

Seroprevalence of anti-SARS-CoV-2 antibodies in Indore, Madhya Pradesh: A community-based cross-sectional study, August 2020

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ABSTRACT

Background: In India, laboratory diagnosis of SARS - CoV-2 infection has been mostly based on real-time reverse transcriptase-polymerase chain reaction (RT-PCR). Studies have shown that Viral titres peak within the first week of symptoms but may decline later hampering RT-PCR-based diagnostic strategies. Exact estimate is difficult under high-risk screening strategy with evidences of having large number of asymptomatic cases. This has prompted a call for adoption of antibody testing as potential source of data. **Materials and Methods:** A cross-sectional study with a sample size of 7000 was conducted for 15 days including all the 85 wards under Indore Municipal Corporation. Stratified Random Sampling was used to collect the samples. Trained teams collected basic sociodemographic information and serum samples which were tested for the presence of specific antibodies to COVID-19 using ICMR-Kavach IgG ELISA kits. The data collected was compiled and analysed using appropriate statistical software. **Results:** Overall weighted seroprevalence of the study population was found to be 7.75%. The prevalence in males and females was comparable (7.91% vs 7.57%). Highest seropositivity (10.04%) was seen among individuals aged more than 60 years. Total number of infections in the population were estimated to be 2,03,160. Overall Case Infection Ratio was found to be 27.43. **Conclusion:** The current seroprevalence study provides information on proportion of the population exposed, but the correlation between presence and absence of antibodies is not a marker of total or partial immunity. It must also be noted that more than 90 percent of the population is still susceptible for COVID-19 infection. Hence, non-pharmaceutical interventions like respiratory hygiene, physical distancing, hand sanitization, usage of personal protective equipment such as masks and implementation of public health measures need to be continued.

Keywords: Community based, Indore, infection, SARS-CoV-2, seroprevalence

Introduction

The first case of infection with the novel coronavirus, subsequently designated as SARS-CoV-2, emerged in Wuhan, China on 31st December 2019.^[1] In the wake of rapid increase in

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the number of COVID-19 cases worldwide, a global pandemic was declared by the World Health Organisation (WHO) on 12 March 2020.^[2] Despite intensive containment efforts, there has been a rapid international spread such that as of 28 August 2020, there have been a cumulative of more than 15 million cases worldwide, with more than 630,000 deaths.^[3] In India, the first case of COVID-19 was detected on 30 January 2020, in the southern state of Kerala.^[4] Since then the number of cases has exceeded one million with deaths nearing thirty thousand. Containment efforts by India have mostly relied on mass quarantine or 'lock-down' measures to control and restrict population movement, thereby reducing person-to-person contacts.^[5] This lockdown, which continued for at least 68 days for the entire country, has arguably been one of the largest mass-disruptions of human movement internationally in response to COVID-19, restricting free movement of 130 million people across the country.^[6]

In India, laboratory diagnosis of infection has been mostly based on real-time reverse transcriptase-polymerase chain reaction (RT-PCR). Diagnostic RT-PCR typically targets the viral ribonucleic acid (RNA)-dependent RNA polymerase (RdRp) or nucleocapsid (N) genes using swabs collected from the upper respiratory tract (nose and throat).^[7] However, the requirement for specialist equipment and reagents, skilled and trained laboratory and collection staff, and an inordinate but necessary time-gap between sampling and generation of test results act as potential bottlenecks. Therefore, clinical care and public health containment efforts are hampered by diagnostic delays, and further restricted by a lack of wider testing strategies including both mass screening, and specific high-risk groups. In addition to this, the issues of quality/timing of collection, transportation time of the samples have an impact on the outcomes of rt-PCR. Viral titres peak within the first week of symptoms in the upper respiratory tract, but may decline post this time frame, thereby hampering RT-PCR based diagnostic strategies.^[8,9] With evidences of having large number of asymptomatic cases and the fact that the patients would turn out negative on RT-PCR in post-recovery period, exact estimate of Covid cases is difficult under high-risk screening strategy. These reasons have prompted the call for adoption of antibody testing as a potential source of data to address the gap in data and inform public health and governance policies oriented towards COVID-19. Antibody-based tests would be capable of detecting both ongoing as well as past SARS-CoV-2 infections due to their capacity to detect IgG and IgM, thereby providing valuable clues about asymptomatic infections in the community.^[10] India has indigenously developed its own IgG-based ELISA test for COVID-19, with a reported sensitivity of 92.37% and a specificity of 97.9%.^[11]

