Antibodies to SARS-CoV-2 in *All of Us* Research Program Participants, January 2-March 18, 2020

Keri N. Althoff PhD, MPH[1], David J. Schlueter, PhD[2], Hoda Anton-Culver PhD[3], James Cherry PhD[4], Joshua C. Denny MD, MS[5], Isaac Thomsen MD, MSCI,[6] Elizabeth W. Karlson MD, MS[7], Fiona P. Havers MD, MHS[8], Mine S. Cicek PhD[9], Stephen N. Thibodeau PhD[9], Ligia A. Pinto PhD[10], Douglas Lowy MD[4], Bradley A. Malin PhD[6], Lucila Ohno-Machado, MD, PhD, MBA[11], Carolyn Williams PhD, MPH[12], David Goldstein PhD[13], Aymone Kouame MSDA, MSIS[6], Andrea Ramirez, MD, MS[5], Adrienne Roman, PhD[6], Norman E. Sharpless, MD[4], Kelly A. Gebo, MD, MPH*[14], Sheri D. Schully PhD*[5] on behalf of the *All of Us* Research Program

*co-senior authors

1. Department of Epidemiology, Johns Hopkins Bloomberg School of Public Health, Baltimore, MD & Consultant to the *All of Us Research* Program, National Institutes of Health, Bethesda, MD
2. National Human Genome Research Institute, National Institutes of Health, Bethesda, MD
3. Department of Medicine, School of Medicine, University of California, Irvine, Irvine, CA
4. National Cancer Institute, National Institutes of Health, Bethesda, MD
5. *All of Us Research* Program, National Institutes of Health, Bethesda, MD
6. Vanderbilt University Medical Center, Nashville, TN
7. Harvard Medical School, Brigham and Women's Hospital, Boston, MA
8. Centers for Disease Control and Prevention, Atlanta, GA
9. Mayo Clinic, Rochester, MN
10. Frederick National Laboratory for Cancer Research, National Cancer Institute, Frederick, MD
11. School of Medicine, University of California, San Diego, San Diego, CA

12. National Institute of Allergy and Infectious Disease, National Institutes of Health, Bethesda, MD
13. Columbia University Medical Center, New York, NY
14. Department of Medicine, Johns Hopkins University School of Medicine, Baltimore, MD

Corresponding author:

Keri N Althoff

615 N Wolfe St., Rm E7142

Baltimore, MD 21093

410-614-4914

kalthoff@jhu.edu

Summary: 7 individuals had detectable SARS-CoV-2 immunoglobulin G prior to the first confirmed case in the states of Illinois, Massachusetts, Wisconsin, Pennsylvania, and Mississippi, suggesting that SARS-CoV-2 infections were occurring weeks prior to recognized cases in at least 5 US states.

ABSTRACT

Background: With limited SARS-CoV-2 testing capacity in the US at the start of the epidemic (January – March), testing was focused on symptomatic patients with a travel history throughout February, obscuring the picture of SARS-CoV-2 seeding and community transmission. We sought to identify individuals with SARS-CoV-2 antibodies in the early weeks of the US epidemic.

Methods: *All of Us* study participants in all 50 US states provided blood specimens during study visits from January 2 to March 18, 2020. A participant was considered seropositive if they tested positive for SARS-CoV-2 immunoglobulin G (IgG) antibodies on the Abbott Architect SARS-CoV-2 IgG ELISA and the EUROIMMUN SARS-CoV-2 ELISA in a sequential testing algorithm. Sensitivity and specificity of the Abbott and EUROIMMUNE ELISAs and the net sensitivity and specificity of the sequential testing algorithm were estimated with 95% confidence intervals.

Results: The estimated sensitivity of Abbott and EUROIMMUN was 100% (107/107 [96.6%, 100%]) and 90.7% (97/107 [83.5%, 95.4%]), respectively. The estimated specificity of Abbott and EUROIMMUN was 99.5% (995/1,000 [98.8%, 99.8%]) and 99.7% (997/1,000 [99.1%, 99.9%), respectively. The net sensitivity and specificity of our sequential testing algorithm was 90.7% (97/107 [83.5%, 95.4%]) and 100.0% (1,000/1,000 [99.6%, 100%]), respectively. Of the 24,079 study participants with blood specimens from January 2 to March 18, 2020, 9 were seropositive, 7 of whom were seropositive prior to the first confirmed case in the states of Illinois, Massachusetts, Wisconsin, Pennsylvania, and Mississippi.

Conclusions: Our findings indicate SARS-CoV-2 infections weeks prior to the first recognized cases in 5 US states.

