ASYMPTOMATIC TRANSMISSION OF CORONAVIRUS DISEASE 2019 (COVID-19) AT A WORKPLACE IN BANGKOK METROPOLITAN REGION, THAILAND

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Abstract. Coronavirus disease 2019 (COVID-19) caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has a wide clinical spectrum, ranging from asymptomatic infection to multi-organ system failure. Patients with COVID-19 generally spread the virus during the symptomatic phase. However, data on COVID-19 transmission from patients who did not develop symptoms in the whole course of illness is still limited. The study described an outbreak consisting of six asymptomatic COVID-19 cases in an office workplace, which was not related to family transmission or healthcare settings. The first asymptomatic COVID-19 case was identified by positive SARS-CoV-2 nasopharyngeal swab RT-PCR assay during an active surveillance in the community. Close contact tracing among coworkers of the index case identified five other asymptomatic positive nasopharyngeal swab RT-PCR COVID-19 subjects. SARS-CoV-2 serology assays identified another 15 nasopharyngeal swab RT-PCR negative asymptomatic coworkers on the same floor as the six infected cases. No officer workers on different floors were SARS-CoV-2 positive by serology assays and no family members of the first six asymptomatic COVID-19 cases were positive by nasopharyngeal swab RT-PCR assay. The findings highlight the importance of SARS-CoV-2 transmission from asymptomatic patients in a non-healthcare setting and of serology testing in risk individuals with negative nasopharyngeal swab SARS-CoV-2 RT-PCR assay results.

Keywords: asymptomatic COVID-19, COVID-19, SARS-CoV-2 assay, transmission

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INTRODUCTION

Coronavirus disease 2019 (COVID-19) caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has spread throughout the world since January 2020 (WHO, 2021). Clinical presentation varies from no symptoms to upper respiratory tract symptoms or severe respiratory distress (Wong *et al*, 2020). It is estimated that 30-80% of all infected patients are asymptomatic throughout their illness, and 80% of symptoms are mild (Arons *et al*, 2020; Guan *et al*, 2020; Oran and Topol, 2020; Nishiura *et al*, 2020). Patients in the pre-symptomatic phase, especially Days 1-3 prior to onset of symptoms, are able to transmit SARS-CoV-2 (Wei *et al*, 2020). However, information regarding transmission of the virus from patients who are asymptomatic throughout the course of illness is limited, with only a few reports such as of asymptomatic SARS-CoV-2 transmission in a family cluster and school (Bei *et al*, 2020; Wong *et al*, 2020).

In January 2020, the Department of Disease Control and the Ministry of Public Health, Thailand issued a guideline recommending SARS-CoV-2 testing only for suspected and/or symptomatic patients (MOPH, 2020). However, active surveillance has also been implemented to identify asymptomatic cases in communities where infected individuals have been confirmed. The aim of this study is to explore the extent of the outbreak of SARS-CoV-2 from asymptomatic patients in non-healthcare and non-family settings.

MATERIALS AND METHODS

Study population

The first case of COVID-19 was identified during an active surveillance survey in a community in the Bangkok Metropolitan area. Family members and coworkers of the index case were then identified and examined for SARS-CoV-2 infection. Inclusion criteria were: (i) coworkers who worked at an Election Commission Office of Thailand where the COVID-19 index case was identified, (ii) coworkers in close contact with the index case during 14 days prior to the date of positive nasopharyngeal swab RT-PCR test for SARS-CoV-2, and (iii) family members of the index case and other infected COVID-19 cases. Exclusion criterion was participants with incomplete data.

Study protocol was approved by the Institutional Review Board, Faculty of Medicine Ramathibodi Hospital, Mahidol University (approval number: MURA2020/960). Prior written informed consent for the study was waived due to the nature of the COVID-19 outbreak.

Study design

A retrospective, descriptive study was carried out among coworkers and family members of COVID-19 cases from an outbreak at an office of the Election Commission of Thailand in the Bangkok Metropolitan Region by the Institute for Urban Disease Control and Prevention, Department of Disease Control, Ministry of Public Health, Nonthaburi Province, Thailand, from 18 April - 11 May 2020.

Data collection

Demographic data, clinical data, data on COVID-19 assessment exposure risks, radiological findings, and laboratory data of nasopharyngeal swab SARS-CoV-2 RT-PCR and serology tests were collected.

Specimen collection

Nasopharyngeal swab specimens were collected and tested using SARS-CoV-2 RT-PCR assay kits on the same day of collection. Blood samples for SARS-CoV-2 serology tests were drawn from close contacts one week after identifying COVID-19 index case and at least a week after diagnosis of SARS-CoV-2 infection of all six subsequent SARS-CoV-2 infected cases.

