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COVID-Inconfidentes - SARS-CoV-2 seroprevalence in two Brazilian urban areas in the fourth quarter 2020: study protocol and initial results

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COVID-Inconfidentes - SARS-CoV-2 seroprevalence in two Brazilian urban areas in the fourth quarter 2020: study protocol and initial results

COVID-Inconfidentes - seroprevalencia de SARS-CoV-2 en dos áreas urbanas brasileñas en el cuarto trimestre de 2020: protocolo del estudio y resultados iniciales

Authors list at the end of the article

Abstract

Objective: To describe study protocol and initial results of research project COVID-Inconfidentes. **Method:** This paper described the methodological procedures adopted and the prevalence of the SARS-CoV-2 infection in the population. A household survey was conducted between October and December 2020, in two historic cities of Brazil's mining region. Anti-SARS-CoV-2 antibody was detected using the Wondfo® rapid test. The face-to-face interview consisted of administration of a questionnaire containing registration data, sociodemographic and economic variables, living habits, general health condition, mental health, sleep habits, and eating and nutrition. **Results:** We evaluated 1,762 residents, of which 764 (43.4 %) were in Mariana and 998 (56.6 %) in Ouro Preto. For both cities, 51.9 % of the interviewees were female, with a predominance of the age range 35 to 59 years old (47.2 %). The prevalence of the SARS-CoV-2 infection was 5.5 % in both cities, 6.2 % in Ouro Preto, and 4.7 % in Mariana (p-value > 0.05). **Conclusion:** The study was effective to estimate the seroprevalence of infection by the virus and its findings will enable further analyses of the health conditions of the population related to social isolation and the risk of infection with SARS-CoV-2.

Descriptors: Coronavirus Infections; Pandemics; Health Surveys; Epidemiology; COVID-19 Serological Testing.

Resumen

Objetivo: Describir el protocolo de estudio y los resultados iniciales del proyecto de investigación COVID-Inconfidentes. **Método:** Este artículo describe los procedimientos metodológicos adoptados y la prevalencia de la infección por SARS-CoV-2 en la población. Se realizó una encuesta domiciliaria entre octubre y diciembre de 2020, en dos ciudades históricas de la región minera de Brasil. El anticuerpo anti-SARS-CoV-2 se detectó mediante la prueba rápida Wondfo®. La entrevista cara a cara consistió en la administración de un cuestionario que contenía datos de registro, variables sociodemográficas y económicas, hábitos de vida, estado general de salud, salud mental, hábitos de sueño, alimentación y nutrición. **Resultados:** Se evaluaron 1.762 habitantes, de los cuales 764 (43,4 %) estaban en Mariana y 998 (56,6 %) en Ouro Preto. Para ambas ciudades, el 51,9 % de los entrevistados eran del sexo femenino, con predominio del rango de edad de 35 a 59 años (47,2 %). La prevalencia de la infección por SARS-CoV-2 fue del 5,5 % en ambos municipios, del 6,2 % en Ouro Preto y del 4,7 % en Mariana (p > 0,05). **Conclusión:** El estudio fue efectivo para estimar la seroprevalencia de infección por el virus y sus hallazgos permitirán analizar más las condiciones de salud de la población relacionadas con el aislamiento social y el riesgo de infección por SARS-CoV-2.

Palabras clave: Infecciones por Coronavirus; Pandemias; Encuestas de Salud; Epidemiología; Pruebas Serológicas de COVID-19.

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1. Introduction

Emerging diseases, such as COVID-19, represent a major threat to society. Besides causing impacts on health systems, these diseases have the ability to change the social dynamics and harm the economy in its various segments. As well as the high mortality rate and/or the physical and psychological sequels that result from the involvement of organs and systems, limiting people's productive capacity (Carvalho et al., 2022; Morais et al., 2020; Palma et al., 2022; Sousa et al., 2021). Thus, the control of these diseases requires efforts that contribute to prevent the spread of infectious agents allied to epidemiological surveillance.

Since its emergence in December 2019, COVID-19 have generated ongoing challenges for governments and health authorities due to the rapid spread of the disease-causing virus and the risk of overloading health services and deaths from the severity of the disease (Bontempi, 2021; Liu et al., 2020; World Health Organization, 2020b).

