

Title page:

The Change in Seroprevalence in the US plus Puerto Rico between May and September of SARS-CoV-2 Antibody in the Asymptomatic Population

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Objective: Determine the seroprevalence and 3 month temporal change of SARS-COV-2 in the US and Puerto Rico in an asymptomatic well population.

Results: The prevalence of SARS-CoV-2 in the asymptomatic population has doubled during the May to September time period, going from 3.0% to 6.6%. Positive serology to SARS-COV-2 was similar in males and females but varies by age. The greatest increase was in the youngest population. Based on the September prevalence it is estimated that there are 11.1 million (bootstrap 95% CI: 10.8 – 11.5 million) asymptomatic SARS-CoV-2 infections in the US, which is 1.95 times the cumulative number of cases in the US reported to the CDC as of September 1, 2020.

Conclusions: The number of new SARS-COV-2 cases more than doubled in this 3

32 months period. The seroprevalence by age group is high in the youngest (<20,
33 7.8%) and low in the oldest (> 70, 2.0%). Over the three month study period the rate
34 of increase in prevalence for SARS-COV-2 serology varies by state with a range
35 from 0.84 (State of New York) to more than 12 (Tennessee). This study identifies
36 states where public health mitigation has blunted the spread and also areas where
37 additional levels of intensified public education, surveillance and targeted
38 intervention may be warranted.

39 Manuscript

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41 **CoV-2 Antibody in an Asymptomatic Population**

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43 Objective: Determine the rate of change in prevalence of SARS-CoV-2 antibody in
44 a self-reported asymptomatic well population over a three month period.

45

46 Method: Between 12 May and 25 September we examined SARS-CoV-2 antibody
47 results on 113,369 consecutive life insurance applicants who were having blood
48 drawn for the purpose of underwriting mortality risk.

49

50 Results: In May the seroprevalence of SARS-CoV-2 was 3.0%(547/18,441),
51 3.1%(981/31,822) in June and 6.6%(4180/63,103) in September. Positive serology
52 to SARS-CoV-2 was similar in males and females but varies by age. Geographical
53 distribution revealed a very high level of variation between states. Using 2019 US
54 Census state population data to adjust state specific rates of positivity, it is
55 estimated that the September level of seropositivity would correspond to 11.1
56 million asymptomatic SARS-CoV-2 infections in the US, which is roughly 2 times the
57 cumulative number of cases in the US reported to the CDC as of September 1,
58 2020.

59

60 Conclusions: The prevalence of positive SARS-CoV-2 serology increased more
61 than 2 fold during the three month period from May to September 2020 going from
62 3.0% to 6.6%. During this period state specific rates of increase in prevalence vary
63 wildly from 0.8(New York) to more than 12 fold (Tennessee). The estimated number
64 of total SARS-CoV-2 infections based on positive serology is substantially higher
65 than the total number of cases reported to the CDC. This study identifies states
66 where public health mitigation has blunted the spread and also areas where
67 additional levels of intensified public education, surveillance and targeted
68 intervention may be warranted.

Introduction

In early 2020 a novel coronavirus emerged in Hubei Province, China¹. The causative agent was a betacoronavirus most closely related genetically to zoonotic viruses found in bats, and clinically similar to recent emergent epidemic coronaviruses which caused Severe Acute Respiratory Syndrome (SARS) and Middle Eastern Respiratory Syndrome (MERS)². Since then, the virus has become a worldwide pandemic, infecting over 37 million persons and causing more than 1 million deaths as of this writing³. The first case in the United States occurred on January 20th, 2020⁴. And since then the Centers for Disease Control and Prevention has recommended that all states report laboratory-confirmed cases⁵. Case counts have been closely tracked by the CDC, the press, and academic institutions. However, because the illness caused by SARS-CoV-2 may be asymptomatic or minimally symptomatic⁶, these counts of cases may underestimate the number of persons who have been infected. Various studies of seroprevalence in the United States^{7,8} have shown different results based on timing and locality, but have been consistent in showing that seroprevalence is higher than would be implied by simple case counts based on viral antibody testing.

