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Post-Hajj 2024: Molecular Detection and Variant Profiling of SARS-CoV-2 in Ivorian Travelers

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ABSTRACT

The COVID-19 pandemic has been a major threat to global public health, particularly in mass gatherings such as the Hajj. This paper explores the detection and molecular characterization of SARS-CoV-2 infections in Ivorian pilgrims who participated in the Hajj in 2024.

Methods: *RT-PCR for the detection of SARS-CoV-2, followed by sequencing of the Spike protein gene was performed on nasopharyngeal swabs from Ivorian pilgrims returning from the Hajj 2024. Descriptive and phylogenetic analyses were performed to identify socio-demographic factors and SARS-CoV-2 lineages present in these pilgrims.*

Results: *A significant percentage of pilgrims tested positive for SARS-CoV-2. subjects aged over 65 were affected, with a predominance of women. Phylogenetic analysis highlighted the link between viruses detected in pilgrims returning from Mecca and strains circulating in Saudi Arabia, confirming the source of contamination.*

Conclusion: *The study highlights the need for rigorous monitoring of the elderly and appropriate vaccination campaigns for pilgrims. The data collected provide a valuable basis for anticipating future epidemic outbreaks and reinforcing health safety at religious gatherings.*

Keywords
COVID-19, Hajj, SARS-CoV-2, Molecular characterization.

Introduction
A trip to Mecca for the Hajj (pilgrimage) is no ordinary undertaking for many Muslims. The Hajj represents the culmination of years of spiritual preparation and planning [1]. Once the pilgrimage is completed, pilgrims receive the honorary title of Hajji. The pilgrimage is one of the five pillars of Islam, and is therefore compulsory for all adult Muslims who can afford to make the journey and are in good health. Every year, the pilgrimage attracts over 2 million Muslims from all parts of the world [2]. Such a high population density presents a risk of local epidemics and the spread of infectious agents worldwide. Acute respiratory infections (ARI) are the main cause of admission to Saudi hospitals during the pilgrimage [3]. Of the literature on respiratory viruses isolated

in pilgrims [4], it was noted that the respiratory infections most frequently isolated by PCR from patients during the Hajj were rhinovirus, followed by influenza virus, and human coronaviruses the majority of which were coronavirus 229E.

The last quarter of 2019 saw the appearance of a new virus in the coronavirus family, affecting all continents. This virus has been dubbed sarscov2 and the disease is known as coronavirus 2019 (COVID-19) [5]. This situation has presented the Kingdom of Saudi Arabia with public health challenges, given that it is the sole host of the pilgrimage. In order to control this pandemic, the Saudi authorities limited the Hajj in 2020 to a few pilgrims residing in Saudi Arabia, in strict compliance with health guidelines and protocols [6]. As a result of this decision, outside countries had to cancel the travel of their nationals.

In Côte d'Ivoire, the authorities have also suspended the Hajj 2020 and 2021 for Ivorian pilgrims, due to restrictions in the host country. So after two (02) successive years of interruption, Ivorian pilgrims were once again invited to travel to Saudi Arabia in 2022. When the hajj resumed, only 4527 pilgrims were actually transported to the Holy Land this year. Subsequently, a total of 10,000 Ivorian pilgrims were authorized to perform the hajj for the years 2023 and 2024 respectively. As for the 2024 edition, it was marked by over a thousand deaths of different nationalities. The main reason cited by the Saudi authorities was the heat, with temperatures reaching 50°C in the shade. However, the presence of respiratory syndrome among the cases of death was also observed. Against this backdrop, Côte d'Ivoire's Ministry of Health, Public Hygiene and Universal Health Coverage, noting the resurgence of cases of covid19 in certain countries of the sub-region on return from the 2024 hajj, has set up a health information and control system to test travelers returning from the 2024 pilgrimage.

The aim of this study is to assess the presence of SARS-CoV-2 in Ivorian pilgrims returning from the Hajj in 2024, by combining molecular detection and genetic characterization approaches, in order to confirm imports of the virus and identify possible new variants in Côte d'Ivoire. Our study aims to estimate the prevalence of COVID-19 in Ivorian pilgrims returning from the Hajj 2024, as well as their epidemiology, and predominant genotypes.

