SARS-CoV-2 Seroprevalence before Delta Variant Surge, Chattogram, Bangladesh, March–June 2021

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A March–June 2021 representative serosurvey among Sitakunda subdistrict (Chattogram, Bangladesh) residents found an adjusted prevalence of severe acute respiratory syndrome coronavirus 2 antibodies of 64.1% (95% credible interval 60.0%–68.1%). Before the Delta variant surge, most residents had been infected, although cumulative confirmed coronavirus disease incidence was low.

Through November 9, 2021, Bangladesh had reported >1.57 million COVID-19 cases and 27,904 deaths (*1*), with incidence and mortality rates substantially lower than in many other countries. Without performing population-based seroprevalence estimates, it is difficult to know whether differences in rates of illness and death result from undercounts because of limited surveillance and healthcare seeking or reflect actual differences in incidence resulting from interventions or different biological responses to infection. In early March 2021, cases across Bangladesh began to rise at the same time as the Delta variant was detected in neighboring India. Publicly available sequencing data (*2*) indicate that the SARS-CoV-2 Delta variant was first detected in the Chattogram region of Bangladesh in mid-May 2021, and 99% (98/99) of the viral genomes submitted during July 1–October 1, 2021 have been of the Delta variant, similar to national trends.

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The Study

We conducted a representative serosurvey to understand the prevalence of total SARS-CoV-2 antibodies in residents of the Sitakunda subdistrict (Chattogram district) of Bangladesh, a region with an urban-to-rural gradient that includes Chattogram, Bangladesh's second largest city. We conducted the survey over 2 periods, March 27–April 13 and May 23–June 13, because of a national COVID-19 lockdown (April 14–May 30). We used 2-stage sampling based on digitized satellite imagery by first dividing the Sitakunda subdistrict into 1 km^2 grid-cells (or clusters) and randomly selecting grid-cells proportional to the estimated number of households in each, with replacement. We then randomly selected structures weighted by whether they were multistory or single-story. We attempted to enroll all persons ≥1 year of age in each household.

We tested participant serum for total antibodies (IgA, IgM, and IgG) against the receptor-binding domain of SARS-CoV-2 using the SARS-CoV-2 Ab ELISA (Wantai BioPharm, https://www.ystwt.cn), following manufacturer instructions. We corrected seroprevalence estimates for imperfect test performance, household clustering, and individual-level covariates (e.g., age) using a Bayesian modeling approach documented elsewhere and stratified results to match the target population (*3*). Our study was approved by the icddr,b research and ethics review committee and the Johns Hopkins Bloomberg School of Public Health institutional review board.

Given limited data on the immunoassay's performance in south Asia and performance months after infection, we conducted a validation study to estimate its sensitivity and specificity by testing samples from 214

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healthy participants from a 2014 cholera vaccine study and 81 from 52 symptomatic PCR-confirmed SARS-CoV-2–infected patients; none of the positive controls had been hospitalized or vaccinated for COVID-19. We collected samples 3–275 days after symptom onset. We estimated specificity at 99.1% (95% CI 96.7%–99.9%, n = 212/214) and sensitivity at 87.7% (95% CI 78.5%–93.9%, n = 71/81) for detecting previous infection with little evidence of sensitivity decreasing over time after infection (Appendix Table 4, https://wwwnc.cdc.gov/EID/article/28/2/21-1689-App1.pdf).

We enrolled 580 households and 2,307 participants who provided a blood sample. Most participants (54%, n = 1,235/2,307) were female and the median age was 28 (interquartile ratio 16–45) years; most reported working at home (37%), going to school (29%), or conducting business outside of their home (20%) as their main occupation in the month before enrollment. Among all participants, 22 (0.95%) reported ever having a COVID-19 test; 3/22 had positive results (all 3 were also seropositive in the study). Of 2,307 participants, 125 (5.4%) reported being vaccinated (15–144 days before interview) with ≥1 dose of SARS-CoV-2 vaccines, including 117 with a CoviShield ChAdOx1 (Serum Institute of India, https://www.seruminstitute.com) vaccination card and 1 with a Pfizer/BioNTech BNT162n2 (https://www.pfizer.com) vaccination card. As of June 19, 2021, 6 days after the end of the survey, 6.0% of the entire Chattogram district population was reported to have received ≥1 dose of any vaccine; 4.6% had received 2 doses (*4*).