Key Words: SARS-CoV-2 antibodies; United States; *All of Us* Research Program

INTRODUCTION

Among the first 12 known cases of SARS-CoV-2 infection in the United States (US), the earliest recognized symptoms onset date was January 14, 2020, and all 12 cases had recently traveled to mainland China or were close contacts of recent travelers.[1] Domestic testing for SARS-CoV-2 began in mid-January 2020 and the Food and Drug Administration granted Emergency Use Authorization for real-time reverse transcriptase polymerase chain reaction (RT-PCR) testing at the Centers for Disease Control and Prevention (CDC) on February 4 , 2020.[2] With limited testing capacity, testing was focused on symptomatic patients with a positive travel history throughout February. Many states recognized their first confirmed cases in the last week of February or the first week of March 2020.

Infectious disease epidemiology principles indicate that low-level circulation of the pathogen prior to the recognized outbreak was likely.[3] Phylogenetic analyses suggested evolution of the SARS-CoV-2 virus between October and December 2019.[4–6] The deaths of two California residents infected with SARS-CoV-2 virus on February 6 and 17, 2020, and an infected passenger or crew member of a cruise ship that left San Francisco on February 11, suggest that the virus was present in California in February, although a signal in syndromic surveillance was not detected until the end of February.[7] A recent study in blood donation specimens collected among residents of 9 states between December 13 and January 17 found antibodies to SARS-CoV-2 in the US as early as mid-December 2019.[8]

Determining the presence and location of SARS-CoV-2 in the earliest days of the US pandemic, together with other information on the spread and severity of COVID-19 illness, is important for understanding of the emergence of the virus, the epidemiology of this virus, and informing simulation models used to predict cases, deaths, and healthcare utilization and subsequently guide future pandemic planning, policy development and resource allocation.

Serologic evidence of SARS-CoV-2 antibodies can identify individuals who have been infected with SARS-CoV-2, including those who were asymptomatic or had subclinical illness, who are typically undetected at the start of an infectious disease epidemic. Serologic assays differ in their targets, and in their sensitivity and specificity; previous studies have compared commercially available assays using the same positive and negative control specimens.[9] The CDC recommends minimizing false positive antibody test results using a sequential testing approach ("employing two independent tests in sequence when the first test yields a positive result"), particularly when the prevalence of SARS-Cov-2 is expected to be low.[10] The objective of our study was to determine seroprevalence of SARS-CoV-2 immunoglobulin G (IgG) antibodies among participants in the *All of Us* cohort, from whom a blood specimen was collected during study visits occurring at the start of 2020 (January 2 to March 18, 2020).

METHODS AND MATERIALS

The *All of Us* Research Program is an observational cohort study that, beginning in May 2018, enrolls diverse adults (≥18 years of age) in the United States from study site locations in all 50 states, with a goal of enrolling at least 1 million participants, and has been described elsewhere.[11] Briefly, the *All of Us* Research Program includes genomic measurements and the large sample size needed for precision medicine research. Participants are enrolled after an informed consent process at clinics and regional medical centers that compose the *All of Us* Research Program network. Biospecimens collected from participants during *All of Us* study visits occurring from the first day in 2020 (January 2) to the day when in-person visits were paused due to the SARS-CoV-2 public health emergency (March 18, 2020). Participants who had not withdrawn from the *All of Us* study and had a serum specimen deemed acceptable by the biobank were eligible to participate in our nested study. Alaskan Native and American Indian participants were excluded from this analysis, at the request of tribal leaders. The *All of Us* internal review board approved our nested study.

We withdrew frozen serum specimens from the *All of Us* biorepository and prepared for serologic testing, starting with specimens collected in approximately 2-week intervals from March 18, 2020 moving backwards in time until there was a week with no positive specimens or January 2, 2020, whichever came first. This strategy was agreed upon prior to testing any specimens and was necessary due to the costs associated with testing this large sample size. The testing was conducted through a contract by Quest Diagnostics in an environment that meets the Clinical Laboratory Improvement Amendments (CLIA) regulations. We selected the Abbott Architect SARS-CoV-2 IgG ELISA (target: nucleocapsid; manufacturer's sensitivity=89.3% 95% confidence interval [82.6, 93.7%]; manufacturer's specificity=99.6% [99.0%, 99.9%]) and the EUROIMMUN SARS-CoV-2 ELISA (target: spike protein; manufacturer's sensitivity=90.0% [74.4%, 96.5%]; manufacturer's specificity=100% [95.4%, 100%]). Laboratory testing began in May 2020 and continued through January 2021; we analyzed the results as they were completed.

