








# BMJ Open COVID-19 among staff and their family members of a healthcare research institution in Bangladesh between March 2020 and April 2021: a test-negative case-control study

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## ABSTRACT

**Objective** To identify factors associated with COVID-19 positivity among staff and their family members of icddr, a health research institute located in Bangladesh.

**Setting** Dhaka, Bangladesh.

**Participants** A total of 4295 symptomatic people were tested for SARS-CoV-2 by reverse-transcription PCR between 19 March 2020 and 15 April 2021. Multivariable logistic regression was done to identify the factors associated with COVID-19 positivity by contrasting test positives with test negatives.

**Result** Forty-three per cent of the participants were tested positive for SARS-CoV-2. The median age was high in positive cases (37 years vs 34 years). Among the positive cases, 97% were recovered, 2.1% had reinfections, 24 died and 41 were active cases as of 15 April 2021. Multivariable regression analysis showed that age more than 60 years (adjusted OR (aOR)=2.1, 95% CI 1.3 to 3.3;  $p<0.05$ ), blood group AB (aOR=1.5, 95% CI 1.1 to 2;  $p<0.05$ ), fever (aOR=3.1, 95% CI 2.6 to 3.7;  $p<0.05$ ), cough (aOR=1.3, 95% CI 1.1 to 1.6;  $p<0.05$ ) and anosmia (aOR=2.7, 95% CI 1.3 to 5.7;  $p<0.05$ ) were significantly associated with higher odds of being COVID-19 positive when compared with participants who were tested negative.

**Conclusions** The study findings suggest that older age, fever, cough and anosmia were associated with COVID-19 among the study participants.

## INTRODUCTION

The COVID-19 pandemic is a global health challenge the likes of which the world has never been experienced so far on this scale. Since its first documentation in December 2019 in the Wuhan City, Hubei Province, China, this disease has spread across all over the world with deadly consequences.<sup>1</sup>

## STRENGTHS AND LIMITATIONS OF THIS STUDY

- ⇒ This manuscript used a growing database of employees from a health research institute who underwent COVID-19 tests.
- ⇒ Information was collected in real-time processes as per the directive of the institute management.
- ⇒ Reverse-transcription PCR tests for COVID-19 were done in the Virology laboratory at icddr, a state-of-the-art laboratory in Bangladesh.
- ⇒ Data on the presence of chronic diseases, bacille Calmette-Guerin (BCG) vaccination, and usual physical activities were collected over telephone interviews from only 65% of the participants.
- ⇒ This study did not address the variants of SARS-CoV-2 circulating in the region or the possible modifications of symptom presentations depending on the variant infecting the patients.

The SARS-CoV-2 is the aetiological agent of this illness.<sup>2</sup> COVID-19 was avowed as a global pandemic on 11 March 2020, by WHO.<sup>3</sup> As of 22 September 2021, the disease accounts for 230 446 504 confirmed cases and 4 725 210 deaths worldwide.<sup>4</sup> The first case of COVID-19 in Bangladesh was officially detected on 8 March 2020. As of 22 September 2021, a total number of 1 545 800 confirmed cases were detected with 27 277 deaths in the country.<sup>5</sup> Although some countries have responded quickly enough to contain the disease, we generally witnessed a somewhat casual response on a global scale.<sup>1,2</sup> Resource-limited countries did not have had the means to respond most effectively due to the lack of large-scale testing facilities, available testing kits, adequate infrastructure as well as intensive care support for all and

proper quarantine measures.<sup>5</sup> These efforts were further hampered by poor living conditions, high population density and substandard health services, subsequently, facilitating the mass spread of the disease.<sup>3</sup>

The typical presenting symptoms of COVID-19 are fever, dry cough, sore throat, dyspnoea or fatigue coupled with the recent history of exposure.<sup>6–9</sup> Many studies have already reported different factors associated with COVID-19 infection. Most commonly observed factors are older age, male sex, presenting symptoms, for instance, cough, fever, loss of smell, close relationship with index case and family members of patients with COVID-19 positive.<sup>10–12</sup> Studies with a larger sample size showed that smoking and physical inactivity are also associated with COVID-19 infection and mortality.<sup>13</sup>

Existing evidence showed that the presence of chronic disease is a risk factor for both the susceptibility to infection and progression of COVID-19 to severe disease.<sup>14</sup> It was observed that the severity of COVID-19 outcome is higher among patients with hypertension, obesity, type 2 diabetes mellitus (DM) and other chronic diseases like chronic lung disease, chronic kidney disease and coronary heart disease.<sup>14–16</sup> Recent studies also reported a relationship between blood group types and positivity as well as the severity of COVID-19 disease.<sup>17–19</sup> Few studies suggest that the bacille Calmette-Guerin (BCG) vaccination could be protective against COVID-19 infection as countries with compulsory BCG vaccination had fewer COVID-19 cases.<sup>20–24</sup>

Although many papers were published on factors associated with COVID-19 positivity, there remains a scarcity of data collected from countries where the data repository systems are not properly developed.<sup>25</sup> Despite commendable efforts so far in Bangladesh to contain the disease within manageable level considering its high population density, there has been a paucity of data on the epidemiology of COVID-19, particularly involving high-quality sources.<sup>26</sup> However, icddr,b, a well-renowned health research institute based in Bangladesh, has been maintaining a high-quality database for its staff and their family members since the inception of COVID-19 in the country. The current analysis took the opportunity of the COVID-19 staff database of icddr,b to explore the factors associated with COVID-19 infection.