In Brazil, the federal government's neglect and mismanagement in articulation of actions to pandemic control has led many municipalities to make important epidemiological surveillance decisions, in isolation, to avoid the spread of the virus in their territories. In this context, the difficulties inherent in the diagnosis and notification process, scarcity of human resources in the health services and epidemiological surveillance sectors, and lack of diagnostic tests have made it extremely difficult to understand the actual situation with respect to the frequency of both the disease and the infection (Guimarães-Ximenes-Neto et al., 2021; Hallal, 2020; Li et al., 2020; Qiu, 2020; Sousa et al., 2022).

Other difficulties related specifically to the disease include: the greater probability that only people with severe symptoms seek health care services and undergo diagnostic tests, the high percentage of infected people with mild symptoms or who are asymptomatic, and the social determination of the disease itself (Hallal, 2020; Li et al., 2020; Qiu, 2020; Boas et al., 2022). These factors can interfere with the dynamics of transmission, giving rise to underreporting, and the masking of the actual COVID-19 data. Therefore, it is extremely relevant to assess the viral circulation situation at the local level, especially in smaller cities, where resources are even more scarce.

Thus, a population study is an important tool that can be used to understand the actual epidemiological scenario of the disease in different territories, identify its magnitude, understand its transmission dynamics, and its demographic, geographical, and social distribution. It allows for the construction of valid indicators capable of describing the prevalence of the infection and predicting the risk of disease. Therefore, immunochromatographic tests for antibody detection represent an important tool for epidemiological surveillance services enabling the identification and interruption of the virus transmission chain, subsidizing the health surveillance actions that aim to increase monitoring and individual care, such as the adoption of prevention measures (Deeks et al., 2020; Mina et al., 2020; Nota Técnica no 5/2020).

In this context, was conducted a seroepidemiological survey (COVID-Inconfidentes Project) to determine the prevalence and dynamics of SARS-CoV-2 infection in the population of two Brazilian cities in the fourth quarter 2020 and subsequent socioeconomic and health effects.



This paper aimed to describe study protocol and initial results of research project COVID-Inconfidentes.

2. Methods

2.1 Study design

In this population-based serological study, a household survey was conducted in two historic cities of Brazil's mining region, between October and December 2020. The survey was carried out in three stages, with intervals of 21 days, in which different census sectors were evaluated in each stage.

The study design followed the recommendations of the seroepidemiological investigation protocol for SARS-CoV-2 infection of the World Health Organization (World Health Organization, 2020b). All the procedures adopted by this study, followed the Declaration of Helsinki (World Medical Association, 1997) and the Brazilian guidelines and standards for research involving humans (Resolução no 466/2012). The study project was approved by the Research Ethics Committee of the Federal University of Minas Gerais under Opinion No. 4,292,475 dated 09/22/2020, Certificate of Submission for Ethical Consideration No. 32815620.0.1001.5149.

2.2 Study location

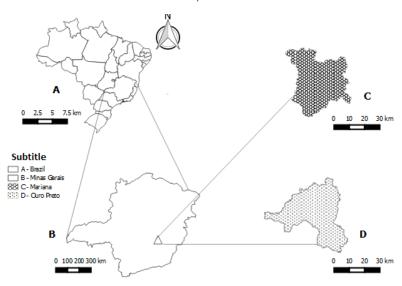
Mariana and Ouro Preto are two neighboring, medium-sized cities located in the Serra do Espinhaço, a central-southern region of the Brazilian state of Minas Gerais (Figure 1), known as the Iron Quadrangle. This metallurgical area is considered to be the largest national producer of iron ore. Since these cities have historical, sociodemographic, and geographical characteristics that are different from other Brazilian municipalities, and further, since surveys conducted in other regions of Brazil may not reflect the real prevalence of infection in the resident population, it is important to understand the dynamics of the epidemic locally.

The main economic activities of the municipalities revolve around the mining and metallurgical industries, tourism, and services, especially those related to the Federal Institution of Higher Education which is based in the city. These generate high population mobility due to the flow of workers and students (Instituto Brasileiro de Geografia e Estatística, 2013).



Figure 1

Geographic location of the municipalities in COVID-Inconfidentes study. Mariana and Ouro Preto - Minas Gerais, Brazil, 2020.