We estimated the sensitivity, specificity and Clopper-Pearson exact binomial 95% confidence intervals of the Abbott and EUROIMMUNE ELISAs using blinded positive control specimens from individuals who were hospitalized or discharged and convalescing with RT-PCR-confirmed SARS-CoV-2 infection (median [interquartile range] days since symptom: 14 [11, 18] days for n=18 positive controls from Brigham and Women's Hospital, Boston, MA; 45 [41, 55] days for n=44 positive controls from Vanderbilt University Medical Center, Nashville, TN; and unknown for n=45 positive controls from the Mayo Clinic, Rochester, MN). Negative control specimens came from *All of Us* participants who enrolled and had blood collected in the year prior to the emergence of SARS-CoV-2, specifically January to March 2019 (n=1,000). Up to 8 duplicate positive control specimens and 2 duplicate negative control specimens were used to ensure there were at least one positive and one negative control specimen on each plate.

Given the anticipated low prevalence of SARS-CoV-2 seropositive participants from January 2 – March 18, 2020, our definition of seropositive was that an individual must be seropositive on

Abbott and subsequently seropositive on EUROIMMUN, consistent with the CDC guidance for sequential testing; for this testing algorithm, net sensitivity and specificity and Clopper-Pearson exact binomial 95% confidence intervals were estimated.[10] In a sensitivity analysis of potential false positive results, SARS-CoV-2 IgG nucleocapsid and spike protein titers were quantified at a National Cancer Institute research, non-CLIA laboratory using in-house developed ELISAs. Additionally, to estimate the probability of false positives, we simulated 1 million participants and applied the lower 95% confidence interval bounds of the sensitivity and specificity for the Abbott and EUROIMMUN assays and mimicked the sequential testing algorithm to estimate the maximum probability of a false positive result with our definition of seropositive (see **Supplement** for details of the simulation).

We report the date of specimen collection, state of residence, and characteristics (age at the time of specimen collection, sex at birth, race and ethnicity) of those meeting our definition of seropositive using the sequential testing algorithm. An exception was granted to the *All of Us* program's Data and Statistics Dissemination (DSD) policy to report individual test results.[12] We also reviewed the electronic health records data and the participant-reported survey data of those who met our seropositive definition for information regarding recognized respiratory illness.

RESULTS

A total of 24,079 *All of Us* participants who had a biospecimen collected in our study period, had not withdrawn from the study, and had an acceptable specimen for testing were included. Participants were predominately female sex at birth (57%) and 49% were non-Hispanic White, 24% non-Hispanic Black/African American, and 17% Hispanic, Latino or Spanish (**Table 1**). Participants resided in 50 states with the largest number of participants residing in California (14%), Massachusetts (11%), Alabama (10%), Illinois (10%), Pennsylvania (8%), Arizona (8%), New York (8%), Wisconsin (7%), Florida (6%) and Michigan (5%, **Figure 1**).

Using our positive and negative controls, the estimated sensitivity of Abbott and EUROIMMUN was 100% (107/107 [96.6%, 100%]) and 90.7% (97/107 [83.5%, 95.4%]), respectively. Specificity of Abbott and EUROIMMUN was 99.5% (995/1,000 [98.8%, 99.8%]) and 99.7% (997/1,000 [99.1%, 99.9%), respectively. The net sensitivity and specificity of our sequential testing algorithm was 90.7% (97/107 [83.5%, 95.4%]) and 100% (1,000/1,000 [99.6%, 100%]), suggesting a low probability of false positives. Of the 1,000 negative controls, 5 were false positives on Abbott and 3 were false positives on EI; no negative control samples were false positive in our sequential method of testing (that is, negative controls were false positive on both Abbott and EUROIMMUN).

Of the 147 *All of Us* participants with a positive Abbott result, there were 9 individuals with samples that were subsequently positive on EUROIMMUNE and met our definition of seropositive (**Table 2**). Seven of the 9 seropositive individuals were detectable prior to the first confirmed cases in the states of their residence. These included individuals with specimens collected January 7 from Illinois, January 8 from Massachusetts, February 3 from Wisconsin, February 15 from Pennsylvania, and March 6 in Mississippi (**Figure 2**). Of the two seropositive participants who responded to the *All of Us* COVID-19 Participant Experience (COPE) survey, one reported experiencing fever, cough, sore or painful throat and a belief they had COVID-19 within a reasonable time frame (two weeks after) specimen collection given the survey was administered in May 2020. Review of electronic health record data during the relevant time frame revealed two seropositive participants had illnesses compatible with mild COVID-19 (e.g., fatigue and mild respiratory symptoms), but additional testing was limited and no diagnosis was confirmed. The other seven seropositive participants had no evidence of health care utilization in their electronic health record data.

