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## Seroprevalence of SARS-CoV-2 antibodies in blood donors during the third wave of infection in Campeche Mexico

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### ABSTRACT

At the end of 2020 and previous to the second wave of COVID-19 in Mexico, the seroprevalence in unvaccinated people in the state of Campeche, Mexico was below 15 %. The prevalence changes with ongoing pandemic and per geographical areas. Blood donors screening is a powerful and cost-effective alternative to monitor the population's infection exposure.

The aim of the study was to estimate IgG anti-SARS-CoV-2 seropositivity in the Campeche's main blood bank in Mexico.

This was a cross-sectional study carried out at the main blood bank of the State of Campeche, located in the Yucatan peninsula, Mexico for the period of August through September 2021 during the third wave of the epidemic. A blood sample from 479 blood donors were included in the study, the overall seropositivity for SARS-CoV-2 IgG antibodies was 69.1 %. Vaccinated donors represented 69.9 % versus 29.4 % unvaccinated. The seropositivity in unvaccinated represented 42.5 % and 81.17 % in vaccinated. The seroconversion in vaccinated donors after first shot was 79 % for Astra-Zeneca-vaccine and 90 % for Pfizer-vaccine.

In conclusion 69.1 % of blood donors are seropositive for SARS-CoV-2 and 42.5 percent unvaccinated people are already also seropositive.

### 1. Introduction

The SARS-Cov-2 pandemic has represented a challenge for all the countries in the world, becoming a public health problem. There has not been a single common health policy to address this pandemic among the countries around the world. Some countries applied strict health policies while others were more relaxed. In Mexico, the implemented health polices to address the pandemic can be considered relaxed in comparison to those in countries who closed their frontiers, restricted international flights, as well as imposing restrictions on individual rights and a prolonged shut-down. The pandemic may be maintained in an active stage by the infected symptomatic and asymptomatic individuals who are spreading the infection. The asymptomatic individuals could represent up to 81 % [1] and they are the engine for the transmission of the pandemic, but at the same time represent the heard immunity that

could diminish the virus transmission. Mexico and many others countries have not established a screening protocol of the population to monitor exposure to virus and detection of asymptomatic cases in order to obtain more detailed epidemiologic data.

Spite of selection bias, blood donors' screening has been used to monitor the population's exposure to infections and indirectly to know the herd immunity level in an exposed population. The identification of SARS-CoV-2 seroprevalence can give an estimate of community transmission of virus infection. [2–4]. In Norway the third wave of infection peaked in March 2021 and a study that was carried out in blood donors showed that 1131/11,792 (9.5 %) were seropositive and 62 % of the donors who had seropositivity due to natural SARS-CoV-2 infection had some degree of COVID-19 symptoms, whereas 38 % did not report symptoms [5]. From a single blood center located in the northwest region of India at Jaipur, Rajasthan, during the period of mid-December

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2020 through January 2021 out of 534 healthy voluntary blood donors, 42.88 % (229) were found to be seropositive [6]. Whereas in Delhi, India at the transfusion medicine department of the tertiary care center, from September to October 2020, the overall seropositivity for SARS-CoV-2 IgG antibodies was 27.57 % (294/1066). In addition, the highest seropositivity 46.6 % (137/492) was seen in the age group 26–35 years [7]. At the Department of Transfusion Medicine, Odisha State, India; from November 2020 to January 2021, the overall IgG seropositivity for SARS-CoV-2 was 35.9 % (70/1032), which included 303 donors (29.36 %) with neutralizing antibody titer of SARS–COV2 IgG antibodies [8]. The above data suggests that prevalence of SARS-CoV-2 infection changes with the ongoing of the pandemic, vaccination programs, and per geographical areas.

In México, the vaccination program started in December 2020 and ran through February 2021 for health workers, from February through May 2021 for seniors, from May through July for people 40–59 years old, and from July 2020 to March 2022 for the rest of population. In Campeche state young people received their first shot between July and September 2021 [9].

Unfortunately, in Mexico there have been few publications in base-population to capture enough epidemiological data; two works at national level describe the transmission dynamics during 2020, indicating a national prevalence of 24–33 % by the end 2020 [10,11]. Two more other studies were carried out in cities located in the northern and southeastern part of Mexico during the same year of 2020; the seroprevalence was 27 % and 21 %, respectively by the end of the same year [12,13].

In the last census 2020, Campeche state had a population of 928, 363 and 55 % of them concentrated in two main cities (Campeche and Carmen), the rest are dispersed in small urban settlements and rural villages, and 70 % are over 18 years old [14].