## METHODS

This is an observational test-negative design, including data from the staff and their family members of icddr,b, Dhaka, Bangladesh. We reported this study by following Strengthening the Reporting of Observational studies in Epidemiology statement checklist for the case-control studies.<sup>27</sup>

### Study design

This test-negative case-control study used clinical, socio-demographic and laboratory data from the COVID-19 staff database of icddr,b, a health research institute in

Dhaka, Bangladesh. Here cases were icddr,b staff or family members who had symptoms suggested of COVID-19, contacted icddr,b staff clinic and tested positive for SARS-CoV-2. In contrast, controls are patients from the same population with similar symptoms who underwent the same tests for the COVID-19 at the icddr,b facility and tested negative. Since controls are the same group of patients who present for testing but test negative, a test-negative design is very helpful to control for factors that are usually challenging to estimate in an observational study particularly care-seeking behaviour and access to care. However, some of the contacts were symptomless and tested positive included in the analysis as cases and some contacts were tested negative considered as controls. The study was conducted between 19 March 2020 and 15 April 2021, during the SARS-CoV2 pandemic.

### Study premise

icddr,b is one of the leading public health research organisations in Bangladesh. Since 19 March 2020, icddr,b started a system to prevent and protect its ~4000 employees and their family members (~12 000) against COVID-19. All staff with any clinical symptom (fever, cough and cold or respiratory distress) suggesting COVID-19 were instructed to contact icddr,b staff clinic. Subsequently, staff clinic doctors instructed the suspected individual to provide a nasopharyngeal swab to be tested at icddr,b Virology Laboratory using reverse-transcription PCR (RT-PCR). All contacts of COVID-19-positive staff were isolated or quarantined and tested accordingly. Besides, all the relevant information from the individual has been entered into the database in collaboration with the Staff Clinic, Dhaka Hospital at icddr,b, Virology Laboratory and Human Resources. Not to mention, we have used the data from this database to conduct our analysis.

### Study population

icddr,b employees and their family members who contacted staff clinic with symptoms suggestive of COVID-19 before 16 April 2021 provided nasopharyngeal swabs and tested for COVID-19 were considered as the study population. For individuals tested more than once, only the first instance was considered.

### Sample collection and laboratory assay

From all symptomatic staff and family members, a nasopharyngeal swab was collected by a trained nurse, and the swab was sent to the Virology Laboratory at icddr,b to be analysed using a real-time reverse transcriptase-PCR (rRT-PCR). In brief, total RNA was extracted from nasopharyngeal swabs using the chemagic Viral NA/gDNA (PerkinElmer, Massachusetts) Kits. RNA was tested for SARS-CoV-2 by rRT-PCR targeting ORF1ab- and N-gene specific primers and probes following the protocol of the Chinese Center for Disease Control and Prevention (briefly as China CDC). A positive case was determined if the CT values of two targets (ORF1ab and N) were <37 in the same specimen. If CT values of any sample were

37–40 or a single target was positive, it was resampled and retested. If the CT values were still 37–40 and the amplification curves had obvious peaks, the sample was considered positive.

### Data collection and staff database

Data were extracted from icddr,b staff database and additional data on chronic disease, blood groups, and lifestyle factors were collected by interview over phone. icddr,b COVID-19 staff database has been carefully documenting all basic information related to SARS-CoV-2 infection and COVID-19 disease among icddr,b staff and their family members. This includes age, sex, area of residence, history of contact, travel history, presenting symptoms and assay result for COVID-19 positivity and compliance of quarantine/isolation.

Additionally, through telephone interviews, data on blood group, routine physical activity, history of BCG vaccination, pre-existing chronic disease like DM, hypertension, chronic obstructive pulmonary disease (COPD), asthma, ischaemic heart disease, cancer or kidney disease were collected using a short case report form. Data on routine physical activities were collected using pretested 'International physical activity questionnaire (IPAC)-short form' ([www.ipaq.ki.se](http://www.ipaq.ki.se)), and this questionnaire was already validated.<sup>28</sup> Based on the last 7 days, recall data physical activities were categorised as no, mild, moderate and vigorous categories. To minimise bias, all names of the employees were removed from the Microsoft Access-based study database. Consent to participate in this study was collected in electronic media like email, short message service (SMS) or WhatsApp based on availability and accessibility.

### Variables

This study was done to explore the factors associated with COVID-19 positivity. The outcome variable was COVID-19 positivity based on RT-PCR assay and the explanatory variables were age, sex, presenting symptoms, area of residence, travel history, history of contacts, presence of chronic disease, smoking, blood group, BCG vaccination and physical activities.

### Operational definitions

Recovery: icddr,b staff and/or family members who were tested positive to COVID-19 were released from isolation based on the following conditions and considered recovered. Symptomatic and non-hospitalised cases were considered recovered 10 days after onset of symptom and if they were without fever for the last 3 days and also there was a significant improvement of their respiratory symptoms. Hospitalised patients were considered recovered 21 days after onset of symptoms and if they were without fever at least for 3 days without the use of antipyretics and there was a significant improvement of respiratory symptoms. For asymptomatic RT-PCR-positive cases were considered recovered 10 days after sample collection. This can be

noted that testing for COVID-19 using RT-PCR was not required for release from isolation.

