








# SARS-CoV-2 antibody seroprevalence rates among Egyptian blood donors around the third wave: Cross-sectional study

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## Abstract

**Background:** Seroprevalence studies may provide a more representative situation of the disease burden and population-level immunity in a country.

**Aim:** The aim of this study was to determine the prevalence of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) antibodies among asymptomatic blood donors attending the Cairo University blood bank services at various points in time around the third wave.

**Methods:** This cross-section study included 3058 eligible blood donors, representing a demographically and socially heterogeneous healthy population and categorized as: Group 1, 954 donors in the period from March 20 to 30/2021; Group 2, 990 donors in the period from June 3 to 10/2021. These two groups were tested for IgG against SARS-CoV-2 nucleocapsid antigen (NC) to detect qualitative reactivity. Group 3, 1114 donors in the period from July 20 to 30/2021 were tested by the SARS-CoV-2 IgG II Quant assay for the quantitative detection of IgG antibodies, including neutralizing antibodies (antispikes antibodies).

**Results:** Donors' age ranged between 18 and 59 (mean  $33.9 \pm 9$ ) years. There was no significant correlation between seroprevalence and gender, area of residence, ABO or Rh blood types, and occupation or education. Antibody prevalence was found to be 13.2% in Group 1, 19.2% in Group 2 (overall 16.2%), and 66% in Group 3. There were only 49 included cases vaccinated against COVID-19.

**Conclusion:** We concluded that the significant increasing trend in seroprevalence rates during the third wave, March, June, and July, in Egypt, reflects a high cumulative incidence of seroconversion that mirrored the epidemic curve in its rise, fall, and nadir.

## KEYWORDS

Egyptian blood donors, SARS-CoV-2 antibody, seroprevalence, third-wave COVID infection

## 1 | INTRODUCTION

On the 11th of March 2020, the World Health Organization declared COVID-19 to be a global pandemic.<sup>1</sup> The particular coronavirus responsible for this pandemic is the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2).<sup>2</sup>

Since the beginning of the pandemic, there were controversial views about the infectiousness of asymptomatic patients. It was reported that approximately 40%–45% of patients will remain asymptomatic. Knowing the seroprevalence rates of SARS-CoV-2 among apparently healthy asymptomatic individuals is important in determining the fraction of a population that may constitute a significant source of virus transmission.

Earlier on within the pandemic, a global survey for determining seroprevalence rates had been carried out in 48 participating blood transfusion centers. This allowed for tracking of infection rates in donors over time. Such surveys are done for the purpose of directing public health policies and implementing strategies for the reduction of viral transmission.<sup>3</sup>

Seroprevalence studies may provide a more representative situation of the disease burden and population-level immunity in a country. These studies may play an important role in ensuring that the health system in a country remains operative without failure and may help in making decisions for the procurement of adequate doses of vaccines.<sup>4</sup>

Several studies addressing the issue of host humoral immune response to SARS-CoV-2 among COVID-19-confirmed patients have been published.<sup>5,6</sup> It is now well accepted that total antibody to SARS-CoV-2 (T Ab) is the earliest and most sensitive serologic biomarker, rising from 1 to 2 weeks of symptom onset, and thereon, all patients remained reactive. IgM and IgG seroconversion occurs usually in the second or third week, where there is a quick decline of IgM levels and a longer persistence of IgG. Antibodies to nucleocapsid antigen (NC) are more abundant and decay more quickly,<sup>6</sup> while antibodies to spike receptor-binding domain (RBD) (S) inhibit binding to ACE2 receptor, hence generating a strong viral-neutralizing response and viral clearance.<sup>7</sup>

Currently, there is not much available data on serologic screening of asymptomatic people. Published data showed that seroprevalence of SARS-CoV-2 antibodies ranged from 1.6% to 4.1% among different populations.<sup>8–10</sup>

### 1.1 | Aim of work

The aim of this study was to determine the prevalence of SARS-CoV-2 antibodies among asymptomatic blood donors who attended the Cairo University blood bank (CUBB) services at various points in time (during the third wave of the COVID-19 pandemic and beyond).

## 2 | MATERIALS AND METHODS

A cross-sectional study consisting of serological testing for SARS-CoV-2 in healthy asymptomatic volunteer blood donors attending CUBB.

