

COVID-19 Cluster Linked to Aerosol Transmission of SARS-CoV-2 via Floor Drains

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Background. Recently, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) transmission through exposure to aerosols has been suggested. Therefore, we investigated the possibility of aerosol SARS-CoV-2 transmission within an apartment complex where residents reported testing positive for SARS-CoV-2 despite having no direct contact with other SARS-CoV-2–infected people.

Methods. Information on symptom onset and exposure history of the patients was collected by global positioning system (GPS) tracking to investigate possible points of contact or spread. Samples collected from patients and from various areas of the complex were analyzed using RNA sequencing. Phylogenetic analysis was also performed.

Results. Of 19 people with confirmed SARS-CoV-2 infection, 5 reported no direct contact with other residents and were from apartments in the same vertical line. Eight environmental samples tested positive for the virus. Phylogenetic analyses revealed that 3 of the positive cases and 1 environmental sample belonged to the B.1.497 lineage. Additionally, 3 clinical specimens and 1 environmental sample from each floor of the complex had the same amino acid substitution in the ORF1ab region.

Conclusions. SARS-CoV-2 transmission possibly occurs between different floors of an apartment building through aerosol transmission via nonfunctioning drain traps.

Keywords. aerosol transmission; coronavirus; outbreak investigation; severe acute respiratory syndrome coronavirus 2; COVID-19.

The coronavirus disease 2019 (COVID-19) caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) threatens public health worldwide. As of 13 July 2021, the outbreak had resulted in 187 million confirmed cases and 4 million deaths [1]. SARS-CoV-2 is transmitted by direct contact through droplets and airborne transmission [2, 3]. In addition, aerosol transmission without direct contact is unpreventable by current quarantine policies, and its transmission path is difficult to prove based on epidemiological surveys alone [4, 5]. Therefore, understanding aerosol transmission of SARS-CoV-2 is vital for public health [6].

Despite the lack of definitive evidence, possible airborne transmission of SARS-CoV via floor drains was reported in an apartment complex in Hong Kong during the 2003 severe acute respiratory syndrome (SARS) epidemic [7–9]. Similar to SARS-CoV, the possibility of cluster infections with SARS-CoV-2 via aerosol transmission has also been suggested. COVID-19 cases

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in other cities have been reported to be associated with plumbing defects and aerosol transmission from excrement [10, 11].

In this study, we investigate COVID-19 cases in an apartment complex in South Korea between 10 and 27 January 2021. In addition, we explore the possibility of aerosol transmission without direct contact between infected individuals using epidemiological surveillance and molecular phylogenetic analysis.

METHODS

Epidemiologic Investigation

A cluster of COVID-19 cases was reported in Apartment Complex A in a city in South Korea between 10 and 27 January 2021. The index patient (patient 1) resided in Apartment 202 of the apartment building during a business visit from 3 to 9 January 2021. After experiencing a sore throat on 5 January, the patient underwent voluntary testing on 9 January and was confirmed to be positive for SARS-CoV-2 on 10 January. Patient 2 lived in Apartment 502 and underwent voluntary testing on 11 January after developing cough and chills on 10 January; the patient was then confirmed to be positive for SARS-CoV-2 on 12 January. Patients 3 and 4, who lived in Apartment 402, developed symptoms of cough and throat pain on 20 and 23 January 2021, respectively; they underwent SARS-CoV-2 testing on 26 January and were confirmed positive on 27 January. Patient 5, who lived in the same apartment as patients 3 and 4, developed symptoms on 23 January 2021, and was confirmed to be

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SARS-CoV-2 positive. This individual was identified through contact tracing.

We collected clinical and epidemiological information from patients 1 to 5. To investigate the possibility of transmission and prevent the spread of COVID-19, sophisticated communitybased contact tracing and epidemiological investigations were conducted with in-depth interviews regarding symptom onset and exposure history. When we identified the cluster, global positioning system (GPS) tracking was performed to investigate contact history, possibility of spread, and exposure history between confirmed COVID-19 patients through mobile phone data [12]. Apart from the confirmed patients, all residents living in 2 vertical lines (vertical lines 1 and 2) in the apartment building underwent SARS-CoV-2 reverse transcription polymerase chain reaction (RT-PCR) testing on 27 January 2021 [13]. Environmental samples collected on 28 January and 1 February 2021, were subjected to SARS-CoV-2 RT-PCR testing [14]. Moreover, field and environmental investigations of the apartments were conducted through architectural schematics and on-site measurements. We investigated the airflow around drains and vents in the bathroom by using an anemometer and visualized the airflow using a smoke generator. The investigation was carried out on behalf of the public health authority and institutional review board approval was not required.