Mild disease: when a COVID-19 test-positive case had mild clinical symptoms and with no sign of pneumonia on imaging was considered a mild disease. The presence of any one symptom or in a combination of symptoms like cough, fever, malaise, sore throat, muscle pain or headache without shortness of breath was considered mild clinical symptoms.

Moderate disease: when a COVID-19 test-positive patient presented with signs of pneumonia, with a respiratory rate of  $\leq 30$  breaths /min, and peripheral capillary oxygen saturation (SpO<sub>2</sub>) of more than 93 at room air was considered moderate COVID-19 disease.

Severe disease: when a COVID-19 test-positive case developed respiratory distress ( $>30$  bpm), a peripheral capillary oxygen saturation (SpO<sub>2</sub>) of  $\leq 93\%$  at rest and a ratio of arterial oxygen partial pressure (PaO<sub>2</sub> in mm Hg) to fractional inspired oxygen (PaO<sub>2</sub>/FiO<sub>2</sub>) of  $\leq 300$  mm Hg, or lung infiltrates of  $\geq 50\%$  in chest X-ray, was considered severe COVID-19 disease.

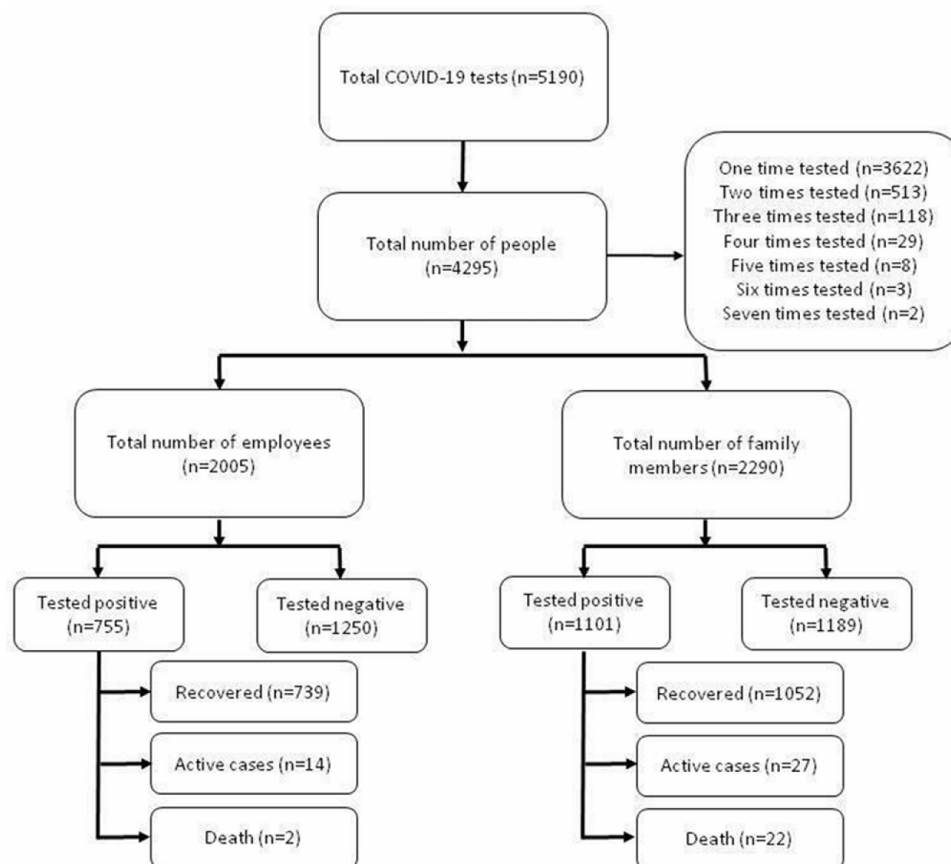
Reinfection: for this analysis, reinfection was defined as any symptomatic study participant who was tested positive for COVID-19 at least 2 months after a positive test result and who was clinically recovered from the initial infection.

### Data analysis

At first, we described baseline characteristics of the study population, including age, sex, area of residence, symptoms, dates of disease diagnosis and comorbidities. We reported categorical variables as number (%) and continuous variables as median (IQR). To compare the categorical variables,  $\chi^2$  or Fisher's exact tests were done, as appropriate. To explore the factors associated with COVID-19 positivity, binary logistic regression was carried out. Bivariate associations between each independent variable with COVID-19 positivity were initially performed. In the multivariable model, to remove overfitting, we selected variables that demonstrated a p value of  $<0.2$  in bivariate analysis. The final multivariable model was also adjusted for seasonality. We calculated seasonality using the formula  $\sin(2m\pi/12)+\cos(2m\pi/12)$ , where 'm' is the calendar month.<sup>29</sup> Multicollinearity was checked by calculating the variance inflation factor (VIF) and variables considered in the final model had a VIF of 2 or less. A p value of less than 0.05 was regarded as statistically significant and all analyses were done in STATA (V.15.1 StataCorp).