### 2.1 | Study subjects

The study subjects included all eligible donors who attended CUBB for blood donation at the time in which the kits for SARS-CoV-2 antibody testing are available. They were demographically and socially heterogeneous healthy population from different areas of Egypt because CUBB is considered the biggest blood bank, which serves all Cairo University hospitals and is a referral hospital from all over Egypt.

For candidates to be accepted as blood donors, they had to comply with all the donation eligibility criteria set by the Egyptian Ministry of Health and the American Association of Blood Banks. Recently, some criteria regarding COVID-19 have been included as: donors could not have had flu-like symptoms within the 30 days before donation nor had close contact with suspected or confirmed COVID-19 cases or traveled abroad in the past 30 days. Candidates presenting with fever (forehead temperature > 37.8°C) on the donation date were also deferred. Thus, individuals included in the study had no symptoms of COVID-19 and were healthy and asymptomatic individuals. COVID-19 vaccine was introduced in Egypt by the end of February 2021 to healthcare workers only and then extended to all adult population a month later. Thus, this study included only 49 vaccinated donors against COVID-19. The study subjects consisted of a total number of 3058 healthy blood donors in the period from March 20 to 30/2021 (Group 1), from June 3 to 10/2021 (Group 2) and then from July 20 to 30/2021 (Group 3).

All included subjects answered the questionnaire and signed a consent. The questionnaire included the following items: name, age, gender, area of residence (within the capital or other municipalities), occupation, education level, medical history [diabetes, hypertension, asthma, cancer, cardiac] history of previous cough, chest oppression, previous clinical examination, laboratory tests, travel during the last 3 months, and exposure to a frank COVID patient. The study was approved by Cairo University Research Ethical Committee (REC) on 14 May 2020.

### 2.2 | Sample collection

The plasma samples used for mandatory testing in the donated units were also used for SARS-CoV-2 antibody testing. At the beginning of blood donation, samples were collected and barcoded for each donor.

### 2.3 | Antibody testing

For study Groups 1 and 2, the Abbott Architect SARS-CoV-2 IgG assay (chemiluminescent microparticle immunoassay) was used, which was the antibody assay available at the time in our center. This assay measures IgG against the SARS-CoV-2 and was used to detect qualitative reactivity.

Based on the manufacturer's recommendation, a cut-off ratio of  $\geq 1.4$  was considered positive.

For study Group 3, the Abbott SARS-CoV-2 IgG II Quant assay was used for the qualitative and quantitative determination of IgG antibodies, including neutralizing antibodies, to the RBD of the S1 subunit of the spike protein of SARS-CoV-2 in plasma (Antispike antibodies). This was the assay method introduced later in the study. The sample cut-off was 50 AU/ml (arbitrary units per milliliter).<sup>11,12</sup>

## 2.4 | Statistical analysis

Data were tabulated in an Excel spreadsheet, with donor demographic characteristics reported by code, so that their individual identity would remain anonymous.

Data were analyzed using SPSS version 24: for the qualitative variables, the percentage was used, while the mean and standard deviation was used for quantitative variables.  $\chi^2$  was used to determine whether there is a statistically significant difference between the observed frequencies in the studied groups. The level of significance was  $p < 0.05$ .

## 3 | RESULTS

The overall seroprevalence was 34.43% (1053 subjects out of 3058 studied cases). They were categorized as:

Group 1: Nine hundred and fifty-four donors in the period from March 20 to 30/2021.

Group 2: Nine hundred and ninety donors in the period from June 3 to 10/2021 were tested for IgG against SARS-CoV-2 nucleocapsid antigen (NC) to detect qualitative reactivity. The antibody prevalence was found to be 13.2% and 19.2%, respectively, in the studied Groups 1 and 2 (Figure 1). The overall prevalence of antibodies in those two batches was 16.2% with a minimum qualitative index  $> 1.4$ . The mean  $\pm$  SD antibody level is shown in Table 1.

Group 3: One thousand one hundred and fourteen donors in the period from July 20 to 30/2021 were tested by the SARS-CoV-2 IgG II Quant assay for the quantitative detection of IgG antibodies, including neutralizing antibodies to the RBD of the S1 subunit of the spike protein of SARS-CoV-2 in plasma. The seroprevalence rate was 66%. The mean  $\pm$  SD antibody level is shown in Table 1 and Figure 1.

The characteristics of the included blood donors (for all groups) are summarized in Table 2. There were significantly more males than females in the studied population. The ages ranged between 18 and 59 years with a mean of  $33.9 \pm 9$ .

There was no significant correlation between seroprevalence and gender, area of residence, ABO or Rh blood types, and occupation or education.