There were 1,443 (63%) seropositive participants. Nearly all (98%) who reported having been partially (47/49) or completely vaccinated (75/76) were seropositive. In 85% of enrolled households, ≥1 participant was seropositive and an average of 62% of participants in each household were seropositive. We estimated that 31% of the total variability in seropositivity in the community was attributable to variation in seropositivity between households (intraclass correlation coefficient 0.31, 95% CI 0.27–0.36). We found evidence of a gradient in seropositivity associated with population density. Participants living in higher population density areas were significantly more likely to be seropositive: 69% of participants living in the most population-dense areas were seropositive compared with 52% of participants living in the least population-dense areas (p<0.0001; Appendix Table 1). We found similar results using alternative metrics related to urbanicity (Appendix Table 1). Among seropositive participants, 57% (815/1,442) reported having had ≥1 COVID-consistent symptom since April 2020 and 58% (474/812) of these participants reported seeking healthcare.

Adjusting for age, sex, household clustering, and test performance, we estimated the seroprevalence of SARS-CoV-2 in Sitakunda to be 64.1% (95% credible interval [CrI] 60.0%–68.1%) among all participants and 63.4% (95% CrI 59.2%–67.6%) when considering only unvaccinated participants (Table; Appendix Table 3). We estimated a 7% (95% CrI 1%–13%) higher risk of being seropositive in men compared with women. Risk generally increased with age, with those <10 years of age having the lowest risk, including a ≥34% lower risk of being seropositive compared with those 25–34 years of age (Table; Appendix Table 3). We found similar adjusted seroprevalences in the population recruited before the lockdown (63.1%, 95% CrI 56.2%–69.8%; n = 665) and after the lockdown (65.3%, 95% CrI 60.6%–69.9%; n = 1,643). In between the 2 survey rounds, during the

Table. Overview of SARS-CoV-2 seropositivity, seroprevalence and relative risk seropositivity in Sitakunda subdistrict, Chattogram district, Bangladesh*

Variable	Observations, no.	Positive, no. (%)	Negative, no. (%)	Adjusted seroprevalence, % (95% CrI)	Adjusted relative risk, % (95% CrI)
Age, y					
1–4	90	37 (41.1)	53 (58.9)	47.1 (37.0–57.3)	0.66 (0.51–0.81)
5–9	174	71 (40.8)	103 (59.2)	45.0 (37.1–52.9)	0.63 (0.51–0.74)
10–14	258	140 (54.3)	118 (45.7)	58.8 (52.0–65.3)	0.83 (0.73–0.94)
15–24	482	305 (63.3)	177 (36.7)	67.2 (61.7–72.6)	0.96 (0.88–1.05)
25–34	381	258 (67.7)	123 (32.3)	69.7 (64.5–75.0)	Referent
35–44	325	225 (69.2)	100 (30.8)	74.0 (68.3–79.5)	1.07 (0.97–1.17)
45–54	250	180 (72.0)	70 (28.0)	73.8 (67.2–80.3)	1.06 (0.96–1.17)
55–64	208	132 (63.5)	76 (36.5)	69.0 (62.1–75.8)	0.99 (0.88–1.10)
≥65	139	95 (68.3)	44 (31.7)	73.6 (65.8–81.1)	1.06 (0.94–1.19)
Sex					
M	1,072	690 (64.4)	382 (35.6)	66.7 (62.2–71.3)	1.07 (1.02–1.13)
F	1,235	753 (61.0)	482 (39.0)	61.3 (56.9–65.6)	Referent
Overall	2,307	1,443 (62.5)	864 (37.5)	64.1 (60.0–68.1)	NA

*Adjusted estimates account for sex, age, household clustering, and test performance among all vaccinated and unvaccinated participants. CrI, credible interval; NA, not applicable.

lockdown, the number of clinical cases district-wide decreased and, likely as a result of the Delta variant, began to increase during the end of the second round of data collection (*4*).