### RESULT

Between 19 March 2020 and 15 April 2021, a total number of 5190 testing for SARS-COV-2 were done at icddr,b where 4295 symptomatic people provided their nasopharyngeal swab. Among them, 47% were icddr,b employees and rest were the family members. Overall, 43% were RT-PCR positive for COVID-19 (figure 1). In order to collect data on lifestyle factors, physical activities, presence of chronic disease, blood grouping and BCG vaccination, telephone interview



**Figure 1** Study profile.

was successfully done among 3382 participants. The monthly distribution of COVID-19 testing and number of test positives are illustrated in figure 2. The first case was detected in March 2020. The highest testing was done in 20 June 2020 and we observed the highest positivity rate (54%) on 21 April 2021. We observed the lowest numbers of positive cases between December 2020 and February 2021. As of 15 April 2021, 96% of all COVID-19 positive patients were recovered and there are 41 active cases. Among all COVID-19 test positives, 94.7% were mild or asymptomatic, 2.4% had moderate disease and 2.9% had a severe or critical disease. The reinfection rate was 2.1% and a total of 24 deaths including two employees and 22 family members.

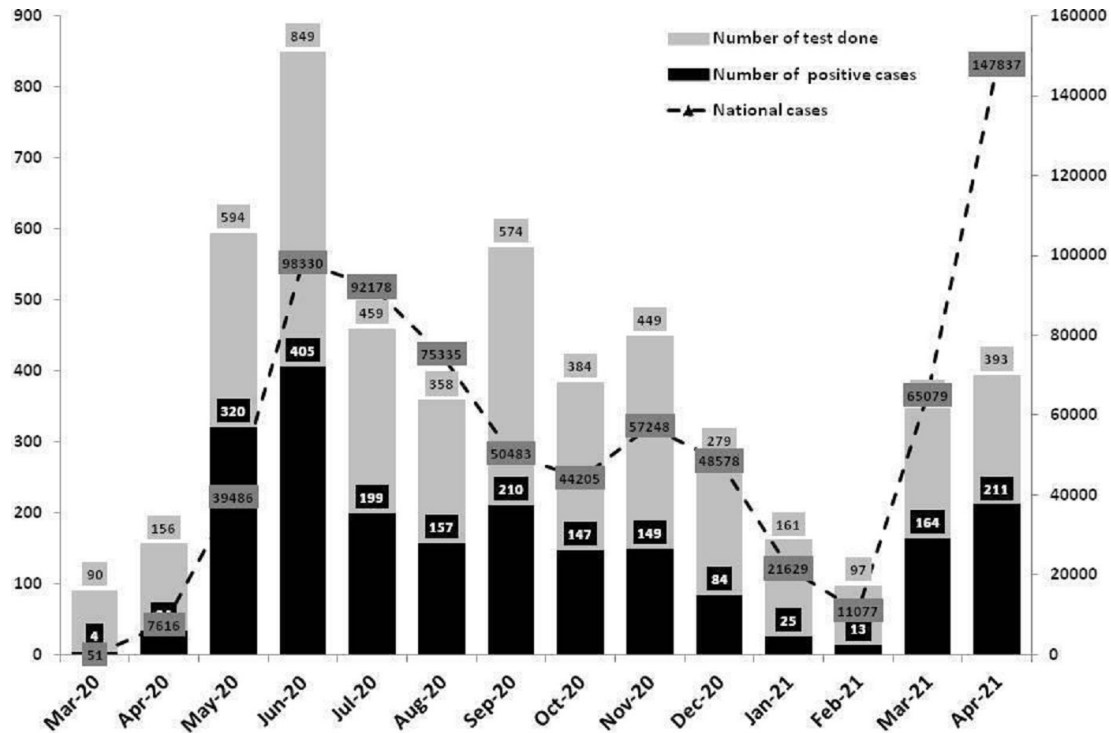
The median age of COVID-19-negative cases was 34 years, which was ranged from 2 months to 100 years and the median age of positive cases was 37 years ranged from 4 months to 88 years. Among the test-positive cases, 10% of them were less than 18 years, and this was 14% among test negatives. Age distribution of both the test positives and negatives was almost equally distributed between 18 years and 60 years. However, there were more 60+ years old people in test positives than in test negatives (10% vs 5%). Regarding sex distribution, 48% of all COVID-19 positives were women and 82% of all interviewed participants had BCG scars in their left upper arm. Regarding ABO blood groups, 23% were blood group A, 33% were blood group B and 34% were blood group O. Blood group AB was present in 11% of COVID-19-positive and 8% of negative

cases (table 1). Distribution of these above-mentioned baseline characteristics was similar in non-hospitalised test positives and negatives (online supplemental table 1).

We were able to collect additional data on presence of chronic diseases, BCG vaccination and usual physical activities through telephone interviews from 2894 participants. It was due to the fact that many were unavailable over phone during the telephone calls were made. Among all participants, 11% had a pre-existing respiratory illness. Hypertension was higher among COVID-19-positive cases. Hypertension prevalence was 22% for all COVID-19 positives compared with 17% in COVID-19 negatives. DM was more in positive cases than the negatives (15% vs 12%). The prevalence of ischaemic heart disease (4%), chronic liver disease (1%), hypothyroidism (4%) and chronic kidney disease (2%) were almost equally distributed (table 1).