Next-Generation Sequencing

To obtain the SARS-COV-2 complete genome sequences, viral RNA was extracted using the QIAamp viral RNA mini kit (Qiagen), and depletion of human ribosomal RNA was achieved using the NEBNext rRNA Depletion Kit (New England Biolabs) to isolate SARS-CoV-2 RNA from the samples. Library preparations were performed using the TruSeq RNA Sample Prep Kit v2 (Illumina) protocol. The enriched libraries were quantified using the Kapa Library quantification kit (Roche) and sequenced using the MiSeq reagent kit v2 (300 cycles; Illumina). Sequencing output was analyzed using CLC

Genomics Workbench 10 (Qiagen). Base-called reads were trimmed and aligned to a reference genome (NCBI Reference, NC_045512). The complete genome sequences determined for patient 3 and floor drain in Apartment 402 were termed hCoV-19/South Korea/KUMC127/2021 and hCoV-19/South Korea/KUMC124/2021, respectively.

Phylogenetic Analysis

A SARS-CoV-2 complete sequence set (n = 128) was prepared to analyze the phylogenetic relationship between patient samples (n = 3, patients 1, 2, and 3) and an environmental sample (n = 1, floor drain of Apartment 402). In total, 126 genome sequences were randomly collected from GISAID EpiCoV on 28 February 2021 (No. of sequences: South Korea = 108, including patients 1 and 2; reference sequences = 18; https:// www.gisaid.org). The sequences were concatenated-only open reading frames (ORF1ab, S, ORF3a, E, M, ORF6, ORF7a, ORF8, N, and OFR10). The sequence set was aligned using MAFFT version 7.471 and a phylogenetic analysis was conducted in MEGA X using the maximum likelihood method and a general time reversible model (GTR + G + I substitution) [15, 16]. A phylogeny test was performed using the bootstrap method with 1000 replications, followed by visualization of the maximum likelihood tree using Figtree version 1.4.3 (http://tree.bio.ed.ac. uk/software/figtree).

RESULTS

Epidemiologic Surveillance and Field Investigation

The epidemic curve based on the date of initial symptom onset in cases from Apartment Complex A is shown in Figure 1. In total, 19 confirmed COVID-19 cases were related to the apartment complex. There were 5 confirmed cases from the 3 apartments in vertical line 2, while there were no confirmed cases in vertical line 1 (Table 1). The other 14 confirmed COVID-19 cases occurred as a result of close or direct contact-based





Table 1. RT-PCR Results of the Residents of Apartment Complex A

| Vertical line 1 | | | | Vertical line 2 | | | |
|--------------------|---------------------|-----------------|--------------------------|--------------------|---------------------|-----------------|------------------------------------|
| Apart- ment No. | No. of Residents | Date of Testing | No. Tested and Result | Apart- ment No. | No. of Residents | Date of Testing | No. Tested and Result |
| 101 | 0 | | | 102 | 0 | | |
| 201 | 1 | 27 January 2021 | 1 Negative | 202 | 1 | 9 January 2021 | 1 Positive (patient 1) |
| 301 | 3 | 27 January 2021 | 3 Negative | 302 | 4 | 27 January 2021 | 4 Negative |
| 401 | 1 | 27 January 2021 | 1 Negative | 402 | 3 | 26 January 2021 | 3 Positive (patients 3, 4, and 5) |
| 501 | 2 | 27 January 2021 | 2 Negative | 502 | 3 | 11 January 2021 | 1 Positive (patient 2), 2 negative |

transmissions linked to patient 2, by social contact unrelated to the apartments [17].

The apartment complex was built in October 1986 and comprises 260 apartments distributed between 9 buildings. The apartment building that we investigated has 5 floors with 6 apartments on each floor; thus, there are 6 vertical lines of 5 apartments in the building. Two vertical lines of apartments share a single entrance and a staircase. There are no elevators; thus, residents of the 2 vertical lines use the same stairs to move between floors. The 6 apartments on each floor face each other in pairs, making 3 on each side, with the front doors approximately 2 m apart. Each apartment has an area of 68 m² and consists of 3 bedrooms, a living room, a kitchen, and a bathroom. The kitchen is located next to the bathroom, and there is a space between the veranda and bathroom, where the drainpipe and ventilation duct for the bathroom pass vertically. For the plumbing scheme of the bathroom, apartments on different floors are connected by a black water pipe attached to the toilet bowl and a gray water pipe



