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Research Article

Seroprevalence of SARS-CoV-2 IgG antibodies among rural children aged 6-14 years in a selected block of West Bengal, India

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Abstract

***Background and objectives**:* Children comprised a significant part of the population during the second and third waves of the COVID-19 pandemic. The objectives of this study were to estimate the seroprevalence of COVID-19 IgG antibody among the children aged 6 to 14 years and to determine, if any, the factors associated with seropositivity.

***Methods**:* This cross-sectional study was conducted in a selected block of West Bengal, India over a period of 1 year (April 2022-March 2023) among children. Thirty villages in the block were selected by cluster sampling technique. COVID-19 IgM/IgG Rapid Antibody Test Kit (ICMR approved) was used for the detection of SARS-CoV-2 IgG antibodies. Data were analyzed by appropriate statistical tests.

***Results**:* Total 600 children were enrolled in the study. SARS-CoV-2 IgG antibody was positive in 57.2% children. The seropositivity rate (91.8%) was significantly ($p<0.001$) high among children of age group 12 to 14 years. Seropositivity rate was not significantly different between male and female children (46.4% *vs*. 53.6%; $p>0.05$).

***Conclusion**:* SARS-COV-2 IgG antibody was positive in a high proportion of children residing in rural areas indicating asymptomatic coronavirus infections among rural population. Socio-demographic factors such as higher age group and father's education were significantly associated with seropositivity.

Introduction

Children are the foundation of any nation, and the health and welfare of its child population determines the progress of any country. The COVID-19 pandemic, which hasn't been formally declared over yet, has led to some significant advancement in the worldwide health care industry. Since children constituted a significant portion of the unprotected population during the second and third waves of the COVID-19 pandemic, their vulnerability was an important consideration. Children and adolescents are also susceptible to

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the infection and thus form a part of the transmission chain. In late 2021, different nations had reported COVID-19 outbreaks in schools and child care facilities. What is more striking is that children were often reported to have asymptomatic infections than adults in case of COVID-19 [1].

Even though SARS-CoV-2 was thought to impact children and adolescents more mildly than adults, it nonetheless affects a variety of systems, with the cardiovascular signs being most noticeable [2]. In addition to being extremely unwell and necessitating Intensive Care Unit (ICU) admissions, child death rate, particularly in those with Multisystem Inflammatory Syndrome in Children (MIS-C), have been reported as high as 9% [3]. According to the World Health Organization (WHO), children under five years of age represented 2% of reported global COVID-19 cases during January 2020 to October 2021 and older children (5 to 14 years) accounted for 7% of the cases [4]. There was limited seroprevalence data among children in late 2021. Also, the antibody response to SARS-CoV-2 among children was poorly characterized.

Very few studies related to SARS-CoV-2 antibody detection among children were carried out in India in the years 2020 and 2021 and literature from West Bengal was scarce [5]. Following the second wave of COVID-19 cases in 2021, George *et al*. conducted a study in a rural area of Karnataka, India, and found that children's seroprevalence of antibodies to SARS CoV-2 was 45.9% [6]. In 2021, a multicenter study conducted by Misra *et al*. [7] found the prevalence of SARS-CoV-2 antibody among under-18-year-olds in both urban and rural areas as 55.7%, with a higher seropositivity rate among females. In another study, about 48.3% of children aged 5 to 17 in both urban and rural Kerala were found positive for COVID-19 antibody [8]. But there was no significant association with gender. In Delhi, India, seroprevalence of immunoglobulin G antibodies against SARS-CoV-2 among children aged 5 to 17 rose from 52.8% in January 2021 to 81.8% in September and October 2021, according to a repeated cross-sectional study [9]. Age and seropositivity correlated positively, but not with gender.

There was a dearth of information about the status of seroprevalence of SARS-COV-2 IgG antibody among people of rural Bengal, especially among children. Therefore,, the present study was conducted in a block of West Bengal, India with objectives to estimate the seroprevalence of SARS-CoV-2 IgG antibody among rural children aged 6 to 14 years and to find the factors associated (if any) with seropositivity among them.