In the catchment area of this serosurvey, only 1 healthcare facility (Bangladesh Institute of Tropical and Infectious Diseases) provided SARS-CoV-2 PCR testing. Among the 2,400 participants who had a reverse transcription PCR test during April 2020–May 31, 2021, a total of 705 (29%) tested positive. By crudely extrapolating our serologic estimates by multiplying the estimated population size by the adjusted seroprevalence among those who were unvaccinated, we estimated that >200,000 infections occurred during the same period in Sitakunda. Assuming all positive cases were from Sitakunda and not neighboring areas, this corresponds to a minimum of 300 infections per medically confirmed case, a much higher proportion than has been documented in most settings across the world (*5*,*6*).

Conclusions

These results illustrate that prior to the June 2021 surge in COVID-19 cases in Bangladesh fueled by the Delta variant, most of the population in Sitakunda had already been infected despite a relatively low incidence of reported virologically confirmed SARS-CoV-2 infections. Key limitations to these results include the relatively small geographic area covered by the survey and that we only assessed circulating antibodies to a single SARS-CoV-2 epitope, which does not fully capture the immune profile of participants.

In Bangladesh, where cases captured by surveillance are limited by healthcare seeking, even in population-dense settings, representative seroprevalence surveys can help with continuing to track the evolution of this pandemic. In addition to providing important validation data on a widely used immunoassay, our results help lay the foundation for understanding the role of variant strains on key epidemiologic parameters, including our understanding of reinfection, and help set expectations for SARS-CoV-2 control in the months to come, in the study area and beyond.

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About the Author

Dr. Bhuiyan is a scientist at icddr,b. His research interests include immunology, vaccinology, and enteric infections.

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Appendix

Appendix Table 1. Descriptive statistics for 2,307 serosurvey participants in Sitakunda Upazila by seropositivity*

Characteristics†	Negative, no. (%)	Positive, no. (%)	Total	P value
N	864	1,443	2,307	NA
Sociodemographic				
Age, median (range)	23 (1–92)	31 (1–97)	28 (1–97)	NA
Age, y				<0.0001
1–4	53 (59)	37 (41)	90	
5–9	103 (59)	71 (41)	174	
10–14	118 (46)	140 (54)	258	
15–24	177 (37)	305 (63)	482	
25–34	123 (32)	258 (68)	381	
35–44	100 (31)	225 (69)	325	
45–54	70 (28)	180 (72)	250	
55–64	76 (37)	132 (63)	208	
≥65	44 (32)	95 (68)	139	
Sex				1.0
M	482 (39)	753 (61)	1,235	
F	382 (36)	690 (64)	1,072	
Main activity in previous mo				0.2
Business outside home	137 (30)	319 (70)	456	
Child	54 (56)	42 (44)	96	
Farmer	34 (42)	46 (57)	80	
Homemaker	293 (35)	555 (65)	848	
Not worked (adult)	30 (42)	41 (58)	71	
Other	20 (24)	65 (76)	85	
Student	296 (44)	374 (56)	670	
Highest educational attainment				<0.0001
No schooling	167 (44)	209 (56)	376	
Primary	304 (42)	413 (58)	717	
Lower secondary	238 (35)	446 (65)	684	
Upper secondary	125 (30)	285 (70)	410	
Bachelors	24 (26)	67 (74)	91	
Postgraduate	4 (15)	23 (85)	27	
Household monthly income‡, USD				0.6
<12	0 (0)	0 (0)	0	
12–35	22 (25)	66 (75)	88	
35–59	21 (46)	25 (54)	46	
59–83	18 (36)	32 (64)	50	
83–118	79 (42)	107 (58)	186	
118–236	348 (38)	559 (62)	907	
>236	376 (37)	654 (63)	1,030	
Household size‡, median (range)	5 (2–18)	5 (1–18)	5 (1–18)	NA
Measures of urbanicity				
Friction§,¶,#				<0.0001
0.001–0.0012	166 (31)	371 (69)	537	
0.0012–0.0014	210 (36)	374 (64)	584	
0.0014–0.0018	189 (32)	410 (68)	599	
0.0018–0.0023	299 (51)	288 (49)	587	
Population density, per 1 km^2††,‡‡				<0.0001
517–2,354	250 (48)	273 (52)	523	
2,354–3,708	237 (39)	371 (61)	608	