Additionally, in sensitivity analyses of SARS-CoV-2 IgG antibody titers, 7 of the 9 seropositive participants had titers above the limit of detection, including 1 with nucleocapsid IgG titers, 4 with spike IgG titers, and 3 with nucleocapsid and spike IgG titers. Finally, we estimated the mean probability of false seropositive results given the low prevalence of SARS-CoV-2 during the study

period through simulations. Using our prevalence of 0.00037 (9/24,079), the estimated mean probability that all 9 were false positives was 0.00001 across 1000 replications of the simulation study; there was a 0.00019, 0.00210, 0.01405, 0.06251, 0.19487, 0.43841, 0.72959, and 0.93492 probability that there were at least 8, 7, 6, 5, 4, 3, 2, and 1 false positive results, respectively.

DISCUSSION

Our retrospective study of blood specimens from *All of Us* participants collected January 2 to March 18, 2020 suggests evidence of SARS-CoV-2 infection weeks prior to recognition of the virus in Illinois, Wisconsin, Pennsylvania, Mississippi, and Massachusetts. As recommend by the CDC, our study used a rigorous sequential testing definition of seropositive to minimize false positive results.

The median time from SARS-CoV-2 infection to IgG antibody presence is 14 days.[13–15] Assuming individuals who were seropositive according to our sequential definition were infected at least 2 weeks prior to biospecimen collection, our findings suggests the virus may have been present in Illinois as early as December 24, 2019. Our findings of suspected SARS-CoV-2 infection in January in Wisconsin (n=1 participant) and Massachusetts (n=1) corroborate a recent retrospective study of antibodies that were reactive in microneutralization with live SARS-CoV-2 (USA-WA1/2020) in Wisconsin (n=3) and Massachusetts (n=16) in blood donations made December 30, 2019-January 17, 2020.[8] We found evidence of SARS-CoV-2 antibodies in Mississippi in early March before the state's first case on March 11. A study of blood donors from California, Connecticut, Iowa, Massachusetts, Michigan, Oregon, Rhode Island, Washington, and Wisconsin used other laboratory approaches, including microneutralization tests and a receptor binding domain/ACE2 blocking activity assay. Our estimates of seropositive individuals using a sequential testing approach with commercial assays triangulate the findings from the blood donors, suggesting the robustness of the conclusions of seropositive individuals in both studies before the first state-recognized cases.

Further, our findings expand the knowledge of undetected SARS-CoV-2 infections likely occurring in early January in Illinois and early February in Pennsylvania. These data suggest SARS-

CoV-2 infection in states far from the initial hotspots, and originally considered points of entry to the US of Seattle, WA, and New York City, NY. Although the virus was presumed to be circulating in New York City, Seattle, and the state of California, none of the *All of Us* participants in these states tested positive, perhaps due to the low (albeit increasing) and highly-localized transmission from January through mid-March (and the smaller numbers of *All of Us* participants from the state of Washington).

Given the disproportionate burden of the subsequent US COVID-19 epidemic in minority populations, it is noteworthy that 7 of the 9 seropositive individuals were from older minority participants.[16] Although the *All of Us* study is enriched with populations that are underrepresented in biomedical research, there was a disproportionate increased burden of seropositivity in Black/African Americans (5/9) and Hispanic, Latino or Spanish (2/9) compared to the race and ethnicity distribution in the *All of Us* study population. Although the numbers are limited, these findings reinforce scientific hypotheses of the impact of social factors on viral circulation, including structural discrimination against racial and ethnic minority groups.

This study contributes to the evidence of low-level circulation of SARS-CoV-2 in many states at the start of the US epidemic. Federal testing recommendations included travel to a geographic area with known SARS-CoV-2 transmission or contact with a confirmed SARS-CoV-2 case until cases were confirmed in most states; the June 13, 2020 consolidation of testing recommendations did not include the travel epidemiologic link.[17] These epidemiologic links were particularly important at the beginning of the US pandemic when testing capacity was limited. Although our data suggest antibody evidence of SARS-CoV-2 infection weeks prior to the first confirmed cases in 5 states, the CDC's first report of community transmission (i.e. a confirmed infection in a person without a travel history or exposure to a confirmed SARS-CoV-2) did not occur until February 26, 2020.[18] The epidemiologic links in the testing recommendations may have been in place too long, obscuring the geographic spread of SARS-CoV-2 found in our results. Future pandemic management should

carefully consider the impact of epidemiologic links in testing recommendations and reduce testing restrictions as early as possible..