Because SARS-CoV-2 is novel, the presence of antibodies in the blood likely indicates a history of infection since the pandemic began, and serologic testing can be used to estimate the overall rate of infection, even in those who had minimal symptoms or who were never tested despite symptoms.

In this study, a convenience sample of blood specimens submitted to a commercial laboratory was used to conduct a survey of seroprevalence over a three month study period. The goal was to estimate the seroprevalence and overall rate of increase or decrease in positivity nationally and state by state.

Method: In the United States, the process of purchasing life insurance often involves a brief physical examination by paramedical professionals, the collection of height, weight and blood pressure measurements, and the testing of blood and urine specimens for common laboratory test related to overall health. Such tests are seldom, if ever, performed on individuals below age 16 years or above 85 years.

Between May12th and September 25, 2020, 113,369 consecutive individuals were tested for antibodies to SARS-CoV-2. Individuals were part of a convenience sample from a pool of life insurance applicants who had blood tests performed as part of life insurance underwriting. This sample represents approximately one fifth of all samples tested at the facility during that time. All applicants self-reported that they were well at the time of application. The antibody tests were performed using the Roche Elecsys Anti-SARS-CoV-2 kit on the Roche e602 analyzer, with a stated sensitivity of 100% and specificity of 99.8%. Other information available on test subjects included state of residence, age and sex.

The differences in continuous variables between the antibody-positive and negative groups were tested for significance with the Mann-Whitney U test, while differences in categorical variables were tested using the chi-square test. All statistical analyses were performed using R (version 3.6.1)⁹ and R-studio (version1.2.1335)¹⁰.

To estimate the total burden of SARS-CoV-2 infections in the US the total 2019 estimated census population was multiplied by the US population proportion between the ages of 16 and 80 (75.5%). Then, the state-specific proportion of positive tests was applied from our sample. Confidence limits were estimated by generating 5000 bootstrap samples (with replacement) of our data and recalculating the total number of US cases. Under and over-representation of states was determined by a ratio between the proportion of individuals living in a given state to the proportion of tests performed in that state.

Results

The overall sample included 113,369 individuals with a median age of 42 years (IQR: 33-54), 55.5% of whom were male. Geographical distribution deviated

somewhat from the overall population distribution of the US, with some under-representation from Maine, West Virginia, Vermont and Oklahoma, and over-representation from Nebraska, Hawaii, and Utah. Characteristics of the study population are displayed in Table 1. The antibody positive group tended to be slightly younger (median age 41) vs. the antibody negative group (median age 42). The distribution by decade of age is displayed in Table 2. This study estimated the seroprevalence of SARS-CoV-2 antibodies in a geographically diverse sample of adults in the US within a 3-month collection period ending the 25th of September 2020. The rate of positivity ranged from 0% to 14.7% by state and from 2-7.8% across age, displayed in Figure 1. While slightly more females were positive the difference is not statistically significant. The choropleth map of seropositivity roughly corresponds to the areas where the most SARS-COV-2 cases were reported during this period of time displayed in Figure 2. Our results suggest that many more infections occurred than were reported. This is likely due to asymptomatic or minimally symptomatic infections for which care was not sought or symptomatic infection for which testing was not available or obtained.

Various studies have been published, both before and after peer review, which have reported seroprevalence of SARS-CoV-2 antibodies in the US. Most notably, Havers et al¹¹. evaluated a convenience sample (n=16,025) of serological tests on sera submitted to 2 commercial laboratories from 10 US regions. Their estimates of seroprevalence ranged from 1% to 7%, with the highest rates occurring in the New York City metro area, Louisiana and Connecticut. The timeframe of this testing differed by region and was earlier than the current study. The authors estimated that the seroprevalence implied that between 6 and 24 times the number of infections had occurred in the studied regions than had been reported.