Laboratory tests

SARS-CoV-2 RT-PCR assays

RNA was extracted from nasopharyngeal swabs using two nucleic acid extraction kits, namely, the NX-48S Viral NA kit (Genosolution Inc, Seoul, South Korea) and the MagDEA®Dx SV kit (Precision System Science Co Ltd, Chiba, Japan) and subjected to SARS-CoV-2 RT-PCR assay using two commercially available SARS-CoV-2 nucleic acid diagnostic kits, namely, Novel Coronavirus (2019-nCoV) Nucleic Acid Diagnostic (PCR-Fluorescence Probing) Kit (Sansure Biotech Incorp, Changsha, PR China) targeting open reading frame 1a and 1b (*ORF1ab*) and nucleocapsid (N) genes, and Allplex™ 2019-nCoV Assay Kit (Seegene Incorp, Seoul, South Korea) targeting N, RNA-dependent RNA-polymerase (RdRp) and envelop (E) genes. Procedures, including use of internal control of each assay, were carried out following the manufacturers' instructions. The negative threshold cycle (Ct) values for both kits were >40 for all genes.

SARS-CoV-2 serology assays

Serum from each blood sample was stored at -20°C until used. Three SARS-CoV-2 serology tests were employed: (1) an Abbott SARS-CoV-2 IgG assay (Abbott Architect Instrument; Abbott Core Lab, Chicago, IL) is a chemiluminescent microparticle immunoassay for qualitative detection of serum IgG against SARS-CoV-2 N protein, and result is reported as an index value, with index value <1.4 considered negative and ≥1.4 positive; (2) a Wondfo SARS-CoV-2 antibody assay kit (Guangzhou Wondfo Biotechnology Co Ltd, Guangzhou, PR China) employs a lateral flow method that detects total antibodies against SARS-CoV-2 N protein and appearance of two bands indicates a positive result, one band for presence of total specific antibodies and the other a positive control; (3) a Superbio SARS-CoV-2 antibody assay kit (Jiangsu Superbio Biomedical Co Ltd, Nanjing, PR China) employs a colloidal gold method for detecting IgM and IgG against SARS-CoV-2 N and spike (S) proteins, and appearance of two bands indicates a positive result, one positive band for presence of IgM and/or IgG and the other a positive control band.

Statistical analysis

Data are reported as frequency and 95% confident interval (CI) using a StataCorp LLC Version 13 software (StataCorp LP, College Station, TX).

RESULTS

Among 250 participants from an active surveillance to identify SARS-CoV-2 infected individuals in a community in the Bangkok Metropolitan region during 18 April - 11 May 2020, WT (the first index case) was the only individual detected on 20 April 2020 with a positive nasopharyngeal swab SARS-CoV-2 RT-PCR result (Table 1). WT worked at the Office of the Election Commission of Thailand where 926 individuals were employed. Three hundred and thirty office close contacts of WT during the 14 days prior to positive test result were identified and five more individuals (JB, SG, SW, UC, and WK) were tested positive by a nasopharyngeal swab SARS-CoV-2 RT-PCR assay (1.5%; 95% confidence interval (CI): 0.4-3.5).

UC (second index case) was diagnosed on 27 April 2020, having had contact with WT on 7, 14, 21 and 22 April 2020, and on 22 April 2020 UC attended a meeting for at least 5 minutes in an air-conditioned office with five coworkers, all of whom did not wear face masks during the meeting. Four others (JB, SG, SW, and WK) were tested positive by a nasopharyngeal swab SARS-CoV-2 RT-PCR assay during the subsequent quarantine period commencing 27 April 2020 (Table 1). Ct values for RT-PCR of SARS-CoV-2 N gene of all six infected cases was in the range of 31-38 (Table 1). Repeat nasopharyngeal swab SARS-CoV-2 RT-PCR tests conducted 4-37 days following the first positive results were negative (Table 1). All six infected patients remained asymptomatic during their quarantine period and chest radiological findings were negative.

Serological tests (Abbott and Wondfo SARS-CoV-2 antibody kits) performed on the six infected patients 9-33 days following the first positive nasopharyngeal SARS-CoV-2 RT-PCR results were positive in five patients (except SG who had the highest Ct value) (Table 1). Serological tests conducted among office close contacts (n = 223/330) of index case one week following detection of infection (three serological tests namely, Abbott, Superbio and Wondfo SARS-CoV-2 antibody kits) conducted on 217 samples and two tests (Abbott and Wondfo kits) on six samples revealed 15 individuals with at least one positive serology test result (7%; 95% CI: 4-11), among whom seven (3%; 95% CI: 1-6) were positive in all three serology tests, two (1%; 95% CI: 0-3) positive in two serological tests (Abbott and Wondfo kits) and six (3%; 95% CI: 1-6) positive in only one serology test (two and four with Superbio SARS-CoV-2 IgG and Abbott SARS-CoV-2 IgG test kit respectively) (Table 1). These 15 close contacts worked on the same floor as UC and possibly had contact with the latter some period prior to diagnosis. However, all 15 serology positive subjects were negative for SARS-CoV-2 RT-PCR tests conducted 1-2 weeks apart (Table 1). No individual working on different floors from the infected cases displayed positive serology test results.