Materials and methods

This descriptive cross-sectional study was carried out in Budge-Budge II block, West Bengal over a period of 12 months from April 2022 to March 2023. The study was approved by Institutional Ethics Committee (IPGME&R/IEC/2022/006, dated 21.01.2022). For children 7-11 years, informed oral assent in presence of parents and for children 12 - 14 years old, informed written assent was taken. Informed written consent was taken from all parents.

Study population: Children aged 7 to 14 years who had been residing with their families in the block for last one year or more were included. Those who had a laboratory confirmed COVID-19 infection in the past or who had any symptoms of COVID-19 infection during the time of data collection were excluded.

Sample size and sampling method: Considering 61.1% seroprevalence of anti- SARS-CoV-2 IgG antibody rate [10] and at 95% confidence interval (CI) and with 10% margin of error, the total sample size was calculated as 591 (after multiplying by 2 for design effect for cluster sampling and adding 20% as inconclusive). A total of 30 clusters were selected. Therefore, from each cluster (village) 591/30=19.7≈20 children were enrolled. Thirty villages were selected from a total of 61 villages using probability proportional to size method.

Data collection: Before commencing data collection, an orientation cum training session was conducted involving the Block Medical Officer, Accredited Social Health Activists (ASHA), Auxiliary Nurse Midwives (ANMs) and other health workers followed by pretesting among 20 children of the same age group who were not included in the final sample. Information on socio-demographic, clinical,

COVID-19 exposure related questions and vaccination details were collected in a predesigned, pretested, and structured questionnaire. Socio-demographic variables included: age, gender, type of family, socio-economic status, as per Modified BG Prasad Scale 2022 [11], parents' education, and occupation. Information on COVID-19 related infection included: history of COVID-19 infection in family, vaccination status of family members, and number of doses of vaccine received.

Collection of blood and test: About 30µL of whole blood was collected aseptically by finger prick and tested immediately for SARS-CoV-2 IgM or IgG antibodies by Oscar Covid-19 Rapid Antibody Test Kit (ICMR approved). The measurement range of the assay was from 0.40 U/ml to 250 U/ml. Levels of <0.80 and ≥0.80 U/ml were considered as negative and positive respectively according to the manufacturer's recommendations. Blood sample was placed in the specimen well of the test kit. Two drops (100µL) of buffer solution (provided with the kit) were added to the specimen. The results were read after 15 minutes. If a coloured line appeared at the IgG level along with the control line, the results were interpreted as positive.

Data analysis: Descriptive statistical measures such as frequencies, mean, standard deviation and confidence interval (CI) were determined. Z test for proportion was applied to test for significant difference between age groups and gender. Multivariable binary logistic regression was performed to find predictors of IgG positive test among the study population.

Results

Out of 600 children, about 40.5% belonged to age group of 12-14 years and their mean age was 10.36 ± 2.53 years. A little more than half were females (54.7%) and more than 80% followed Hinduism by faith. About 86.3% resided in joint households and half of the families belonged to upper middle class (50.2%). There was a health care worker in only 1.2% of the families (Table-1). None of the children had undergone any kind of COVID-19 detection test prior to the study. A small proportion (64, 10.7%) of the families had a laboratory confirmed history of COVID-19 infection within last one year in at least one of the members. Out of those who tested positive, 4 required hospitalizations and 1 of them died (Table-2).

Table-1: *Distribution of the study participants according to the socio-demographic profile (N=600)*

Characteristics		Number (%)
Age group	6 to 8 years	164 (27.3)
	9 to 11 years	193 (32.2)
	12 to 14 years	243 (40.5)
Gender	Male	272 (45.3)
	Female	328 (54.7)
Religion	Hindu	486 (81.0)
	Muslim	114 (19.0)
Family type	Joint	518 (86.3)
	Nuclear	82 (13.7)
Socio-economic status	Upper	0
	Upper Middle	301 (50.2)
	Middle	297 (49.5)
	Lower Middle	2 (0.3)
	Lower	0
Education of father	Illiterate	5 (0.8)
	Primary	69 (11.5)
	Middle school	197 (32.8)
	Secondary	237 (39.5)
	HS	68 (11.3)
	Graduate and above	24 (4.0)
Occupation of father	Farmer/Nursery	357 (59.5)
	Daily wage labourer	111 (18.5)
	Tailor	81 (13.5)
	Driver	23 (3.8)
	Private job or service	16 (2.7)
	Shop owner	5 (0.8)
	Teacher	5 (0.8)
	Boatman	2 (0.3)
Education of mother	Illiterate	0
	Primary	70 (11.7)
	Middle school	251 (41.8)
	Secondary	205 (34.2)
	Higher Secondary	74 (12.3)
	Graduate and above	0
Occupation of mother	Homemaker	577 (96.2)
	Papad making	20 (3.3)
	Tailor	3 (0.5)
Any health care worker in family	Yes	7 (1.2)
	No	593 (98.8)

