(RESEARCH ARTICLE)

Check for updates

Epidemiological Survey of COVID-19 in the municipality of Rio Paranaíba, Minas Gerais, Brazil, After Pandemics

Maria Luiza dos Santos Leocádio [1], Giovanna Ferreira da Fonseca [1], Fabiano Bezerra Menegídio [2], Caroline Garcia [3], Karine Frehner Kavalco [1] and Rubens Pasa [1, *]

[1] Laboratory of Molecular Diagnosis, Federal University of Viçosa, campus Rio Paranaíba. Route MG 230, km 7, 38810-000, Rio Paranaíba, MG, Brazil.
[2] Laboratory of Bioinformatics and Omic Sciences, University of Mogi das Cruzes, Mogi das Cruzes, SP, Brazil.
[3] Cytogenetics Laboratory, State University of Southwest Bahia, Jequié, BA, Brazil.

Abstract

COVID-19 is a respiratory infection caused by the Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2), a pathogen characterized by high mutation and transmission rates. The rapid circulation of the virus is exacerbated by asymptomatic infections, generating new lineages with genetic variations. These factors contribute to reduced treatment and vaccination efficacy and alterations in disease severity levels, among other consequences. Mass testing is one of the most effective methods for epidemic control, contributing to the monitoring of active cases and describing the epidemiological profile of infections. The city of Rio Paranaíba hosts one of the campuses of the Federal University of Viçosa, where the Laboratory of Molecular Diagnostics (LMD) is located, a specialized center for COVID-19 testing serving various municipalities in the region. Thus, the present study aimed to investigate the epidemiological scenario of COVID-19 in the municipality of Rio Paranaíba (MG). Study participants provided saliva samples, which were sent for processing at the LMD, where RNA was extracted, and the presence of SARS-CoV-2 was detected by RT-qPCR. A total of 324 tests were conducted throughout May and June 2023, with an average of 40 saliva samples collected per week. The majority of participants were in the 20-29 age group (56.8%), female (55.9%), and had received 3 doses of the COVID-19 vaccine (54.6%). The tests yielded 321 negative samples and 3 positive samples, all of which were from symptomatic participants, with only one having a comorbidity (chronic lung disease). The majority of positive cases were recorded in men in the 20-29 age group who had received 2 doses of the COVID-19 vaccine.

1. Introduction

Emerging infectious diseases, such as Severe Acute Respiratory Syndrome (SARS), are considered a significant public health challenge. According to publications by the United States Centers for Disease Control and Prevention (CDC), these are defined as infections that have recently appeared in a population or have existed previously but are rapidly increasing in incidence and geographical range. They may occur through the introduction of an etiological agent into other species or as a variant of an existing human infection, followed by its establishment and rapid dissemination within a new host population. Their occurrence is also prompted by the recognition of a previously undetected infection already present in the population and the infectious origin of an established disease (1).

Consequently, infectious agents are continuously adapting, giving rise to new diseases or causing existing ones to propagate in new waves of infections. Factors such as habitat degradation and fragmentation, global climate change, the

advent of globalization, and increased international travel facilitate vector access and the emergence of etiological agents capable of infecting humans (2). When a disease spreads locally within a population, it is characterized as an epidemic. When this epidemic reaches a global scale, also affecting social and economic factors, it becomes a pandemic (3).

The outbreak of Severe Acute Respiratory Syndrome (SARS) in 2003, caused by a coronavirus identified as SARS-CoV (Severe Acute Respiratory Syndrome Coronavirus), surprised the world by spreading rapidly across continents. It resulted in over 8,000 infections with approximately 10% mortality and had a devastating effect on the economies of several countries, establishing itself as the first pandemic of the 21st century (4). In contrast, COVID-19 in 2020 accounted for about 100,000 deaths just one month after the announcement by this institution, resulting in 10 times more cases than SARS in a quarter of the time (5).

Coronaviruses (CoVs) can be categorized into three groups based on genetic and antigenic criteria: α-CoVs, β-CoVs, and γ-CoVs. CoVs primarily infect birds and mammals, with α-and β-CoVs capable of causing infections in humans. The infection predominantly affects the upper respiratory tract, resembling a common cold, but can also compromise the lower respiratory tract, leading to severe acute respiratory syndrome (SARS) (6).

