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Research Article

COVID-19 Dynamics in Rajshahi Division: An RT-qPCR-Based Study

Islam MJ[1,2], Hossain MS[1], Tama RT[1], Hilaly MR[3], Parvin A[1*], Hossain MS[4], Rahman MM[1*]

[1]Department of Biotechnology and Genetic Engineering, Islamic University, Kushtia-7003
[2]Mediacal Technologist (Lab), Department of Virology, Rajshahi Medical College, Rajshahi.
[3]Department of Computer Science, Maharishi International University, Fairfield, Iowa-52557, USA
[4]Department of Statistics, Jagannath University, Dhaka-1100

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***Corresponding authors**

Parvin A, E-mail: anzana@btge.iu.ac.bd
Rahman MM, E-mail: mmrahman@btge.iu.ac.bd

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A B S T R A C T

On March 8, 2020, the case of COVID-19 patient was first reported in Bangladesh. Since then, the Directorate General of Health Services (DGHS) has announced that COVID-19 is being identified daily in Bangladesh. This study was conducted at Rajshahi Medical College, Bangladesh, from July 2022 to June 2023. The COVID-19 epidemiology and demography study were performed in the Rajshahi division, comprising four major regions (Rajshahi, Joypurhat, Natore, and Chapai Nawabganj). An analysis was conducted to determine the correlation between three variables: age, gender, and various demographic regions of the patients. A total (N=7979) COVID-19-positive cases were confirmed with reverse transcription polymerase chain reaction (RT-qPCR) at Rajshahi Medical College. Out of the age groups, the 20-29 age group exhibited the highest infection rate among the deceased COVID-19-positive cases, accounting for 21.7% (n = 1731) of the total confirmed patients. Men were more affected by COVID-19, with 61.6% (n = 4914) of the cases being male, compared to 38.4% (n = 3065) of female cases. Rajshahi district reported the highest number of positive COVID-19 cases, comprising 69.7% (n = 5564) of the total cases, followed by the remaining three regions, Joypurhat, Natore, and Rajshahi Medical College region (RMC), respectively. Furthermore, compared to rural inhabitants, urban individuals were found to be more susceptible to contracting SARS-CoV-2 infection. This study provides valuable insights for health professionals and policymakers to devise effective strategies for controlling and reducing the spread of COVID-19 in the region.

INTRODUCTION

In Wuhan City, Hubei Province, China, several cases of pneumonia—thought to be viral in nature—have been documented as of December 2019 (Yang *et al*., 2020). The disease was initially identified as the novel coronavirus, or 2019-nCoV disease, but following the Global Agreement, the World Health Organization renamed it coronavirus disease 2019 (COVID-19) (Siordia and Juan 2020) . Acute respiratory distress of severe magnitude SARS-CoV-2, also known as the coronavirus, is the virus that causes respiratory infections. COVID-19 Coronaviruses are encapsulated, zoonotic, single-stranded RNA viruses that are members of the Coronaviridae family (Ciotti *et al*., 2019). SARS-CoV-2 is more deadly than previously emerging coronaviruses like SARS-CoV and MERS-CoV, in addition to other important coronavirus characteristics. Affected individuals can spread the virus through respiratory droplets, and transmission from asymptomatic carriers increases the virus's lethality (Li *et al*., 2020). Although the virus first appeared in Wuhan, China, it has since spread throughout the world. According to recent research, the clinical transmission of COVID-19 may differ globally based on geographic location and ethnic origin (Xie *et al*., 2020). WHO formally designated COVID-19 as a global "pandemic" on March 11, 2020; Bangladesh declared its first confirmed virus case on March 8, 2020 (Mina *et al*., 2020). Initially, specific preventive measures like wearing masks, keeping social distance, and shunning crowds were strictly enforced. However, limitations are currently being loosened due to the sustained decline in the frequency of the