Based on self-reporting data using the 'IPAC', we identified that in the preceding 7 days before interviews, overall 58% of the participants did not perform any physical activities, 35% performed mild physical activities, 5% had moderate and 3% had vigorous physical activities. Except for the vigorous physical activities, there was no difference in physical activities between COVID-19-positive and negative cases. Negative cases performed more vigorous physical activities than the positives ( $p < 0.05$ ).

Considering the symptoms before testing for SARS-CoV-2, fever was the most frequent presenting symptom followed



**Figure 2** Monthly distribution of COVID-19 test result from 19 March 2020 to 15 April 2021. Note: The bar diagram is showing monthly test results for the current study and the interrupted line diagram is showing the incidence of COVID-19 disease in Bangladesh in the same period along the second Y-axis in the right.

<sup>i</sup>The bar diagram is showing monthly test results for the current study and the interrupted line diagram is showing the incidence of COVID-19 disease in Bangladesh in the same period along the second Y-axis in the right

by cough. Fever was the most frequent presenting symptom among COVID-19 positives when compared with negative cases (70% vs 47%). Cough was present in 50% of positives and 47% of all negatives. Anosmia was a presenting symptom for 2% COVID-19-positive cases compared with 0.7% of negative cases. Sore throat was higher in COVID-19 negatives (9%) than the COVID-19 test positives (6%). Similarly, shortness of breath was higher in test negatives (4% vs 2%). Other presenting symptoms like body ache (3%), headache (0.5%) and loose motion (1%) were equally present in both the groups (table 1).

### Factors associated with COVID-19 positivity

To identify factors associated with COVID-19 positivity, multi-variable logistic regression was performed. The adjusted analysis showed that participants older than 60 years had higher odds of being COVID-19 positive than those who were younger than 18 years old (adjusted OR (aOR) 2.1, 95% CI 1.3 to 3.3;  $p < 0.05$ ) and participants with blood group AB had higher odds of being test positive than the blood group A (aOR 1.5, 95% CI 1.1 to 2;  $p < 0.05$ ). Similarly, participants presented with fever (aOR 3.1, 95% CI 2.6 to 3.7;  $p < 0.05$ ), cough (aOR 1.3, 95% CI 1.1 to 1.6;  $p < 0.05$ ) and anosmia (aOR 2.7, 95% CI 1.3 to 5.7;  $p < 0.05$ ) had higher odds of being COVID-19 positive and participants presented with sore throat were found inversely related to COVID-19 test positive (aOR 0.5, 95% CI 0.4 to 0.7;  $p < 0.05$ ) (table 2).

### DISCUSSION

The analysis showed that older age, blood group AB compared with blood group A, and presence of fever, cough and anosmia before sample collection were associated with an increased risk of COVID-19 test positivity when compared with test negatives. On the other hand, the presence of sore throat during sample collection was found negatively associated with COVID-19 test positivity.

Consistent with other published studies, older age has been one of the most common factors that have been associated with COVID-19 positivity.<sup>30-33</sup> The major presenting symptoms among COVID-19 test positives were fever and cough followed by anosmia. Other reported symptoms were cold, shortness of breath, body aches, headache, weakness, sore throat and loose motion. This finding was consistent with a recently reported retrospective cohort study from Bangladesh where they observed that the major three symptoms among COVID-19 positive patients were fever, cough and anosmia.<sup>34</sup> Although in the absence of a test negative, comparison group that study was not able to ascertain that these factors were associated with positivity.<sup>34</sup> Shortness of breath and sore throat were more common in COVID-19 test-negative patients, which were also observed in other studies.<sup>35</sup> A recent study that used COVID-19 data from five continents showed that over 50% of COVID-19 positives were asymptomatic.<sup>36</sup> The most common presenting symptom was fever (>50%), which was trailed by dry cough (45%), tiredness (38%) and

**Table 1** Baseline characteristics of staff and family members

Characteristics	N/n for each characteristic	COVID-19	
		Negative	Positive
Age group, n (%)	4284		
<18 years	4284/529	335 (14%)	194 (10%)
18–30 years	4284/1169	693 (29%)	476 (26%)
31–40 years	4284/1025	589 (24%)	436 (24%)
41–50 years	4284/723	405 (17%)	318 (17%)
51–60 years	4284/520	276 (11%)	244 (13%)
>60 years	4284/314	132 (5%)	182 (10%)
Female sex, n (%)	4295/1996	1102 (45%)	894 (48%)
BCG scar*, n (%)	2845/2347	1299 (82%)	1048 (83%)
ABO blood group†, n (%)	2689		
A	2689/630	359 (24%)	271 (23%)
B	2689/897	482 (32%)	415 (35%)
AB	2689/254	121 (8%)	133 (11%)
O	2689/908	525 (35%)	383 (32%)
Pre-existing chronic disease			
COPD‡/asthma/respiratory illness, n (%)	2894/292	169 (11%)	123 (9%)
Hypertension, n (%)	2894/557	269 (17%)	288 (22%)
Ischaemic heart disease (IHD), n (%)	2893/127	59 (4%)	68 (5%)
Chronic liver disease (CLD), n (%)	2893/36	20 (1%)	16 (1%)
Diabetes mellitus (DM), n (%)	2893/389	194 (12%)	195 (15%)
Hypothyroidism, n (%)	2893/114	59 (4%)	55 (4%)
Chronic kidney disease (CKD), n (%)	2892/53	23 (1%)	30 (2%)
Physical activity	2846		
No	2846/1668	931 (59%)	737 (58%)
Mild	2846/980	529 (34%)	451 (35%)
Moderate	2846/126	63 (4%)	63 (5%)
Vigorous	2846/72	48 (3%)	24 (2%)
Presenting symptoms	4295		
Fever, n (%)	4295/2436	1140 (47%)	1296 (70%)
Cough, n (%)	4295/2075	1145 (47%)	930 (50%)
Cold, n (%)	4295/342	201 (8%)	141 (8%)
Shortness of breath, n (%)	4295/149	105 (4%)	44 (2%)
Body ache, n (%)	4295/134	68 (3%)	66 (4%)
Headache, n (%)	4295/21	11 (0.5%)	10 (0.5%)
Sore throat, n (%)	4295/314	208 (9%)	106 (6%)
Weakness, n (%)	4295/12	6 (0.3%)	6 (0.3%)
Anosmia, n (%)	4295/50	16 (0.7%)	34 (2%)
Loose motion, n (%)	4295/38	20 (1%)	18 (1%)
Runny nose, n (%)	4295/19	14 (0.6%)	5 (0.3%)