The WHO global research map for COVID-19 and others recommend population-level seroepidemiological studies to generate data on the levels of infection in populations and recommend containment measures accordingly.^[12] The first national-level sero survey was carried out in India by ICMR in May 2020 which reported a seroprevalence of 0.78%.^[13] Following

this, serosurveys have been conducted in various cities of India to guide the public health policy and action.^[14] In the state of Madhya Pradesh, the cities of Bhopal, Indore and Ujjain have emerged as the epicentres of COVID-19, together accounting for almost half the total number of cases from Madhya Pradesh.^[15,16] Although the number of cases detected in Madhya Pradesh are relatively low (around 25,000 as on July 24, 2020), the testing rates (tests/million) remain lower than the national average.^[17] It might be argued, therefore, that a substantial number of positive cases remain undiagnosed. Indore is also known as the commercial capital of Madhya Pradesh and through this study, we aim to address crucial unknowns regarding the extent of transmission and rate of infection in Indore. Encouraging practices for community-level management with this background for reducing COVID-19 transmission and diagnosing asymptomatic spreaders to minimize the overall impact of this pandemic remains the primary objective of the study.

Materials and Methods

Study design and sample size

A cross-sectional study was conducted in all the 85 wards of Indore Municipal Corporation (IMC) between 11 and 23 August 2020. In May 2020, ICMR considered seroprevalence of 5% in hotspot areas,^[18] therefore, hypothesizing 1% rise each for June and July and 0.5% for first half of August 2020, prevalence of 7.5% was used for sample size estimation for the study. Open Epi was used for sample size calculation using a confidence level of 95%, absolute precision of 1% and design effect of 2.5, sample size of 6656 was reached, which was approximated to 7000. Additional 20% households were included in the sample size to meet the attrition and non-consent/non-response rate.

The sample size, thus calculated, was divided proportionately across 85 wards under Indore Municipal Corporation according to their population and stratified random sampling method was used to collect the samples. The three strata included; males aged 18 and above, females aged 18 years and above and children and adolescent aged 1 to 18 years. The sample in each ward was equally divided among the three strata. The list of households available with the revenue department of IMC was used to select the households randomly through computer-generated random numbers. Kish Grid method was used to randomly identify each candidate based on age and gender.

The Ethics review committee of the institution approved the protocol. The protocol was approved on 7th August, 2020, by the ethics committee of MGM Medical college.

The survey team visited the household and after explaining the purpose of the study, took written informed consent and assent in case of individuals less than 18 years of age and collected the requisite information from eligible study participants. Witnesses assisted participants who were not able to read or write in English or Hindi.

Basic socio-demographic information was collected using an android based mobile application developed by the Indore COVID Control room. Phlebotomists used standard venipuncture technique to collect 2-3 ml blood from consenting participants. Blood was collected using vacutainer with gel clot activator each vacutainer was first labelled with the corresponding unique alpha-numeric ID. The samples were transported to the designated laboratory. ICMR approved ELISA kit (COVID Kavach) with sensitivity of 92.1% and specificity of 97.7% was used for antibody estimation.

Detection of SARS-CoV-2 antibodies

The in-house laboratory of the Medical College, Indore was involved in ELISA testing of samples. Proper bio-safety precautions were followed in the laboratory while handling the blood samples. The samples were processed following the laboratory standard operating procedures. A unique lab ID was given to each sample and testing was performed in an anonymized manner. Serum is separated in eppendorf vials by centrifugation technique. Samples which were hemolysed, contaminated and of insufficient quantity were rejected. Anti-SARSCoV-2 IgG antibodies were assessed using a commercially available ELISA (COVID Kavach™; Ahmedabad, India) targeting the whole-cell antigen of SARS-CoV-2. Sera diluted at 1:100 were processed and ELISA Microplate reader (Multiskan, Labsystem) was used for OD values at 450 nm. The kit manufacturer's recommended cut-off for positivity ($>Av.NC+0.2$) had a sensitivity of 92.1% and a specificity of 97.7%.