condition (Ferdous *et al*. 2020). However, as awareness wanes and a new round of attacks emerges, the situation becomes noticeably more difficult (Ghanbari 2020). The unpredictable effects and complex issues around public safety, uncertainty, isolation, and distance-keeping, as well as the economic fallout from COVID-19, also trigger personal psychological aspects. As a result, a variety of psychological consequences on people and groups could occur, such as emotional reactions and, in severe cases, the onset of mental health illnesses (Mina *et al.*, 2021). Most molecular laboratories in Bangladesh that use RT-PCR to identify SARS-CoV-2 were founded after the outbreak started the Real-time reverse transcription polymerase chain reaction (RT-PCR) testing volumes for SARS-CoV-2 on suspected COVID-19 patients and separated healthcare professionals increased at these sites, when faced with this unique coronavirus, some employees in public health labs ignored worries about the possibility of contracting SARS-CoV-2 from work-related exposure, one of the biggest worries during the COVID-19 pandemic is protecting laboratory workers, yet there is still a lack of sufficient information on this topic (Iwen *et al.*, 2020). Since its discovery, SARS-CoV-2 has become a pandemic. As of October 29, 2023, there have been 697,085,418 reported cases and 6,932,422 deaths worldwide (Worldometer 2020). As of October 29, 2023, there are 2,045,892 confirmed cases and 29,477 deaths in Bangladesh (Uddin *et al*., 2023). On the other hand, the Rajshahi division, which is in Bangladesh's northwest, borders India to the north and west. As one of Bangladesh's eight first-class administrative areas, it has an area of 18,174.4 square kilometers (7,017.2 square miles) and is home to 20,353,119 people (as of the census taken in 2022). The Rajshahi division, which consists of eight districts (Natore, Rajshahi, Sirajganj, Pabna, Bogra, Chapai Nawabganj, Naogaon, and Joypurhat), is unique due to its diverse population and geography (Ferdousi and Narzis 2022). These regions have different demographic, socio-economic, and cultural characteristics. Furthermore, these differences in characteristics play an important role in the epidemiology of SARS-CoV-2 as it affects populations. For economic and educational reasons, based on regional location and socioeconomic status, different groups of citizens from different places flock to the sector and participate in unrestricted freedom of movement. Taking consideration of all the information, such as the Rajshahi Division's population features, location, and demography, it is necessary to classify the many aspects related to the COVID-19 epidemic and infection trends. The aim of this work is to perform an epidemiological investigation of COVID-19-positive cases, with a particular emphasis on risk factors and molecular verification. It additionally aims to compare various parameters and look into factors that affect COVID-19 severity. By concentrating on patients who underwent testing at Rajshahi Medical College, this study investigates the prevalence of COVID-19 in Bangladesh's Rajshahi Division. It is believed that the findings will guide the development of preventive measures, assist in managing and containing the disease's spread, and enhance knowledge of the pandemic's evolution in the northwest Bangladeshi districts of Rajshahi division.

METHODOLOGY

To determine the prevalence of SARS-COVID-19 infection among the people of the Rajshahi division in 2022-2023, we need to use appropriate epidemiological methods. Here is a step-by-step guide on how to conduct this prevalence study.

Study Area

The present research was carried out in the COVID-19 diagnostic lab at the Department of Virology, Rajshahi Medical College, situated in the Rajshahi division of Bangladesh. Samples from four districts within Rajshahi were included in this study.

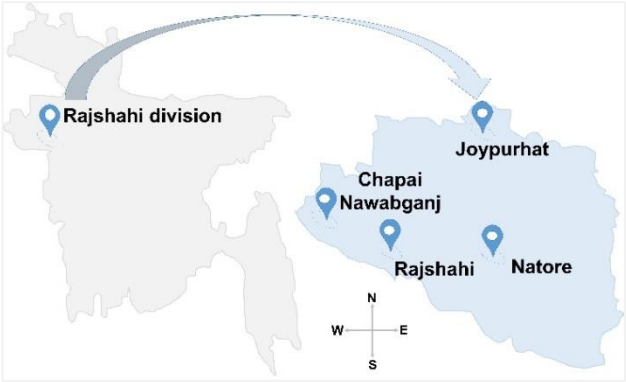

Figure 1. Diagrammatic representation of the study area (Rajshahi division of Bangladesh). Location icons indicate diverse geographical districts. Source: modified from Google Image (www.google.com)

Study Design

Choose a cross-sectional study design, as this will provide a snapshot of the prevalence of COVID-19 in the Rajshahi division during the specified time frame.

Study Population

Patients of all ages and both sexes are clinically suspected patients of COVID-19 in different districts of the Rajshahi division. We classified the population into eight (19 or lower, 20-29, 30-39, 40-49, 50-59, 60-69, 70-79, and 80 or above) distinct groups based on age. Another two (male and female) population groups were created based on sex of the patients. Based on district of Rajshahi division, the population was classified into five (Rajshahi, Joypurhat, Natore, Chapai Nawabganj and RMC) groups.