Stadlbauer et al reported on longitudinal changes in seroprevalence in New York City between late February and mid-April 2020¹². Over this period of time seroprevalence increased from 2.2% to 10.1%. Rosenberg et al also reported on seroprevalence in the New York metro area¹³.

The collection period was from April 19 to 28, 2020, and the estimate was 22.7%. The higher estimate than the current study, despite being performed in an earlier time period, is likely due to a geographical distribution that is more localized to the highest prevalence metro region, rather than the entire state of New York. We report that, around the time of study, the number of infections in the US was nearly 2 times higher than cases of infection reported suggesting a much more widespread pandemic, but with a smaller rate of hospitalization, complications and deaths. Weaknesses of the study include the imbalanced representation of the US states, as well as the lack of samples from those under age 16 or over age 80. The age distribution is also more heavily weighted to the young adult years, which is not representative of the US population. Although the sample size was large, it was not large enough to stratify by both age and geography when estimating population seroprevalence. Finally, the life insurance-buying population tends to be both healthier and mid-income or wealthier than average, and this could also bias the results in an indeterminate direction. Even with these stated limitations the study validates the need for population wide surveillance; it is in keeping with the AJPH editor on Accurate Statistics on COVID-19 Are Essential for Policy Guidance and Decisions¹⁴ clearly define the need for population wide seroprevalence surveys to collect the information necessary for sound national and public health policy decisions.

Conclusion

The rate of SARS-CoV-2 seropositivity in this population of self-reported well insurance applicants implies a burden of infection approximately 2 times higher than the number of reported cases. State specific rates of increase identify regions where public health mitigation strategies are working and areas where additional effort or changes in strategies are required. First public education must be intensified in regions where the rate of increase is the highest; the temporal prevalence data presented in this report should provide the “facts to support” these changes.

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Western IRB reviewed the study under the Common Rule and applicable guidance and determined it is exempt under 45 CFR § 46.104(d)(4) using de-identified study samples for epidemiologic investigation.

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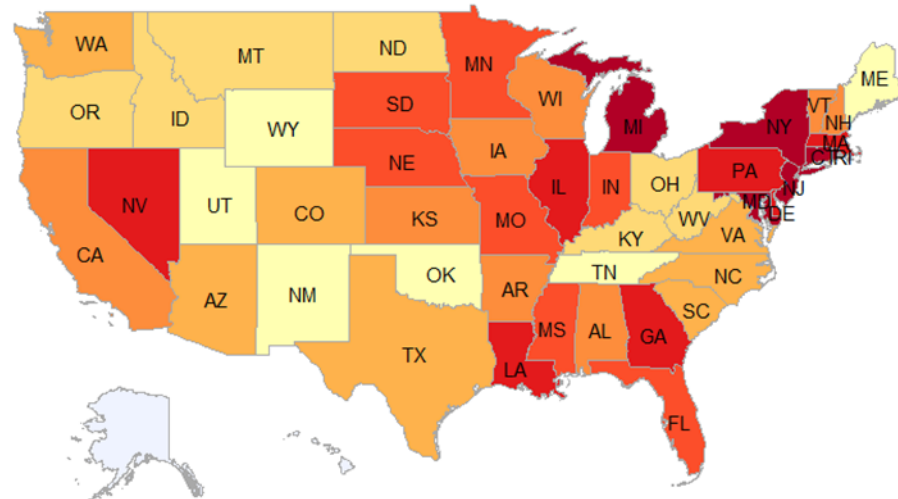
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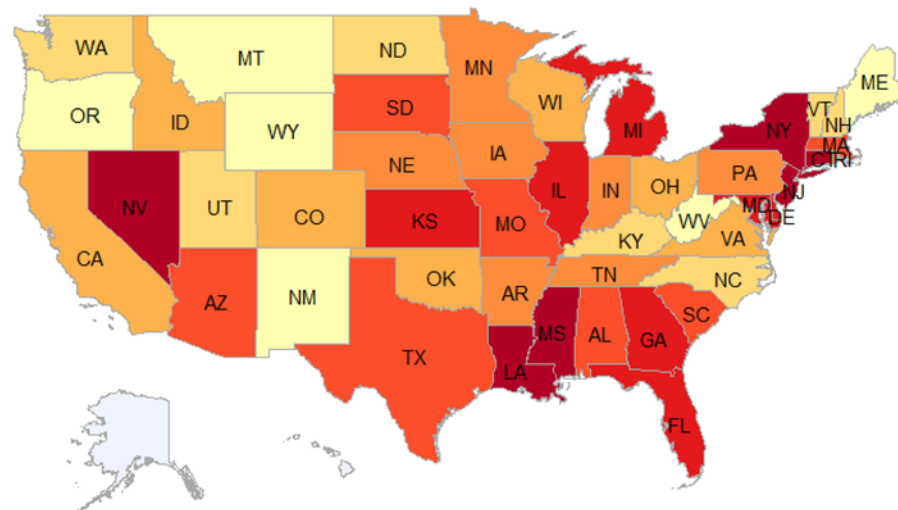
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Figure 1