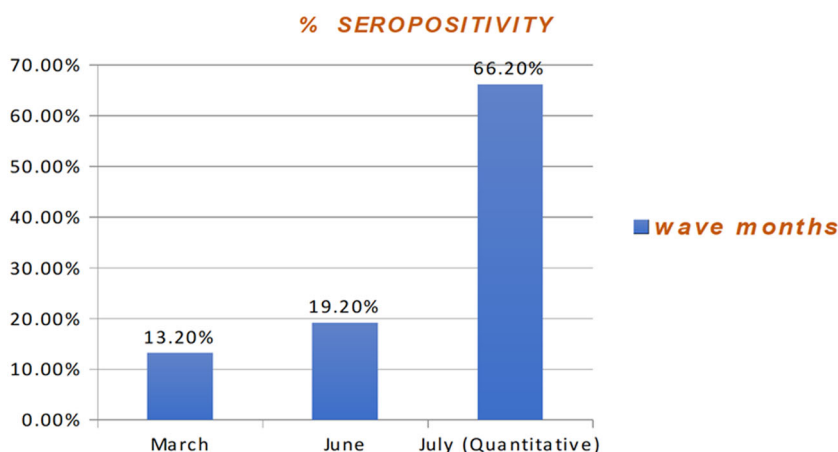
COVID-19 vaccine was introduced in Egypt by the end of February 2021. Most of the vaccinated population (42/49 cases, 85.7%) were seropositive; however, the vaccinated, seropositive cases represent only 4% of the all-seropositive studied population (42/1053 cases) (Table 2). Group 1 comprised no vaccinated cases.

**TABLE 1** SARS-CoV-2 serum antibodies among blood donors.

Methods	Positive				
	Mean	Standard deviation	Median	Minimum	Maximum
Qualitative [NC] cut-off ratio (n = 1944)	3.17	1.54	2.70	1.40	8.34
Quantitative [RBD] (AU/ml) (n = 1114)	596.98	1575.44	225.95	50.00	27,678.70

Abbreviations: AU/ml, arbitrary units per milliliter; NC, nucleocapsid antibody; RBD, receptor-binding domain.

**FIGURE 1** Seroprevalence rates in studied groups,  $\chi^2 = 788.84$ .



**TABLE 2** Sociodemographic parameters in relation to seroprevalence of SARS-CoV-2 antibodies in the studied blood donors (total no. = 3058).

Sociodemographic parameter	Total studied blood donors		Antibody testing				$\chi^2$	p value
			Negative (no. = 2005)		Positive (no. = 1053)			
	No. = 3058	%	No.	%	No.	%		
Sex								
Female (16.4%)	503	16.4	308	15.4	195	18.5	5.007	0.025*
Male (83.6%)	2555	83.6	1697	84.6	858	81.5		
Residence								
Capital	2536	82.9	1668	83.2	868	82.4	0.282	0.595
Noncapital	522	17.1	337	16.8	185	17.6		
Occupation								
Student	323	10.6	196	9.8	127	12.1	4.083	0.253
Civil	792	25.9	525	26.2	267	25.4		
Noncivil	1241	40.6	815	40.6	426	40.5		
None	702	23.0	469	23.4	233	22.1		
Education								
Higher	1654	54.1	1060	52.9	594	56.4	3.489	0.062
Lower	1404	45.9	945	47.1	459	43.6		
Vaccine (rolled out in Feb 2021)								
No	3009	98.4	1998	99.7	1011	96.0	58.002	<0.001*
Yes	49	1.6	7	0.3	42	4.0		
WAVE								
March (Qual)	954	31.2	828	41.3	126	12.0	788.843	<0.001*
June (Qual)	990	32.4	800	39.9	190	18.0		
July (Quant)	1114	36.4	377	18.8	737	70.0		

Abbreviations: Qual, qualitative; Quant, quantitative; no., number; %, percentage.

\*Significant.

The characteristics of the 49 vaccinated studied donors were shown in (Table 3). Most (73.5%) of the vaccinated population had a higher education than the nonvaccinated cases (Table 3). Most of the vaccinated seropositive subjects (35/42 subjects) were included in Group 3 and had antispikes antibodies; however, only 7/42 vaccinated, seropositive subjects were included in Group 2 and had NC antibodies (Table 4).