Statistical analysis

Data analysis was done using statistical software [STATA; survey analysis module, STATA SE 12 (64 bit)]. Crude values and population estimates (weighted proportions) have been reported for describing the survey participants. Prevalence was estimated with 95% confidence interval using the survey data analysis module in STATA software. Appropriate sampling weights were used to adjust the seroprevalence estimates. Sampling weights (survey weights) are positive values associated with the observations in the dataset, used to ensure that metrics derived from a data set are representative of the population (the set of observations). In this study, the sampling weights were calculated by taking the number of samples from each ward divided by the ward population. Different weights were calculated based on the population of various wards of Indore. Finite population correction was calculated as the inverse of the sampling weights. Seroprevalence was estimated by adjusting the results against sensitivity and specificity. We applied the adjusted stratum specific seroprevalence to the total population of each stratum projected

for the year 2020 using 2011 census data to estimate the number of infections in each strata. The published literature indicates that IgG antibodies against SARSCoV2 infection start appearing by end of first week and most cases are IgG positive by end of second week. We therefore considered the number of reported COVID-19 cases on 27 July (2 weeks before the initiation of serosurvey) to estimate the plausible range of the number of infections. We defined the infection to case ratio as number of individuals with SARS-COV-2 infection (IgG detection) per number of RT-PCR reported cases of COVID-19.

Results

In the study, overall 7,103 blood samples collected were found to be satisfactory as per lab standards and results of these samples have been utilized for estimation of weighted seroprevalence of SARS-CoV2 antibodies in general population of Indore. The wards of the city of Indore were classified into three categories (tertiles) based on the case prevalence; high (28 wards), medium (28 wards) and low (29 wards) risk categories based on the case prevalence of more than 30, 15–30, less than 15 cases per 10,000 population respectively. Table 1 depicts the seroprevalence estimates for the three risk categories. An overall weighted seroprevalence of 7.75% was reported. The estimated seroprevalence in low, medium and high case prevalence areas was 4.25, 7.5 and 11.67 percent respectively.

Table 2 shows the seroprevalence by gender and different age groups. The prevalence in males and females was comparable (7.91% vs 7.57%). Highest seropositivity (10.04%) was seen among individuals aged more than 60 years. It was observed to be comparable in other age groups.

Based on the overall seroprevalence, the estimated number of total infections were calculated to be 2,03,160 for the study population. Based on seroprevalence observed in different wards, these were categorized into low (<6%), medium (6-10%) and high (>10%) seroprevalence zones.

Figure 1 shows a comparison of COVID-19 Case prevalence and SARS-CoV-2 antibodies seroprevalence. Marked difference in seroprevalence was observed in different wards. Seroprevalence of more than 30% was observed in hotspot areas like Bambai bazaar (Ward number 68). Four other hotspots namely Somnath, Haji Colony, Jawahar Marg, Ranipura (Ward number: 46, 38, 69, 60 respectively) had seroprevalence ranging between 20-25%. Overall 25 wards (29.2% of the population) had seroprevalence of above 10%, 13 wards (14.4% of the population) reported

Table 1: Seroprevalence of SARS-CoV-2 antibodies by wards (low, medium and high case burden)

Wards categorized by case prevalence	Total samples	Samples tested positives	Crude prevalence (in percent)	Weighted prevalence (95% CI)
Overall	7103	548	7.72	7.75% (7.14-8.36)
High	2251	262	11.65	11.67% (10.38-13.00)
Moderate	2375	179	7.54	7.50% (6.46-8.55)
Low	2477	107	4.32	4.25% (3.46-5.03)

