15:83-95 (PMID: 15737 151).

18. Doremalen NV, Bushmaker T, Munster VJ. Stability of Middle East respiratory syndrome coronavirus (MERS-CoV) under different environmental conditions. Euro Surveill 2013;18(38): 20590 (PMID: 24084338).

19. Li Y, Leung GM, Tang JW, et al. Role of ventilation in airborne transmission of infectious agents in the built environment-a multidisciplinary systematic review. Indoor Air 2007;17:2-18 (PMID: 17257148).

20. López JH, Romo ÁS, Molina DC, et al. Detection of Sars-Cov-2 in the air of two hospitals in Hermosillo, Sonora, México, utilizing a low-cost environmental monitoring system. Int J Infect Dis 2020;102:478-82 (PMID: 33157295).

21. Wang X, Pan Z, Cheng Z. Association between 2019-nCoV transmission and N95 respirator use. J Hosp Infect 2020;105(1): 104-5 (PMID: 32142885).

22. Herfst S, Böhringer M, Karo B, et al. Drivers of airborne human-to-human pathogen Transmission. Curr Opin Virol 2017;22:22-9 (PMID: 27918958).

23. Ho PL, Tang XP, Seto WH. SARS: hospital infection control and admission strategies. Respirology 2003;8:41-5 (PMID: 15018 133).

24. Reed D, Kemmerly SA. Infection control and prevention: a review of hospital-acquired infections and the economic implications. Ochsner J 2009;9:27-31 (PMID: 21603406).

25. Tong X, Xu H, Zou L, et al. High diversity of airborne fungi in the hospital environment as revealed by meta-sequencing-based microbiome analysis. Sci Rep 2017;7:39606 (PMID: 28045065).

26. Sivagnanasundaram P, Amarasekara RWK, Madegedara RMD, Ekanayake A, Magana-Arachchi DN. Assessment of Airborne Bacterial and Fungal Communities in Selected Areas of Teaching Hospital, Kandy, Sri Lanka. Biomed Res Int 2019;2019:7393926 (PMID: 31309114).

27. Mirhoseini SH, Nikaeen M, Khanahmd H, Hatamzadeh M, Hassanzadeh A. Monitoring of airborne bacteria and aerosols in different wards of hospitals-Particle counting usefulness in investigation of airborne bacteria. Ann Agric Environ Med 2015;22(4): 670-3 (PMID: 26706974).

28. Scaltriti S, Cencetti S, Rovesti S, Marchesi I, Bargellini A, Borella P. Risk factors for particulate and microbial contamination of air in operating theatres. J Hosp Infect 2007;66:320-6 (PMID: 17 655973).

29. Wu HL, Huang J, Zhang CJP, He Z, Ming WK. Facemask shortage and the coronavirus disease (COVID-19) outbreak: Reflection on public health measures. EClinicalMedicine 2020;21: 100329 (PMID: 32292898).

30. Chang CC, Ananda-Rajah M, Belcastro A, et al. Consensus guidelines for implementation of quality processes to prevent invasive fungal disease and enhanced surveillance measures during hospital building works, 2014. Intern Med J 2014;44:1389-97 (PMID: 25482747).

31. Loh TP, Horvath AR, Wang CB, et al. Laboratory practices to mitigate biohazard risks during the COVID-19 outbreak: an IFCC global survey. Clin Chem Lab Med 2020;58(9):1433-40 (PMID: 32549123).

32. Loh TP, Horvath AR, Wang CB, et al. Operational considerations and challenges of biochemistry laboratories during the COVID-19 outbreak: an IFCC global survey. Clin Chem Lab Med 2020; 58(9):1441-9 (PMID: 32549122).