There are limitations to our study. First, *All of Us* participants were not confirmed to be infected with SARS-CoV-2 via molecular diagnostic tests or paired acute-convalescent sera. The strength of serology studies, however, is the capture of potentially asymptomatic individuals and those with sub-clinical illness who do not seek or were unable to obtain testing. Second, it is possible that we have detected pre-existing, non-SARS coronavirus antibodies that bind to SARS-CoV-2 nucleocapsid and spike protein in these 9 individuals, rendering these individuals as false positives.[9,19,20] Previous studies have shown low cross-reactivity of the S1 domain of the spike protein (target for EUROIMUNE) and low-level cross-reactivity of the S2 domain of the spike protein and the nucleocapsid protein (target for Abbott) and a strong correlation between antibodies against spike protein and neutralization; neutralization experiments were not performed on specimens from the 9 seropositive individuals due to this evidence.[9,19,21,22] Our sensitivity analyses of nucleocapsid and spike protein IgG titer quantification provided additional supporting evidence that it is unlikely all 9 of these seropositive individuals are false positives (estimated probability=0.00001) and was aligned with the probability of 2 false positives (7 of the 9 seropositive individuals had quantified IgG titers and the estimated probability of 2 false positives was 0.72959); we did not believe further depleting the specimen for neuralization results was warranted. The sequential testing algorithm net specificity was 100% (95% CI 99.6, 100%); none of the negative control specimens were positive on both Abbott and EUROIMMUNE. It may be more likely that we have misclassified those with low titers as seronegative; the positive controls used to estimate Abbott and EUROIMMUN sensitivity were individuals who were hospitalized, or recently discharged, with RT-PCR-confirmed SARS-CoV-2 infection. Third, we do not yet know if these SARS-CoV-2 antibodies were the result of infections acquired during travel or within the participant's community. Fourth, the findings likely cannot be extrapolated to broader seroprevalence estimates as participants in a longitudinal study enriched for under-represented minorities in biomedical research may not be

generalizable, particularly in the setting of a low prevalence of infection. A clear strength of our study is that it is nested in the *All of Us* Research Program, which provides the opportunity to investigate a critical time in the SARS-CoV-2 US epidemic within a single-protocol-driven, longitudinal study with large-scale geographic, race, and ethnicity breadth, and ongoing follow-up with participants.

Our findings highlight the importance of diverse longitudinal cohort studies that collect biospecimens for conducting retrospective studies to expand the understanding of the epidemiology of SARS-CoV-2, particularly spread of the virus before recognized cases in 5 US states, and inform public health surveillance testing strategies, computational models of the entrance of the novel virus into susceptible populations, and subsequent intervention and mitigation efforts.

NOTES

Acknowledgment

The *All of Us* Research Program would not be possible without the partnership of its participants.

Disclaimer

The findings and conclusions in this report are those of the authors and do not necessarily represent the views of the U.S. Centers for Disease Control and Prevention.

Funding

This work was supported by the National Institute of Health, Office of the Director, and the National Cancer Institute. The *All of Us* Research Program is supported (or funded) by the National Institutes of Health, Office of the Director: Regional Medical Centers: 1 OT2 OD026549; 1 OT2 OD026554; 1 OT2 OD026557; 1 OT2 OD026556; 1 OT2 OD026550; 1 OT2 OD 026552; 1 OT2 OD026553; 1 OT2 OD026548; 1 OT2 OD026551; 1 OT2 OD026555; IAA #: AOD 16037; Federally Qualified Health Centers: HHSN 263201600085U; Data and Research Center: 5 U2C OD023196; Biobank: 1 U24 OD023121; The Participant Center: U24 OD023176; Participant Technology Systems Center: 1 U24 OD023163; Communications and Engagement: 3 OT2 OD023205; 3 OT2 OD023206; and Community Partners: 1 OT2 OD025277; 3 OT2 OD025315; 1 OT2 OD025337; 1 OT2 OD025276.

Potential conflicts

Dr. Gebo initiated this work during her role as the Chief Medical and Scientific Officer of the *All of Us* Research Project. Dr. Althoff reports consultancy fees from TrioHealth (paid to her). Dr. Goldstein reports consultancy feeds from AstraZeneca, Gilead Sciences, GoldFinch Bio, and Goassamer Bio (paid to him) and is an equity holder of Q State Biosciences, Praxis Therapeutics, and Apostle Inc. No other authors report a potential conflict of interest.