2.3 Study population and sample

Residents in permanent households in the urban areas of the cities of Ouro Preto and Mariana, aged 18 years or older, were considered eligible for this study. According to data from the 2010 demographic census, there were 44,569 and 33,902 inhabitants, distributed in 17,753 and 14,078 households, in Ouro Preto and Mariana, respectively (Instituto Brasileiro de Geografia e Estatística, 2013). The protocols adopted and described below were identical for both the municipalities and considered the population data described above.

2.4 Sample size calculation

The sample size calculation was based on the population estimate for each city, taking into consideration a confidence level of 95 %, a design effect equal to 1.5, and the parameters presented in Table 1, which also shows the sample distribution by stage of survey. Considering the losses resulting from refusals, absence of residents selected for the study, and the existence of closed households during the visit, a percentage of 20 % of recomposition was added to the sample size in each city.



Table 1
Sample size calculation for the stages of the survey COVID-Inconfidentes.
Mariana and Ouro Preto - Minas Gerais, Brazil, 2020.

Stage of the Survey	% estimated infection	Precision	Sample Ouro Preto	Sample Mariana	Sample Total
1	3 %	3.0 %	186	186	372
2	5 %	3.5 %	223	223	446
3	10 %	4.0 %	323	323	646

A three-stage conglomerate sampling design was adopted: census sector (selected for stages 1, 2, and 3 of survey, randomly and without replacement), households (selected from a systematic sampling process), and residents (one resident selected randomly, using the "Sorteador de Nomes®" applicative) (Aramburu, 2018).

2.5 Training the data collection team

The data collection team was composed of researchers, interviewers, and phlebotomists. The interviewers were recruited from the academic environment, with those interested being students who were enrolled regularly in undergraduate or graduate courses. The candidates underwent interviews in which their availability and interest were evaluated. After the selection, the students received a digital document, a video that explained the research objectives.

The interviewers were subdivided into teams coordinated by the six researchers. Each team received online training conducted by the coordinators/researchers, in which the project objectives and data collection methodology were addressed, and the interview questionnaire was disseminated. Subsequently, face-to-face training sessions for interviewers were conducted at the Medical School of the Federal University of Ouro Preto, on the use of the digital questionnaire (<u>Appendix 1</u>) and how to approach the interviewees, ethical aspects, and biosafety during data collection.

During the face-to-face training, the research team and medical infectologist responsible for the team's biosafety were present. The health monitoring of the interviewers was conducted through periodic evaluation, prior to the beginning of each stage of the survey, using health questionnaires associated with the results of serological testing (immunochromatography reaction).

The phlebotomists also received digital documents and videos explaining the importance of their work and the research methodology. In a second session, these professionals were trained, in person, by a nurse who addressed the venipuncture technique for blood collection and biosafety measures.



The videos that explained the theme of the project were also disseminated to community health agents who were responsible for guiding the coordinators in the census sectors of the areas of greatest social vulnerability, which were randomly selected in the first stage of the survey in both cities. All the videos were created in a language accessible to the target audience and addressed the role of each professional in the data collection team.

2.6 Data Collection

The data collection process included listing and approaching households, recruiting participants, the collection of venous blood, and interviews. The activities were carried out during weekends (Friday, Saturday, and Sunday), aiming to enhance the participation of residents who worked during the week and increasing the representativeness of this population group.

In the week prior to the collection, the research team carried out the enrollment of households in preselected census sectors and developed plans to raise awareness in the population through the distribution of folders and posters in public places as well as the dissemination of the survey via local radio stations, social media (Instagram and Facebook), the University TV, WhatsApp groups, and religious celebrations.

An informative folder was attached in the door to randomly selected household, indicating that household had been selected for the collection which would take place during the weekend. Taking into consideration the possibility of loss (refusal or closed household), an informative folder was attached to the neighbors on the left and right of the selected household (<u>Appendix 2</u>).

On the data collection days, the entire field team met at a support spot, where they donned personal protection equipment, received general instructions and were informed about the census sectors would be visited that day. The teams then moved to the census sectors where each interviewer approached the household and under the supervision of the researchers, selected the adult to be interviewed. In the case of a closed household, the absence of the resident selected or in the case of a refusal, the team would approach the neighbor to the right and, in the case of failure there, the household to the left of the one selected. If the refusal persisted, or if there were no residents in the house, the team moved on to the next house selected. There was no return to closed homes.