In this survey on antibody responses for SARS-CoV-2 among Egyptian blood donors, an increasing trend in the prevalence rates was seen throughout the study period: 13.2% during March, 19.2% during June, and 66% during July ( $p < 0.001$ ) (Figure 1). This trend mirrors the World Health Organization (WHO) epidemic curve, where the curve has been shown to be on the rise during March, April, and May, and then it started a downwards trend in June. The nadir was reached by the end of July.<sup>13</sup>

## 4 | DISCUSSION

To date, this is the first large study in Egypt addressing the seroprevalence of SARS-CoV-2 in asymptomatic, healthy blood donors in CUBB.

Since the evolution of the pandemic, many countries applied screening of COVID-19 antibodies, as an indication of immunity acquired by naturally exposed or vaccinated individuals. SARS-CoV-2 seroprevalence studies are thus fundamental to effectively monitor the extent of the COVID-19 epidemic and support authorities in making informed decisions.

Blood bank infrastructure allows rapid feasible random screening of regional populations to monitor seroprevalence. To collect samples from adults above 18 years is simple and is accessible from blood donors.

**TABLE 3** Donor characteristics of nonvaccinated versus vaccinated donors.

	Vaccine				$\chi^2$	p value
	No (3009)		Yes (49)			
	No.	%	No.	%		
Sex						
Female	497	16.5	6	12.2	0.640	0.424
Male	2512	83.5	43	87.8		
Residence						
Capital	2496	83.0	40	81.6	0.059	0.808
Noncapital	513	17.0	9	18.4		
Occupation						
Student	317	10.5	6	12.2	6.585	0.086
Civil	772	25.7	20	40.8		
Noncivil	1226	40.7	15	30.6		
Non	694	23.1	8	16.3		
Education						
Higher	1618	53.8	36	73.5	7.533	0.006*
Lower	1391	46.2	13	26.5		

Abbreviations: No., number; %, percentage.

\*Significant.

In our survey, we found an increasing trend in prevalence throughout the study period: 13.2% during March (Group 1), 19.2% during June (Group 2), and 66% during July (Group 3) ( $p < 0.001$ ) (Figure 1). Seropositive donors had humoral evidence of immunity against SARS-CoV-2.

Dynamics of the third peak in SARS CoV-19 in Egypt, according to WHO daily cases in Egypt,<sup>13</sup> are described in our study. We observed a peak of 66% seropositivity at the end of the wave. This dramatic rise was seen among healthy blood donors in parallel with widespread intracommunity transmission of the disease.

Our work reflects a high cumulative incidence of infection and seroconversion, which might be explained by the decline in infection rates in June. The decline in the spread of the virus went along with the high population seropositivity. A nadir in infection rates was observed by the end of July.

In this study, the antibody responses for SARS-CoV-2 were 34.43% (1053/3058 studied cases), which is higher than the previously published data that ranged from 1.6% to 4.1% among different populations.<sup>8–10</sup> These higher seropositive results may not entirely be representative of the general population in Egypt because the included blood donors were aged 18–59 years only with the exclusion of children and the elderly. Also, in this study,

there was an increasing trend in the seropositive prevalence rates throughout the study period: 13.2% during March, 19.2% during June, and 66% during July ( $p < 0.001$ ) (Figure 1). The WHO epidemic curve mirrors this trend, where the curve has been shown to be on the rise during March, April, and May, and then it started a downwards trend in June. The nadir was reached by the end of July.<sup>13</sup>

Also, the low seropositivity during the period from March to June may be due to the method of detection of the qualitative nucleocapsid antibodies that disappear early (short-lived), resulting in low seroprevalence rates.<sup>14–17</sup> On the other hand, the quantitative detection of the neutralizing antibodies is long-lived and probably demonstrates protection against further infection, particularly if the titer is high.<sup>18</sup> Another reason for the higher seroprevalence could be the change in the assay. Vaccinated individuals in Period 3 will have increased the seroprevalence detectable by the S assay. Since only 5% were vaccinated, it would not be inflated by more than that.

In this study, there was no significant correlation between seroprevalence and area of residence, Rhesus blood groups, or ABO blood types. On the other hand, there was no significant higher seropositivity among the less educated subjects. This observation was not expected, as these less-educated subjects usually do not strictly comply with social distancing, especially in their jobs. Their houses are also crowded and no clean water sources are available for proper hygiene.