Characteristics†	Negative, no. (%)	Positive, no. (%)	Total	P value
3,708–5,382	178 (34)	349 (66)	527	
5,382–11,360	199 (31)	450 (69)	649	
Household distance from Chittagong port**, m				<0.001
7,799–15,367	178 (32)	375 (68)	553	
15,367–26,595	212 (36)	375 (64)	587	
26,595–37,003	222 (39)	348 (61)	570	
37,003–46,579	252 (42)	345 (58)	597	
COVID-19–related factors				
Symptoms in previous mo§§	54 (34)	107 (66)	161	NA
Symptoms after 14 Apr 2020				0.2
0	374 (37)	627 (63)	1,001	
1	120 (36)	211 (64)	331	
2	166 (36)	294 (64)	460	
3	138 (39)	216 (61)	354	
4	46 (41)	65 (59)	111	
5	8 (26)	23 (74)	31	
6	4 (80)	1 (20)	5	
7	2 (29)	5 (71)	7	
Doctor or hospital care for symptoms after 14 Apr 2020				0.2
No	228 (40)	344 (60)	571	
Yes	264 (36)	474 (64)	738	
COVID-19 testing and vaccination				
Ever tested for COVID				0.3
No	857 (38)	1,427 (62)	2,284	
Once	6 (29)	15 (71)	21	
Multiple times	1 (100)	0 (0)	1	
Test result				0.2
Negative	6 (33)	12 (67)	18	
Positive	0 (0)	3 (100)	3	
Inconclusive	0 (0)	0 (0)	0	
Received COVID-19 vaccine¶¶				<0.0001
No	861 (39)	1,320 (61)	2,181	
1 dose	2 (4.1)	47 (96)	49	
2 doses	1 (1.3)	75 (99)	76	
Unknown	0 (0)	1 (100)	1	
Vaccine type				<0.0001
CoviShield/ChAdOx1	2 (1.7)	115 (98)	117	
Pfizer	0 (0)	1 (100)	1	
COVID-19–related behaviors after 14 Apr 2020				
Mask use				1.0
No	274 (40)	413 (60)	687	
Yes	589 (36)	1,030 (64)	1,619	
Mask frequency in previous week				0.5
Never	9 (50)	9 (50)	18	
1–2 times	92 (35)	173 (65)	265	
3–5 times	120 (40)	178 (60)	298	
Almost every day	366 (35)	667 (65)	1,033	
Public transportation use change##				0.2
No change	62 (50)	63 (50)	125	
Less use	345 (34)	672 (66)	1,017	
1–2 more times per day	0 (0)	0 (0)	0	
3–5 more times per day	1 (50)	1 (50)	2	
Continued change in public transportation use	335 (34)	663 (66)	998	NA

*NA, not applicable
†Chi-squared tests for trend were performed on categorical variables with numerically increasing categories and Pearson chi-squared tests of homogeneity were performed on all other categorical variables.
‡Household level proportions.
§Data from the Malaria Atlas Project (https://malariaatlas.org).
¶Sampling cluster level measures (1km^2).
#Minutes to travel 1 m.
**Lower no. = more urban.
††Data sourced from WorldPop (https://www.worldpop.org).
‡‡Lower no. = more rural.
§§COVID-like symptoms include: fever, cough, shortness of breath, loss of taste/smell, nausea, diarrhea, and vomiting.
¶¶Only 245 participants confirmed their vaccination status, whether having received it or not, by vaccination card. 94% (117/125) of participants that reported $\geq$1 dose of vaccination confirmed their vaccination status by a vaccination card.
##Change compared to use before April 14th, 2020. This field is restricted to only those who said they used public transport before April 14, 2020.