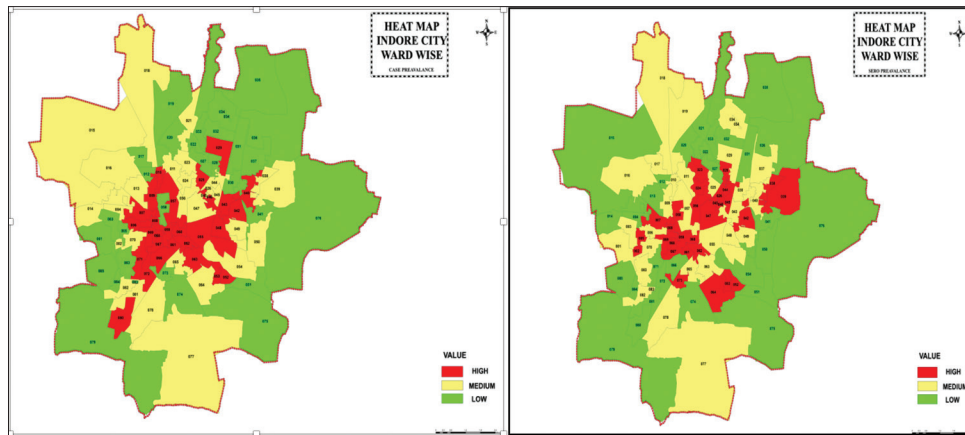


Figure 1: Heat map of Indore city COVID-19 Case prevalence (left) and SARS-CoV-2 antibodies Seroprevalence (right) as on 31st July 2020

Table 2: Seroprevalence of SARS-CoV-2 antibodies by gender and age groups

Variable	Samples collected	Crude prevalence In percent	Weighted prevalence (95% CI)
Gender			
Male	3680	7.90	7.91% (7.06-8.76)
Female	3423	7.50	7.57% (6.70-8.44)
Age group			
<18 years	2435	1.77	7.26% (6.24-8.28)
18 yrs and above	4668	7.95	7.97% (7.21-8.74)
18-45	3098	7.13	7.11% (6.22-8.01)
45-60	1106	10.03	10.04% (8.28-11.80)
>60	464	8.41	8.40% (5.87-10.92)

seroprevalence between 6-10% and 47 wards (56.4% of the population) had less than 6%. Overall Case Infection Ratio was 27.43 was observed as shown in Table 3. Infection fatality ratio of the study was found to be 1 death per 579 expected infections.

Discussion

Overall weighted seroprevalence of the study population was found to be 7.75%. Similar comparative findings have been reported from studies conducted internationally during recent times in Spain, Italy, Iran, United States, Brazil and Germany with seroprevalence of 5%, 4.6%, 22%, 10%, 21–30%, 4% and 15%, respectively.^[19-25] There has been a wide range of reports on seroprevalence of SARS-CoV2 antibodies from various places across India as well, including 57% in urban slums of Mumbai to 23.5% among general population of Delhi.^[14] A similar kind of serosurvey of Haryana,^[26] conducted in August 2020, showed overall sero-prevalence of 8% which is comparable to our study. Bhubaneswar reported the sero-prevalence of 5.15% of the sero-survey conducted across 25 wards on August 28 and 29 by RMRC (Regional Medical Research Centre) under the ICMR prescribed guidelines.^[27]

It has to be noted that a SEROCov-POP prospective study conducted in Switzerland reported that most of the population of Geneva remained uninfected during the wave of the

pandemic between April -May 2020, despite the high prevalence of COVID-19 in the region.^[23] It can be thus considered that the range of seroprevalence is quite variable throughout the world, mostly due to the different stages of the pandemic in different places. Even in our study seroprevalence varied greatly between different wards such as in few selected hotspot areas seroprevalence of almost five times that in low-risk areas was seen. Highest prevalence is in population of 45-60 years age group followed by more than 60 years age group like a study conducted in Switzerland which observed a significantly lower seroprevalence for children aged 5–9 years and adults older than 65 years as compared with those aged 10–64 years.^[23]