The novel Coronavirus, order Nidovirales, family Coronaviridae, subfamily Orthocoronavirinae, was designated as severe acute respiratory syndrome-coronavirus-2 (SARS-CoV-2) by the International Committee on Taxonomy of Viruses (ICTV), and the World Health Organization (WHO) declared COVID-19 (coronavirus disease) as the name for this new disease in February 2020. SARS-CoV-2 is a β-coronavirus, which is a non-segmented, positive-sense RNA virus with an envelope. It has other representatives that have caused epidemics in the past, such as SARS-CoV and MERS-CoV (7,9).

In late December 2019, residents of Wuhan (Hubei province, China) began to be admitted to hospitals with symptoms of pneumonia and other complications involving the respiratory tract. The patients had a history of contact with a seafood market, either as traders or frequent visitors, which also traded various other live wild animals. Following etiological and epidemiological analyses, the new betacoronavirus capable of infecting humans was detected for the first time, named severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the agent responsible for COVID-19 (coronavirus disease 2019) (8,9).

Some CoVs were initially discovered as causes of enzootic infections limited to animals but progressed to establish zoonotic disease in humans. The genome sequence of SARS-CoV-2 was found to be 96.2% identical to a bat CoV RaTG13, while sharing 79.5% identity with SARS-CoV. Based on viral genome sequencing results and evolutionary analysis, it is suspected that bats are the natural host of origin for the virus, and SARS-CoV-2 may be transmitted from bats through unknown intermediate hosts to infect humans (6,9). Furthermore, it exhibits the exact molecular mechanism of infection observed in human COVID-19, involving the binding of angiotensin-converting enzyme 2 (ACE-2), which acts as a cellular receptor for the virus (6).

The disease was declared a pandemic on March 12, 2020, by the World Health Organization (WHO), and more than three years later, on May 5, 2023, the end of the Public Health Emergency of International Concern regarding COVID-19 was declared. By the end of June 2023, over 768 million confirmed cases and 6 million deaths due to COVID-19 had been recorded worldwide (10).

The global spread of SARS-CoV-2 impacted all sectors of human life, including medicine, economics, education, and research, among others, abruptly altering daily habits of social interaction. The clinical presentation of infected patients is generally characteristic of influenza-like illness (ILI), which may progress to severe acute respiratory syndrome (SARS) when major complications occur, such as respiratory failure or secondary infections (11,12).

One method of identifying viral infection is through laboratory diagnosis using RT-qPCR (Reverse Transcription Quantitative Real-Time Polymerase Chain Reaction). Other diagnostic methods exist, such as serological tests or non-laboratory methods like clinical-epidemiological examinations. Each presents advantages and disadvantages: serological testing, for example, is a rapid test that detects antibodies produced by the body to combat the virus. The antibody formation process takes approximately 14 days after the onset of infection, meaning the test is only effective when used in patients who are already in the recovery phase (WHO, 2020). Clinical diagnosis can be imprecise, as the initial clinical manifestations of COVID-19 resemble other viral respiratory infections and some endemic diseases (14).

The molecular nucleic acid amplification test is the most indicated for diagnosing infection in patients in the acute phase, according to the WHO (WHO, 2020). The RT-qPCR technique performs two processes: reverse transcription of viral

RNA to cDNA, followed by amplification of this genetic material through polymerase chain reaction (15). For analysis, it is necessary to collect a biological sample from the patient, typically involving the respiratory tract, where viruses show greater affinity with host cells. These can be nasopharyngeal or oropharyngeal swabs, sputum, saliva, among others (WHO, 2020). The most commonly used method is swabbing via nasopharyngeal swabs, an effective but invasive method. Studies such as Rodrigues da Silva et al. (2022) and Rao M et al. (2021) demonstrated that saliva is also a method with good sensitivity for viral detection, with the added benefit of being collected by patients themselves, mitigating personal discomfort and potential contamination of professionals involved in the collection (16,17).

Mass testing is an efficient method for monitoring viruses. The earlier infected individuals are detected, the greater the chances of controlling propagation, especially in cases where a portion of infections are asymptomatic (18). When there are no symptoms, the individual is not referred for testing and does not apply all transmission prophylaxis methods, resulting in greater virus dissemination. This increases the incidence of COVID-19 cases, a disease that presents high transmissibility (18). Therefore, surveillance and epidemiological surveys are of great importance for local epidemic control, contributing to reducing virus circulation in a given region.

Uncontrolled circulation is dangerous, as more excellent circulation increases the chances of virus mutations that can lead to the emergence of different variants and lineages, which may be related to decreased treatment and vaccination efficacy, increased transmission capacity, and disease severity, among other factors (11). Epidemiology, as a public health instrument, analyzes effective forms of combating an epidemic, locating contamination foci and describing the current epidemiological picture of communicable diseases.