Appendix Table 2. Descriptive statistics for unvaccinated, serosurvey participants (n = 2,181) by seropositivity in Sitakunda Upazila. This table includes sociodemographic factors, measures of urbanicity, COVID-like symptoms, COVID testing and vaccination, and COVID-19–related behaviors.

Characteristics†	Negative, no. (%)	Positive, no. (%)	Total, no.	P value
N	268	371	2,181	NA
Sociodemographic				
Age, median (range)	23 (1–92)	29 (1–97)	26 (1–97)	NA
Age, y				<0.0001
1–4	53 (59)	37 (41)	90	
5–9	103 (59)	71 (41)	174	
10–14	118 (46)	140 (54)	258	
15–24	176 (37)	303 (63)	479	
25–34	123 (33)	249 (67)	372	
35–44	100 (34)	198 (66)	298	
45–54	69 (32)	147 (68)	216	
55–64	76 (44)	97 (56)	173	
≥65	43 (36)	78 (64)	121	
Sex				1.0
M	480 (41)	700 (59)	1,180	
F	381 (38)	620 (62)	1,001	
Main activity in previous mo				0.2
Business outside home	136 (32)	284 (68)	420	
Child	54 (56)	42 (44)	96	
Farmer	34 (45)	42 (55)	76	
Homemaker	291 (37)	494 (63)	785	
Not worked (adult)	30 (49)	31 (51)	61	
Other	20 (27)	53 (73)	73	
Student	296 (44)	373 (56)	669	
Highest educational attainment				<0.0001
No schooling	166 (45)	199 (55)	365	
Primary	302 (44)	392 (56)	694	
Lower secondary	238 (37)	414 (63)	652	
Upper secondary	125 (34)	248 (66)	373	
Bachelors	24 (32)	51 (68)	75	
Postgraduate	4 (20)	16 (80)	20	
Household monthly income‡, USD				0.5
<12	0 (0)	0 (0)	0	
12–35	22 (28)	58 (72)	80	
35–59	21 (49)	22 (51)	43	
59–83	18 (36)	32 (64)	50	
83–118	78 (43)	102 (57)	180	
118–236	347 (40)	517 (60)	864	
>236	375 (39)	589 (61)	964	
Household size‡, median (range)	5 (2,18)	6 (1,18)	5 (1,18)	NA
Measures of urbanicity				
Friction§,¶,#				<0.0001
0.001–0.0012	165 (33)	331 (67)	496	
0.0012–0.0014	209 (38)	348 (62)	557	
0.0014–0.0018	189 (34)	375 (66)	564	
0.0018–0.0023	298 (53)	266 (47)	564	
Population density, per 1 km^2††,‡‡				<0.0001
517–2,354	249 (50)	250 (50)	499	
2,354–3,708	236 (40)	347 (60)	583	
3,708–5,382	178 (36)	313 (64)	491	
5,382–11,360	198 (33)	410 (67)	608	
Household distance from Chittagong port**, m				<0.001
7,799–15,367	177 (35)	336 (65)	513	
15,367–26,595	212 (37)	355 (63)	567	
26,595–37,003	221 (40)	325 (60)	546	
37,003–46,579	251 (45)	304 (55)	555	
COVID-19–related factors				
Symptoms in previous mo§§	54 (36)	97 (64)	151	NA
Symptoms after 14 Apr 2020				0.4
0	372 (39)	570 (61)	942	
1	120 (38)	197 (62)	317	
2	166 (38)	266 (62)	432	
3	138 (41)	200 (59)	338	
4	46 (43)	60 (57)	106	