Methods

Sample and Data Collection

Between June 25 and July 4, 2024, nasopharyngeal swab samples were collected from Ivorian pilgrims coming back from the Hajj pilgrimage. This sampling happened as soon as they arrived at the international airport, in line with the national COVID-19 surveillance protocols. Skilled healthcare staff conducted the collections using standardized procedures to maintain the integrity and reliability of the samples. Each specimen was quickly placed in viral transport medium and sent under cold chain conditions to the reference lab for further molecular analysis. The socio-demographic data (age, sex) and geographical location of origin of each pilgrim were recorded on an epidemiological form and entered into an Excel database. Information on COVID-19 vaccination, notably vaccination status, was also collected from vaccination records if available, or from study participants if they were unable to provide records during the interview.

RNA Extraction

RNA from nasopharyngeal samples was extracted and purified manually using the Viral RNA Mini kit (QIAgen Cat. No. 52904) in accordance with the manufacturer's instructions. RNA was eluted in 60 µL of AVE buffer. Extracts were assayed and analyzed for purity using a spectrophotometer (NanoDropTM). The concentration and purity of extracts is obtained by measuring absorbance at 260 nm and 280 nm respectively. RNA extracts were stored at -80°C for analysis.

Detection of SARS-CoV-2 in Nasopharyngeal Swabs by RT-qPCR

To confirm the presence of SARS-CoV-2 in the samples, a multiplex RT-qPCR test was performed using the STANDARD M nCoV Real-Time Detection SD BIOSENSOR kit (Ref. M-NCOV-01). This kit detects a fragment of the E gene and a fragment of the ORF1ab gene of SARS-CoV-2. The internal control (IC) was used as an inhibition control for RT-qPCR. A QuantStudio 5 thermal cycler (Thermo Fisher Scientific) was used to perform the assay, and results were analyzed using Design and Analysis 2.6.0 software (Thermo Fisher Scientific). Reagent preparation protocols and amplification programs were carried out in accordance with the kit manufacturer's instructions.

Sequencing of the SARS-CoV-2 Spike Protein

Library Preparation and Sequencing with MinION Mk1C

We used the sequencing protocol for SARS-CoV-2 S gene amplicons obtained using the LP-471- reverse transcriptase PCR (RT-PCR) procedure for the SARS-Cov-2 S gene developed by the CDC (Center for Disease Control and Prevention). This protocol was used with reagents from New England BioLabs (NEB, Ipswich, MA, USA) and Oxford Nanopore Technology (Oxford Nanopore Technologies, Oxford, UK). Each sequencing run included positive samples and a negative control. The sequencing library was prepared using the Ligation Sequencing Kit (SQK-ND114.96, Oxford, UK) and individually coded using native barcodes (EXP-NBD114) following the manufacturer's instructions. For cDNA synthesis, two separate PCR reactions were performed for each SARS-CoV2-positive sample. The first PCR was performed with the SC1 primer pool, while the second PCR was performed with the SC2 primer pool. Master mix preparation for cDNA amplification was carried out using the SARS-Cov-2 S gene reverse transcriptase PCR (RT-PCR) kit containing both primer pools. The reaction mixture for 1 sample consisted of 4.25 µl of nuclease-free water, 5 µl of SC1 pool or SC2 pool, 12.5 µl of 2X SSIV reaction mix and 0.25 µl of RT SuperScript IV mix. The reaction mix was incubated in an Eppendorf thermocycler (Applied Biosystems) according to the following amplification program: 50°C for 10 minutes, 98°C for 2 minutes, followed by a 40-cycle phase comprising denaturation at 98°C for 10 seconds, hybridization at 60°C for 10 seconds and initial elongation at 72°C for 1 minute 15 seconds, followed