\*BCG scar: the bacille Calmette-Guerin vaccination confirmed by the presence of a scar in the left upper arm

†ABO blood group: reported A, B, O or AB Blood group

‡COPD: known case of chronic obstructive pulmonary disease

**Table 2** Socio-demographic and clinical factors associated with COVID-19 positivity

Characteristics	OR (95% CI)	P value	aOR (95% CI)*	P value
Age in years	Reference:<18 years			
18–30 years	1.1 (0.87 to 1.39)	0.419	1.1 (0.82 to 1.49)	0.518
31–40 years	1.07 (0.84 to 1.37)	0.563	1.22 (0.89 to 1.66)	0.215
41–50 years	1.24 (0.96 to 1.6)	0.106	1.33 (0.95 to 1.87)	0.100
51–60 years	1.33 (1.01 to 1.75)	0.044	1.45 (0.98 to 2.13)	0.062
>60 years	2.2 (1.6 to 3.03)	0.000	2.05 (1.28 to 3.27)	0.003
Female sex	1.18 (1.03 to 1.35)	0.019	1.13 (0.95 to 1.34)	0.157
BCG scar	1.04 (0.86 to 1.27)	0.660		
Blood group	Reference: A group			
B group	1.14 (0.93 to 1.4)	0.209	1.13 (0.9 to 1.4)	0.287
AB group	1.46 (1.09 to 1.95)	0.012	1.46 (1.07 to 2)	0.017
O group	0.97 (0.79 to 1.19)	0.745	0.97 (0.78 to 1.21)	0.775
Pre-existing chronic disease				
COPD/asthma	0.89 (0.69 to 1.13)	0.335		
Hypertension	1.41 (1.17 to 1.7)	0.000	1.2 (0.94 to 1.53)	0.135
Ischaemic heart disease	1.44 (1.01 to 2.06)	0.045	1.13 (0.73 to 1.75)	0.578
Chronic liver disease	0.99 (0.51 to 1.91)	0.966		
Diabetes mellitus	1.28 (1.03 to 1.59)	0.023	0.9 (0.69 to 1.18)	0.452
Hypothyroidism	1.16 (0.8 to 1.68)	0.446		
Chronic kidney disease	1.63 (0.94 to 2.81)	0.083	1.29 (0.69 to 2.41)	0.430
Physical activity	Reference: No			
Mild	1.08 (0.92 to 1.26)	0.359	0.99 (0.82 to 1.18)	0.871
Moderate	1.26 (0.88 to 1.81)	0.206	1.47 (0.99 to 2.18)	0.058
Vigorous	0.63 (0.38 to 1.04)	0.071	0.64 (0.37 to 1.09)	0.102
Presenting symptoms				
Fever	2.85 (2.47 to 3.29)	0.000	3.09 (2.61 to 3.66)	0.000
Cough	1.3 (1.13 to 1.49)	0.000	1.34 (1.14 to 1.58)	0.000
Cold	0.99 (0.76 to 1.3)	0.955		
SOB	0.62 (0.43 to 0.91)	0.014	0.66 (0.42 to 1.03)	0.065
Body ache	1.21 (0.84 to 1.75)	0.295		
Head ache	2.19 (0.79 to 6.04)	0.130	1.7 (0.54 to 5.37)	0.366
Sore throat	0.66 (0.5 to 0.86)	0.003	0.52 (0.38 to 0.71)	0.000
Weakness	1.57 (0.48 to 5.17)	0.454		
Anosmia	2.65 (1.36 to 5.17)	0.004	2.69 (1.26 to 5.72)	0.010
Loose motion	0.98 (0.41 to 2.34)	0.968		

\*This model was adjusted by seasonality.

aOR, adjusted OR; BCG, bacille Calmette-Guerin ; COPD, chronic obstructive pulmonary disease; SOB, shortness of breath.

sore throat (30%).<sup>36</sup> A systematic review showed that the common symptoms were fever (83%), cough (61%), fatigue (34%), myalgia (21%), dyspnoea (22%), headache (11%) and diarrhoea (7.5%).<sup>37</sup> Similar findings were observed in other systematic reviews and studies done in other countries.<sup>89,38</sup> Therefore, inarguably fever and cough are the most common discriminatory feature of COVID-19 compared with test negatives. Loss of smell (anosmia) was the next most important clinical feature in patients with COVID-19 in our

study. Several studies also observed the similar feature that patients presented with anosmia had a higher probability of being tested positive.<sup>34,39,40</sup> Nevertheless, these results represented discriminating features between COVID-19 positives and COVID-19 suspects.