Thus, one of the campuses of the Federal University of Viçosa located in the municipality of Rio Paranaíba, in the Alto Paranaíba region of the state of Minas Gerais (MG), with a territorial extension of 1,352,353 km² and an estimated population of 14,532 people (19), has been conducting disease diagnostic tests since May 2020 in a laboratory accredited by the RedeLab COVID-19. The Laboratory of Molecular Diagnostics (LDM) has performed over 25,000 examinations from residents of various municipalities in the Regional Health Superintendence of Patos de Minas (MG), in addition to promoting research related to the virus's activity in the region, being a strong ally in confronting the health crisis caused by SARS-CoV-2 (20). The city of Rio Paranaíba recorded 3,189 confirmed cases of COVID-19 from the beginning of the pandemic until the end of April 2023 (21).

The primary aim of this study is to monitor the epidemiological situation of COVID-19 in the municipality of Rio Paranaíba (MG) between May and June 2023. This research seeks to contribute to the ongoing surveillance efforts and provide valuable insights into the local dynamics of COVID-19 transmission and prevalence

2. Material and methods

The study was conducted at the Laboratory of Molecular Diagnostics (LMD) at the Federal University of Viçosa (UFV), Rio Paranaíba campus, from March to June 2023. All sample collection and processing steps were performed using recommended personal protective equipment (PPE) for each stage, including laboratory coats, gowns, gloves, caps, and surgical/PFF2/N95 masks. Equipment was frequently sterilized with 70% alcohol and UV light during and after procedures, and autoclaved filter tips were used to prevent contamination.

Study participants were volunteers who provided a biological sample along with a self-reported questionnaire containing variables for analysis: age, sex, presence of flu-like symptoms such as fever, cough, fatigue, shortness of breath, and rhinorrhea, among others, with the possibility of multiple or no symptoms; time since symptom onset; comorbidities and/or habits, including diabetes, hypertension, chronic lung diseases, obesity, and smoking, among others; and number of COVID-19 vaccine doses received.

This study was approved by the Human Research Ethics Committee of the Federal University of Viçosa (protocol number 5,991,500). After hand sanitization with antiseptic gel, participants were provided with a sterile rayon swab for self-collection of saliva following prior instruction and under supervision. The swab was inserted into a sterile 1.5mL Eppendorf tube containing 1mL of saline solution, identified, and stored in thermal boxes with ice. Samples were sent to the LMD for screening and processed in a class II biosafety cabinet. Screening consisted of sanitizing the collection container surface with 70% alcohol, vortexing for 30 seconds, and removing the swab, followed by storage at 4°C until RNA extraction.

RNA was extracted using the Loccus Extracta 32 automated extractor with the Extracta Kit - Viral RNA and DNA (MVXA-P016 FAST) from Loccus, following the manufacturer's RNA extraction protocol. Extracted nucleic acid was stored at -20°C for subsequent analysis, and the remaining sample was stored in an ultra-freezer at -80°C to maintain viability in

case process repetition was necessary. Subsequently, amplification and detection of the viral gene (E gene) were performed using RT-qPCR with the SARS-CoV-2 MOLECULAR KIT (EDx) from Bio-Manguinhos in a CFX 96 Real-Time PCR thermocycler (Bio-Rad), following the manufacturer's instructions. Excess viral RNA was also stored in the ultra-freezer at -80°C. Using the data provided in the questionnaire and the test results, absolute and relative frequency tables were created using RStudio and Microsoft Office Excel 2013 to conduct the epidemiological survey of COVID-19 in the municipality of Rio Paranaíba during the evaluated period.

3. Results and Discussion

On average, 40 saliva samples were collected and examined weekly over 8 weeks between May and June 2023. In total, 162 samples were analyzed from the urban area of Rio Paranaíba municipality and 162 samples from the UFV campus, amounting to 324 samples. Of these, 321 tested negative for COVID-19, while the remaining 3 samples yielded positive results. The mean age of participants was 26.5 years, with a minimum of 9 years and a maximum of 69 years. Table 1 presents the distribution of participants according to age, sex, biological sample collection site, number of COVID-19 vaccine doses received, and test results. The majority of study participants were aged 20-29 years (56.8%), female (55.9%), and had received 3 doses of the COVID-19 vaccine (54.6%).