by final elongation at 72°C for 5 minutes. After amplification, electrophoresis of the amplified products was performed, and only the 2500-bp amplicons for PCR performed with primer pool SC1 and the 2200-bp amplicons for PCR performed with primer pool SC2 were selected and pooled for further sequencing. For end preparation, we used the NebNext Ultra II End Repair/dA-Tailing module (NEB, Ipswich, MA, USA), as follows: 1.2 µl of Ultra II End Prep reaction buffer (NEB, Ipswich, MA, USA), 0.5 L of Ultra II End Prep enzyme mixture (NEB, Ipswich, MA, USA), 3.3 µl of nuclease free water (NEB, Ipswich, MA, USA), and 3 µl of amplicons from the previous step. The reaction mixture was incubated in the thermocycler for 15 minutes at 25 C, 15 minutes at 65 C and cooled to 4 C. Amplicons were barcoded with a 1. 25 µl of EXP-NBD114 (barcodes ONT, Oxford, UK), 5 µl of Blunt TA Ligase Master Mix (M0367, NEB, Ipswich, MA, USA), 3 µl of nuclease free water (NEB, Ipswich, MA, USA), and 0.75 uL of the reaction mixture from the previous step, then incubated as follows: 20 min at 25 C, 10 min at 65 C, and cooled for 1 min at 4 C. Next, 10 µl of all barcoded samples were pooled and purified using 0.4 uL of AMPure XP magnetic beads (BeckmanCoulter, Brea, CA, USA). For adapter ligation, approximately 30 ng of barcoded samples were mixed with 10 µl of NEbNext Quick Ligation Reaction Buffer (NebNext Quick Ligation Module, E6056, NEB, Ipswich, MA, USA), 5 µl of MIX Adapter (AMII, ONT, Oxford, UK), and 5 µl of Quick T4 DNA Ligase (NebNext Quick Ligation Module, E6056, NEB, Ipswich, MA, USA). The mixture was incubated at room temperature for 20 minutes, followed by purification with AMPure XP magnetic beads (Beckman Coulter, Brea, CA, USA). The final library was quantified on a Qubit 4.0 spectrophotometer, using the Qubit dsDNA HS Assay kit (Thermo Fisher Scientific). Approximately 15 ng of the library was loaded in a final volume of 75 uL onto a flongle-type Flow-cell (FLO-FLG001, R.10.2) inserted into the MinION Mk-1C device (Oxford Nanopore Technologies, Oxford, UK) using an adapter following the instructions. Base calling and demultiplexing were performed using MinKNOW software (ONT, Oxford, UK) (with strict parameters to ensure that amplicons had barcodes at both ends), which is integrated into the MinION Mk1C (Oxford Nanopore Technologies, Oxford, UK). When the run is complete, the Fastq-pass file is retrieved for sequence analysis.

Bioinformatics Analysis

Raw data analysis of the fastq pass file was carried out on the EPI2ME bioinformatics platform (epi2me.nanoporetech.com/), developed by Metrichor Ltd. (Ox ford, UK), version 2022.04.26-13521, which uses the Artic, Nextclade, Pangolin subsections (Artic + Nextclade + Pangolin-v3.3.1). The Artic software studies the depth of coverage for each barcoded sample and can be used to explore individual amplicons that may not have been amplified with both primer pools. Nextclade software identifies genetic variants in relation to the reference genome and provides quality control data [7]. Pangolin indicates the sample lineage [8]. The Wuhan SARS-CoV-2 virus was used as the reference genome (MN908947).

Data Analysis

The prevalence of COVID-19 was deduced by dividing the number of patients testing positive for the specific virus by the number of participants. Rates were weighted by date of arrival. Descriptive data analysis was performed for demographic and epidemiological characteristics of patients with COVID-19 and influenza virus infection, using numbers and percentages. Patients positive and negative for SARSCoV-2 and influenza were compared using chi2, with a significance level of < 0.05. These analyses were performed using Rstudio software version (4.4.2).

Phylogenetic Analysis

For the construction of the phylogenetic tree, 18 SARS-CoV-2 sequences from the year 2024 from Saudi Arabia and those from Bangladesh (02 sequences), Taiwan (02 sequences), Bhutan (01 sequence), Philippines (02 sequences), Indonesia (02 sequences), Singapore (01 sequence) and Yunnan (01 sequence) were downloaded through the GISAID platform (see Appendix A: additional data). These sequences were selected on the basis of their collection date (June to July 2024) within the platform and their geographical location (Northern Hemisphere). These sequences were then aligned with the SARS-CoV-2 genomic sequences in our study. This alignment was carried out using MAFFT (Multiple Sequence Alignment) Version 7, [9]. Phylogenetic trees were constructed using the maximum likelihood (ML) method, using MEGA 7 software [10]. Bootstrap values of 1000 replications were used to assess the robustness of the tree topology.

