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Full Length Research Paper

Epidemiology, prevalence and seroprevalence of COVID-19 among vulnerable people in Thies region, Senegal

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Cases of pneumonia linked to a new Coronavirus have been noted in Wuhan city, Hubei province in China. This epidemic caused by the new Coronavirus called COVID-19 by the WHO, quickly became a global pandemic. This study is to assess the epidemiological situation as well as the prevalence and seroprevalence of COVID-19 in 213 vulnerable patients in the Thies region. This is a prospective study, targeting vulnerable people recruited in the Thies region from June to September 2022. For each patient, a serological test and a search for the SARS CoV-2 genome was carried out using a blood sample and a nasopharyngeal swab. 213 participants, vulnerable to COVID-19, living in the Thies were include. The age of the patients varied between 12 and 97 years and the age (61-80) were the most represented group. The majority were diabetic (26.3%), followed by hypertensives (23%). The elderly represented 14.6%, and the patients with cardiopathy 5.2%. Pregnant women represented 5.2% of our study and 12% of patients presented other comorbidities. Furthermore, 34% of our study population were vaccinated. RT-PCR to detect SARS-CoV-2 RNA was positive for 11 patients, representing a prevalence of 5.2%. Serological tests showed that 15.49% carried IgM anti-SARS-CoV-2 antibodies, probably indicating an ongoing infection. And 76.52% had anti-SARS-CoV-2 antibodies of the IgG type. However, 11.73% tested positive for both classes of antibodies. The circulation of the virus is still present in Thies region, particularly among vulnerable people. The high IgG seroprevalence could indicate immunity in the population studied.

Key words: SARS-CoV-2, comorbidities, genomic marker, seroprevalence, Thies.

INTRODUCTION

The new Coronavirus 2019 (COVID-19) disease, caused by severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2), appeared in the city of Wuhan, capital of Hubei province in China in December 2019 (Mohan and Nambiar, 2020; Zhu et al., 2020; World Health Organization, 2020). It spread very rapidly to many countries around the world and was declared a pandemic by the WHO on March 11, 2020 (Sohrabi et al., 2020; Hamid et al., 2020). This new viral strain is characterized by rapid human-to-human transmission, with a basic

reproduction rate estimated between 2 and 4 (Liu et al., 2020). Worldwide, as of 2 November 2023, 771,679,618 confirmed cases of COVID-19, including 6,977,023 deaths, have been reported to WHO (https://COVID-19.who.int). Senegal, which confirmed its first case of COVID-19 on March 2, 2020 (Diouf et al., 2020), had 89,022 confirmed cases of COVID-19 with 1,971 deaths by the same date (https://COVID-19.who.int).

Clinically, the disease can range from asymptomatic, with mild symptoms, to severe pneumonia, which can progress to the major complication of acute respiratory distress syndrome (ARDS), a potentially fatal outcome (Guan et al., 2020). According to the WHO, 80% of people infected with SARS-CoV-2 present mild, uncomplicated forms of the disease. However, 20% of those infected are vulnerable to COVID-19 and will develop severe or even critical forms that may require hospitalization or intensive care (Surveillances, 2020). This identified category includes, but is not limited to, elderly subjects (Wu and McGoogan, 2020; Du et al., 2020), people with co-morbidities such as diabetes (Wu and McGoogan, 2020; Wang et al., 2020), hypertension (Wu and McGoogan, 2020; Wang and Zhang 2020), cardiovascular disease (Wu and McGoogan, 2020; Wang and Zhang 2020), cancer (Wu and McGoogan, 2020; Liang et al., 2020), chronic respiratory disease (Wu and McGoogan, 2020), cerebro-vascular disease (Du et al., 2020; Wang and Zhang 2020), obesity, chronic kidney disease, smoking, asthma (Grasselli et al., 2020). Although current data show no evidence of vertical transmission of SARS-CoV-2, many pregnant women are infected worldwide.

In Senegal, SARS-CoV-2 was detected in suspected cases and symptomatic contacts by RT-PCR from nasopharyngeal swabs. Asymptomatic patients were not systematically diagnosed. Serological testing, especially on a large scale in people vulnerable to COVID-19, had not yet been carried out. It was against this background that we conducted this study to assess the prevalence of COVID-19 in this vulnerable segment of the population.