Case detection with respect to the expected infection based on estimated seroprevalence in Indore as on July 31, 2020 is 1:23 in low seroprevalent zones and 1:25 in medium zones but is as high as 1:32 in High seroprevalent zones [Table 3]. Low rate of case detection in High seroprevalence is >10% could probably be indicating the higher prevalence of asymptomatic cases in these wards in turn leading to higher rate of transmission and thus higher rate of seroprevalence. In other words, wards with low/medium seroprevalence could be hypothesized to have lower proportion of asymptomatic cases whereas wards with high seroprevalence have significantly higher proportion of asymptomatic cases. Previously published literature indicates that the IgG antibodies against SARSCoV-2 infection start appearing by the end of first week after the onset of symptoms and in almost all cases IgG is positive by the end of second week, and thus shall be positive by the end of third week or during the fourth week after exposure to the virus.^[28] Hence, the estimate of the total number of people infected around two weeks prior to our study was calculated to be 2,03,160. Reported number of COVID cases as on 27 July in the city was 7,058 and hence the effective Case: Infection ratio turns out to be 29 expected infections per reported case. This implies that a very large portion of the infected population is asymptomatic, quite comparable to many studies elsewhere in the world.^[29]

Seroprevalence and primary care practice

An insight into the findings of this study, as well as similar studies

Table 3: Case Infection Ratio by wards categorized into tertiles based on case prevalence

Categorization based on sero prevalence	Population (Percentage)	Positive cases as on 31 st July, 2020	Expected Infections	Case Infection ratio
High	765403 (29.2%)	2757	89323	32
Moderate	377791 (14.4%)	1139	28334	25
Low	1478224 (56.4%)	2688	62972	23

supplement the knowledge of the local practitioners. As there are only a few studies on seroprevalence in India so far (mainly catering to small pockets of population), seroprevalence in the local community setting of primary care practitioners remains less clear. Progressive dynamics of the pandemic makes it even more complex. Since the actual cases reported overall are the lab-confirmed COVID-19 positive individuals (mostly comprised of symptomatic cases and those found through contact tracing), many asymptomatic cases who can potentially spread the disease are missed out. Primary care practitioners, being the first point of contact with the community play a major role in triage. Background of local seroprevalence and the epidemic dynamics through the knowledge of the timely trends of COVID-19 disease progression in various clusters of different stages/phases of the pandemic would help in guiding the primary care practitioners in obtaining a better picture at the local community level. This would also help creating priority groups for implementation of various interventions for risk communication, behavioural change communication, conducting contact tracing programs, suspecting a case of COVID-19 during asymptomatic or mild symptomatic phase followed by early treatment initiation and isolation, implementation of vaccination programmes in future and help monitor their effectiveness.

Limitations

Limitations of the study include non-response from individuals less than 18 years of age, non-inclusion of clinical and exposure variables. The seroprevalence estimates were adjusted to test performance characteristics. The test performance characteristics might influence the seroprevalence estimates. It is possible that the ELISA may exhibit cross-reactivity with antibodies to other coronaviruses leading to false-positive results. The current seroprevalence study provides information on proportion of the population exposed, but however, the correlation between presence and absence of antibodies is not a marker of total or partial immunity. Follow-up studies need to be conducted to understand the duration and nature of protection provided by these antibodies and also seroprevalence in targeted groups need to be conducted to get further insights.

Conclusion

In conclusion, it must be noted that more than 90 percent of the population is still susceptible for COVID-19 infection. Hence, COVID-19 appropriate behavior (non-pharmaceutical interventions) including respiratory hygiene, physical distancing, hand sanitization, usage of personal protective equipment such as masks and implementation of public health measures need to be continued. Non-Pharmaceutical interventions with focus on

high-risk groups (HRG) need to be continued to further prevent further spread of infection.

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Conflicts of interest

There are no conflicts of interest.