The current study was carried out at the main blood bank of the State of Campeche at the “Centro Estatal de la Transfusión Sanguínea de Campeche”, located in Campeche city in the Yucatan peninsula, in the southeast part of Mexico, for the period of August through September 2021 during the third wave of the epidemic

The objective was to estimate the prevalence of anti-SARS-CoV-2 IgG antibodies during the third wave of the pandemic at the main Blood Bank of Campeche state.

## 2. Method

### 2.1. Study design and subjects

This is a cross-sectional survey for anti-SARS-Cov-2 IgG seroprevalence on residual serum samples from blood donors who attended Centro Estatal de la Transfusión Sanguínea de Campeche, Mexico during the period of August through September 2021. Centro Estatal de la Transfusión Sanguínea is the main blood bank in the State of Campeche and receives 600–800 donors each month from all over the State and it is the main blood supply center for the hospital located in the City of Campeche, Fig. 1. All donors underwent a medical review according to Mexican Official Norm NOM-253-SSA1–2012.

### 2.2. Data collection

Once the donation was approved, they were offered to voluntarily enroll in the current study, afterwards a written consent was signed by the donors and they completed a survey. The blood collection was done every two weeks for two months, from August to September of 2021.

There were 3 subjects who rejected to participate out of a total of 479 (0.62 %); usually the donation is between 600–800 blood donors per month, but for the period of the study, the blood donation decreased.

The collected data included age, gender, place of residence, whether they had been diagnosed with COVID-19, vaccinated for COVID-19 and the type of vaccine they received. The serum sample was kept at  $-30^{\circ}\text{C}$  and analyzed no more than 5 days later.

### 2.3. IgG serology against SARS-CoV-2

The IgG detection was made using a home-made ELISA against SARS-CoV-2 (2019-nCoV) Spike S1-His recombinant protein (40,588-V08B,



Fig. 1. Map of Mexico: arrow indicate Campeche state in the Yucatan peninsula in the gulf of Mexico.

SINO BIOLOGICAL). The ELISA plate was coated with 50 ng/well of antigen in carbonate/bicarbonate buffer pH 9.6 and sat overnight at 4 °C. After that, the plate was extensively washed with PBS-Tween 20 0.05 % then it was blocked with PBS-Tween 20- skimmed milk 3% for 1 h. Next, the serum sample was added at 1:200 diluted in PBS-Tween 20- skimmed milk 3% and incubated for 2 h at 37 °C followed by extensive washing with PBS-Tween 20. Later, anti-human IgG-peroxidase conjugate was added at 1:20,000 (SSA002, Sino Biological), sat for 1 h. and followed with extensive washing, as mentioned above. Finally, the reaction was developed by adding the substrate (peroxide hydrogen at 0.01 %) and chromogen (o-phenyl di-amine OPD at 0.05 %), and then stopped with H<sub>2</sub>SO<sub>4</sub> 2.5 N and the reading was 492 nm.

The home-made ELISA was validated against Abbot SARS-CoV-2-IgG chemiluminescent microparticle immunoassay [15,16]. In brief, we used as reference samples: fifty positive and forty negative which were previously verified with Abbot SARS-CoV-2-IgG chemiluminescent microparticle immunoassay and run them in our home-made ELISA. We observed 47 positives out 50 positive reference samples and all forty negative sample was correctly identified. The resulted concordance index, sensitivity and specificity values were 92 %, 94 % and 100 % respectively. In all assays, positive and negative control was included.

2.4. Statistical analysis

Frequencies, Kappa index concordance, sensitivity, specificity values, percentages, and the categorical data were analyzed using Chi-Square or Fisher’s exact test with SPSS software [SPSS 26, SPSS Inc, Chicago, IL].

2.5. Ethical consideration

The project was approved by the Ethics Committee, (DGPI “Estudio de seroprevalencia IgG contra SARS-Cov-2 en donadores de banco de sangre durante el inicio de segundo pico de la pandemia en Campeche” Universidad Autonoma Campeche).

3. Results

The overall seropositivity for SARS-CoV-2 IgG antibodies in blood donors was 69.1 %. The male frequentness was overrepresented (83 %),

although, IgG against SARS-CoV-2 seropositivity was lower in males (67 %) versus females (76 %), but without statistical significance (Chi-Square 2.5 and p = 0.11). Fig. 2

The mean age of blood donors was 33.5 years old (SD+/- 10.5) and fifty percent of them were between 18 and 32 years old, but 67.8 % under 37 years old. If we compare donors below 30 years old vs donors over 31 years old and seropositivity, there were no differences (Chi-Square 0.86 and p 0.35). The group of 28–32 years old showed the lowest IgG reactivity to SARS-CoV-2 (52 %) and statistically significant when compared to donors over 38 years old (Chi-Square 9.4 and p = 0.001). but the rest of blood donors, independent of their age did not show differences. Table 1. This data suggests that a great portion of blood donors population is already immune to SARS-CoV-2, independent of their age.