Previous studies investigated the association between human ABO blood groups and different infectious agents.<sup>41</sup> This is plausible that blood group antigens can increase host susceptibility by acting as a receptor or



coreceptor for microorganisms and viruses.<sup>41</sup> As a part of the innate immune system, ABO blood group has previously been shown to work against some enveloped viruses carrying ABO-active antigens such as SARS.<sup>41</sup> An association was reported between a higher risk for COVID-19 infection and mortality with blood group A and a lower risk of infection and mortality with blood group O.<sup>17 42</sup> However, a recent US-based multicentre study observed that patients with blood group B and AB had higher likelihood for a COVID-19 positive test result and blood type O had higher likelihood for a negative test result.<sup>19</sup> Our finding is partially consistent with the US studies as we observed participants with the AB group were more likely to test positive for SARS-CoV-2 than participants with blood group A. However, several meta-analyses and systematic reviews were published on this, and surprisingly, the results were counterintuitive.<sup>43–45</sup> One meta-analysis showed that people with blood group A are more vulnerable to COVID-19 infection and blood group AB is less susceptible to getting infected with SARS-CoV-2,<sup>43</sup> while another meta-analysis observed that both blood groups A and AB are linked to COVID-19 infection and individuals with blood group O are relatively less vulnerable.<sup>44</sup> Therefore, the association between blood group and COVID-19 positivity is still enigmatic.

Reports showed that nations with mandatory BCG vaccination had fewer numbers of patients with COVID-19.<sup>20 22</sup> Therefore, induction of trained immunity through BCG vaccination was thought to be a potentially effective approach to protect against SARS-CoV-2 infection.<sup>20–24</sup> We did not observe any association between COVID-19 infection and BCG vaccination. BCG vaccination coverage is high in Bangladesh and we observed that 82% of both the COVID-19 positives and negatives had BCG scars in the upper arm. We think a limited power could be the reason behind this non-association.

We observed that 20% of all participants had hypertension and 14% had DM. Surprisingly, around 58% of respondents did not have any physical activities, and only 34% performed mild physical activities in the preceding 7 days (table 1). According to the 'IPAC' used in this research to evaluate usual physical activities by the respondents, mild activities include only walking and do not include running or vigorous activities or exercise. Therefore, by combining 'no' and 'mild' physical activities, we can see that 92% of the participants who provided data on physical activities did not perform any physical exercise. Although we did not observe any association between COVID-19 positivity and the presence of chronic disease or physical activities, we thought this was still a very important finding. Another probable reason for this lack of association could be most of the cases were mild. Compared with national prevalence (8%–12%), the prevalence of DM is higher in this population.<sup>46</sup> The prevalence of hypertension and DM was similar to a recently published Bangladeshi study among patients with COVID-19 positive, where they also observed that these comorbidities were associated with hospitalisation.<sup>34</sup> Studies showed that the presence of chronic disease is associated with a higher risk of infection

and also increased COVID-19-associated hospitalisation.<sup>34</sup> Another reason why the current study did not show chronic conditions associated with a higher risk of COVID-19 infection is probably the test-negative case-control design of the study; since the control group was also symptomatic patients, their chance of having chronic conditions may be higher compared with the general population. If the control group was average healthy people, the results might be different.

This study was housed in a health research institute. The current staff headcount in icddr,b is 4383 with a diverse group of employees from different socioeconomic strata. These include international scientists, local scientists, doctors and senior management staff to drivers, security guards, health attendants and their families. Due to nationwide lock downs, only essential staff had been attending office in-person except those who worked in the hospital, laboratories and support services. Therefore, it was not possible to pinpoint the major source of infection. Although the data indicated that most of the infections were originated from the community.

An important concern is a high percentage of positivity (43%) in the test performed in this research which is above the global trend. Overall, the percentage of positivity is less than 10% for most of the countries.<sup>47</sup> During the pick of the pandemic, in Bangladesh, this was around 25%.<sup>48</sup> The high percentage of positivity in the current study was maybe due to a strong screening process before testing done by experienced physicians in a population who are related to health-care delivery services.

Since this study was conducted among employees and their families of an organisation, this data might not be representative of the general population of Dhaka city. However, the pattern of monthly distribution of test positivity in the current study followed a similar trend with the national test positivity rate (figure 2). Despite a considerably large sample size, the absence of any standard sampling technique for the selection of study participants is also prone to different biases.<sup>49</sup> Moreover, telephone interviews to collect data on chronic disease and physical activities were performed only on 65% of the population during the study period. There is a possibility that population characteristics may differ in 35% of the participants whose data on chronic disease are not available. This is also a limitation of the study. To address this, we compared the basic characteristics of this group with the remaining participants who had telephone interview data available and the result showed it was comparable between the groups (online supplemental table 1). The selection of variables to be studied was based on data available from the earlier period of the pandemic. Over the period, infections by new variants caused a change in disease manifestation.<sup>50</sup> There is a possible time bias in the knowledge of the population and health professionals about some symptoms not initially related to COVID-19. For example, the variable anosmia is studied but not ageusia. Another limitation is we could not adjust disease severity in a multivariable model due to the unavailability of data. It can be noted that controlling for severity could be helpful to address residual bias in