Place of Study

All the samples and relevant information were collected from the suspected patients of COVID-19 in different districts of the Rajshahi division. COVID-19 Rapid tests were carried out in Upazilla Health Complex and Sador Hospital of different districts of the Rajshahi division and RT-PCR was performed in the Molecular laboratory, Department of Virology, Rajshahi Medical College, Rajshahi.

Sample Collection and Processing

The authorities of reported health complexes collected nasopharyngeal and oropharyngeal swabs from the suspected individuals by following the WHO guidelines (Organization 2020). The swab samples were preserved in normal saline-

containing collection tubes. Immediately after the collection of samples, a proper cold chain was preserved and sent to the PCR lab at the Department of Virology, Rajshahi Medical College, Rajshahi, Rajshahi division, Bangladesh.

Storage of Sample

At the special COVID-19 diagnostic laboratory, specimens are kept at 2–8 °C for a maximum of 4 days or, in accordance with WHO guidelines, preserved in dry ice at −70 °C or below (Organization 2020). The samples are then processed in accordance with WHO interim recommendations using real-time RT-PCR, a laboratory method for qualitative in-vitro detection of COVID-19. When there is likely to be a delay in specimens reaching the laboratory, the use of a viral transport medium is strongly recommended. The collected specimens were frozen to -20°C and shipped on dry ice. It was important to avoid repeated freezing and thawing of specimens.

Packaging and Shipment of Clinical Specimens

Specimens for virus detection should reach the laboratory as soon as possible after collection. Correct handling of specimens during transportation is essential (Organization 2020). Specimens delivered promptly to the laboratory for stored and shipped at 2-8°C.

RNA Extraction and PCR Reaction

Viral RNA was recovered from patient-derived specimens using the SanSure Biotech Sample Release reagent (SanSure Biotech, China), which made use of quick RNA release technology in compliance with the manufacturer's instructions. To avoid any residue building up at the rim, a 200 µL aliquot of the material was put into a 1.5 mL Eppendorf (EP) tube, centrifuged at 12,000 rpm for 5 minutes, and the supernatant was carefully withdrawn. After that, each tube received 50 µL of sample reagent, mixed for five seconds by vortexing. The RT-PCR reaction was conducted using the lysed material directly. Following the kit instructions, 20 µL of the lysed sample and 30 µL of the master mix were added to a 0.2 mL PCR tube, which was used as the template. 26 µL of PCR mix (primers (4.62%), probes (1.15%), dNTPs (3.85%), MgCl2 (0.77%), RNasin (0.48%), and PCR buffer (89.13%)) was included in this master mix. and 4 µL of an enzyme mixture contains 37.5% Taq enzyme and 62.5 percent RT enzyme. The final PCR reaction had a total volume of 50 µL.

Real-time Polymerase Chain Reaction

The designed primers were used to amplify target specific complementary sequences of the virus genome. Using RNA-dependent DNA polymerase, cDNA copies were produced. In RT-PCR, this amplification was monitored using various fluorescence dyes or DNA probes labeled with fluorescence molecules that target a specific part of the genome. RT-PCR can be used as a one-step process or as a two-step process. Generally, the one-step RT-PCR procedure is preferred for virus detection because the whole reaction is run in a single tube with the primers and reagents in the one-step procedure (Maniruzzaman *et al.* 2022). So, we used a one-step RT-PCR procedure for the detection of the viral genome.

Primers and Probe Designing

For primer and probe designing a covid-19 testing kit was used where the prime and probe were designed. These were created to detect the SARC-CoV-2 virus. For this nucleocapsid (N), spike (S), ORF1a, ORF1b, or ORF8, RNA-dependent RNA polymerase (RdRP), and envelope (E) genes were created.

RT-PCR for the SARS-CoV-2 Detection Methods

The RT-PCR Diagnostic Panel (RT-PCRDP) is the COVID-19 PCR test method developed by the Centers for Disease Control and Prevention (CDC). Before starting, nucleic acid was isolated from clinical samples and combined with the master mix in accordance with the CDC procedure for the RT-PCRDP testing kit. The mixture was incubated for 30 minutes at 50 °C in an RT-PCR thermocycler. Then, incubation temperatures were modified to conduct experiments. Reverse transcription took place at 50 °C for 15 minutes after a 2-minute incubation period at 25 °C. Enzyme activation took place at 95 °C for 2 minutes, and amplification took place at 95–55 °C for 3–30 seconds (with probe cleavage for quenching fluorophores). It recorded real-time amplification progress and continuously checked fluorescence signals when used in conjunction with the thermocycler. We carried out separate investigations for every target, manually modifying threshold parameters. Every clinical sample had the presence of the human RNAse P (RNP) gene checked in order to guarantee specimen quality. We got positive outcomes in 35 cycles.