About 73% of the families had all their members vaccinated against COVID-19. Out of the 64 people infected with COVID-19 in past one year, 49 had received two doses of vaccine prior to getting the infection. Most common reason for not taking the vaccine was fear of side effects (Table-3).

Out of 600 children, 343 (57.2%) were SARS-CoV-2 IgG antibody positive by rapid antibody test kit. There were no IgM positive cases and none of the tests were inconclusive. The seropositivity was found to be significantly high (91.8%; p<0.001)) in 12 to 14 years old children compared to other age groups. Sero-positivity rate did not differ significantly between male and female across all age groups (p >0.05; Table- 4).

On performing Pearson's Chi Square test, socio-demographic categories such as age group and father's education were found to be significantly associated with seroprevalence. The variables which were considered biologically plausible to be included for multivariable binary logistic regression were age group, family type, father's education, father's occupation, and mother's education. However, on multiple regression analysis only higher age group, 9 to 11 years and 12 to 14 years had statistically significant higher adjusted odds ratio of seropositivity (Tables-5 and 6).

Table-2: *Distribution of the study participants according to COVID-19 related information (N=600)*

Information sought		Number (%)
Ever got tested for COVID-19?	Yes	0
	No	600 (100.0)
Ever got tested positive with COVID-19?	Yes	-
	No	-
Ever had any direct contact with a COVID-19 infected patient?	Yes	0
	No	600 (100.0)
Any family member infected with COVID-19 in the past?	Yes	64 (10.7)
	No	536 (89.3)
If yes, how many family members got infected with COVID-19? (n=64)	1 member	49 (76.5)
	2 members	14 (21.8)
	3 members	0
	4 members	1 (1.6)
Was any infected family member hospitalized? (n=64)	Yes	4 (6.2)
	No	60 (93.8)
Out of those hospitalized, did you lose any member? (n=4)	Yes	1 (25.0)
	No	3 (75.0)

Table-3: *Distribution of the study participants according to COVID-19 vaccination related information (N=600)*

Information sought		Number (%)
Have all your family members been vaccinated against COVID-19?	Yes	438 (73.0)
	No	162 (27.0)
How many members got infected with COVID-19 after vaccination? (n1=64)		49 (76.6)
How many doses had they received prior to getting infected? (n=49)	2 doses	49 (100.0)
How many members in your family have still not taken the vaccine? (n=162)	1 member	97 (59.9)
	2 members	49 (30.2)
	3 members	15 (9.3)
	4 members	1 (0.6)
Reasons for not taking vaccine (n=162)	Fear of side effects	138 (85.2)
	Children are not infected much	14 (8.6)
	Not required	6 (3.7)
	Vaccine does not work	3 (1.9)
	Scared of injection	1 (0.6)

Table-4: *Seroprevalence of SARS-CoV-2 IgG antibodies among the study participants (N=600)*

Age groups (Years)	Male		Female		Total	
	Number	SARS-CoV-2 IgG Positive n (%)	Number	SARS-CoV-2 IgG positive n (%)	Number	SARS-CoV-2 IgG positive n (%)
6-8	78	28 (35.9) (CI: 25.3 - 47.5)	86	12 (14.0) (CI: 7.14 - 23.1)	164	40 (24.4) (CI: 18.0 - 31.7)
9-11	91	35 (38.5) (CI: 28.4 - 49.2)	102	45 (44.1) (CI: 34.3 - 54.3)	193	80 (41.5) (CI: 34.4 - 48.7)
12-14	103	96 (93.2) (CI: 86.5 - 97.2)	140	127 (90.7) (CI: 84.6 - 94.9)	243	223 (91.8) (CI: 87.6 - 94.9)
Total	**272**	**159 (46.4)** (CI: 40.9 - 51.7)	**328**	**184 (53.6)** (CI: 48.2 - 59.0)	**600**	**343 (57.2)** (CI: 53.1 - 61.1)