Table 1 Distribution of participants according to variables: age, sex, collection site, vaccination status, and test results. Values are represented in absolute frequency (N) and relative frequency (%)

Variables	n	%
Age		
0-9	1	0.30%
10-19	60	18.50%
20-29	184	56.80%
30-39	39	12.00%
40-49	24	7.40%
50-59	10	3.10%
60-69	6	1.90%
Sex		
Males	143	44.14%
Females	181	55.86%
Location		
UFV	162	50%
City	162	50%
Vaccin shots (COVID-19)		
None	2	0.62%
1 shot	2	0.62%
2 shots	84	25.93%
3 shots	177	54.63%
4 or more shots	59	18.20%
Results (COVID-19)		
negative	321	99.07%
positive	3	0.93%

Certain factors are associated with an increased risk of disease in individuals, including advanced age, sex, and comorbidities such as diabetes and heart disease (22). In Brazil, the profile of COVID-19 fatalities predominantly featured males (55.5%) in the 60-69 age group (23.4%) (23). Studies indicate that men are more susceptible to COVID-19, in addition to exhibiting less attention to individual health and social distancing, resulting in a higher mortality rate

(22,24). Comparing vaccination data and sex of the project participants, 75.69% (n=137) of women had received 3 or more vaccine doses, while this frequency decreased to 69.26% (n=99) for men. Among positive participants, the majority (n=2) were men with 2 vaccine doses, aged 20-29 years, with samples collected at the UFV campus. Beyond these factors, economic, social, and geographical characteristics also impacted infection and mortality rates in the country, particularly affecting populations with socioeconomic vulnerability (25).

During the completion of the questionnaire, participants declared whether they had flu-like symptoms or not. In total, 225 (69.44%) subjects reported being asymptomatic at the time of biological sample collection, while the remaining 99 (30.56%) reported one or more flu-like symptoms. Among the participants, the most prevalent symptoms were rhinorrhea, cough, sore throat, and headache. All participants who tested positive for COVID-19 were symptomatic (Table 2).

In addition to symptoms, participants also declared whether they had any comorbidities or habits associated with worsening clinical outcomes in COVID-19-positive patients. A total of 201 (62.04%) subjects reported having no comorbidities, while 123 (37.96%) reported having one or more comorbidities. The most prevalent comorbidities and habits in the study were chronic lung diseases (including asthma), smoking, and obesity (Table 3).

Table 2 Symptoms presented by participants, comparing positive and negative results for COVID-19, represented in absolute frequency and relative frequency (%)

	Negative (n=321)	**Positive (n=3)**
Symptoms	96 (29.91%)	3 (100%)
Fever	6 (1.87%)	2 (66.67%)
Cough	44 (13.71%)	3 (100%)
Fatigue	7 (2.18%)	2 (66.67%)
Shortness of breath	1 (0.31%)	1 (33.33%)
Sore throat	29 (9.03%)	2 (66.67%)
Muscle pain	12 (3.74%)	2 (66.67%)
Headache	24 (7.48%)	2 (66.67%)
Rhinorrhea	63 (19.63%)	1 (33.33%)
Vomiting	2 (0.62%)	0
Diarrhea	3 (0.93%)	0

Table 3 Comorbidities presented by participants, comparing positive and negative results for COVID-19, represented in absolute frequency and relative frequency (%)

	Negative (n=321)	**Positive (n=3)**
Comorbidities	122 (38.01%)	1 (33.33%)
Diabetes mellitus	6 (1.87%)	0
Cardiovascular diseases	2 (0.62%)	0
Hepatic disease	1 (0.31%)	0
Chronic pulmonary diseases	46 (14.33%)	1 (33.33%)
Obesity	13 (4.05%)	0
Arterial hypertension	9 (2.8%)	0
Tabacco uses disorder	33 (10.28%)	0
Others	12 (3.74%)	0

The limited number of positive cases precluded a more in-depth discussion regarding the association between COVID-19 and symptomatology and comorbidities in the municipality. The literature indicates that the most common symptoms among infected individuals are fever, cough, and fatigue (26). The three participants who tested positive in this study presented cough as a common symptom. Symptomatic participants who tested negative primarily reported rhinorrhea and cough, symptoms that may be related to other viral infections or climatic conditions during the study period (27). Regarding comorbidities, those that indicate the highest risk for COVID-19 are cardiovascular diseases, respiratory diseases, and diabetes, in addition to smoking habits (28). Only one positive participant presented a comorbidity (chronic lung disease), and during their infection, eight symptoms were reported, including shortness of breath.