The general objective of this study was to investigate virological and immunological markers of SARS-CoV-2 (genome and specific antibodies) in people vulnerable to COVID-19 in the Thies region over a period of four months.

METHODOLOGY

Type and setting of study

A descriptive and cross-sectional study was conducted over four months, which was carried out during the year 2021. Participants were enrolled in the internal medicine, gynaecology and laboratory departments at the Regional Hospital Center, Thies, Senegal. Samples were processed, stored and serologically tested at the National Public Health Laboratory. RT-PCR was performed at the Regional Laboratory of Saint-Louis, Senegal.

Study population

A total of 213 people vulnerable to COVID-19, living in Thies, were enrolled in this study. The inclusion criteria were people aged over 60 years, and/or presenting a vulnerability factor (heart disease, chronic kidney disease, sickle cell anaemia, etc.) or pregnant women.

We excluded patients who did not meet the criteria for vulnerability to COVID-19 and those who had not consented to participate in this study. Participants were seen for a consultation or hospitalized in the medical ward, emergency department or medical biology laboratory for analyses. Beforehand, however, all participants signed a consent form after a clear and detailed explanation of the study.

A data collection form was completed for each participant, to collect various information such as socio-demographic data, underlying pathologies, history of Covid-19 episodes, anti-Covid-19 vaccination.

Following a favourable opinion from the National Health Research Ethics Committee, the study was authorised by the Senegalese Ministry of Health and Social Action under number 0000075/MSAS/CNERS/SP on 30 March, 2022.

Specimen collection, processing and storage

We collected whole blood in EDTA tube and nasopharyngeal secretions from each participant using a swab in a tube containing 3 ml of virus transport medium. The samples were then transferred to the National Public Health Laboratory where the blood samples were centrifuged, the plasma collected, aliquoted and stored at -20°C.Nasopharyngeal samples were aliquoted under type 2 SHP and stored at -80°C. Each aliquot was assigned an anonymous identification code to ensure confidentiality.

PCR test

Extraction of viral RNA

The viral genome was extracted with the Zymo-research kit, using silica columns, according to the manufacturer's recommendations. Briefly, 200 µl of Viral RNA Buffer were added to 100 µl of nasopharyngeal sample in a sterile tube, then the mixture was transferred to a column with a collection tube and centrifuged at 10,000 rpm for 2 min. The supernatant was then discarded. Then 500 µl of ethanol were added to the column and centrifuged at 14,000 rpm for 1 min to precipitate the viral genome. After two washing steps with Viral Wash Buffer to remove cellular debris, the genome bound to the column was eluted in 15 µl of DNase/RNase-free water. The eluted RNA was stored at -80°C until use. For quality control, 100 µl of RNA/DNase free water was extracted in

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Table 1. Breakdown of the susceptible general population by age group in Thies.

Age Groupe (year)	Groupe size	Percentage (%)
0-20	4	1.9
21-40	58	27.4
41-60	60	28.3
61-80	71	33.5
≥81	19	9
Total	212	100

the same way simultaneously as the samples.

Amplification and viral genome detection

Detection of the viral genome was carried out by ANDIS FAST SARS-CoV-2 RT-qPCR® multiplex real-time RT-PCR kit, at the Regional Laboratory, Saint-Louis, Senegal. This test simultaneously targets 3 genes, 2 of which are specific to SARS-CoV-2. It detects the ORF1ab and N genes (specific to SARS-CoV-2), and the E gene for all sarbecoviruses, including SARS-CoV-2. RNase P is used as an internal control for sample collection, nucleic acid extraction and amplification. The viral RNA is first reverse transcribed into cDNA, then amplified and detected in real time by PCR machines. The ORF1ab, N and E target genes and the internal control were revealed in the FAM, ROX, VIC and Cy5 channels respectively. Handling was carried out in appropriate, certified biosafety cabinets by trained personnel. The following thermal profile was used; 50°C for 2 min, 95°C for 2 sec, 95°C for 1 sec (41 cycles), and 60°C for 13 sec (41 cycles). Signal acquisition was carried out at the last stage.