Figure 2. Probable route of airborne transmission from Apartment 202. A, Layout of the apartment building. B, Layout of Apartment 402. C, Diagram of the toilet structure and plumbing system of the apartment building.

connected to the bathtub, wash basin, and floor drain (Figure 2). A vent pipe is connected to the black and gray water pipes. Schematics show P or bell traps (P-traps are U-shaped pipes that hold a small amount of water at the bottom of the curve whereas bell traps are a water reservoir cup with a bell shape that dips into the cup to form a water seal, over it) connected to the bathtub, washbasin, toilet bowl, and floor drain. At the time of the field survey, the bathtubs in Apartments 202, 402, and 502 had all been removed, with their drains blocked, during interior remodeling. Although each bathroom had an air vent hole, individual ventilator fans or backdraft dampers were not installed in any apartment except in Apartment 502. In Apartments 202 and 402, the air vent in the bathroom had been blocked with a sheet. After removing the sheet, an inward airflow into the bathroom via the bathroom air vent was observed. When the kitchen hood was activated, the air inflow increased by approximately 0.7 m/s. Additionally, a bell trap was reported in the floor drain during the field survey; however, a smoke experiment revealed reverse flow, indicating that the trap was malfunctioning. Furthermore, when the kitchen hood was turned on, there was an even stronger reverse flow caused by negative pressure.

In the first set of environmental samples, 4 tested positive for SARS-CoV-2 (3 doorknobs and a washbasin). Therefore, another set of samples was collected from the bathroom. Of the 8 samples in the second set, 4 samples tested positive for SARS-CoV-2 (1 washbasin and 3 from the floor drain; Table 2).

Phylogenetic Analysis of Clinical and Environmental Specimens

To investigate COVID-19 outbreak within Apartment Complex A, an in-depth epidemiological analysis was performed using the MiSeq platform. The complete SARS-CoV-2 genome was determined based on next-generation sequencing (NGS) from a clinical specimen (patient 3) and the inner side of the lid of the floor drain in Apartment 402 toilet. Viral sequences from patients 1 and 2 were obtained from the GISAID EpiCoV database; however, the samples of nonapartment contacts for patient 2 were not sequenced. Subsequently, we identified the genetic relationship between clinical (patients 1, 2, and 3) and environmental specimens (lid of the floor drain in Apartment 402) based on the phylogenetic tree, which showed that KDCA1641 (patient 2), KDCA1634 (patient 1), KUMC127 (patient 3), and KUMC124 (lid of the floor drain in Apartment 402 toilet) clusters belonged to the B.1.497 lineage, GH clade (Figure 3). Furthermore, 3 clinical specimens and 1 environmental sample from each floor in the apartment complex had the same amino acid substitution (A6914V) in the ORF1ab region, which is not commonly present in sequences from South Korea, indicating that they were of the same origin.

DISCUSSION

We investigated the possibility of aerosol transmission of SARS-CoV-2 based on reports of COVID-19 within Apartment

Table 2. qRT-PCR Results of Environmental Samples Collected in Apartment 402 During Field and Environmental Investigations

| Environmental Sample | Ct Value |
|---|--------------|
| First set of samples collected 28 January 2021 | |
| Fuse box | -,- |
| Clock | -,- |
| Doorknob of veranda door | 36.01, 36.95 |
| Doorknob of utility room door | 37.05, 35.98 |
| Doorknob of room 1 door | -,- |
| Doorknob of room 2 door | 37.77, 38.26 |
| Doorknob of room 3 door | -,- |
| Doorknob of room 4 door | -,- |
| Doorknob of front door | -,- |
| Kitchen shelf | -,- |
| Kitchen faucet | -,- |
| Inner doorknob of toilet door | -,- |
| Outer doorknob of toilet door | -,- |
| Toilet faucet | -,- |
| Vent in toilet | -,- |
| Washbasin in bathroom | 27.38, 27.12 |
| Shower handle | -,- |
| Toilet bowl handle | -,- |
| Inner side of toilet bowl | -,- |
| Water from toilet bowl | -,- |
| Seat of toilet bowl | -,- |
| Second set of samples collected 1 February 2021 | |
| Washbasin in bathroom | 32.99, 32.90 |
| Water overflow drainage hole in washbasin | -,- |
| Surface of washbasin | _,_ |
| Outer side of washbasin faucet | -,- |
| Inner side of washbasin faucet | _,_ |
| Outer side of floor drain lid in toilet | 31.74, 31.68 |
| Inner side of floor drain lid in toilet | 27.79, 28.02 |
| Floor drain hole | 28.38, 28.63 |

Abbreviations: -, negative on RT-PCR testing; Ct (E,RdRp), E and RdRp gene cycle threshold; qRT-PCR, quantitative reverse transcription polymerase chain reaction.