Results

A total of 1,567 Ivorian pilgrims were sampled and tested for SARS-CoV-2 from June 26 to July 04, 2024. The average age was 57, with nearly 95% of pilgrims over 50, and 56% women. Of all participants, 53 (3%) tested positive for SARSCoV2 (Table 1). Of the pilgrims included, 1224 (78%) had received a vaccine against COVID prior to departure for the Hajj. During the testing period, the prevalence of SARSCoV2 infection was highest (5 and 6%) on June 28 and 29, in contrast to July 02 and 03, when there were no Sarscov2 positives. Of the Sarscov2-positive patients, 31 (58%) were female versus 22 (42%) males. The average age of SARSCoV2-positive patients was 57. Among pilgrims vaccinated against sarscov2, 17 were infected with influenza, as opposed to 5 non-vaccinated pilgrims. The average age of the flu-positive pilgrims was 55. No significant differences were found between Sarscov2- or flu-positive and flu-negative pilgrims with regard to age group, sex and Covid vaccination history. However, a significant difference was noted on the day of arrival (Table 1).

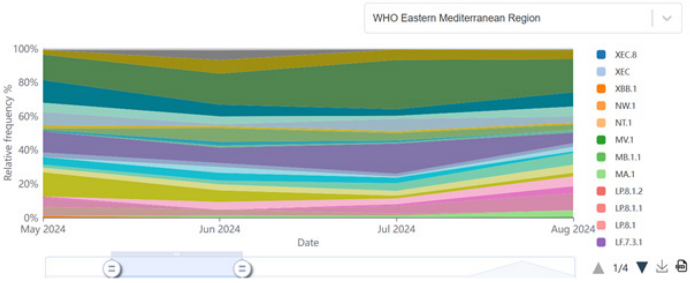
Genotyping

Of 53 Sarscov2-positive cases, 8 (15.09%) SARS-CoV-2 positives were sequenced. These positives were all Omicron variants. The clades obtained included 62.5% of 24A, and 12.5% of 24H, 24B and 24G respectively. Different lineages could be determined with JN.1.16.1 (25%), KS.1.1.2 (25%), and 12.5% respectively for KP2.33, LF.7.1, KP,2,2, and MA,1 (Figure 1).

Table 1: Characteristics of Ivorian pilgrims returning from the Hajj 2024.

	Number of tests n (%)	Positive SARS-Cov2 n (%)	Negative SARS-Cov2 n (%)	P value
Arrival date				
June 25	69 **(4.4)**	2 **(2.9)**	67 **(97.1)**	
June 26	465 **(29.7)**	22 **(4.7)**	443 **(95.3)**	
June 27	82 **(5.2)**	1 **(1.2)**	81 **(98.8)**	
June 28	212 **(13.6)**	11 **(5.2)**	201 **(94.8)**	
June 29	82 **(5.2)**	5 **(6.1)**	77 **(93.9)**	*< 0,005*
June 30	184 **(11.7)**	6 **(3.3)**	178 **(96.7)**	
01-July	88 **(5.6)**	3 **(3.4)**	85 **(96.6)**	
02-July	199 **(12.7)**	0	199 (100)	
03-July	78 **(5.0)**	0	78 **(100)**	
04-July	108 **(6.9)**	3 **(3.8)**	105 **(97.2)**	
Gender				
Male	679 **(43.3)**	22 **(3.2)**	657 **(96.8)**	
Female	884 **(56.5)**	31 **(4.5)**	853 **(96.5)**	*< 0,005*
Missing	4(0.2)	0	4 **(100%)**	
Average age	57,6	57,6	57,8	
Age				
01 _ 24	4 **(0.2)**	0	4 **(100%)**	
25_ 40	63 **(4.0)**	4 **(6.3)**	59 **(93.7)**	*< 0,005*
41_ 65	628 **(40.1)**	19 **(3.0)**	609 **(97.0)**	
> 65	213 **(13.6)**	10 **(4.7)**	203 **(95.3)**	
Missing	659 **(42.1)**	20 **(3.0)**	639 **(97.0)**	
Immunization				
Vaccine	1224 **(78.1)**	38 **(3.1)**	1186 **(96.9)**	*0.3272*
Non Vaccine	343 **(21.9)**	15 **(4.4)**	328 **(95.6)**	

a)