For amplification the master mix consisted of 8.5 µl of RT-PCR Reaction Mix and 1.5 µl enzyme mix. Next, 10 µl of master mix was dispensed into each well of the plate. Next, 10 µl of sample or positive control or RNase-free water (for the negative control) was added to the corresponding wells of the PCR plate. The plates were then centrifuged at 3000 RPM for 30 sec and analysed using a real-time RT-PCR machine (Mbow et al., 2022). Results were considered positive if viral RNA was detected at threshold cycle (Ct) values ≤ 35 and negative at Ct values >38. Positive, negative and internal controls were used for quality control.

Serological tests

For the detection of antibodies, we used the STANDARD Q COVID-19 IgM/IgG Plus ® serological kits.

This test is based on the immunochromatographic method and allows qualitative detection of antibodies specific to SARS-CoV-2 (anti-nucleocapsid) (N) protein antibodies and anti-spike antibodies present in human serum, plasma or whole blood. This test is intended for professional in vitro diagnostic use of SARS-CoV-2 infection in the convalescent phase in a patient with clinical symptoms of SARS-CoV-2 infection. It only provides an initial screening test result. More specific alternative diagnostic methods must be performed in order to obtain confirmation of SARS-CoV-2 infection.

Data collection and processing

A questionnaire created from Google document was used for data collection. The patient's sociodemographic, clinical characteristics, and vaccination status for COVID-19 were collected. The data were then corrected using Excel software. Statistical tests were performed using SPSS 20.0 software.

RESULTS

Characteristics of the study population

A total of 212 participants, vulnerable to COVID-19, living in the Thies region were enrolled in this study. The mean age was 55.06 years and the gender ratio (M/F) 0.67. Patients ranged in age from 12 to 97 years, with the age group [61-80] being the most represented. The distribution of the study population according to age and gender is shown in Table 1 and Figure 1.

In Table 2 majority of participants were diabetics (26.4%), followed by hypertensives (23,1%). The elderly accounted for 14.6%, heart disease for 5.2%, asthma and sickle cell disease for 3.8% and 4.2% respectively. Kidney failure accounted for 5.7 % of the study population. Pregnant women accounted for 5.2% of our study population, and 11,8% of patients had other comorbidities.

In terms of COVID-19 vaccination, 34% of the study population had been vaccinated, with a predominance of elderly subjects, hypertensives and diabetics. Figure 2 illustrates the vaccination status of the study population.

Prevalence of COVD-19

RT-PCR enabled us to detect SARS-CoV-2 RNA in 11 patients, giving an overall prevalence of 5.2% in Table 3.

Among the vulnerable groups studied, we noted a prevalence of COVID-19 at 18.18% and 11.11% in cardiopathy and sickle cell disease respectively. Among hypertensives and diabetics, the prevalence was 8.16% and 5.35% respectively. The prevalence of other comorbidities was 3.84%. Figure 3 illustrates the prevalence of COVD-19 according to the patient's condition.

Seroprevalence of anti-SARS-CoV-2 antibodies

Serological analyses (immunochromatographic method)

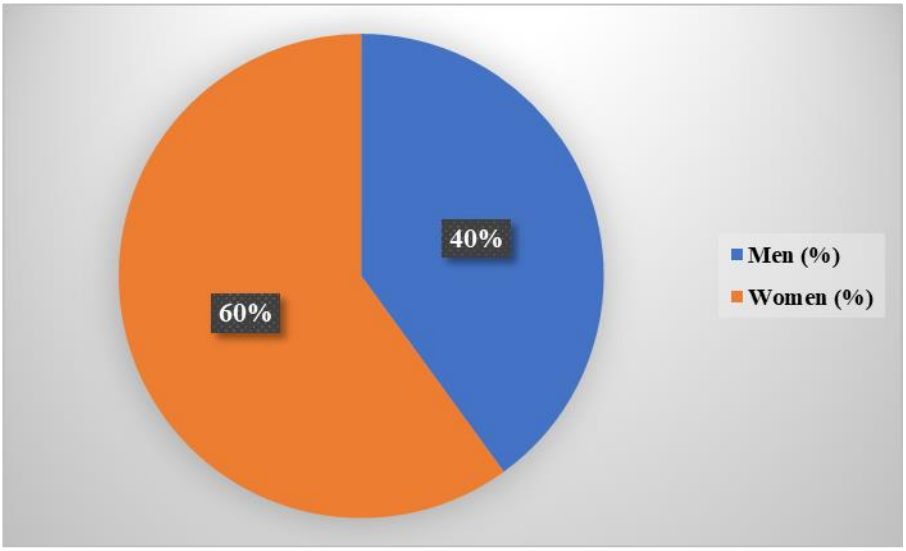

Figure 1. Breakdown of study population in the susceptible general population by gender in Thies.