After the presentation of the research process and the participant's acceptance, they were asked to sign the informed consent form and the phlebotomist collected the venous blood with a venipuncture in the antecubital fossa region, the participant seated, and the arm supported on an appropriate support. For the blood collection, a 7.5 mL S-Monovette® (Sarstedt) serum gel tube was used to obtain the serum which was used for the COVID-19 rapid test and the other biochemical analyses. A 2.7 mL S-Monovette® (Sarstedt) collection tube containing sodium fluoride/EDTA was used to obtain whole blood for the molecular biology analyses.

At the end of the blood collection process, the interviewer commenced the face-to-face interview, which lasted between 30 to 45 minutes, varying according to the perception and dexterity of the interviewee in answering the questions. Data collection was conducted using the DataGoal® application that was



installed on the tablets to capture the geographic coordinates (latitude and longitude) of the interviewee's home.

All the recommendations of the national protocols to fight the new coronavirus were adopted, with an emphasis on hand washing, the use of personal protection equipment (apron, cap, disposable surgical mask, and goggles), changing every shift, approaching the research participants preferably in the peridomestic area and, when this was not possible, in a large, ventilated room (Nota Técnica GVIMS/GGTES/ANVISA n° 04/2020).

The interviewers kept a minimum distance of 1.5 m from the interviewees, and physical contact was restricted to only the moment of biological material collection.

2.7 Instruments and Data Collection

The face-to-face interview consisted of the administration of a questionnaire, in electronic format, that contained registration data, sociodemographic and economic variables, life habits, general health condition, mental health, sleep habits, and food and nutrition (<u>Appendix 1</u>), as follows:

- **Registration data:** sex, individual taxpayer registry (CPF) number, filiation, date of birth, telephone number, and symptoms in the last 15 days.

Sociodemographic and economic variables: race/color, marital status, filiation, education, occupation, and family income prior to and during the pandemic.

- **Lifestyle:** routine activities and behaviors, sun exposure, lifestyle, participation in religious celebrations prior to and during the pandemic
- **Health conditions:** Self-perception of health, presence of physical pain, use of medications/supplements, weight, height, and perception of COVID-19.
- **Mental health:** Assessment of anxiety and depression symptoms using the General Anxiety Disorder-7 (GAD-7) (Moreno et al., 2016) and the Patient Health Questionnaire-9 (PHQ-9)6 (Santos et al., 2013) scales.
- **Sleep habits:** Sleep quality was measured using the Pittsburgh Sleep Quality Index (PSQI) scale (Bertolazi et al., 2011).
- **Food and nutrition:** Daily frequency of food consumption, perceived food prices, comfort food consumption, food accessibility, and availability of food at home, adapted from the research on Surveillance of Chronic Diseases by Telephone Inquiry VIGITEL, from the Ministry of Health (Ministério da Saúde, 2020).

The questions about personal information were aimed at obtaining information for the registration and reporting of serological test results to the Ministry of Health, through the electronic system of epidemiological surveillance for notifications to the Unique Health System (e-SUS VE), to assist in the monitoring and analysis of the epidemiological situation of COVID-19 in the municipalities. The other



questions assessed the living conditions of the population, allowing us to perform a situational analysis of the determinants of health.

Data collection was performed using offline devices and, at the end of the day, in a place with a stable internet connection, the devices were synchronized and the information that was collected, was sent to the storage cloud of the software used (DataGoal®). The devices were then loaded for use on the next collection day.

2.8 Laboratory Analysis - immunochromatography test for antibody detection

At the end of each shift (morning and afternoon), the blood samples were checked, packed in a thermal box, and transported to the Epidemiology Laboratory of the Medical School of the Federal University of Ouro Preto for processing and storage by the technical team. In the laboratory, the serum tubes were centrifuged at 2500 rpm for 15 min, and a yield of approximately 3 mL of serum per sample was obtained. Then, the serum was aliquoted in different polypropylene tubes and portioned as follows: one tube with a 50 μ L aliquot for testing COVID-19 and the rest in aliquots of approximately 500 μ L to 1000 μ L for use in the other biochemical analyses. These were stored in a freezer at -80°C until required. The EDTA tube was stored in a -20 °C freezer for the subsequent extraction of the genetic material.

Serological screening for anti-SARS-CoV-2 antibodies was performed by trained professionals using immunochromatographic reaction-based tests. The diagnostic One Step COVID-2019 Test (Guangzhou Wondfo Biotech®, China) was used to determine IgM/IgG antibodies in whole blood, serum, or human plasma, qualitatively, without distinguishing the immunoglobulin type (Celer, 2020).