RESULTS

Prevalence rates of COVID-19 with different Age groups: Distribution of COVID-19 in Rajshahi Division (2022-2023)

At the COVID-19 diagnostic laboratory of Rajshahi Medical College, Rajshahi division, Bangladesh, 7,979 COVID-19-positive patients were tested to determine the prevalence of SARS-CoV-2 positive cases across various age groups. The results showed greater prevalences than those in other age groups, with 21.7% (n = 1,731) in the 20 – 29-year age group and 19.8% (n = 1,578) in the 30 – 39-year age group (Table 1 and Figure 2). Moreover, 53.8% of the patients were younger than 40 years old.

Table 1. Prevalence of COVID-19 infection in different age groups.

Frequency distribution for different age groups of COVID-19 infection				
Age Groups	**Frequency**	**Percent**	**Valid Percent**	**Cumulative Percent**
19 or lower	981	12.3	12.3	12.3
20-29	1731	21.7	21.7	34.0
30-39	1578	19.8	19.8	53.8
40-49	1266	15.9	15.9	69.6
50-59	1114	14.0	14.0	83.6
60-69	783	9.8	9.8	93.4
70-79	390	4.9	4.9	98.3
80 or above	136	1.7	1.7	100.0
Total	7979	100.0	100.0	

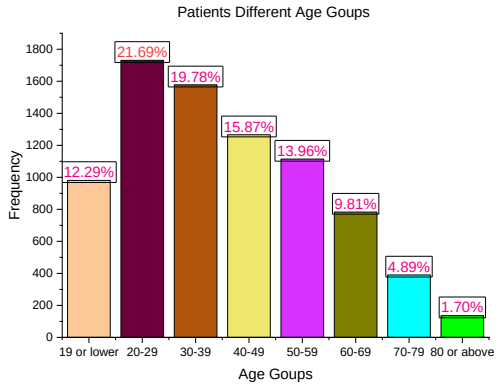

Figure 2: Percentage of COVID-19 infection in different age groups.

Gender Disparity (2022-2023)

Males exhibited a 23.2% greater incidence of positive cases than females, according to the gender-specific prevalence of 61.6% (n = 4,914) in males and 38.4% (n = 3,065) in females (Table 2 and Figures 3 and 4).

Table 2. Frequency and percentage of COVID-19 infection between males and females.

Frequency distribution for different age groups of COVID-19 infection				
Sex of the patients	**Frequency**	**Percent**	**Valid Percent**	**Cumulative Percent**
M	4914	61.6	61.6	100.0
F	3065	38.4	38.4	38.4
Total	7979	100.0	100.0	

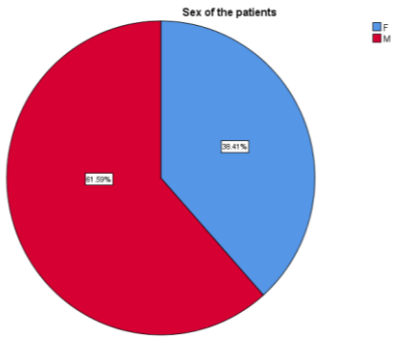

Figure 3: Percentage of COVID-19 infection between males and Females.

Demographic analysis of COVID-19 in Rajshahi division

The frequency of circulating SARS-CoV-2 positive cases varied throughout other districts in the Rajshahi division among the 7,979 persons who tested positive for COVID-19 at the COVID-19 diagnostic laboratory of Rajshahi Medical College. With a prevalence of 69.7% (n = 5,564), Rajshahi was the most prevalent, followed by Joypurhut, Natore, and Chapai Nawabganj, with respective prevalence rates of 22.3% (n = 1,782), 3.9% (n = 315), and 3.9% (n = 311). (Table 3 and Figure 4).

Table 3. Frequency and percentage of COVID-19 infection between different districts in Rajshahi Division.