Table 2. Breakdown of the susceptible general population by type of vulnerability in Thies.

Vulnerability	N (%)
Diabetes	56 (26.4)
Hypertension	49 (23.1)
Elderly	31 (14.6)
Renal failure	12 (5.7)
Pregnancy	11 (5.2)
Heart disease	11 (5.2)
Sickle cell disease	9 (4.2)
Asthma	8 (3.8)
Other	25 (11.8)
Total	212 (100)

showed that 15.6% (33 patients) carried IgM-type anti-SARS-CoV-2 antibodies, probably indicating ongoing infection. A further 76.9% (163 participants) had IgG anti-SARS-CoV-2 antibodies. However, 11.79% (25 patients) tested positive for both serological tests at the same time (Table 4).

IgG seroprevalence was significantly higher than IgM seroprevalence in our study population. Among diabetics, 73.21% carried IgG anti-SARS-CoV-2 antibodies and 10.71% IgM anti-SARS-CoV-2 antibodies. For the elderly, 77.42% had IgG antibodies to SARS-CoV-2 and 22.58% had IgM antibodies to SARS-CoV-2.

Among hypertensive patients, 83.67% had IgG antibodies to SARS-CoV-2 and 16.33% had IgM antibodies to SARS-CoV-2. In heart disease patients, 7.73% carried IgG anti-SARS-CoV-2 antibodies and 36.36% had IgM anti-SARS-CoV-2 antibodies. Among people with renal failure, 91.67% carried IgG antibody to SARS-CoV-2 and no IgM antibody to SARS-CoV-2.

For people with sickle cell disease, 88.89% carried IgG antibody to SARS-CoV-2 and among pregnant women, 90.91% were carriers of IgG anti-SARS-CoV-2 antibodies and only 9% had IgM anti-SARS-CoV-2 antibodies.

And finally, among people with other comorbidities, 64% carried IgG anti-SARS-CoV-2 antibodies and 20% IgM anti-SARS-CoV-2 antibodies.

DISCUSSION

In this prospective study of 212 patients recruited at the Thies regional hospital, our general objective was to

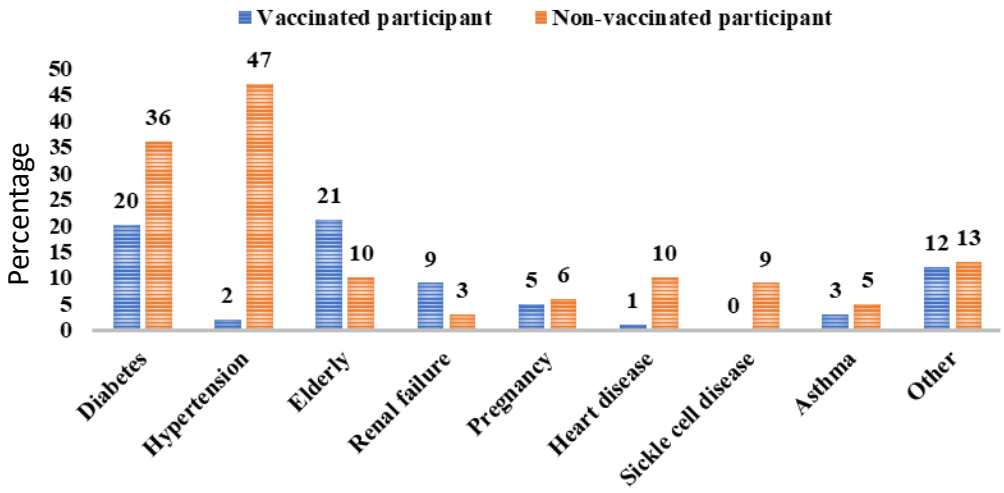

Figure 2. Vaccination status of the susceptible general population by type of vulnerability in Thies.