healthcare-seeking behaviour. Because residual confounding due to health-seeking behaviour may still be present in the non-hospitalised cases and controls, we have compared baseline characteristics between the non-hospitalised cases and controls, and the data were almost identical to the baseline data of all COVID-19 positives and negatives (online supplemental table 2). Finally, one more limitation of the current study is the possible change in symptoms depending on circulating variants of SARS-CoV-2 was not addressed here. Before the Omicron variant, Bangladesh observed the third wave of COVID-19 pandemic and faced a record uprising from June 2021 to September 2021, powered by the highly contagious Delta variant.<sup>48</sup> Unfortunately, the study period for this report was between March 2020 and April 2021. We first started testing for variants in January 2021.<sup>51</sup> At that time the pre-existing variant was Hu-Wuhan-like variants which were dominated till the first week of March 2021.<sup>52</sup> The Alpha variant (B.1.1.7) was discovered first in January and it gradually increased over time and became the most dominant variant in the first week of March 2021.<sup>52</sup> Since, March 2021, the SARS-CoV-2 was dominated by the Beta variant (B.1.351) which replaced almost all other variants until the emergence of the Delta variant at the beginning of May 2021.<sup>52</sup> Since we have the data on variants for only 4 months, we could not adjust this in our analysis.

Nevertheless, this study reports on factors associated with COVID-19 in a sizeable population using a high-quality growing database. The findings might not be a surprise to our recent knowledge on COVID-19, still, there has been a paucity of similar data in this part of the world. Moreover, this study also confirms that some findings like older age, fever, cough and anosmia are almost universal presentations of COVID-19 and features like the presence of chronic disease, BCG vaccination and blood groups with COVID-19 infection need more research.

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Supplementary Table 1. Characteristics of participants based on phone call success for telephone interviews

Characteristics	Phone call success	
	Yes (n=2894)	No (n=1401)
Covide-19 confirmed case	45%	40%
Age group		
< 18 years	13%	11%
18 – 30 years	28%	26%
31 – 40 years	24%	25%
41 – 50 years	17%	17%
51 – 60 years	11%	14%
> 60 years	7%	8%
Female sex	46%	48%
Fever	56%	58%
Cough	48%	48%
Cold	7%	10%
Shortness of Breath	4%	3%
Body ache	4%	2%
Headache	0.5%	0.5%
Sore throat	7%	7%
Weakness	0.3%	0.3%
Anosmia	1%	1%
Loose motion	0.6%	1%
Runny nose	0.5%	0.3%

Supplementary Table 2. Baseline characteristics of non-hospitalized COVID-19 test positives and test negatives

Characteristics	n	COVID-19	
		Negative	Positive
Age group, n (%)	4149		
< 18 years		335 (14%)	194 (10%)
18 – 30 years		689 (29%)	472 (27%)
31 – 40 years		579 (24%)	418 (23%)
41 – 50 years		404 (17%)	299 (17%)
51 – 60 years		266 (11%)	209 (12%)
> 60 years		125 (5%)	159 (9%)
Female sex, n (%)	4159	1088 (45%)	853 (49%)
BCG scar, n (%)	2772	1285 (82%)	1007 (83%)
ABO Blood group, n (%)	2619		
A		355 (24%)	262 (23%)
B		477 (32%)	397 (35%)
AB		116 (8%)	127 (11%)
O		520 (35%)	365 (32%)
<b>Pre-existing chronic disease</b>			
COPD/Asthma/Respiratory illness, n (%)	2815	164 (11%)	115 (9%)
Hypertension, n (%)	2816	259 (16%)	262 (21%)
Ischemic heart disease (IHD), n (%)	2814	57 (4%)	57 (5%)
Chronic liver disease (CLD), n (%)	2814	20 (1%)	15 (1%)
Diabetes mellitus (DM), n (%)	2816	186 (12%)	167 (13%)
Hypothyroidism, n (%)	2814	57 (4%)	53 (4%)
Chronic kidney disease (CKD), n (%)	2813	23 (1%)	27 (2%)
Physical activity	2772		
No		926 (60%)	710 (58%)
Mild		515 (33%)	428 (35%)
Moderate		62 (4%)	59 (5%)
Vigorous		48 (3%)	24 (2%)
<b>Presenting symptoms</b>	4159		
Fever, n (%)		1125 (47%)	1218 (70%)

Cough, n (%)	1133 (47%)	886 (51%)
Cold, n (%)	199 (8%)	137 (8%)
Shortness of Breath, n (%)	104 (4%)	38 (2%)
Body ache, n (%)	68 (3%)	64 (4%)
Headache, n (%)	11 (0.5%)	9 (0.5%)
Sore throat, n (%)	207 (9%)	102 (6%)
Weakness, n (%)	6 (0.3%)	6 (0.3%)
Anosmia, n (%)	16 (0.7%)	33 (2%)
Loose motion, n (%)	20 (0.8%)	17 (1%)
Runny nose, n (%)	14 (0.6%)	5 (0.3%)