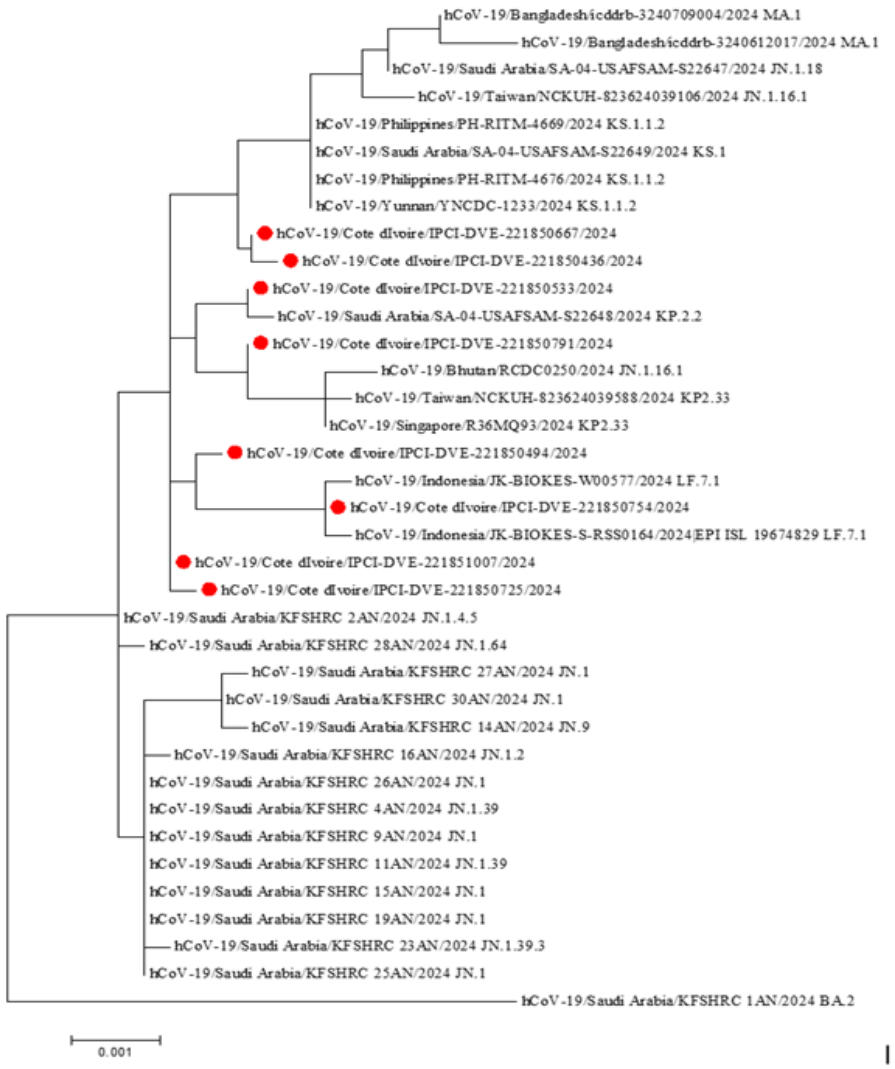


b)

Figure 1: Representation of Ivorian and Mediterranean pilgrim lineages.
a) Proportion of Omicron lines obtained from Ivorian pilgrims.
b) Representation of SARS-CoV-2 variants in the Mediterranean region from May 2024 to August 2024.