Characteristics†	Negative, no. (%)	Positive, no. (%)	Total, no.	P value
5	8 (28)	21 (72)	29	
6	3 (75)	1 (25)	4	
7	2 (33)	4 (67)	6	
Doctor or hospital care for symptoms after 14 Apr 2020				1.0
No	228 (41)	321 (59)	549	
Yes	263 (38)	432 (62)	695	
COVID-19 testing and vaccination				
Ever tested for COVID				0.5
No	854 (40)	1,308 (60)	2,162	
Once	6 (35)	11 (65)	17	
Multiple times	1 (100)	0 (0)	1	
Test result				0.2
Negative	6 (43)	8 (57)	14	
Positive	0 (0)	3 (100)	3	
Inconclusive	0 (0)	0 (0)	0	
COVID-19–related behaviors after 14 Apr 2020				
Mask use				1.0
No	272 (41)	385 (59)	657	
Yes	588 (39)	935 (61)	1,523	
Mask frequency in the previous week				0.9
Never	9 (53)	8 (47)	17	
1–2 times	92 (36)	163 (64)	255	
3–5 times	120 (41)	170 (59)	290	
Almost every day	365 (38)	591 (62)	956	
Public transportation use change¶¶				0.2
No change	61 (51)	58 (49)	119	
Less use	344 (37)	598 (63)	942	
1–2 more times per day	0 (0)	0 (0)	0	
3–5 more times per day	1 (50)	1 (50)	2	
Continued change in public transportation use	334 (36)	590 (64)	924	NA

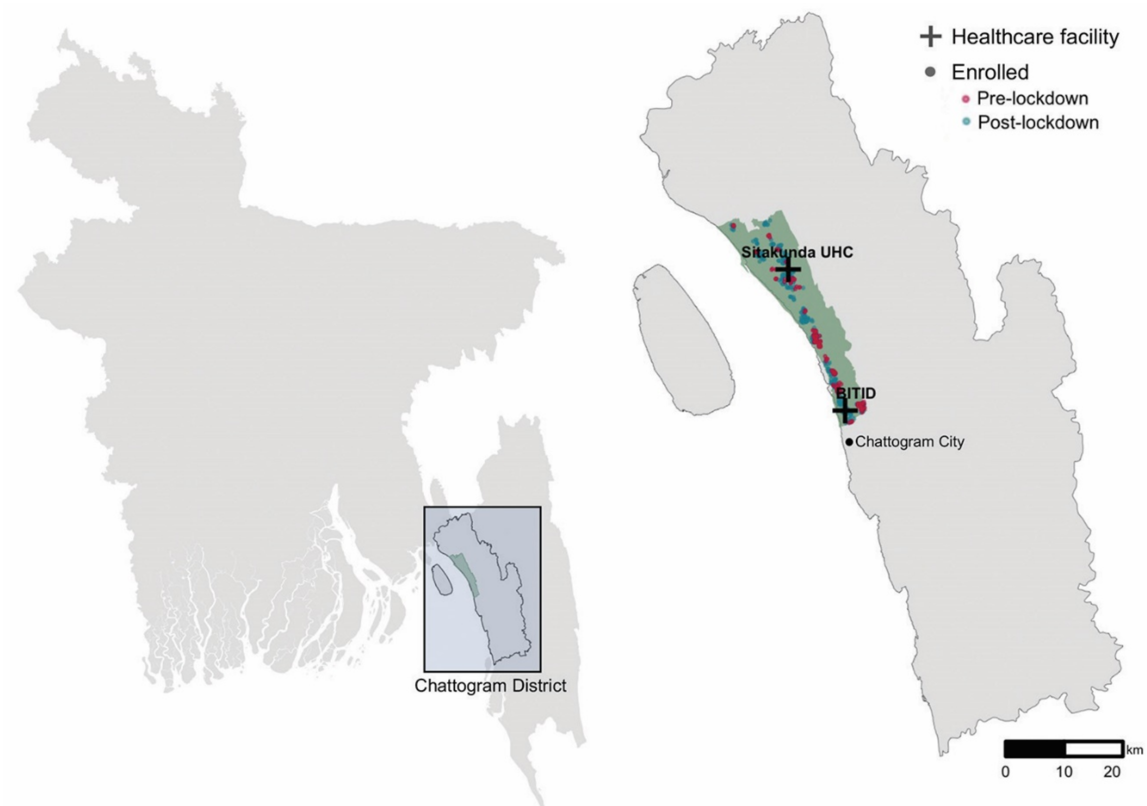
*NA, not applicable
†Chi-squared tests for trend were performed on categorical variables with numerically increasing categories and Pearson chi-squared tests of homogeneity were performed on all other categorical variables.
‡Household level proportions.
§Data from the Malaria Atlas Project (6).
¶Sampling cluster level measures (1km^2).
#Minutes to travel 1 m.
**Lower no. = more urban.
††Data sourced from WorldPop (https://www.worldpop.org).
‡‡Lower no. = more rural.
§§COVID-like symptoms include: fever, cough, shortness of breath, loss of taste/smell, nausea, diarrhea, and vomiting.
¶¶Change compared to use before April 14th, 2020. This field is restricted to only those who said they used public transport before April 14, 2020.