According to the Rio Paranaíba City Hall, the last recorded case of COVID-19 in the municipality before the commencement of the present study was dated April 24, 2023, when the official epidemiological bulletin was released (21). From the beginning of the project, weekly reports were sent to the city hall to update the test results, and when positive results were obtained, they were disclosed in the subsequent bulletin. Since the onset of the COVID-19 pandemic, the Federal University of Viçosa (Rio Paranaíba campus) has been assisting in the diagnosis of the disease in various municipalities in the region through the Laboratory of Molecular Diagnostics. At a time when combating false information and discussing the importance of scientific dissemination was essential, Brazilian universities proved to be an important tool in public health, acting seriously to combat the advancement of SARS-CoV-2 infections (29,30).

The COVID-19 pandemic afflicted Brazil and the world for more than three years. Surveillance studies and epidemiological surveys contribute to a greater understanding of the disease, especially considering that the virus was previously unknown. Addressing mass testing of the population becomes crucial in cases of infections, such as those caused by SARS-CoV-2, which presents a proportion of asymptomatic cases and the potential for mutation into other variants.

Test results were reported weekly to the municipal authorities, and when a positive case was identified, it was disclosed to the population through the epidemiological bulletin, contributing to the transparency of case reporting for disease control.

4. Conclusion

In the municipality of Rio Paranaíba, throughout May and June 2023, the disease situation was under control, with three positive cases detected by this study. All three cases were symptomatic, and the positive participant who presented the most symptoms reported a comorbidity of chronic lung disease. This individual's infection was the most symptomatic among the positive cases. The majority of positive cases were men with two doses of the COVID-19 vaccine in the 20-29 age group.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

References

[1]   Morse SS. Factors in the emergence of infectious diseases. Emerg Infect Dis. 1995;1(1):7–15.

[2]   Akin L, Gözel MG. Understanding dynamics of pandemics. Turk J Med Sci. 21 de abril de 2020;50(SI-1):515–9.

[3]   Martens P. How Will Climate Change Affect Human Health? Am Sci. 1999;87(6):534.

[4]   LeDuc JW, Barry MA. SARS, the First Pandemic of the 21st Century1. Emerg Infect Dis. November 2004;10(11):e26.

[5]   Khalil OAK, Khalil S da S. SARS-CoV-2: taxonomy, origin and constitution. Rev Med. 2020 Dec 10;99(5):473–9.

[6]   Guo YR, Cao QD, Hong ZS, Tan YY, Chen SD, Jin HJ, et al. The origin, transmission and clinical therapies on coronavirus disease 2019 (COVID-19) outbreak - an update on the status. Mil Med Res. 13 de março de 2020;7(1):11.

[7]   Kim D, Lee JY, Yang JS, Kim JW, Kim VN, Chang H. The Architecture of SARS-CoV-2 Transcriptome. Cell. 14 de maio de 2020;181(4):914-921.e10.

[8]   Wu F, Zhao S, Yu B, Chen YM, Wang W, Song ZG, et al. A new coronavirus associated with human respiratory disease in China. Nature. 1º de março de 2020;579(7798):265–9.

[9]   Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, et al. A Novel Coronavirus from Patients with Pneumonia in China, 2019. N Engl J Med. 20 de fevereiro de 2020;382(8):727–33.

[10]   WHO WHO. Coronavirus Disease (COVID-19) Situation Reports [Internet]. 2023 [citado 20 de novembro de 2024]. Disponível em: https://www.who.int/emergencies/diseases/novel-coronavirus-2019/situation-reports

[11]   CDC. Influenza (Flu). 2024 [citado 20 de novembro de 2024]. Similarities and Differences between Flu and COVID-19. Disponível em: https://www.cdc.gov/flu/about/flu-vs-covid19.html

[12]   Solomon DA, Sherman AC, Kanjilal S. Influenza in the COVID-19 Era. JAMA. 2020 Oct 6;324(13):1342–3.

[13]   World Health Organization (WHO). (2020). Diagnostic tests for SARS-CoV-2. Available at: apps.who.int/iris/bitstream/handle/10665/334254/WHO-2019-nCoVlaboratory-2020.6-por.pdf. Accessed in: May 2023

[14]   State Department of Health (MG). COVID-19 diagnostic manual - Version 5. 2021.