Figure 2 shows the evolutionary relationships between different hCoV-19 strains from different countries. The geographical diversity of sequences comes from several countries, including Saudi Arabia, Côte d'Ivoire, Indonesia, Bangladesh, the Philippines and Taiwan. Sequences are grouped according to their evolutionary proximity. Sequences from Côte d'Ivoire clearly show connections to Asian and Saudi strains. The phylogenetic tree shows significant genetic diversity of hCoV-19, with clear geographical groupings. The analysis highlights clusters illustrating the evolution and dispersal of the virus

Figure 3 shows the distribution of the number of positive cases by locality gave Abobo West (6 cases) and Abobo East (5 cases), with Yopougon and Cocody Bingerville each having 4 cases**.** Several localities, notably Treichville Marcory, Korhogo, Grand Bassam, Bouaké Sud, Adjamé, Plateau Attécoubé and Abengourou, had 3 cases. In sum, major urban areas such as Abobo, Yopougon, Cocody and Treichville Marcory are the most affected. Inland towns such as Korhogo, Bouaké and Abengourou are also reporting cases, albeit in smaller numbers. Several localities have only one positive case, including Touba, Oumé, Man, Kouto, Kani, Guiglo, Cocody (another locality), Bondoukou, Anyama and Agboville. Unspecified" indicates a case from an unknown or unspecified location.



Ivorian pilgrims' sequences

Figure 2: Phylogenetic tree illustrating the SARSCOV2 strains identified during the return of Ivorian pilgrims from June 25 to July 04. Nucleotide sequences of the S(Spike) gene were used to construct an unrooted tree. The maximum likelihood method under the general reversible model in MEGA 7 was used. Values at branch nodes indicate bootstrap resampling results after 1000 iterations, while scale bars indicate nucleotide frequency in the trees. The nation and years of virus collection and analysis structure are shown from left to right. Ivorian sequences are indicated by a red circle.

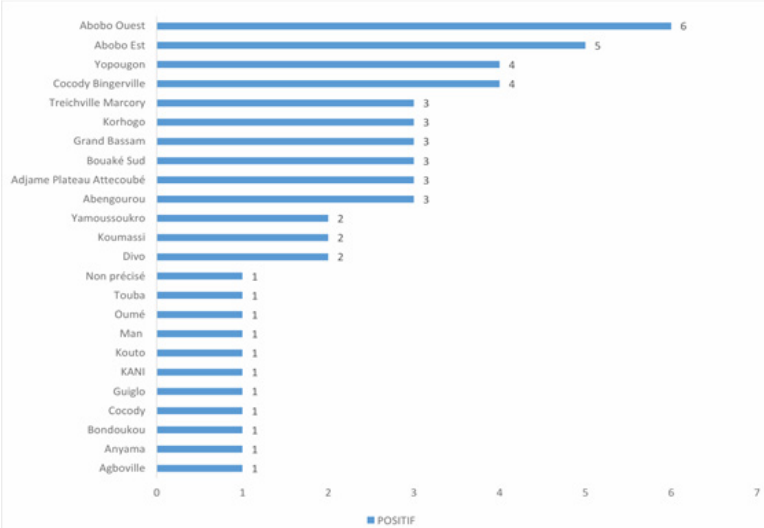


Figure 3: Geographical distribution of positive cases among pilgrims.