In this study, according to the manufacturer's instructions, $10~\mu L$ of serum and three drops of the buffer solution were added to the reaction well in the test device. The sample was absorbed by capillary action and mixed with the SARS-CoV-2 antigen-dye conjugate, which flowed through the pre-coated membrane. In cases where the level of SARS-CoV-2 antibodies in the sample was at or above the cut-off range (minimum detection limit of the test), antibodies that were bound to the antigen-dye conjugate, were captured by the human anti-IgG antibody and the anti- μ -chain antibody complex and immobilized in the test region of the device, producing a colored test band, indicating a positive result.

When the level of the SARS-CoV-2 antibody in the sample was zero or below the cut-off, there was no visible colored band formed in the test region of the device, indicating a negative result. As a control for the procedure, a colored line appeared in the control region, indicating that the test was working correctly. The minimum waiting time for the reading was 15 min, while the maximum time that was allowed was 20 min.

According to the manufacturer's validation data, the sensitivity of the One Step COVID-2019 Test (Wondfo®) was 86.43% (95 % CI: 82.58% - 89.58%) and the specificity was 99.57% (95 % CI: 97.92% - 99.92%). It is important to note that the negative results did not exclude a SARS-CoV-2 infection, and the positive results cannot be used as absolute evidence of the presence of SARS-CoV-2. These results only indicate a previous exposure to the infectious agent (Celer, 2020).



2.9 Issuing and Delivery of Reports

After the serological testing for anti-SARS-CoV-2 antibodies, a report was issued and sent to each study participant. The reports served as information for the participants and teams of the COVID-19 Confrontation Committees in the municipalities.

The reports were issued by professionals from the laboratory team, using Microsoft Excel® spreadsheets, which contained information provided by the participant (full name, date of birth, and contact phone number) and the results of the serological test (reactive or non-reactive). In addition, the reports contained information regarding the interpretation of the tests and the precautions to be taken, as well as the contacts of the Health Surveillance Departments of each city, so that the study participants could ask questions and receive guidance on the procedures to be adopted, especially those whose results were positive.

The reports were issued within seven days after collection of the data, in a pdf file, via WhatsApp messaging application. Participants who did not use the application, received the results by telephone and, for those who did not have a telephone, the report was delivered to the home.

Finally, the Microsoft Excel® spreadsheets with the participants' data and test results were forwarded to the Mariana and Ouro Preto Health Secretariats for case monitoring and management.

2.10 Database consolidation and consistency and sample weight

At the end of the collection period, the database was consolidated by merging the three stages of the survey conducted in the two cities. A consistency analysis was then carried out to verify the consistency of the data, the presence of typing errors, or the absence of data. The sample weight of each selected unit (census tract, household, and individual) was calculated separately for each city, taking into consideration the inverse of the probability of selection, according to the sampling plan of the study and according to data from the 2010 demographic census. An adjustment was applied to compensate for the loss of interviews due to non-response and the weight of the household and the selected resident was calibrated, so that the population totals by sex and age group (18 to 29 years, 30 to 49 years, 50 to 59 years, and 60 years and older) were consistent with the 2019 population projections (DATASUS) (Ministério da Saúde, 2011).

2.11 Data Analysis

The relative frequencies of the sociodemographic variables were estimated for each stage of the survey (1, 2, and 3) and for all cities. The prevalence rate of seropositivity for SARS-Cov-2 and the confidence intervals (95 % CI) were estimated for each stage of the survey, per city, and for both cities. Pearson's Chisquare test was used to compare the proportions of the sociodemographic variables and the prevalence of seropositivity for SARS-Cov-2 between each stage of the survey. The weighting factors were incorporated into the analyses using the svy command of the Stata® software, version 12.0. A significance level of 95 % (p-value \leq 0.05) was adopted.



2.12 Role of the funding source

The funder of the study had no role in study design, data collection, data analysis, data interpretation, or writing of the report. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit for publication.

3. Results

We approached 5,252 households, of which 2,523 (48.0 %) were in the city of Mariana and 2,713 (52.0 %) in the city of Ouro Preto. Of the total, 1,912 (36.4 %) households were closed; in 1,079 (20.5 %), the residents refused; in 267 (5.1 %), the selected residents were absent; and in 1,762 (33.5 %) the residents agreed to participate in the study, of which 764 (43.4 %) were in Mariana and 998 (56.6 %) in Ouro Preto, as described in Table 2.