Appendix Table 3. Estimated seroprevalence of SARS-CoV-2 in Sitakunda Upazila adjusted for sex, age, household clustering, and test performance among unvaccinated participants.

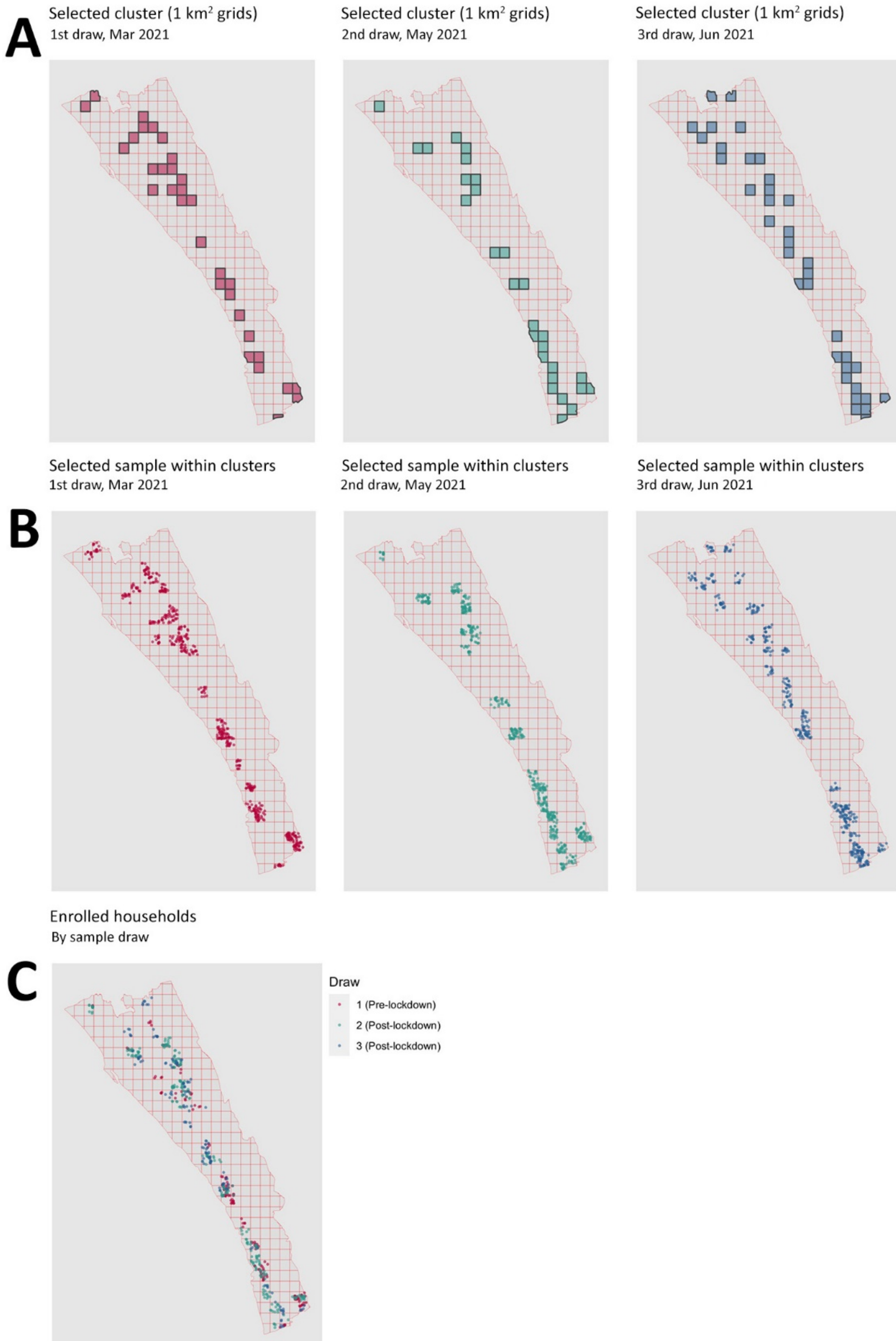
Variable	Observations	Positive	Negative	Adjusted seroprevalence (95% CI)	Adjusted relative risk (95% CI)
Age					
1–4 y	90	37 (41.1)	53 (58.9)	47.3 (37.4–57.5)	0.67 (0.53–0.82)
5–9 y	174	71 (40.8)	103 (59.2)	44.8 (37.1–52.9)	0.63 (0.52–0.74)
10–14 y	258	140 (54.3)	118 (45.7)	59.2 (52.3–66.2)	0.85 (0.74–0.95)
15–24 y	479	303 (63.3)	176 (36.7)	67.6 (62.2–73.1)	0.98 (0.89–1.07)
25–34 y	372	249 (66.9)	123 (33.1)	69.1 (63.7–74.3)	Referent
35–44 y	298	198 (66.4)	100 (33.6)	72.4 (66.4–78.3)	1.05 (0.95–1.15)
45–54 y	216	147 (68.1)	69 (31.9)	71.3 (63.8–78.4)	1.03 (0.92–1.15)
55–64 y	173	97 (56.1)	76 (43.9)	63.8 (55.9–71.4)	0.92 (0.80–1.04)
≥65 y	121	78 (64.5)	43 (35.5)	71.2 (62.6–79.8)	1.04 (0.90–1.17)
Sex					
M	1001	620 (61.9)	381 (38.1)	65.8 (61.0–70.6)	1.07 (1.01–1.13)
F	1180	700 (59.3)	480 (40.7)	60.7 (56.3–65.3)	Referent
Overall	2,181	1,320 (60.5)	861 (39.5)	63.4 (59.2–67.6)	NA