Table 3. Covid19 RT-PCR results in the susceptible general population in Thies.

Variable		Number (%)	95% Confidence interval
PCR	Indetermined	1 (0.5)	0.0-1.4
	Negative	200 (94.3)	91.1-97.2
	Positive	11 (5.2)	2.8-8.5
	Total	212	100.0

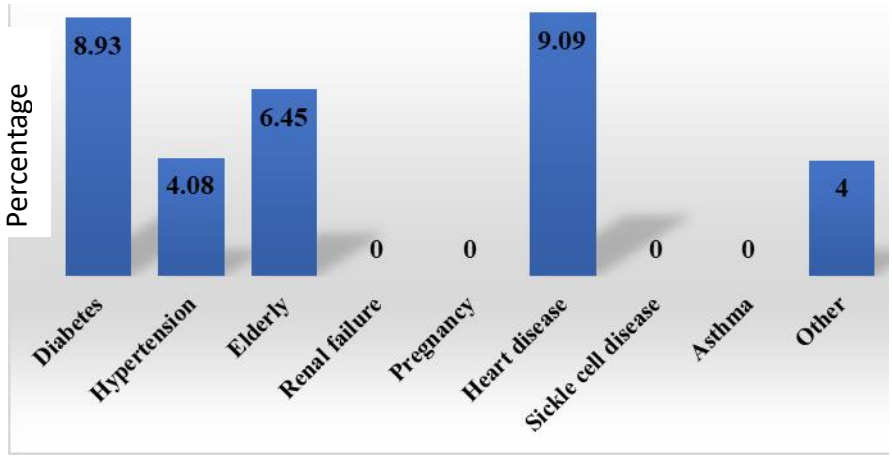

Figure 3. Prevalence of COVD-19 according to vulnerability in the susceptible general population in Thies.

search for markers of SARS-CoV-2 (genome and specific antibodies) in vulnerable individuals over a 4-month period. We were also able to report the epidemiological profile, prevalence and seroprevalence of COVID-19 in the population studied.

Epidemiological data

Age

Age is a major risk factor. In fact, 80% of deaths involve

Table 4. Covid19 serology results in the susceptible general population in Thies.

Variable	IgM (standard Q)	IgG (standard Q)
	N (%)	N (%)
Negative	176 (83)	46 (21.7)
Sample not received	3 (1.4)	3 (1.4)
Positive	33 (15.6)	163 (76.9)
IgM and IgG positives	25 (11.73)	25 (11.73)
Total	212 (100)	212 (100)

people aged over 60 (Phua et al., 2020). The risk of death from COVID-19 rises sharply after the age of 60 and even more so after the age of 64, but younger people can also present severe forms of the disease. Admissions to intensive care for patients with COVID-19 increase from the age of 50, and the most recent studies have confirmed the major role of age (Williamson et al., 2020), independently of any other pathology. The increasing risk of severe forms of the disease increases with age from the 45-64 age group onwards, while deaths are more frequent over the age of 65. All patients in our study were aged over 12 years, with an average age of almost 58 years. The 61-80 age group was the most represented. This result is comparable to that found by Riccardo et al., 2020 in Italy, with an average age of 62 years, but lower than that of Guede et al., 2021 in Côte d'Ivoire, who found an average age of 40 years. In our study, children were not greatly affected by SARS-CoV-2 infection during this period.

Gender

Females accounted for the majority of cases, with 60% and a gender ratio of 0.67. In contrast to a study on the prevalence of co-morbidities and their effects on SARS-CoV-2 (Yang et al., 2020), where the male gender ratio was significantly higher. The study by Lara et al. showed similar results, with women predominating at 61% (Coelho et al., 2022).