Frequency distribution of COVID-19 infection in different district of Rajshahi Division				
Permanent resident of the Patients	**Frequency**	**Percent**	**Valid Percent**	**Cumulative Percent**
Rajshahi	5564	69.7	69.7	99.9
Joypurhat	1782	22.3	22.3	26.2
Natore	315	3.9	3.9	30.2
CN*	311	3.9	3.9	3.9
RMC**	7	0.1	0.1	100.0
Total	7979	100.0	100.0	

*CN=Chapai Nawabganj; **RMC= Rajshahi Medical College Area population

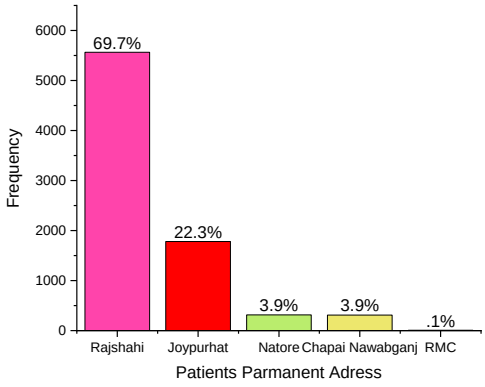

Figure 4: Graphical presentation of the percentage of COVID-19 infection between different districts in the Rajshahi division.

Age and Gender Interaction: COVID Cases in Rajshahi Division

Among the 7,979 COVID-19-positive individuals tested at the COVID-19 diagnostic laboratory of Rajshahi Medical College, Department of Virology, in Rajshahi division, Bangladesh, the prevalence of circulating SARS-CoV-2 positive cases within the age bracket of 20-29 years was 13.75% (n = 1,097) for males and 7.95% (n = 634) for females. Table 4 and Figure 5 show that these rates were higher than those of other age groups.

Table 4. COVID-19 infection of male and female in different age groups.

Age groups and sex of the patient's cross tabulation			
Age groups	**Sex of the patients**		**Total**
	F	**M**	
19 or lower	425	556	981
20-29	634	1097	1731
30-39	560	1018	1578
40-49	537	729	1266
50-59	453	661	1114
60-69	278	505	783
70-79	126	264	390
80 or above	52	84	136
Total	3065	4914	7979

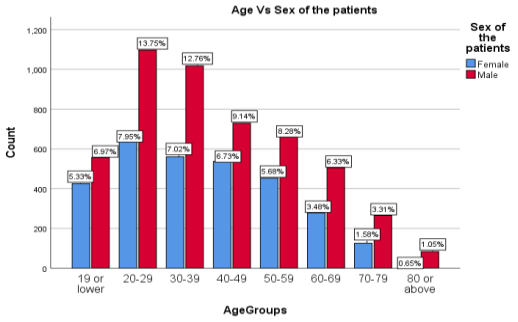

Figure 5: Percentage of COVID-19 infection of males and females in different age groups.