The number of vaccinated blood donors represented 69.6 % versus 29.4 % unvaccinated. In unvaccinated blood donors seropositivity for SARS-CoV-2 IgG antibodies represented 42.5 % (60/141), whereas in the vaccinated group was 80.17 % (271/338). In México, 5 different vaccines were administered, and the vaccination program is still in progress. The two main vaccines donors received were AstraZeneca-vaccine and Pfizer-vaccine, both representing over 95.5 % in all the donors in the study. Additionally, all of them had received their first shoot two to four weeks prior to their blood donation. The seroconversion in donors after their first shot was 79 % for AstraZeneca and 90 % for Pfizer vaccines (Chi-Square 3.79 and p = 0.02). Comparison between non-vaccinated and AstraZeneca vaccinated the seroconversion was

Table 1

IgG seroprevalence against SARS-CoV-2 and age of blood donors in Campeche, Mexico during the third epidemic wave (August-September 2021).

Age	IgG negative(%)	IgG positive(%)	%
18–22	21 (30 %)	50 (70 %)	71 (14.8 %)
23–27	33 (33 %)	66 (66.6 %)	99 (20.6 %)
28–32	34 (48 %)	37 (52 %)	71 (14.8 %)
33–37	28 (33.4 %)	56 (66.6 %)	84 (17.5 %)
38–42	8 (16 %)	42 (84 %)	50 (10.4 %)
43–47	14 (28.6 %)	35 (76.9 %)	49 (10.2 %)
48–52	6 (23 %)	20 (76.9 %)	26 (5.4%)
>53	4 (13.8 %)	25 (86.2 %)	29 (6%)
Total			479

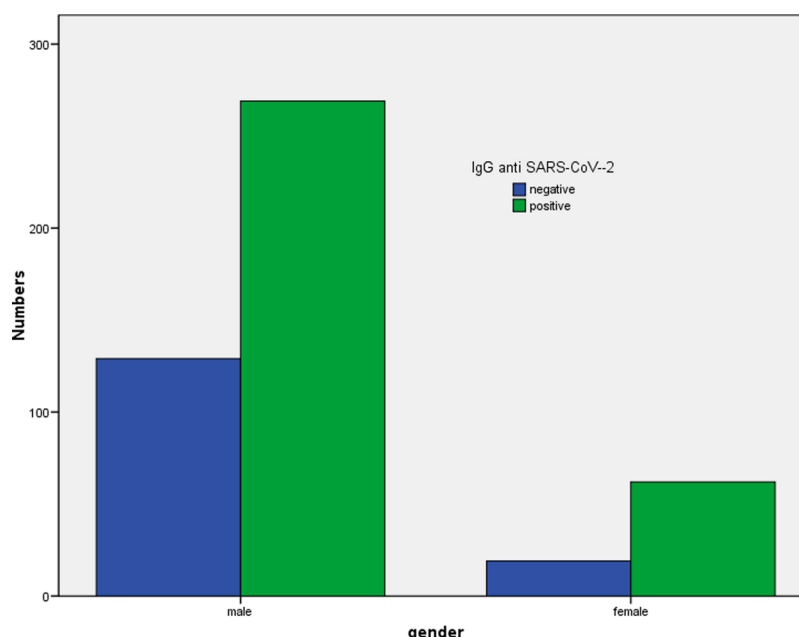


Fig. 2. Gender and IgG seroprevalence against SARS-CoV-2 in blood donors in Campeche, Mexico.

statistically significant (Chi-Square 72.2,  $p < 0.000001$ ) Fig. 3

#### 4. Discussion

Herd immunity is defined as “the indirect protection from infection conferred to susceptible individuals when a sufficiently large proportion of immune individuals exist in a population” [17–19]. The herd immunity in this context is represented by vaccinated and unvaccinated-asymptomatic subjects. For SARS-CoV-2 it was estimated that the minimum percentage of the total population required to halt transmission is 85 % [17–19]. The higher the immunity in the population the less transmission of the virus, this is the one goal vaccination programs around the world want to achieve. Although, the issue of herd immunity in COVID-19 infection is still debatable due majorly to vaccine hesitancy and to the evolving nature of the virus, the goal of the vaccination programs is to reduce the disease and the virus transmission.

By the end of 2020, previous studies in Campeche, Mexico and before the vaccination program started, showed the seroprevalences were between 10%–20% in the unvaccinated population [11]. In the present work 42 % (60/141) corresponded to unvaccinated subjects. These findings could suggest that natural immunity has increased two to four times in unvaccinated people in the last 8 months. Nevertheless, after the vaccination program was implemented, immunity to Sars-CoV-2 has had a positive impact as 69.1 % of blood donors tested is now immune. Although, our results cannot translate to population because our sample was not statistically representative, however our data clearly show that over two-third of blood donors are seropositive and 42 % are seropositive even without vaccine.