References

1. Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, et al. A novel coronavirus from patients with pneumonia in China, 2019. *N Engl J Med* 2020;382:727-33.
2. World Health Organization. WHO announces COVID-19 outbreak a pandemic [Internet]. World Health Organisation. World Health Organization; 2020 [cited 2020 Jul 23]. p. 1. Available from: <https://www.euro.who.int/en/health-topics/health-emergencies/coronavirus-covid-19/news/news/2020/3/who-announces-covid-19-outbreak-a-pandemic>.
3. Worldometer. Coronavirus Cases [Internet]. Worldometer. 2020 [cited 2020 Jul 23]. p. 1-22. Available from: <https://www.worldometers.info/coronavirus/>.
4. Ministry of Health and Family Welfare. Update on Novel Coronavirus: One positive case reported in Kerala [Internet]. Press Information Bureau. 2020 [cited 2020 Jul 23]. p. 1. Available from: <https://pib.gov.in/PressReleaseDetail.aspx?PRID=1601095>.
5. The Lancet. India under COVID-19 Lockdown. *Lancet* 2020;395:1315:[https://www.thelancet.com/pdfs/journals/lancet/PIIS0140-6736\(20\)30938-7.pdf](https://www.thelancet.com/pdfs/journals/lancet/PIIS0140-6736(20)30938-7.pdf).
6. Dash N, Rose W, Nallasamy K. India's lockdown exit: Are we prepared to lock horns with COVID-19 and dengue in the