Nasopharyngeal swab SARS-CoV-2 RT-PCR assays of household contacts (n = 10: one each from JB and SG family, two from SW family and three

Table 1
Timeline of SARS-CoV-2 RT-PCR and serology assays of six asymptomatic subjects working in the same office in Bangkok, Thailand (20 April - 29 May 2020).

Case	RT-PCR assay of nasopharyngeal swab				Serology test		
	Date	Result Threshold cycle (Ct) value	Date	Result	Date	Abbott	Wondfo
WT	20 April 2020	Positive 29 for E, 31 for N, 32 for RdRp	28 April 2020	Negative	13 May 2020	Positive	Positive
UC	24 April 2020	Positive 32 for E, 35 for N, 35 for RdRp	28 May 2020	Negative	28 May 2020	Positive	Positive
SG	25 April 2020	Positive E (negative), 38 for N, RdRp (negative)	13 May 2020	Negative	13 May 2020	Negative	Negative
JB	7 May 2020	Positive 30 for E, 31 for N, 31 for RdRp	13 June 2020	Negative	13 June 2020	Positive	Positive
WK	15 May 2020	Positive 36 for N, 39 for ORF	29 May 2020	Negative	29 May 2020	Positive	Positive
SW	15 May 2020	Positive 36 for N, 40 for ORF	29 May 2020	Negative	29 May 2020	Positive	Positive

E: SARS-CoV-2 E gene (negative, Ct >40); N: SARS-CoV-2 N gene (negative, Ct >40); ORF: SARS-CoV-2 open reading frame gene 1a and 1b (negative, Ct >40); RdRp: SARS-CoV-2 RNA dependent RNA polymerase gene (negative, Ct >40); Abbott: Abbott SARS-CoV-2 IgG test kit; Wondfo: Wondfo SARS-CoV-2 total antibody test kit

Table 2

Dates of assays of household contacts of five asymptomatic SARS-CoV-2 RT-PCR positive workers at same office of index case, Bangkok, Thailand (28 April - 17 May 2020)

Case[a]	Household contact	Date of RT-PCR assay[b]
UC	Spouse	28 April 2020
	Friend 1	28 April 2020
	Friend 2	28 April 2020
SG	Friend	28 April 2020
JB	Spouse	13 May 2020
WK	Spouse	17 May 2020
	Child 1	17 May 2020
	Child 2	17 May 2020
SW	Friend 1	17 May 2020
	Friend 2	17 May 2020

[a]Placed in quarantine on 27 April 2020; [b]Negative result

each from UC and WT family) carried out 2-6 days following diagnosis of the respective subject were negative (Table 2).

DISCUSSION

Investigating the spread of SARS-CoV-2 on the same floor of an office workplace demonstrated transmission of SARS-CoV-2 could occur from individuals who were asymptomatic. This emphasized the importance of identification of asymptomatic persons through active case surveillance, which can mitigate or eliminate transmission of the virus in a community. Had active surveillance not been performed, the first two index cases would not have been identified and quarantined and might have spread the virus to even more people identified in the study.

The second index case likely spread SARS-CoV-2 to four other coworkers on 22 April 2020 when they participated in a long meeting with the former, all of whom did not wear face masks. However, it was also possible that the second index case could have been infected by the first before 22 April 2020 and subsequently spread the infection to others. The two index cases had met with each other on three separate occasions before 22 April 2020. Because SARS-CoV-2 incubation period can be as long as 14 days and be transmitted during the first week after onset of symptoms (Wolfel *et al*, 2020), it is possible that transmission between the two might have occurred on either the first or second encounter. Apart from the second index case, the first case did not have close contacts with any other persons in the workplace and, therefore, was unlikely to spread SARS-CoV-2 to others than the second case.

The study also demonstrated the usefulness of serology tests for diagnosis of COVID-19, especially in subjects with no symptoms and negative nasopharyngeal swab SARS-CoV-2 RT-PCR results. Among this group who had previous contact with coworkers positive for SARS-CoV-2 by nasopharyngeal swab RT-PCR test, 7% were tested positive by at least one of three SARS-CoV-2 serological assays employed in the study and could be considered to have had COVID-19. However, false positive results could not be totally excluded as the serology tests were conducted only once and there was no follow-up serology test to detect possible rise in titer.

It is worth noting no household contacts were tested positive by nasopharyngeal swab SARS-CoV-2 RT-PCR. As the four infected close contact subjects of the second index case were placed in quarantine once infection of the latter was diagnosed, they were still in an incubation period when there was no viral transmission. However, the RT-PCR test was conducted on household contacts only once and there were no follow-ups or were serology assays performed.

In conclusion, this study shows transmission of SARS-CoV-2 from asymptomatic (not pre-symptomatic) cases as viral loads are similarly high among symptomatic, pre-symptomatic and asymptomatic patients (Arons *et al*, 2020). In addition, serology tests proved useful in detecting asymptomatic SARS-CoV-2 infection in persons with negative nasopharyngeal swab RT-PCR results, and thus, serology tests should facilitate tracing of infected people in investigations of outbreaks.

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