REFERENCES

1. Kujawski SA, Wong KK, Collins JP, et al. Clinical and virologic characteristics of the first 12 patients with coronavirus disease 2019 (COVID-19) in the United States. *Nature Medicine*. 2020;26(6):861-868. doi:10.1038/s41591-020-0877-5

2. Hinton D. *2019-Novel Coronavirus (2019-NCoV) Real-Time Reverse Transcriptase (RT)-PCR Diagnostic Panel Centers Emergency Use Authorization for the Centers for Disease Control and Prevention*.; 2020. Available at: https://www.fda.gov/media/134919/download. Accessed December 11, 2020.

3. Lourenço J, Paton R, Ghafari M, et al. Fundamental principles of epidemic spread highlight the immediate need for large-scale serological surveys to assess the stage of the SARS-CoV-2 epidemic. *medRxiv*. March 2020:2020.03.24.20042291. doi:10.1101/2020.03.24.20042291

4. Andersen KG, Rambaut A, Ian Lipkin W, Holmes EC, Garry RF. The proximal origin of SARS-CoV-2. *Nature Medicine* 2020;26:450-452. doi:10.1038/s41591-020-0820-9

5. Li X, Zai J, Zhao Q, et al. Evolutionary history, potential intermediate animal host, and cross-species analyses of SARS-CoV-2. *J Med Virol*. 2020;92(6):602-611. doi:10.1002/jmv.25731

6. van Dorp L, Acman M, Richard D, et al. Emergence of genomic diversity and recurrent mutations in SARS-CoV-2. *Infect Genet Evol*. 2020;83:104351. doi:10.1016/j.meegid.2020.104351

7. Jorden MA, Rudman SL, Villarino E, et al. Evidence for Limited Early Spread of COVID-19 Within the United States, January–February 2020. *MMWR Morb Mortal Wkly Rep*. 2020;69(22):680-684. doi:10.15585/mmwr.mm6922e1

8. Basavaraju S V, Patton ME, Grimm K, et al. Serologic testing of U.S. blood donations to identify SARS-CoV-2-reactive antibodies: December 2019-January 2020. *Clin Infect Dis*.

2020;ciaa1785. doi:10.1093/cid/ciaa1785

9. Patel EU, Bloch EM, Clarke W, et al. Comparative performance of five commercially available serologic assays to detect antibodies to SARS-CoV-2 and identify individuals with high neutralizing titers. *J Clin Microbiol*. 2021;59(2):e02257-20. doi:10.1128/JCM.02257-20

10. US Centers for Disease Control and Prevention. Interim Guidelines for COVID-19 Antibody Testing in Clinical and Public Health Settings. Available at: https://www.cdc.gov/coronavirus/2019-ncov/lab/resources/antibody-tests-guidelines.html. Accessed December 10, 2020.

11. The "All of Us" Research Program. The "All of Us" Research Program. *N Engl J Med*. 2019;381(7):668-676. doi:10.1056/NEJMsr1809937

12. The All of Us Research Program. The All of Us Research Program: Data and statistics dissemination policy. Avaialble at: https://www.researchallofus.org/wp-content/themes/research-hub-wordpress-theme/media/2020/05/AoU_Policy_Data_and_Statistics_Dissemination.pdf. Accessed December 16, 2020.

13. Guo L, Ren L, Yang S, et al. Profiling Early Humoral Response to Diagnose Novel Coronavirus Disease (COVID-19). *Clin Infect Dis*. 2020:1-8. doi:10.1093/cid/ciaa310

14. Zhao J, Yuan Q, Wang H, et al. Antibody responses to SARS-CoV-2 in patients of novel coronavirus disease 2019. *Clin Infect Dis*. 2020. doi:10.1093/cid/ciaa344

15. To KKW, Tsang OTY, Leung WS, 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(5):565-574. doi:10.1016/S1473-3099(20)30196-1

16. Webb Hooper M, Nápoles AM, Pérez-Stable EJ. COVID-19 and Racial/Ethnic Disparities. *JAMA*. 2020;323(24):2466-2467. doi:10.1001/jama.2020.8598

17. US Centers for Disease Control and Prevention. CDC releases consolidated COVID-19 testing recommendations. Available at: https://www.cdc.gov/media/releases/2020/s0613-covid19-testing-recommendations.html. Accessed December 16, 2020.

18. US Centers for Disease Control and Prevention. CDC confirms possible instance of community spread of COVID-19 in the US. Available at: https://www.cdc.gov/media/releases/2020/s0226-Covid-19-spread.html. Accessed December 16, 2020.