Table 2

Number of households approached, households closed, number of refusals, absent sorted dweller and number of households collected. Mariana and Ouro Preto - Minas Gerais, Brazil, 2020.

		Total households approached	Households Closed	Refusals	Absent Sorted Dweller	Households Collected
Mariana						
1° Stage	16 to 18/10	523 (100.0 %)	183 (35.0 %)	112 (21.4 %)	47 (9.0 %)	181 (34.6 %)
2° Stage	06 to 08/11	988 (100.0 %)	377 (38.2 %)	204 (20.6 %)	109 (11.0 %)	298 (30.2 %)
3° Stage	27 to 29/11	1012 (100.0 %)	407 (40.2 %)	244 (24.1 %)	76 (7.5 %)	285 (28.2 %)
Ouro Preto						
1º Stage	30/10 to 01/11	885 (100.0 %)	310 (35.0 %)	181 (20.5 %)	107 (12.1 %)	287 (32.4 %)
2° Stage	20 to 22/11	993 (100.0 %)	369 (37.2 %)	172 (17.3 %)	98 (9.9 %)	354 (35.6 %)
3° Stage	11 to 13/12	851 (100.0 %)	266 (31.3 %)	166 (19.5 %)	62 (7.3 %)	357 (41.9 %)
Total		5252 (100.0 %)	1912 (36.4 %)	1079 (20.5 %)	267 (5.1 %)	1762 (33.5 %)

For both cities, 51.9 % of the interviewees were female, with a predominance of the age range 35 to 59 years old (47.2 %), 51.5 % were married, 67.9 % black or mulatto,71.5 % with more than nine years of schooling and 61.2 % with a family income higher than two minimum wages. No significant differences were observed in the distribution of the sociodemographic variables between each stage of the survey (Table 3).



Table 3

Sociodemographic characteristics of participants in the Covid-Inconfidentes Epidemiological Survey. Mariana and Ouro Preto - Minas Gerais, Brazil, 2020.

Variables	Total (n= 1762)	1º Stage (n= 469)	2º Stage (n= 662)	3º Stage (n= 631)	p- value*
Age Group					
18 - 34 years	34.0 (30.5 – 37.6)	36.4 (29.3 – 44.1)	34.7 (30.3 – 39.2)	30.9(25.4 - 36.9)	
35-59 years	47.2 (43.1 – 51.4)	44.8 (37.8 – 52.1)	46.5 (41.8 – 51.4)	50.3 (41.8 – 58.9)	0.748
≥ 60 years	18.8 (16.0 – 21.9)	18.8 (13.9 – 25.4)	18.8 (14.7 – 23.8)	18.8 (14.4 – 24.2)	
Marital status					
Single	36.1 (32.9 – 39.5)	32.0 (25.8 – 38.9)	37.0 (30.9 – 43.5)	39.3 (35.2 – 43.7)	
Married	51.5 (47.4 – 55.5)	53.5 (44.9 – 62.0)	49.2 (42.7 – 55.8)	51.7 (46.2 – 57.0)	0.214
Othersa	12.4 (10.1 – 15.1)	14.5 (9.9 - 20.6)	13.8 (10.2 – 18.7)	9.0 (6.8 - 11.8)	
Cor					
White	26.1(22.1 - 30.7)	25.9 (19.2 – 33.9)	25.9 (18.3 – 35.1)	26.8 (20.7 – 33.8)	
Black	21.5 (17.7 – 25.8)	19.3 (13.3 – 27.6)	22.1 (15.2 – 31.0)	23.0 (18.0 - 28.9)	0.888
Brown	46.4 (37.3 – 50.1)	47.9 (38.7 – 57.3)	47.5 (39.2 – 56.0)	43.6 (37.3 – 50.1)	0.000
Others	6.0 (4.8 - 9.0)	6.9 (4.3 - 10.8)	4.5(2.8 - 7.1)	6.6 (4.8 - 9.0)	
Scholarity					
Notliterate	1.7(0.8 - 3.5)	2.1 (0.6 - 6.8)	2.1 (0.6 - 6.9)	0.9(.3-2.6)	
Upto 9 years	26.8 (23.2 – 30.6)	30.5 (24.2 – 37.6)	24.2 (19.7 – 29.3)	25.6 (18.8 – 33.8)	0.523
> 9 years	71.5 (67.5 – 75.2)	67.4 (59.9 - 74.0)	73.7 (68.0 - 78.7)	73.5 (65.2 – 80.4)	
Family Income					
≤ 2 MW	38.9 (34.1 – 43.8)	43.5 (33.5 – 54.1)	38.8 (33.3 – 44.7)	34.0 (26.4 – 42.6)	
> 2 a ≤ 4 MW	34.0 (30.4 – 37.8)	27.3 (20.1 – 35.9)	36.7 (32.8 – 40.7)	38.2 (32.6 – 44.2)	0.265
> 4 MW	27.2 (22.6 – 32.2)	29.2 (20.5 – 39.6)	24.5 (18.8 – 31.2)	27.8 (20.0 – 37.1)	