Complex A. Through in-depth interviews, we discovered that all infected individuals in the different apartments of the complex had no direct contact with each other or with other previously confirmed SARS-CoV-2-infected people, as detected by GPS tracking. Therefore, we suspected aerosol transmission of SARS-CoV-2 via air vents or drains connected between apartments [7-11, 18]. First, we considered the possibility of spread via bathroom air vents [18]. At the time of the field survey of Apartment Complex A, the air vent was sealed in the bathroom of Apartment 202 and was not operating properly in Apartment 402. Moreover, in the first set of environmental samples collected from Apartment 402, the bathroom air vent sample tested negative for SARS-CoV-2; thus, the spread of COVID-19 in Apartment Complex A by aerosol transmission via bathroom air vents was considered less likely. In addition, the only way to ventilate was by opening the windows because the apartment did not have any mechanical ventilation system. However, residents rarely opened their windows due to the winter season during the outbreak. Each housing unit



Figure 3. Phylogenetic relationship between severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) samples on the basis of SARS-CoV-2 spread within Apartment Complex A. The sequences of clinical (patients 1, 2, and 3) and environmental samples (floor drain of Apartment 402) used for analysis are indicated in red, and bootstrap values are in different colors on the nodes. The scale bar represents the average number of nucleotide substitutions at each site.

has a floor radiant heating system and a room air conditioner for cooling. Therefore, there was no possibility of air mixing through the air conditioning system. Second, we considered the possibility of transmission via bathroom drains, as identified in a previous case study of an apartment building [7-11]. Among the second set of environmental samples collected from Apartment 402, the floor drain samples tested positive for the virus. In addition, based on the structure and plumbing schematics of the building and the results of the smoke experiment, we found that the traps in the floor drains were not functioning properly. Previous studies imply that the stack effect can cause vertical air movement through drainpipes or air vent ducts [11, 18]. Furthermore, according to the Korea Meteorological Administration, the local average outdoor air temperature was -3.9° C during the index patient's stay at the apartment, and the low outdoor air temperature could have

caused upward air movement in the drainpipes and air vent ducts, even though the apartment building had only 5 stories. Moreover, a kitchen hood fan could have accelerated the air movement and aerosol transmission between apartments in the same line. Third, there is the possibility of transmission through corridors and stairs. Environmental samples were not collected from these places, and the virus was not detected on the front door handle. Furthermore, people usually wear masks on stairs, and the front door is relatively well sealed. Therefore, we consider the possibility of transmission through these spaces to be low. This is supported by the fact that none of the vertical line 1 residents, who share the same stairs and corridor, were infected.

The transmission route was further confirmed by molecular epidemiologic analysis. Phylogenetic analysis showed that both clinical and nonclinical samples were part of the same transmission chain. Consequently, our results provide strong molecular evidence of airborne transmission of SARS-CoV-2 through a toilet floor drain, which suggests that aerosol transmission of COVID-19 occurs between different floors of the same apartment building due to nonfunctioning drain traps.

Our study has some limitations. First, we could not detect viable viruses from environmental samples. Second, data regarding contact and exposure history to COVID-19 may be inaccurate because of recall bias. Third, although the investigation into the effects of corridors and stairs in transmission has not been conducted, the possibility is low.

In conclusion, our study suggests that molecular phylogenetic analysis by whole-genome sequencing can be performed to investigate the transmission route. Thus, if the epidemiological evidence of transmission between SARS-CoV-2-infected patients is unclear, whole-genome analysis may help in identifying the transmission route. In addition, our study underlines the importance of appropriate environmental sampling in clarifying epidemiological links among patients.

We demonstrated that this type of aerosol transmission may lead to the persistence of an epidemic in a community. Therefore, to identify and control the modes of transmission of SARS-CoV-2 and other respiratory viruses, continuous surveillance and research on aerosol transmission is essential [19]. Moreover, traps are installed in floor drains under the Building Code, but it is necessary to check whether the traps function properly (to check whether the water seal in the trap is being maintained) to prevent transmissions. Also, in facilities vulnerable to infection clusters, such as hospitals and prisons, the structures and ventilation systems should be evaluated and monitored, considering the possibility of aerosol transmission [19, 20].

Notes

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