19. Okba N, Muller M, Li W, et al. Severe Acute Respiratory Syndrome Coronavirus 2-specific antibody responses in Coronavirus disease patients. *Emerg Infect Dis*. 2020;26(7):1478-1488. doi:10.1101/2020.03.11.987958

20. Ng K, Faulkner N, Cornish G, et al. Preexisting and de novo humoral immunity to SARS-CoV-2 in humans. *Science*. 2020:370(6522):1343. doi:10.1126/science.abe1107

21. De Assis RR, Jain A, Nakajima R, et al. Analysis of SARS-CoV-2 antibodies in COVID-19 convalescent blood using a coronavirus antigen microarray. *Nat Commun*. 2021;12(6). doi:10.1038/s41467-020-20095-2

22. Khan S, Nakajima R, Jain A, et al. Analysis of serologic cross-reactivity between common human coronaviruses and SARS-CoV-2 using coronavirus antigen microarray. *bioRxiv*. March 2020. doi:10.1101/2020.03.24.006544

Table 1: Characteristics of N=24,079 *All of Us* Research Program participants with blood specimens collected January 2 to March 18, 2020 available for serologic testing

	***All of Us* participants**	
	N= 24,079	
Characteristic	**n**	**%**
Age in years, median (IQR)	53	(37, 65)
Sex at birth		
Female	13,692	57%
Male	10,100	42%
Other sex at birth	8	0%
Skip, prefer not to say, or no answer	279	1%
Race and Ethnicity		
Asian	630	3%
Non-Hispanic Black/African American	5,712	24%
Hispanic Black/African American	81	0%
Non-Hispanic White	11,896	49%
Hispanic White	279	1%
Pacific Islander	14	0%
Hispanic, Latino or Spanish	4,059	17%
Other race	785	3%
Skip, Prefer not to say or no answer	623	3%
Geography of residence		
Northeast	6,953	29%
Southeast	6,092	25%
Midwest	5,612	23%
West	5,408	22%
US territories	14	0%

Abbreviations:
IQR=interquartile range
US=United States
Footnotes:
Alaskan Native and American Indian participants were excluded from this analysis, at the request of tribal leaders.

Table 2: Characteristics of SARS-CoV-2 seropositive *All of Us* participants with specimens collected January 2 – March 18, 2020

ID	Race and ethnicity	Age	Sex at birth	Abbott (cut-off: 1.4)	EI (cut-off: 1.1)
1	Black/African American	45-49	Female	**3.09**	**2.681**
2	Black/African American	50-54	Male	**3.99**	**1.416**
3	Black/African American	55-59	Male	**3.17**	**1.846**
4	Black/African American	55-59	Female	**1.83**	**2.487**
5	Black/African American	60-64	Female	**4.13**	**1.984**
6	Hispanic, Latino, or Spanish	60-64	Female	**4.01**	**1.544**
7	Hispanic, Latino, or Spanish	70-74	Male	**2.43**	**1.125**
8	White	50-54	Female	**3.76**	**1.449**
9	White	65-69	Male	**8.09**	**2.891**

Bold indicates measurement above the cut-off (suggesting the participant was seropositive).
Abbreviations:
EI=EUROIMMUN
LLQ=lower limit of quantification

FIGURE LEGENDS

Figure 1: The number of *All of Us* participants with blood specimens collected January 2 to March 18, 2020 available for serologic testing from each state, N=24,079

Footnotes

There are <20 *All of Us* participants in Alaska and Hawaii.

Figure 2: The date of specimen collection compared to the date of the state's initial confirmed SARS-CoV-2 cases, n=9 SARS-CoV-2 seropositive *All of Us* participants with specimens collected January 2 – March 18, 2020

Footnotes

IL=Illinois
MA=Massachusetts
MS=Mississippi
PA=Pennsylvania
WI=Wisconsin

The first SARS-Cov-2 case in the state of Illinois was confirmed on January 24, 2020 (https://www.dph.illinois.gov/news/city-chicago-announces-first-local-patient-travel-related-case-2019-novel-coronavirus). The first confirmed case's spouse was the second confirmed case on January 30, 2020 (https://www.dph.illinois.gov/news/second-illinois-2019-novel-coronavirus-case-identified). The third case was a presumptive positive case announced on February 29 while awaiting confirmation by the CDC (https://www.dph.illinois.gov/news/state-illinois-public-health-officials-announce-new-presumptive-posi- tive-covid-19-case-illinois).