SARS-CoV-2 Seroprevalence - June



SARS-CoV-2 Seroprevalence - September



Tables 1-3

Table 1: Characteristics of study population by SARS-CoV-2 antibody status.
Numeric values shown as median [IQR].
1 p-value by Pearson Chi-square test.

	SARS-CoV-2 negative n = 106,551	SARS-CoV-2 positive n = 5,623	p
Age (yrs)	42 [34,54]	41 [33,52]	0.000 ¹
18-40	45.6%	52.2%	
41-60	41.1%	39.9%	
61-85	13.3%	7.9%	
Sex, (% male)	55.6%	53.3%	0.002 ¹

Table 2. Sera positivity for SARS-COV-2 by age

	NON (106,551)	REA(5623)	Total
<= 20	1127(92.2%)	95 (7.8%)	1222
21 - 30	16112 (93.3%)	1155 (6.7%)	17267
31 - 40	31219 (94.9%)	1680 (5.1%)	32899
41 - 50	24133 (94.9%)	1305 (5.1%)	25438
51 - 60	19621 (95.4%)	937 (4.6%)	20558
61 - 70	12104 (96.8%)	404 (3.2%)	12508
71+	2049 (98.0%)	41 (2.0%)	2090
NO AGE	1296(93.4%)	91(6.6%)	1387
Total	106365 (95.0%)	5617 (5.0%)	113,369

Table 3: Seroprevalence for September and the rate of increase in prevalence from May to September by state.

STATE	Sept%	RATIO	STATE	Sept%	RATIO	STATE	Sept%	RATIO
NY	14.4	0.8	IN	5.2	2.6	NV	10.1	4.2
RI	3.7	0.9	GA	7.8	2.7	MS	9.3	4.4
MA	4.1	1.0	NC	3.1	3.0	FL	9.3	4.8
CT	5.9	1.0	SD	6.5	3.0	KS	8.2	5.1
NJ	9.5	1.0	CA	4.5	3.0	UT	3.6	5.1
DC	6.4	1.2	LA	11.9	3.0	AL	7.9	5.5
MI	5.5	1.3	IA	5.2	3.1	PR	3.9	5.6
PA	3.9	1.5	CO	4.1	3.1	OH	5.0	5.8
MD	6.2	1.5	MO	6.3	3.4	AZ	7.5	6.3
IL	5.7	1.6	KY	3.5	3.5	SC	6.6	6.4
MN	3.8	1.7	WA	4.1	3.5	TX	8.0	6.9
NE	4.2	2.0	AR	4.9	3.5	ID	5.8	8.1
DE	7.1	2.2	WI	4.8	3.7	OK	5.5	10.7
			VA	4.5	3.8	TN	6.8	12.9

September % is number of positives by state. Ratio is the September prevalence divided by the combined prevalence from May and June. The states of AK, ME, WY, VT, MT, ND, WV, NM, HI, and NH with fewer than 10 positives each were excluded from this analysis. Combined they have 45/3,319 (1.43%) positives.