DISCUSSION

The study conducted at Rajshahi Medical College in Bangladesh's Rajshahi division offers significant insights into the epidemiology of COVID-19 within this specific locality. According to the results of our study, among the various age groups, the cohort aged 20–29 showed the greatest susceptibility to infection, with a prevalence rate of 21.7%, followed by the 30-39 age group at 19.8%. On the other hand, those who were older than 80 years old had the lowest incidence rate (1.7%). Mowla reported comparable outcomes in 2020 (Maniruzzaman *et al.*, 2022). The distribution of age of patients from this study reveals that middle-aged people are as likely to be infected as adults and more susceptible to the virus. However, some studies suggest that their death risk is low as they function as carriers (Mina *et al.*, 2020), (Datta *et al.*, 2020; Mina *et al.*, 2020). While all age ranges are at risk of contracting COVID-19, older individuals are at significant risk of experiencing serious illness as a result of physiological changes arising from aging and possible underlying health conditions (Islam *et al.*, 2020). Bangladesh is predominantly made up of young and middle-aged people, who are usually in the working age group, with a median age of 27.6 years. They are often required to leave their homes in order to pursue their jobs, which increases their vulnerability to infection (Rosenberg *et al.*, 2020). Our findings are consistent with preliminary observations that younger people are less likely to be affected, which have been confirmed by other study projects (Guan *et al.*, 2020; Jahan *et al.*, 2020). Our research findings are consistent with another study conducted in Bangladesh that found a substantial correlation between age and the outcome of the COVID-19 risk factor. Strong evidence of a relationship between age and COVID-19 risk was found in another Bangladeshi study, which is consistent with our own findings (Pan *et al.*, 2020). All of these outcomes point to practical actions to safeguard and contain transmission, thus inhibiting the advancement of SARS-CoV-2 within vulnerable communities. The prevalence of COVID-19 was found to be 61.6% (n = 4,914) in males and 38.4% (n = 3,065) in females, according to our study. This indicates a 23.2% increase in positive cases in males as opposed to females. This data is consistent with the results of other studies carried out in Wuhan, Italy, Oman, and the United States, which suggest that COVID-19 disproportionately affects men (Cummings *et al.*, 2020; Jin *et al.* 2020; Khamis *et al.* 2020; Zhu *et al.* 2020). Further research indicates that men exhibit a higher frequency, with rates of 61.6%, 58.1%, and 67% in China, New York, and accordingly (Jehi *et al.*, 2020; Yang *et al.*, 2020; Zhang *et al.*, 2020). Differences in physiological characteristics, such as the development of less effective viral receptors, metabolic variations, and distinct behavioral tendencies among males and females, may account for the gender gap in infection rates. Comparable results from the Cleveland Clinics in Florida and Ohio suggested that men were more likely to test positive for COVID-19 (Zhang *et al.*, 2020). While most studies have found that it occurs more frequently in men, others have also suggested that the sex distributions are roughly equal (Khamis *et al.* 2020; Mollalo *et al.*, 2020). The study reveals the differences in SARS-CoV-2 infection rates between urban and rural environments in four districts in the Rajshahi division of Bangladesh: Rajshahi, Chapai Nawabganj, Natore, and Joypurhat. Samples collected from these regions were diagnosed with COVID-19 at Rajshahi Medical College in Rajshahi. Among the districts, the Rajshahi district had the highest percentage of SARS-CoV-2 prevalence, at 69.7%, compared to the other districts. The regional variation in SARS-CoV-2 infection rates among the previously listed districts is further clarified by our analysis. In particular, the largest percentage of COVID-19-positive cases was found in the Rajshahi district (69.7%; n = 5,564). Joypurhat, Chapai Nawabganj, and Natore districts had the next-highest percentages, 22.3% (n = 1,782), 3.9% (n = 311), and 3.9% (n = 315), respectively. The Rajshahi district has about 65.8% higher COVID-19 prevalence than the Natore district. Assessing the pandemic's state in relation to a range of criteria is essential in order to understand its prevalence, intensity, geographical spread pattern, possible risk factors, susceptibility, risk thresholds, and infection hotspots. Prior research has demonstrated a positive relationship between the incidence of COVID-19 and environmental, socioeconomic, and demographic factors (Hossain *et al.*, 2020). The COVID-19 crisis is expected to be a long-term phase and just having the correct facts and exercising the prescribed health alerts can help fight against this pandemic. A recent study on knowledge, attitudes and fear of COVID-19 during the rapid increase in Bangladesh reported a high prevalence of self-isolation, positive preventive health behaviors linked to COVID-19, and moderate to high levels of fear among Bangladeshi people (Hossain *et al.*, 2020). In our view, the current research will shed light on the epidemiological dimensions during the crisis era of COVID-19. Although the research shows facts that correlate to those that were identified globally, such findings need further study and surveillance in aspect of Bangladesh's different geographical dimensions.

CONCLUSIONS

Clinical data from the COVID-19 testing facility at Rajshahi Medical College, situated in the northwest region of Bangladesh in the Rajshahi division, is analyzed in this study. According to our research, men—especially those under the age of thirty—who live in cities are more likely to get COVID-19 infections. Among the four districts of Rajshahi division, Rajshahi district exhibits the highest rate of COVID-19-positive cases compared to the other districts. Our research reveals the high incidence of SARS-COVID-19 in the Rajshahi division, underscoring the area's susceptibility to the worldwide pandemic. This study provides a clear understanding of the dynamics of the virus's propagation, even though our knowledge of it is still restricted. As such, it facilitates the implementation of improved containment strategies to counteract the COVID-19 pandemic throughout the nation, including several

districts in the Rajshahi division. Still, a number of factors, including social behavior, differences in immune system responses, and occupational exposure, could contribute to the higher incidence in men. Furthermore, the Rajshahi district showed a significantly higher frequency than other districts, maybe as a result of growing urbanization and population density. Additional investigation into the long-term health consequences of SARS-COVID-19 is needed, with a particular emphasis on any aftereffects on those who have recovered. To strengthen global readiness and responsiveness to new infectious diseases, it is imperative to actively support and engage in international collaboration for the purposes of data sharing, research, and response strategy development. The main conclusion drawn from this research is that the interaction between the COVID-19-causing virus and its geographic setting places the most vulnerable members of society at unequal risk. This indicates that in order to effectively address the issues posed by COVID-19, there might be a need to allocate healthcare resources more fairly across areas.