- rainy season? *Pediatr Res* [Internet]. 2020 [cited 2020 Jul 23];818:21-2. Available from: <http://dx.doi.org/10.1038/s41390-020-1063-7>.
7. Padhi A, Kumar S, Gupta E, Saxena SK. Laboratory diagnosis of novel coronavirus disease 2019 (COVID-19) infection. In: *Coronavirus Disease 2019 (COVID-19)*. Nature Publishing Group; 2020. p. 95-107. Available from: <https://pmc/articles/PMC7189402/?report=abstract>.
 8. To KKW, Tsang OTY, Leung WS, Tam AR, Wu TC, Lung DC, *et al.* Temporal profiles of viral load in posterior oropharyngeal saliva samples and serum antibody responses during infection by SARS-CoV-2: An observational cohort study. *Lancet Infect Dis* 2020;20:565-74.
 9. Wikramaratna P, Paton RS, Ghafari M, Lourenco J. Estimating false-negative detection rate of SARS-CoV-2 by RT-PCR. *medRxiv* [Internet]. 2020 Apr 7 [cited 2020 Jul 24];2020.04.05.20053355. Available from: <https://doi.org/10.1101/2020.04.05.20053355>. <https://doi.org/10.1101/2020.04.05.20053355>.
 10. Li Z, Yi Y, Luo X, Xiong N, Liu Y, Li S, *et al.* Development and clinical application of a rapid IgM-IgG combined antibody test for SARS-CoV-2 infection diagnosis. *J Med Virol* 2020;92:1518-24.
 11. Sapkal S, Shete-Aich A, Jain R, Yadav PD, Sarkale P, Lakra R, *et al.* Development of indigenous IgG ELISA for anti-SARS-CoV-2 IgG. *Indian J Med Res* 2020;151:444-9.
 12. World Health Organization. Coordinated Global Research Roadmap: 2019 Novel Coronavirus; March 2020. Geneva: WHO; 2020.
 13. Prevalence of SARS-CoV-2 infection in India: Findings from the national serosurvey, May-June 2020. *Indian J Med Res* 2020;152:48-60.
 14. Media Bulletin. The Hindu. [Internet]. Available from: <https://www.thehindu.com/news/cities/Delhi/delhis-seroprevalence-study-finds-2348-per-cent-people-affected-by-covid-19/article32147726.ece>. [Last accessed on 2020 Sep 03].
 15. Mekaad S. Madhya Pradesh: 118 test Covid-19 positive in Indore, tally reaches 6,457 | Indore News-Times of India. *The Times of India* [Internet]. 2020 Jul 22 [cited 2020 Jul 24];1. Available from: <https://timesofindia.indiatimes.com/city/indore/madhya-pradesh-118-test-covid-19-positive-in-indore-tally-reaches-6457/articleshow/77115083.cms>.
 16. Times News Network. Bhopal's Covid tally breaches 5,000 mark | Bhopal News-Times of India. *The Times of India* [Internet]. 2020 Jul 24 [cited 2020 Jul 24];1. Available from: <https://timesofindia.indiatimes.com/city/bhopal/bhopals-covid-tally-breaches-5k-mark/articleshow/77135526.cms>.
 17. COVID-19 India Tracker | The Wire Science [Internet]. *The Wire*. 2020 [cited 2020 Jul 24]. p. 1. Available from: <https://science.thewire.in/covid19>.
 18. Kumar MS, Bhatnagar T, Manickam P, Kumar VS, Rade K, Shah N, *et al.* National sero-surveillance to monitor the trend of SARS-CoV-2 infection transmission in India: Protocol for community-based surveillance. *Indian J Med Res* 2020;151:419-23.
 19. Pollán M, Pérez-Gómez B, Pastor-Barriuso R, Oteo J, Hernán MA, Pérez-Olmeda M, *et al.* Prevalence of SARS-CoV-2 in Spain (ENE-COVID): A nationwide, population-based seroepidemiological study. *Lancet* 2020;396:535-44.
 20. Valenti L, Bergna A, Pelusi S, Facciotti F, Lai A, Tarkowski M, *et al.* SARS-CoV-2 seroprevalence trends in healthy blood donors during the COVID-19 Milan outbreak. *medRxiv* 2020. doi: 10.1101/2020.05.11.20098442. <https://doi.org/10.1101/2020.05.11.20098442>.
 21. Shakiba M, Nazari SS, Mehrabian F, Rezvani SM, Ghasempour Z, Heidarzadeh A. Seroprevalence of COVID-19 virus infection in Guilan province, Iran. *medRxiv* 2020. doi: 10.1101/2020.04.26.20079244.
 22. Rosenberg ES, Tesoriero JM, Rosenthal EM, Chung R, Barranco MA, Styer LM, *et al.* Cumulative incidence and diagnosis of SARS-CoV-2 infection in New York. *medRxiv* 2020. doi: 10.1016/j.annepidem. 2020.06.004.
 23. Stringhini S, Wisniak A, Piumatti G, Azman AS, Lauer SA, Baysson H, *et al.* Seroprevalence of anti-SARS-CoV-2 IgG antibodies in Geneva, Switzerland (SEROCoV-POP): A population-based study. *Lancet* 2020;396:313-9.
 24. Amorim Filho L, Szwarcwald CL, Mateos SD, Leon AC, Medronho RD, Veloso VG, *et al.* Seroprevalence of anti-SARS-CoV-2 among blood donors in Rio de Janeiro, Brazil. *Rev Saude Publica* 2020;54:69.
 25. Streeck H, Hartmann G, Exner M, Schmid M. Vorläufiges Ergebnis und Schlussfolgerungen der COVID-19 Case-ClusterStudy (Gemeinde Gangelst)[Preliminary results and conclusions from the COVID-19 case-cluster study (communityGangelst).
 26. Haryana sero survey finds virus antibodies in 8% people | India News, *The Indian Express* [Internet]. [cited 2020 Sep 16]. Available from: <https://indianexpress.com/article/india/haryana-sero-survey-finds-virus-antibodies-in-8-people-6583630/>.
 27. Only 5 per cent sample population of Bhubaneswar has antibodies: Sero survey- *The New Indian Express* [Internet]. [cited 2020 Sep 16]. Available from: <https://www.newindianexpress.com/states/odisha/2020/sep/05/only-5-per-cent-sample-population-of-bhubaneswar-has-antibodies-sero-survey-2192805.html>.
 28. Sethuraman N, Jeremiah SS, Ryo A. Interpreting diagnostic tests for SARS-CoV-2. *JAMA* 2020;323:2249-51.
 29. The Centre for Evidence Based Medicine. COVID-19: What proportion are asymptomatic? [Internet]. Available from: <https://www.cebm.net/covid-19/covid-19-what-proportion-are-asymptomatic/>. [Last accessed on 2020 Sep 03].