*Research Article in Health and Medicine*

# COVID-19 Exposure in Urban and Rural Areas

Megan Wong[1,*], Delbert Cai[1,*], and Raymond Cai[1,*]

[1] Columbia University Mailman School of Public Health, New York, NY 10019, United States

* Correspondence: meganwong311@gmail.com, delbertcai@gmail.com, dogeamazingness@gmail.com

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**Abstract:** The COVID-19 pandemic has majorly impacted the lives of people living in different living environments, such as large cities or small towns. However, there is limited knowledge about the difference in infection prevalence between urban and rural populations. To examine these differences, this study examined SARS-CoV-2 antibody levels in blood serum from 36 unvaccinated subjects of urban and rural backgrounds with no self-reported history of COVID-19 infection. Enzyme Linked Immunosorbent Assays (ELISA) detected the presence of SARS-CoV-2 antibodies in the subjects' blood. IgG and IgM assays were used to distinguish between prior (recovered) and more recent infections, respectively. A t-test was conducted to identify a statistically significant difference, or lack thereof, between the antibody levels detected in urban and rural participants. The results indicated that there was no significant difference between the two experimental groups for both IgG and IgM ($p = 0.09, 0.93$, respectively), although the p-value for IgG was low enough to be notable. Overall, this study suggests that there is no major difference between urban and rural environments that impact susceptibility to SARS-CoV-2 infection, although more research with a greater sample size should be conducted.

**Keywords:** SARS-CoV-2; ELISA; COVID-19; IgG; IgM; rural; urban

## 1. Introduction

Urban and rural conditions vary from each other in many different ways, one of which includes the crowdedness of the towns and cities. The exposure to certain viruses could be amplified depending on the number of interpersonal interactions between people living in the two environments. This study hypothesized that urban exposure rates to SARS-CoV-2 are higher than that of rural areas.

When a foreign substance enters the body, the immune system detects it as alien due to the differences in molecules on its surface to those in the body (Encyclopedia Britannica, 2022). The foreign molecule, termed an "antigen," binds to specialized white blood cells known as B-cells, causing them to divide, grow, and eventually produce millions of specific antibodies. As the antibodies enter the bloodstream, they bind to and neutralize the antigens that resemble the ones that triggered their initial immune response. The presence of antibodies in the bloodstream provides evidence for the subject's prior exposure to the corresponding antigen. This study measured antibodies, also known as immunoglobulins, of the classes IgG and IgM.

The antibodies tested in this study are Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV-2). This virus is part of the Coronavirus lineage, a species of virus which has been known before to present a high pandemic risk. This is the ninth documented coronavirus that affects humans and seventh identified in the last 20 years (Holmes et al. 2010). The first reports of SARS-CoV-2 occurred in December 2019 in Wuhan, China. It quickly spread worldwide, causing a global pandemic and shutdown. Many nations completely shut down their borders and had restrictive laws meant to stop the spread.

Many of the symptoms of SARS-CoV-2 are similar to that of the common flu. This includes fever, cough, tiredness, difficulty breathing, muscle aches, sore throat and more. More uniquely, COVID-19 infection has been associated with loss of taste or smell. The severity of these symptoms varies, as some infected individuals may be asymptomatic, while others have very severe symptoms. People who are older or have existing medical conditions have a higher risk of contracting serious illness. These include serious heart disease, cancer, COPD, diabetes (type 1 and 2), high blood pressure, liver disease and more.

A few earlier studies examined COVID-19-related differences between rural and urban areas. Kitchen et al. (2021) investigated the association between Area Deprivation Index (ADI) and prevalence of COVID-19, comparing the differences in metropolitan and non-metropolitan areas. The results indicated a stronger positive association between ADI and COVID-19 prevalence in rural areas compared to that in urban areas, even though a greater population of Americans reside in urban areas. Some of the main ADI factors that impacted the results included education, property value, and family income.

Paul et al. (2020) conducted a study during the COVID-19 pandemic using crowd-sourced data to compare COVID-19 prevalence rates by county, categorizing them by urban and rural. The results demonstrated that urban counties had significantly higher prevalence rates than rural counties. The study also found that African Americans, adults aged 25-49, smokers, and people with obesity had higher prevalence rates of COVID-19.

The purpose of this study was to determine whether or not SARS-CoV-2 IgG or IgM antibodies are more prevalent in people living in urban or rural settings among subjects who have not displayed symptoms of infection. The hypothesis being tested is that asymptomatic infections, as evidenced by the presence of SARS-CoV-2 antibodies in the blood, will be more prevalent in urban settings opposed to rural settings.

## 2. Research Design, Data collection and analysis Methods, Materials

The serum samples used in this experiment were obtained from blood draws from 36 unvaccinated subjects who reported no history of prior COVID-19 infections living in either urban or rural locations (18 from each). The definitions of urban and rural were based on the zip code of the subjects under the guidance of the U.S. Department of Agriculture Economic Research Service State-Level Maps (*25589_NY*, 2023). Identities and confidentiality of subject information were protected by assigning each participant a code number that was associated with each sample. No personally identifiable information was associated with any data point, blood tube, or downstream analysis. The authors of this report did not personally perform the blood draws, serum purifications, or multiplex antibody studies. Immunoglobulin data about these subjects was extracted from a much larger, NIH funded and IRB approved, dataset and study examining various aspects of COVID-19 infection across differing populations, ethnicities, genders and race.