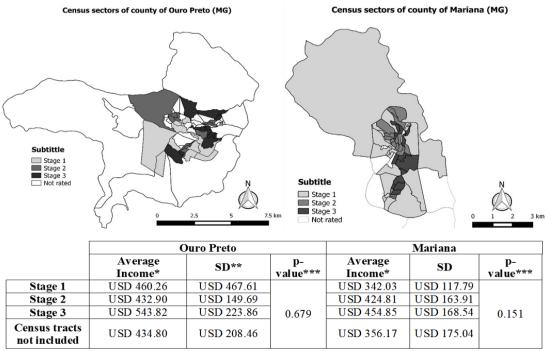
[%] and 95 % CI in parentheses (all such values). *chi-square test. ^a separated/divorced or widowed. MW: minimum wage. Minimum wage value: BRL 1,045.00 ≈ USD 194.25 (1 USD = 5.3797 BRL).

Figure 2 shows the distribution of the census sectors of the urban areas included in each stage of the survey according to data from the IBGE, taking into consideration, the proportionality of the number of households and the average income and ensuring the representativeness of the different socioeconomic levels in the final sample and in each stage of the survey.



Figure 2

Distribution of the census sectors included in the COVID-Inconfidentes survey, in the urban areas of the study cities. Mariana and Ouro Preto - Minas Gerais, Brazil, 2020.



^{* 1} USD = 5.3797 BRL. **SD = standard deviation. ***p-value for the ANOVA-oneway test.

The global prevalence of the SARS-CoV-2 infection was 5.5 %, being 6.2 % in Ouro Preto, and 4.7 % in Mariana (p > 0,05).

3.1 Discussion

The rapid spread of SARS-CoV-2 and the severity of Covid-19 present important diagnostic challenges and require the adoption of techniques capable of ruling out a possible infection, confirming a recent infection, identifying previous infections, and assessing the immune response. In this context, it is essential to understand the prevalence of the virus, so that containment measures can be adopted appropriately (Deeks et al., 2020; Mina et al., 2020).

Since most patients develop antibodies only in the second week after symptom onset, the use of rapid tests for the detection of antibodies to SARS-CoV-2 in clinical practice is not recommended by the World Health Organization (World Health Organization, 2020a; Ying et al., 2020). However, the use of these tests in population studies is relevant, because it allows the identification of a previous exposure to infection and the knowledge of the distribution and characteristics of the population that had contact with the virus, indicating geographical areas with higher viral circulation (Deeks et al., 2020; Mina et al., 2020). Immunochromatographic tests for antibody detection allow for frequent testing because they are inexpensive and easy to perform, making them useful in limiting the spread of the disease. However, these



tests should not replace reference diagnostic tests, such as RT-PCR (Centers for Disease Control and Prevention, 2021; Deeks et al., 2020; Mina et al., 2020; Nota Técnica n° 5/2020). A review study on the efficacy of using immunochromatographic antibody tests in seroprevalence surveys showed that there was a significant increase in the accuracy of the diagnosis of COVID-19 over the first three weeks of symptom onset, with 30 % in the first week, 70 % in the second week, and over 90 % in the third week. The authors also highlighted the low percentage of false-positive results (2 %) observed with these tests (Deeks et al., 2020). Moreover, it is noteworthy that immunochromatographic serology tests perform better when performed with serum or plasma samples, especially in relation to diagnostic sensitivity (Santos et al., 2020).