Vaccination status

Vaccines against COVID-19 protect vulnerable individuals against severe forms of the disease. Vaccination against COVID-19 began in March 2021 with Sinopharm and then AstraZeneca vaccines, and recently Johnson and Johnson. The target groups are medical staff, the elderly and people at risk (co-morbidities). As of 7 August, 2021, the number of people vaccinated in Senegal was estimated at 1,017,529. In terms of vaccination, 34% of the population studied were vaccinated, with a predominance of elderly subjects, hypertensives and diabetics. Our study showed that the majority of the vulnerable population did not vaccinate, contrary to the objectives set.

Vulnerabilities

The comorbidities found in our patients were mainly arterial hypertension and diabetes. In contrast, the comorbidities found in western countries were more serious, essentially cancers, autoimmune diseases, morbid obesity and a history of organ transplants (Kayem et al., 2020). We also noted that the majority of our study population were elderly, which mean that some patients had two vulnerability factors. The comorbidities found in our study population were comparable to the results of a study on SARS-CoV-2 and the prevalence of comorbidities was represented by hypertension, diabetes and cardiovascular disease (Yang et al., 2020). But we also had heart disease, sickle cell disease, renal failure, pregnancy, asthma and other forms of comorbidity which did not have a very high figure.

Prevalence of COVID-19

RT-PCR on nasopharyngeal and/or oropharyngeal swabs is the reference test for diagnosing SARS-CoV-2 infection with very good sensitivity (around 95%) and specificity of 99.9%. In our study, RT-PCR detected viral RNA in 11 patients, representing a prevalence of 5.2 %. In addition, the study by Harboun et al., 2022, carried out in France among residents of 27 nursing homes, after the first wave of COVID-19, found a prevalence of 12.9%. This increase may be linked to differences in the study periods. In Senegal, the number of cases fell considerably during the fifth wave.

In this study, 11 patients tested positive for COVID-19, of whom 9.09 % were cardiopathic, 4.08% hypertensive, 8.93% diabetic, 6.45% elderly and 4% had other comorbidities. The prevalence of heart disease was slightly higher in our study. In a meta-analysis of 32 articles, the comorbidities most frequently observed during SARS-CoV-2 infection were hypertension, diabetes, cardiovascular disease, liver disease, lung disease, malignant tumours, cerebrovascular disease, Chronic obstructive pulmonary disease (COPD), and asthma. Of all these underlying conditions reported, arterial

hypertension had the highest prevalence, estimated at 46% (CI 37%-55%) and asthma had the lowest prevalence of 3% (CI 2-6%) (Javanmardi et al., 2020).

Seroprevalence of anti-SARS-CoV-2 antibodies

Serological tests can detect antibodies produced by the patient in response to the viral infection and are an important tool for assessing post-infection or post-vaccination immunity. In our study, we found a prevalence of 76.9% (163 participants) for IgG class anti-SARS-CoV-2 antibodies. After the first wave of the COVID-19 pandemic, Beaumont et al., (2021) carried out a seroprevalence study in a vulnerable population in France, and found 35.4% seroprevalence. Furthermore, a two-month seroprevalence study carried out in Senegal by Talla et al, at the end of the first wave in the general population, obtained a much lower seroprevalence of 28.4% (24% in Thies region) (Talla et al 2022). Another seroprevalence study by Ahouidi et al., 2022, conducted over four months during the same period, found a slightly lower prevalence of 20.4% (18.3% in Thies region).

Our study showed very high transmission of SARS-CoV-2 among the vulnerable population during the fifth wave of the COVID-19 pandemic in Senegal. This result could be explained by the persistence of antibodies (anti-SARS-CoV-2 IgG) following probable contamination during the first four waves. The second hypothesis that could explain this high prevalence is an increase in prevalence, particularly among vulnerable people.

Conclusion

During the Covid-19 pandemic, elderly and people with comorbidities have paid a heavy price with high mortality rates. This study focused on this vulnerable people to study their genomic and serological profile to Covid-19. Immunity is found in the majority of participants but also circulation of the virus by positive PCR tests and presence of IgM type antibodies.

Today, this disease is considered as an endemic infection like seasonal flu. But surveillance still remains important, particularly surveillance of SARS-CoV-2 variants, but also vigilance when this infection occurs in people with comorbidities.

In countries with limited resources such as Senegal, serological tests are more appropriate for assessing the real exposure of the population and contributing to the development of public policy to fight against this infection.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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