Our findings of 69 % of seroprevalence in blood donors are like a recently published paper for a study carried out in USA from December 2020 to June 2021 on 1,654,487 blood donors where the seroprevalence was 62.17 % [20]. This data demonstrates the impact of program vaccinations and the ongoing of the natural infection. In contrast with a Danish study which reported 2% of seroprevalence in 84,944 blood donors in October 2020 and 7% in February 2021 [21]. This data seems logical because coverage of vaccination reported in February 2021 for the country was 3.1 %. In north Jordan of 1000 blood donors tested between September 2020 and January 2021, the seropositivity reported was 14 % [22]. Whereas, in the northwest region of India between mid-December 2020 and January 2021 of 534 healthy voluntary blood donors the seropositivity was 42.88 % [6]. The above results show high

concordance between blood donors’ seroprevalence and the percentage of vaccination coverage, and likely herd immunity, with differences among geographical regions and population. Despite our sample size not being statistically representative, our data may suggest that population seroprevalence could be close to the one obtained from the blood donors.

Regarding immune response seroconversion with one shot, those vaccinated with Pfizer vaccine showed higher responder (90 %) with statistically significance than those vaccinated with AstraZeneca or Sinovac vaccines (79 and 72 % respectively). In this context it is important to have a complete vaccination schedule.

In conclusion, blood donors in our study are 69 % seropositivity for Sars-CoV-2 and 40 % unvaccinated subjects are already seropositive. 72%–90% of vaccinated subjects produced IgG on their first shot, depending on the vaccine they received.

#### CRedit authorship contribution statement

**Victor Monteón:** Contributions: Conceptualization, formal analysis, investigation, writing original draft, design of the study, analysis and interpretation of data, final approval of the submitted version. **Floribeth Leon Pérez:** Contributions: Conceptualization, formal analysis, investigation, design of the study, analysis and interpretation of data, final approval of the submitted version. **Virginia Peña Hernández:** Contributions: acquisition of data, analysis and interpretation of data, revising it critically for important intellectual content, final approval of the submitted version. **Anatalia Osorio Pacheco:** Contributions: acquisition of data, analysis and interpretation of data, revising it critically for important intellectual content, final approval of the submitted version. **Pedro Fuentes Guzman:** Contributions: acquisition of data, revising it critically for important intellectual content, final approval of the submitted version. **Gicel Ivett Gutiérrez Torres:** Contributions: acquisition of data, analysis and interpretation of data, revising it critically for important intellectual content, final approval of the submitted version.

#### Submission declaration and verification

Our submission has not been published previously, and it is not under consideration for publication elsewhere. Our work in this version is approved by all authors.

It will not be published elsewhere in the same form, in English or in

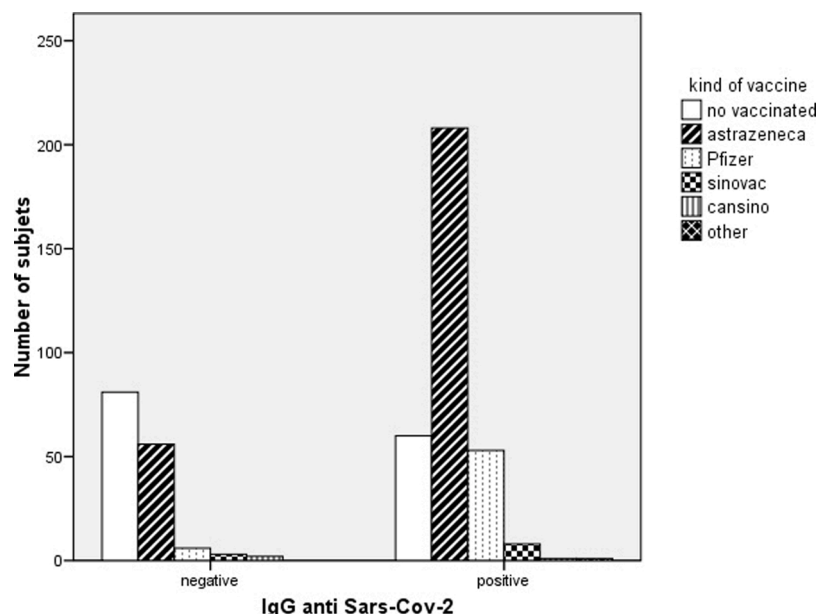


Fig. 3. SARS-CoV-2 vaccines applied in blood donors in Campeche, Mexico and seroconversion.

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