Ethical approval

Head of the Department of Microbiology at Rajshahi Medical College granted approval for this study.

Transparency statement

The data are included with this manuscript; any further information or data needed can be obtained by contacting the corresponding authors.

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Declaration of interests

All authors declare no competing interests.

REFERENCES

Ciotti M, Angeletti S, Minieri M, Giovannetti M, Benvenuto D and Pascarella S 2019: "COVID-19 outbreak: an overview. Chemotherapy." In.: karger. com, https://www. karger. com/Article/Abstract/507423.

Cummings, Matthew J, Matthew R Baldwin, Darryl Abrams, Samuel D Jacobson, Benjamin J Meyer, Elizabeth M Balough, Justin G Aaron, Jan Claassen, LeRoy E Rabbani, and Jonathan Hastie. 2020: 'Epidemiology, clinical course, and outcomes of critically ill adults with COVID-19 in New York City: a prospective cohort study', *The lancet*, **395**: 1763-70.

Datta, A, RM Dipi, SN Mostafa, MN Islam, and PP Das. 2020: 'Evaluation of Diagnostic Performance of Chest CT with RT-PCR in Suspected Coronavirus Patients in Bangladesh', *Mymensingh Medical Journal: MMJ*, **29**: 964-68.

Ferdous, Zannatul M, Islam MS, Sikder MT, Mosaddek ASM, Zegarra-Valdivia JA and Gozal D 2020: 'Knowledge, attitude, and practice regarding COVID-19 outbreak in Bangladesh: An online-based cross-sectional study', *PloS one*, **15**: e0239254.

Ferdousi M and Narzis A 2022: "GIS and Remote Sensing-Based Agricultural and Meteorological Drought Assessment in Rajshahi Division." In *International Conference on Advances in Civil Engineering*, 13-24. Springer.

Ghanbari, Behzad 2020: 'On forecasting the spread of the COVID-19 in Iran: The second wave', *Chaos, Solitons & Fractals*, **140**: 110176.

Guan, Wei-jie, Zheng-yi Ni, Yu Hu, Wen-hua Liang, Chun-quan Ou, Jian-xing He, Lei Liu, Hong Shan, Chun-liang Lei, and David SC Hui. 2020: 'Clinical characteristics of coronavirus disease 2019 in China', *New England journal of medicine*, **382**: 1708-20.

Hossain, Anwar M, Jahid MIK, Hossain KMA, Walton LM, Uddin Z, Haque MO, Kabir MF, Arafat SMY, Sakel M and Faruqui R 2020: 'Knowledge, attitudes, and fear of COVID-19 during the Rapid Rise Period in Bangladesh', *PloS one*, **15**: e0239646.

Islam, Taimur, Talukder AK, Siddiqui N and Islam T 2020: 'Tackling the COVID-19 pandemic: The Bangladesh perspective', *Journal of public health research*, 9: jphr. 2020.1794.

Iwen, Peter C, Karen L Stiles, and Michael A Pentella. 2020: 'Safety considerations in the laboratory testing of specimens suspected or known to contain the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2)', *American journal of clinical pathology*, **153**: 567-70.

Jahan, Yasmin, Rahman S and Rahman A 2020: 'COVID-19: A case report from Bangladesh perspective', *Respiratory medicine case reports*, **30**: 101068.

Jehi, Lara, Xinge Ji, Alex Milinovich, Serpil Erzurum, Brian P Rubin, Steve Gordon, James B Young, and Michael W Kattan. 2020: 'Individualizing risk prediction for positive coronavirus disease 2019 testing: results from 11,672 patients', *Chest*, **158**: 1364-75.

Jin, Jian-Min, Peng Bai, Wei He, Fei Wu, Xiao-Fang Liu, De-Min Han, Shi Liu, and Jin-Kui Yang. 2020: 'Higher severity and mortality in male patients with COVID-19 independent of age and susceptibility'.

Khamis, Faryal, Badria Al Rashidi, Ibrahim Al-Zakwani, Ahmed H Al Wahaibi, and Salah T, Al Awaidy 2020: 'Epidemiology of COVID-19 infection in Oman: analysis of the first 1304 cases', *Oman medical journal*, **35**: e145.

Li, Yanli, Jie Shi, Jianbo Xia, Jie Duan, Lijuan Chen, Xudong Yu, Weishun Lan, Quanfu Ma, Xufeng Wu, and Yichong Yuan. 2020: 'Asymptomatic and symptomatic patients with non-severe coronavirus disease (COVID-19) have similar clinical features and virological courses: a retrospective single center study', *Frontiers in microbiology*, **11**: 1570.