[15]   Menezes ME, Lima LM, Martinello F. Diagnóstico laboratorial do SARS-CoV-2 por transcrição reversa seguida de reação em cadeia da polimerase em tempo real (RT-PCR). Rev Bras Análises Clínicas [Internet]. 2020 [citado 20 de novembro de 2024];52(2). Disponível em: http://www.rbac.org.br/artigos/diagnostico-laboratorial-do-sars-cov-2-por-transcricao-reversa-seguida-de-reacao-em-cadeia-da-polimerase-em-tempo-real-rt-pcr/

[16]   Rao M, Rashid FA, Sabri FSAH, Jamil NN, Zain R, Hashim R, et al. Comparing Nasopharyngeal Swab and Early Morning Saliva for the Identification of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2). Clin Infect Dis Off Publ Infect Dis Soc Am. 4 de maio de 2021;72(9):e352–6.

[17]   Silva NVR da, Pereira SSN, Kavalco KF, Menegídio FB, Medeiros-Santana L, Visôtto LE, et al. Molecular diagnosis of SARS-CoV-2: a validation of saliva samples [Internet]. medRxiv; 2022 [citado 20 de novembro de 2024]. Disponível em: https://www.medrxiv.org/content/10.1101/2022.01.20.22269618v1

[18]   Corrêa PRL, Ishitani LH, Abreu DMX de, Teixeira RA, Marinho F, França EB. The importance of monitoring cases and deaths and the COVID-19 epidemic in Belo Horizonte, 2020. Rev Bras Epidemiol. 2020 Aug 5;23:e200061.

[19]   IBGE. Paranaíba River (MG) | Cities and States | IBGE [Internet]. 2022 [cited November 21, 2024]. Available at: https://www.ibge.gov.br/cidades-e-estados/mg/rio-paranaiba.html

[20]   Visôtto L, Medeiros Santana L, Kavalco K, Pasa R, Good God PI. Extension action in the face of the SARS-CoV-2 pandemic: The role of the UFV-CRP Molecular Diagnostics Laboratory and university leadership. Rev Guará. April 11, 2023;

[21]   Municipal Health Department of Rio Paranaíba. COVID-19 Bulletin [Internet]. 2023 [cited November 21, 2024]. Available at: https://rioparanaiba.mg.gov.br/wp-content/uploads/2023/03/WhatsApp-Image-2023-03-31-at-17.36.28.jpeg

[22]   Rashedi J, Mahdavi Poor B, Asgharzadeh V, Pourostadi M, Samadi Kafil H, Vegari A, et al. Risk Factors for COVID-19. Infez Med. 2020 Dec 1;28(4):469–74.

[23]   Ministry of Health. Special epidemiological bulletin: new coronavirus disease – COVID-19. 2021.

[24]   Global Health. O rastreador de dados desagregados por sexo da COVID-19 | Saúde global 50/50 [Internet]. 2022 [citado 21 de novembro de 2024]. Disponível em: https://globalhealth5050.org/the-sex-gender-and-covid-19-project/the-data-tracker/

[25]   Castro RR, Santos RSC, Sousa GJB, Pinheiro YT, Martins RRIM, Pereira MLD, et al. Spatial dynamics of the COVID-19 pandemic in Brazil. Epidemiol Infect. 25 de fevereiro de 2021;149:e60.

[26]   Grant MC, Geoghegan L, Arbyn M, Mohammed Z, McGuinness L, Clarke EL, et al. The prevalence of symptoms in 24,410 adults infected by the novel coronavirus (SARS-CoV-2; COVID-19): A systematic review and meta-analysis of 148 studies from 9 countries. PloS One. 2020;15(6):e0234765.

[27]   Silva SEL da, Mendonça CROSG, Oliveira AA de, Gabriel RVC, Silva AC, Souza LJN de. Epidemiology and natural history of Coronavirus-associated Severe Acute Respiratory Syndrome. Braz J Dev. 2022 Jan 17;8(1):4351–8.

[28]   Feitoza TMO, Chaves AM, Muniz GTS, Cruz MCC da, Junior I de FC. COMORBIDITIES AND COVID-19. Rev Human and Technological Health Interfaces. 2020 Oct 31;8(3):711–23.

[29]   Nepomuceno GA da M, Ayub GGP, Santiago AF, Dall'Agnol MM, Lima R da C. UNIVERSITY INTEGRATION AND EPIDEMIOLOGICAL SURVEILLANCE IN ADDRESSING COVID-19. Rev FOCO. 2023 Apr 12;16(4):e1557–e1557.

[30]   Silva WNT da, Rosa MFP, Oliveira SV de. Production of epidemiological bulletins as a Health Surveillance strategy in the context of the COVID-19 pandemic. Health Surveillance Under Debate. 2020;8(3):171–7.