Another study was done on 4040 health care workers (HCWs) in Cairo to examine the incidence of SARS-CoV-2 seroconversion, in which the seroconversions were asymptomatic. The seropositivity was three-fold that observed at baseline and the cumulative infections increased by a similar rate, which suggests that the HCW infection reflects what happens in the community.<sup>19</sup>

An immune response usually develops for most people infected within the first few weeks, but how long its duration and its strength are still unclear. Little is known about the level of immune response that translates into protection, how long it may last, or how it may differ from one person to another. Whether the neutralizing antibody response provides the needed protection for preventing new infections remains as yet undefined.<sup>20</sup> This assumed natural immunization rate still does not provide permanent immunity or herd immunity, accordingly part of the population may remain at risk of infection.

Natural reinfections, if humoral and cellular immunity fails, represent a common feature for all human seasonal coronaviruses after the 12th month post infection,<sup>19</sup> demonstrating a nonpermanent protective immunity. This might explain what happens with SARS-CoV-2 infections, unless the spike-specific memory B cells developed during COVID-19 last longer. Unfortunately, protection against SARS-CoV-2 longevity of the immune response is yet to be discovered.<sup>21</sup>

**TABLE 4** Characteristics and seroprevalence rates in donors with regard to study group.

Variables	Wave						Groups	χ <sup>2</sup>	p value
	Qualitative (no. = 1944)				Quantitative (no. = 1114)				
	+(March2021) No. = 954		#(June2021) No. = 990		^(July2021) No. = 1114				
	No	%	No	%	No	%			
Sex									
Female	134	14.0	184	18.6	185	16.6	+/^	2.583	0.108
Male	820	86.0	806	81.4	929	83.4	#/^	1.420	0.233
							+/#	7.318	0.007*
Residence									
Capital	831	87.1	798	80.6	907	81.4	+/^	12.400	0.001*
							#/^	0.225	0.635
Noncapital	123	12.9	192	19.4	207	18.6	+/#	15.121	0.001*
Occupation									
Student	98	10.3	102	10.3	123	11.0	+/^	2.119	0.548
Civil	260	27.3	259	26.2	273	24.5			
Noncivil	385	40.4	390	39.4	466	41.8			
Non	211	22.1	239	24.1	252	22.6	#/^	2.120	0.548
							+/#	1.190	0.755
Education									
High	513	53.8	543	54.8	598	53.7	+/^	0.002	0.966
Low	441	46.2	447	45.2	516	46.3	#/^	0.288	0.591
							+/#	0.226	0.634
Vaccine (rolled out in Feb 2021)									
No	954	100.0	976	98.6	1079	96.9	+/^	-	-
							#/^	6.878	0.009*
Yes	0	0.0	14	1.4	35	3.1	+/#	-	-
Negative	828	86.8	800	80.8	377	33.8	+/^	592.551	0.0001*
Positive	126	13.2	190	19.2	737	66.2	#/^	469.114	0.0001*
							+/#	12.781	0.001*

Abbreviations: +, March 2021; #, June 2021; ^, July 2021; no., number; %, percentage.

\*Significant.

## 5 | CONCLUSION

This study demonstrated the high cumulative incidence of sero-conversion that mirrored the epidemic curve of the WHO in its rise, fall, and nadir. It may also help public health authorities in assessing policies and making informed decisions on controlling COVID-19 pandemic.

### AUTHOR CONTRIBUTIONS

**Nermeen Eldesoukey:** Conceptualization; data curation; investigation; methodology; project administration; resources; writing—

review and editing. **Taghrid Gaafar:** Conceptualization; methodology; project administration; resources; supervision; writing—original draft; writing—review and editing. **Azza Aboul Enein:** Methodology; resources; supervision; writing—original draft; writing—review and editing. **Iman Eyada:** Conceptualization; funding acquisition; methodology; project administration; writing—review—editing. **Sahar Khirat:** Conceptualization; methodology. **Asmaa ElShahawy:** Investigation. **Nehal Diaa:** Investigation. **Ilham Youssry:** Conceptualization; funding acquisition; methodology; project administration; resources; supervision; writing—original draft; writing—review and editing.

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## CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

## ETHICS STATEMENT

This study was approved by the Research Ethics Committee, Faculty of Medicine, Cairo University on 14-5-2020 under the number: N-40-2020. Participants consented to participate in this study. The publication was foreseen in the acceptance of participation.

## TRANSPARENCY STATEMENT

The manuscript is an honest, accurate, and transparent account of the study being reported; that no important aspects of the study have been omitted; and any discrepancies from the study as planned (and, irrelevant, registered) have been explained.

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