Note: CI: Confidence interval

Table-5: *Association between seroprevalence of SARS-CoV-2 IgG antibodies and socio-demographic and other factors (N=600)*

Sociodemographic and other factors		Seropositive n (%)	Seronegative n (%)	Chi square; df	*P* value
Age group	6 to 8 years	40 (24.4)	124 (75.6)	210.24; 2	*P*<0.001
	9 to 11 years	80 (41.5)	113 (58.5)		
	12 to 14 years	223 (91.8)	20 (8.2)		
Gender	Male	159 (58.5)	113 (41.5)	0.338; 1	0.561
	Female	184 (56.1)	144 (43.9)		
Religion	Hindu	276 (56.8)	210 (43.2)	0.148; 1	0.700
	Muslim	67 (58.8)	47 (41.2)		
Caste	General	235 (57.5)	174 (42.5)	0.617; 2	0.734
	SC	41 (53.2)	36 (46.8)		
	OBC	67 (58.8)	47 (41.2)		
Family type	Joint	303 (58.8)	215 (41.5)	2.728; 1	0.099
	Nuclear	40 (48.8)	42 (51.2)		
Socio-economic status	Upper middle	173 (57.5)	128 (42.5)	0.062; 2	0.969
	Middle	169 (56.9)	128 (43.1)		
	Lower Middle	1 (50.0)	1 (50.0)		
Education of father	Up to Middle school	129 (47.6)	142 (52.4)	4.589; 1	0.032
	Secondary and above	128 (38.9)	201 (61.1)		
Occupation of father	Skilled work	66 (50.8)	64 (49.2)	2.774; 1	0.096
	Unskilled work	277 (58.9)	193 (41.1)		
Education of mother	Up to middle school	149 (46.4)	172 (53.6)	3.622; 1	0.057
	Secondary and above	108 (38.7)	171 (61.3)		
Occupation of mother	Homemaker	331 (57.4)	246 (42.6)	0.539; 2	0.763
	Papad making	10 (50.0)	10 (50.0)		
	Tailor	2 (66.7)	1 (33.3)		
Any health care worker in family	Yes	3 (42.9)	4 (57.1)	0.592; 1	0.442
	No	340 (57.3)	253 (42.7)		

df: Degree of freedom

Table-6: *Multivariable binary logistic regression showing predictors of SARS-CoV-2 IgG seropositivity (N=600)*

Independent variables		Seropositive n	COR (95% C.I)	AOR (95% C.I)	*P* value
Age group	6 to 8 years	40	Ref	Ref	
	9 to 11 years	80	2.19 (1.39-3.46)	2.15 (1.35-3.40)	0.001
	12 to 14 years	223	34.56 (19.35-61.73)	33.68 (18.79-60.36)	<0.001
Family type	Nuclear	40	Ref	Ref	
	Joint	303	1.48 (0.93-2.36)	1.09 (0.62-1.95)	0.751
Education of father	Up to middle school	129	Ref	Ref	
	Secondary and above	128	1.42 (1.03-1.97)	1.26 (0.60-2.68)	0.534
Occupation of father	Skilled work	66	Ref	Ref	
	Unskilled work	277	1.39 (0.94-2.05)	0.99 (0.61-1.60)	0.991
Education of mother	Up to	149	Ref	Ref	
	Secondary and above	108	1.37 (0.99-1.90)	0.95 (0.45-2.01)	0.906

COR: Crude Odds Ratio; AOR: Adjusted Odds Ratio; Nagelkerke R^2=0.440

Discussion

In India, the total number of COVID-19 positive children in the second wave was reported higher than that of the first wave [12]. Murhekar *et al*., [10] in a study conducted in 70 districts throughout India, which included both urban and rural areas, reported increased rate of seropositivity to SARS-CoV-2 with age [10]. Antibodies against SARS-CoV-2 were 57.2% children aged 6 to 9 years which was greater than the present study. Seroprevalence was 61.6% among 10 to 17 year old children which was lower than the findings of this study for children aged 12 to 14 years. They also reported higher seroprevalence among females (69.2%) than males (65.8%), a finding similar to the current study. The increased seroprevalence in rural areas indicated that infection in the second wave was widespread in rural areas.