Appendix Table 4. The number of positive controls used to estimate the empirical sensitivity of the Wantai total Ab assay and SARS-CoV-2 positivity by time since symptom onset.

Time post-symptom onset, d	No. samples	% Seropositive (n)
3–13	2	100 (2)
14–30	6	83.3 (5)
31–60	5	100 (5)
61–90	7	100 (7)
91–120	7	85.7 (6)
121–150	10	80 (8)
151–180	12	83.3 (10)
181–210	11	72.7 (8)
211–240	9	100 (9)
241–275	12	100 (12)



Appendix Figure 1. Map of the study population in Sitakunda Upazila (green) in the Chattogram District of Bangladesh. The 580 enrolled households sampled in the serosurvey by enrollment time (pre- versus post-lockdown) and the 2 healthcare facilities in Sitakunda (Bangladesh Institute of Tropical & Infectious Diseases and Sitakunda Upazila Health Complex) are shown on the right side of the figure.



Appendix Figure 2. Map of the sampled clusters and sampled dwellings within clusters in the Sitakunda Upazila by samples drawn at three different periods: first draw (March 2021), second draw (May 2021), third draw (June 2021). Three separate sample draws were conducted because of interruption from the nationally imposed lockdown and a large percentage of nonresidential structures among housing structures sampled from the satellite imagery. A) Clusters were sampled 41 times with 14 structures each during the first 2 draws and 12 structures during the third draw. B) We oversampled the number of structures by 40% to account for nonresidential buildings for a total of 574 sampled structures for the first 2 draws and 492 for the third draw. C) Households were enrolled across the entire subdistrict of Sitakunda during each enrollment period and by sample draw (households enrolled pre-lockdown were only drawn from the first sample).