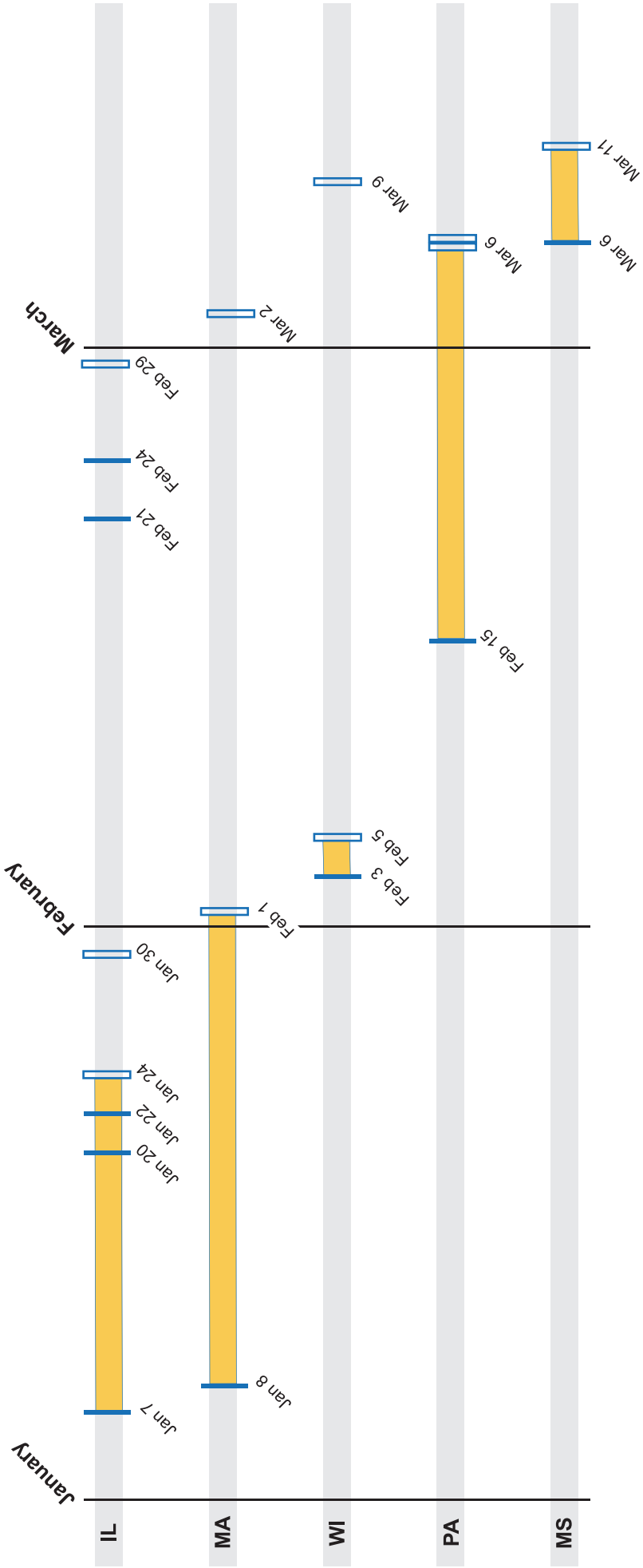
The first SARS-CoV-2 case in the state of Massachusetts was confirmed on February 1, 2020 (https://www.mass.gov/news/man-returning-from-wuhan-china-is-first-case-of-2019-novel-coronavirus-confirmed-in). The second confirmed case was March 2, 2020 (https://www.mass.gov/news/first-presumptive-positive-case-of-covid-19-identified-by-massachusetts-state-public-health).

The first two SARS-CoV-2 cases in the state of Pennsylvania were presumptive positive (while awaiting CDC confirmation) on March 6, 2020 (https://www.governor.pa.gov/newsroom/wolf-administration-confirms-two-presumptive-positive-cases-of-covid-19/).

The first SARS-Cov-2 case in the state of Wisconsin was confirmed on February 5, 2020 (https://www.dhs.wisconsin.gov/news/releases/020520.htm). The second confirmed case was March 9, 2020 (https://www.dhs.wisconsin.gov/news/releases/030920.htm).

The first SARS-CoV-2 case in the state of Mississippi was presumptive positive (while awaiting CDC confirmation) on March 11, 2020 (https://msdh.ms.gov/msdhsite/_static/23,21819,341.html).

Figure1: The number of All of Us participants with blood specimens collected January 2 to March 18, 2020 available for serologic testing from each state, N=24,079



Legend

= Specimen collection date of *All of Us* participant with evidence of SARS-CoV-2 antibodies on both the Abbott and EUROIMMUNE IgG ELISAs

= SARS-CoV-2 cases in the state