In rural Karnataka, George *et al.* [6] conducted a cross-sectional study involving 412 children aged between 11 months to 18 years. The study's findings indicated an overall seroprevalence of antibodies to SARS-CoV-2 as 45.9%. Compared to the current study, the seropositivity to SARS-CoV-2 was comparatively lower in their children. Seroprevalence in the age group 6-10 years was 35.9% while it was 50.4% among 11-15 years old children. In Karnataka, the seropositive status of the parents, any family members who tested positive, and a history of symptoms suggestive of COVID-19 were substantially associated with the seropositivity among children. In the current study, seropositivity was significantly associated with age and father's educational attainment.

A multicentre seroepidemiological study conducted by Misra *et al.* [7] in selected urban and rural areas of five sites selected from four Indian regions (Delhi, Odisha, Uttar Pradesh, Tripura) reported a seroprevalence of 55.7% (using IgG ELISA) among children. This was slightly less than the prevalence found in the current study. The prevalence of SARS-CoV-2 infection among female and male children (58.6% and 53%) in their study was almost same as the current study. Age group wise distribution showed higher seroprevalence among 5-9 years old children (43.8%) compared to this study. About 60.3% of 10-17 years old children were seropositive. Saanu*et al.* [8] also reported COVID-19 antibody seroprevalence of 48.3% in children of Ernakulam district of Kerala. The seroprevalence of COVID-19 antibody was higher among children of mothers with skilled/unskilled occupation, residents of rural area, above poverty line category children, positive history of COVID-19, and those with history of contact with COVID-19-positive patients. None of these factors were found to

have a statistically significant association with seroprevalence in this study. According to a repeated cross-sectional analyses study conducted by Sharma *et al.*[9] in Delhi, India, the seroprevalence of antibodies to SARS-CoV-2 among children aged 5 to 17 climbed to 81.8% in September and October 2021 from 52.8% in January 2021. Jahan *et al.* [13] conducted a meta-analysis on the seroprevalence of IgG antibodies against SARS-CoV-2 in the general population in India from March 2020 to August 2021. The overall pooled seroprevalence was 20.7% in the first wave and 69.2% in the second wave, with higher seroprevalence in urban regions than rural. Also, seroprevalence did not differ by age and gender. In a Bangladesh study among general population by Rahman *et al.* [14], the overall adjusted seroprevalence was 48.3%, which did not differ by gender. Children up to 17 years had a significantly lower seroprevalence 38.6% compared to adults. Increasing age and education were identified as risk factors for seropositivity. A very low IgG seropositivity (3.9% in children aged less than 9 years) was reported from a cross-sectional study in Pakistan by Ahmad *et al.* [15]. Also, the seroprevalence was higher in urban than rural areas. Seropositivity was around twice as common in the older age group (20 years and above) than in the 0–9 age group. In our study the seropositivity rate for SARS-CoV-2 was 91.8% among children aged 12-14 years. Seroprevalence of IgG antibodies to SARS-CoV-2 positively correlated with participants' age. The high rate of seroconversion among children who were not vaccinated suggested the existence of naturally occurring immunity generated by prior exposure to SARS-CoV-2.

The present study had some limitations. The study was conducted in only one block of West Bengal, thus limiting the generalization of the findings on a nationwide scale. A high proportion of children were SARS-CoV-2 IgG positive in the present study. Socio-demographic factors such as higher age group and father's education were significantly associated with seropositivity. In addition to usual infection prevention measures, full vaccination coverage is may be considered as nearly half of the children were still at risk of contracting the disease.

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Authors' contributions

VS, VS MB, AM, MR and RS: Conception, design of the study, analysis, and interpretation of data, drafting and revising the article critically and final approval of the version.

Conflict of interest

There is no conflict of interest.

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