In this sense, we believe that the timing of the collection after exposure is a factor that can interfere with the detection of positive cases during serological population studies. This may explain the differences in seroprevalence observed between the cities of Mariana and Ouro Preto in this study. Nevertheless, it is noted that the prevalence found in these two cities in the Inconfidentes region during the months of October to December 2020 was higher than that described in a population-based survey conducted in the state of Rio Grande do Sul, Brazil, from April 11 to May 11, 2020, which found a prevalence of less than 1 % (Silveira et al., 2020). On the other hand, these results are close to those observed in states in the northeastern region of Brazil during a national household survey conducted between May 14 and June 7, 2020, which demonstrated a high heterogeneity in the prevalence of anti-SARS-CoV-2 antibodies in different regions of Brazil (Hallal et al., 2020).

According to the literature, several factors can explain the differences found in SARS-CoV-2 infection rates between regions and distinct population groups, such as disparities in the stage of infections, low testing, and late detection of cases; divergence in the adoption of control strategies, such as restrictions on the movement of people, local sociodemographic and socioeconomic characteristics, and environmental and meteorological factors, such as humidity and air pollution (Copat et al., 2020; Guimarães-Ximenes-Neto et al., 2021; Boas et al., 2022).

The limitations of our study include the restriction of the sample to two medium-sized Brazilian cities, which represent only 12.2 % of the Brazilian municipalities. Residents in rural areas of the municipalities were excluded from the study. Although the refusal rate was within expectations, the response rate of 33.5 % was lower than that observed in national surveys conducted in Brazil (53 – 54 %) and Spain (60 %) (Hallal et al., 2020; Ministerio de Ciencia e Innovacion and Ministerio de Salud, 2020). In 41.5 % of households it was not possible to collect information from residents because they were absent.

Other studies linked to this project will be developed to further analyze the health conditions of the population studied, related to social isolation and the risk of contamination by SARS-CoV-2, as well as the socioeconomic impacts resulting from them. Therefore, in addition to estimating the prevalence of the SARS-CoV-2 infection in the population, the findings of this study makes it possible to understand the dynamics and profiles of illnesses in the population and their possible determinants, contributing to the improvement of health care, both in the Unified Health System and in the private network.



The results will help deepen the knowledge about social inequalities in the study cities, favoring the implementation of public policies that minimize inequalities and their impacts on health, food, and quality of life of the population.

Furthermore, this study demonstrates that despite the difficulties inherent in facing the Covid-19 pandemic, Brazilian science had effective knowledge, methods, and techniques for obtaining effective information to guide the disease control measures. At that time, the extensive rapid test availability was an important and necessary measure to guide public policies for the prevention and control of the spread of disease throughout the country. In this way, Brazil could have reduce the negative impact of the disease on the population's health and avoided the high number of deaths that occurred in the country.

4. Conclusion

The initial results of research project COVID-Inconfidentes showed that the seroprevalence of SARS-CoV-2 infection in Mariana and Ouro Preto was higher than found in a survey conducted in state of Rio Grande do Sul (April to May, 2020) and similar to observed in states of the northeastern region of Brazil in a national survey (May to June, 2020), indicating a heterogeneous distribution of the infection in the different regions of Brazil at that time.

Despite the inherent limitations in the diagnostic efficacy of the rapid serological test for the detection of antibodies against SARS-CoV-2, its use proved to be effective in verifying the seroprevalence of viral infections in the cities evaluated, allowing researchers to assess the circulation of the virus and to determine the percentage of people exposed in the cities, providing a basis for public managers to make effective decisions, in a socio-health and epidemiological context, to reduce the number of cases and deaths. Furthermore, the future findings of this study will play an important role in providing information with regard the socio-economic and health impacts on the population, resulting from social distancing.

New surveys are needed to understand the pandemic situation in these new phases, including, if possible, rapid antigen tests, which were not yet available at the time of this study was designed. In addition, genotyping is needed to identify circulating viral strains and behavior in the community.

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7. Author contributions

ALM and GLLMC conceived the study and obtained funding for the study. LGL, LAAMJ, HNC, ICSJ, SSM, APD, TSS, AMSR, APB, NNL, BSS, CAS, KFB and CZM supported or conducted data collection. LAAMJ and ICSJ conducted the statistical analyses with support from ACSA. LGL wrote the first version of the manuscript. RDM, JCCC, MCM, RCRMN, LR and WTC reviewed the work substantively. All authors have approved the submitted version.

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