Maniruzzaman M, Islam MM, Ali MH, Mukerjee N, Maitra S, Kamal MA, Ghosh A, Melvin A, Castrosanto, Alexiou A, Ashraf MG 2022: 'COVID-19 diagnostic methods in developing countries', *Environmental Science and Pollution Research*, 29: 51384-97.

Mina, Begum F, Billah M, Karmakar S, Das S, Rahman MS, Hasan MF and Acharjee UK 2021: 'An online observational study assessing clinical characteristics and impacts of the COVID-19 pandemic on mental health: a

perspective study from Bangladesh', *Journal of Public Health*: 1-9.

Mina, Begum F, Billah M, Rahman MS, Das S, Karmakar S, Faruk M and Hasan UK 2020: 'COVID-19: transmission, diagnosis, policy intervention, and potential broader perspective on the rapidly evolving situation in Bangladesh', *J Adv Biotechnol Exp Ther*, **3**: 18-29.

Abolfazl M, Vahedi B and Rivera KM 2020: 'GIS-based spatial modeling of COVID-19 incidence rate in the continental United States', *Science of the total environment*, **728**: 138884.

Organization, World Health 2020: "Laboratory testing for coronavirus disease 2019 (COVID-19) in suspected human cases: interim guidance, 2 March 2020." In.: World Health Organization.

Pan, An, Li Liu, Chaolong Wang, Huan Guo, Xingjie Hao, Qi Wang, Jiao Huang, Na He, Hongjie Yu, and Xihong Lin. 2020: 'Association of public health interventions with the epidemiology of the COVID-19 outbreak in Wuhan, China', *Jama*, **323**: 1915-23.

Rosenberg, Eli S, Elizabeth M Dufort, Debra S Blog, Eric W Hall, Dina Hoefer, Bryon P Backenson, Alison T Muse, James N Kirkwood, Kirsten St. George, and David R Holtgrave. 2020: 'COVID-19 testing, epidemic features, hospital outcomes, and household prevalence, New York State—March 2020', *Clinical Infectious Diseases*, **71**: 1953-59.

Serafini, Gianluca, Gonda X, Canepa G, Pompili M, Rihmer Z, Amore M and Engel-Yeger B 2017: 'Extreme sensory processing patterns show a complex association with depression, and impulsivity, alexithymia, and hopelessness', *Journal of affective disorders*, 210: 249-57.

Siordia Jr, Juan A 2020: 'Epidemiology and clinical features of COVID-19: A review of current literature', *Journal of Clinical Virology*, **127**: 104357.

Uddin, Mahtab, Shawpnil K, Mugdha SBS and Ahmed A 2023: 'Research Article A Statistical Synopsis of COVID-19 Components and Descriptive Analysis of Their Socio-Economic and Healthcare Aspects in Bangladesh Perspective', *People*, 10.

Worldometer, Coronavirus Update. 2020: "Cases and deaths from Covid-19 virus pandemic." In.: Worldometers.

Xie, Yewei, Zaisheng Wang, Huipeng Liao, Gifty Marley, Dan Wu, and Weiming Tang. 2020: 'Epidemiologic, clinical, and laboratory findings of the COVID-19 in the current pandemic: systematic review and meta-analysis', *BMC infectious diseases*, **20**: 1-12.

Yang, Xiaobo, Yuan Yu, Jiqian Xu, Huaqing Shu, Hong Liu, Yongran Wu, Lu Zhang, Zhui Yu, Minghao Fang, and Ting Yu. 2020: 'Clinical course and outcomes of critically ill patients with SARS-CoV-2 pneumonia in Wuhan, China: a single-centered, retrospective, observational study', *The lancet respiratory medicine*, **8**: 475-81.

Zhang, Jin-jin, Xiang Dong, Yi-yuan Cao, Ya-dong Yuan, Yi-bin Yang, You-qin Yan, Cezmi A Akdis, and Ya-dong Gao. 2020: 'Clinical characteristics of 140 patients infected with SARS-CoV-2 in Wuhan, China', *Allergy*, 75: 1730-41.

Zhu, Na, Zhang D, Wang W, Li X, Yang B, Song J, Zhao X, Huang B, Shi W and Lu R 2020: 'A novel coronavirus from patients with pneumonia in China, 2019', *New England journal of medicine*, **382**: 727-33.