Subjects were recruited by word-of-mouth. The average age in the urban group was 34.83 years (standard deviation 14.81) and the average age in the rural group was 35.33 years (standard deviation 13.69). Table 1 shows the population demographics overall and by group.

**Table 1.** Gender, age group, race, and ethnicity composition of participants.

| Category | | Total | | Urban | | Rural | |
|---|---|---|---|---|---|---|---|
| | | Number | Percent | Number | Percent | Number | Percent |
| Gender | Female | 22 | 61.11% | 12 | 66.67% | 10 | 55.56% |
| | Male | 14 | 38.89% | 6 | 33.33% | 8 | 44.44% |
| Age | 16-30 | 15 | 41.67% | 8 | 44.44% | 7 | 38.89% |
| | 31-50 | 15 | 41.67% | 6 | 33.33% | 9 | 50.00% |
| | 51-65 | 6 | 16.67% | 4 | 22.22% | 2 | 11.11% |
| Race | White | 26 | 72.22% | 10 | 55.56% | 16 | 88.89% |
| | African American | 7 | 19.44% | 6 | 33.33% | 1 | 5.56% |
| | Other/Multiracial | 3 | 8.33% | 2 | 11.11% | 1 | 5.56% |
| Ethnicity | Hispanic | 6 | 16.67% | 4 | 22.22% | 2 | 11.11% |
| | Non-Hispanic | 30 | 83.33% | 14 | 77.78% | 16 | 88.89% |

Blood was collected into BD vacutainer red-top tubes with no anticoagulants to extract the serum. The tubes were centrifuged, and the serum was extracted and used for IgG and IgM antibody detection.

The process of determining whether or not the patient had evidence of infection by SARS-CoV-2 was by using an Enzyme Linked Immunosorbent Assay (ELISA). An ELISA is a plated-based assay technique that can detect soluble molecules such as peptides, proteins, antibodies, or hormones. In this case, it was used to test for presence of IgG or IgM antibodies against the spike and nucleocapsid protein of the SARS-CoV-2 virus. Typically, in ELISA, the antigen is immobilized onto a microplate and then complexed with soluble antibody in the subject's serum that is then linked to a second antibody containing a reporter molecule that can be detected spectrophotometrically as a change in color intensity. More recently, ELISA assays can be run in multiplex, meaning multiple tests can be run on the same sample at the same time.

Quantitative results were compared by a two-tailed t-test assuming unequal variances to determine if there was a statistically significant difference in mean signal intensity between the two groups for both IgG and IgM.

## 3. Results

3.1. Statistical Results

**Table 2.** Results from t-tests on IgG and IgM comparing urban versus rural participants. The table shows the mean and standard deviation for optical density detected in the urban and rural groups, as well as the t-test details including the degrees of freedom (df), critical t-value (t Crit), t-statistic (t Stat), and p-value.

| **Category** | **Urban** | | **Rural** | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Mean | Std Dev | Mean | Std Dev | df | t Crit | t Stat | p-value |
| IgG | 2.925 | 6.799 | 0.057 | 0.066 | 17 | 2.110 | 1.790 | 0.091 |
| IgM | 0.191 | 0.116 | 0.194 | 0.106 | 36 | 2.028 | -0.087 | 0.931 |

3.2. Conclusion from Results

Using an alpha cut off of 0.050, no evidence of a statistically significant difference between rural and urban was seen for IgG (p = 0.091). There is no statistically significant or notable difference between rural and urban for IgM (p = 0.931). See Table 2 for details.

## 4. Discussion and Synthesis

The t-test for IgG and IgM was not statistically significant because the p-value was greater than 0.05 (0.091 and 0.931 for IgG and IgM, respectively-see Table 2). This suggests that living in an urban vs. rural environment did not make a notable difference in risk for asymptomatic SARS-CoV-2 infection. However, since the IgG p-value is relatively close to 0.05, these findings suggest that larger sample sizes might improve the accuracy of the results.

The sample size was small at only 36 participants, so the power of the t-test was lower than desired. It is possible that a more significant difference could be detected by increased sample size. Additionally, the study subjects skewed heavily towards white and non-Hispanic race and ethnicity, so may not be representative of the actual population. Using a larger sample size with more methodical sampling methods would address some limitations in this study.

Some possible future studies that can be done to expand the findings of this study include widening the range to other states or countries. This could help gather a more concrete result on whether urban or rural areas influence the asymptomatic infection rate. Other variables, including racial and ethnic subject background, pre-existing health conditions, and income levels, and their correlation with COVID-19 prevalence, could also be further investigated.

This information gleaned from this study could help individuals decide where to live as they are able to better weigh the pros and cons of living in an urban area opposed to a rural area. Being able to know if there are any increased risk factors when living in a certain area is very helpful, especially those that already have underlying health issues. These individuals could potentially already have a higher risk of contracting a viral disease and by moving to an area with higher risk might increase their chance of contracting it. Data could also help governments and communities to decide where to allocate resources and where to focus their treatments for a pandemic. While the COVID-19 pandemic has greatly subsided, many parts of the world are still recovering from the economic recession that it caused. By wisely allocating their resources, it could help countries to stabilize their economy and return to normal faster. More studies should be conducted to explore this further.

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**Conflicts of Interest:** The authors declare no conflict of interest.

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