Discussion

This study presents the results of molecular tests carried out during the resurgence of Covid19 in certain countries of the African sub-region, on the return of their Hajj 2024 pilgrims. The return of Ivorian Hajj 2024 pilgrims took place from June 25 to July 04. During this period, our results enabled us to estimate SARS-CoV-2 infection rates. The prevalence of COVID19 among Ivorian pilgrims was low during the period. In contrast to our results, Egypt showed a significant prevalence of SARS-CoV-2, which was in the same proportion as our data [11] on the return of Egyptian pilgrims from Hajj 2022. This situation can be explained by the rapid evolution of strains, especially that of SARS-CoV-2, and also by changes in population behavior. The occurrence of a wave of COVID-19 during the return from the 2024 pilgrimage could be the sign of a new strain of SARS-CoV-2 [12]. Reports have recently emerged of an increase in the prevalence of COVID-19 in Japan [13] and in China [14], suggesting the emergence of a new wave of the disease. In 2024, the KP.3.1.1 sub-variant of the JN.1 variant has become the majority in France since July 2024, accounting for around 48% of sequences [15]. Although it is more easily transmitted and partially evades the pre-existing immune response, no significant increase in virulence has been observed, and hospitalizations have remained stable. Our results highlighted the important role played by women in the contingent of Ivorian pilgrims. This high proportion can be deduced from a number of specific factors, including culture, religion and social dynamics. In Côte d'Ivoire, more and more women are gaining access to education and economic opportunities. These opportunities enable them to finance their Hajj journey. It's also important to note that in many African societies, elderly women play a central role as spiritual figures and matriarchs. Having often outlived their spouses, and enjoying increased respect in the community, they are more likely to be supported through family fund-raising to perform the Hajj. The over-65 age group was the most affected by sarscov2 in our tests. People aged 65 and over remain particularly vulnerable to severe forms of COVID-19 and are at increased risk of infection by SARS-CoV- 2, due to several factors including a weakened immune system [16]. The immune system undergoes a decrease in efficiency, a phenomenon known as immune senescence. This makes them more susceptible to infections, including COVID-19.

Despite the prevalence of COVID-19 during Hajj 2024 in our study, it remains lower than that observed during other mass gatherings organized before the availability of vaccines [17]. This indicates that vaccines remain effective in preventing COVID-19 infection. However, our study does not report any efficacy of the post-vaccine in preventing COVID-19 infection.

Many countries have begun administering booster doses of vaccine, but there are concerns about their efficacy, durability and possible adverse effects [18]. It has therefore been suggested that the bivalent vaccine, which includes the circulating Omicron sub-variants in addition to the original SARS-CoV-2 strain, be admitted. Recently, the European Union's Committee for Human Medicinal Products (CHMP) recommended authorizing a bivalent vaccine and expanding the arsenal of available vaccines to protect the population against COVID-19, given that the pandemic is continuing and new waves of infections are expected [19] Geographical analysis of positive cases is essential for understanding the dynamics of the spread of an infectious disease, and for guiding control strategies. Results show a breakdown of positive cases by locality, providing a clear picture of the areas most affected. Urban areas record the highest number of positive cases. The high prevalence of cases in urban areas can be explained by several factors, including population density [20]. This may be due to the presence among pilgrims of people from these areas. This trend underlines the importance of raising awareness in places of worship of the importance of sanitary measures.

The sequences of SARS-CoV-2, have shown great genetic diversity across different continents, influenced by adaptive mutations and human-to-human transmission [21]. The resulting phylogenetic tree illustrates the genetic relationships between different hCoV-19 strains collected in different countries, including Côte d'Ivoire, Saudi Arabia, Indonesia, Taiwan and the Philippines. Evolutionary proximity was observed between certain Ivorian strains and those from Indonesia and Saudi Arabia, suggesting intercontinental transmission events. These countries have a very strong Islamic community and are expected to be among the largest contingents at the Hajj 2024 edition.

The Saudi Arabian strains appear highly diversified, which could be the result of widespread propagation and multiple introductions of the virus. In contrast, Asian strains (Taiwan, Philippines) appear more distantly related in the phylogenetic tree, indicating distinct evolutionary lineages. This genetic variability reflects the different dynamics of viral propagation depending on the region and the sanitary measures in place [22]. The emergence of several distinct lineages highlights the virus' ability to accumulate mutations over time. Some mutations may confer adaptive advantages, influencing the transmissibility of the virus or its escape from the immune response [23]. A study by [24] also showed that certain specific mutations, such as the D614G mutation, favored increased virus propagation. The phylogenetic relationships observed in this tree support the hypothesis of virus dispersal via international travel. Such travel has facilitated the introduction and establishment of new strains in various parts of the world [25]. Previous studies have confirmed that international transport routes and mass gatherings such as the Hajj play a key role in the spread of emerging viruses [26]. Although this tree provides valuable information, certain limitations must be taken into account, including the distribution of the strains analyzed could influence the structure of the tree and conclusions on the evolutionary proximity of variants [26]. Also, the nature of the mutations. Further analysis of specific mutations would be required to understand their functional effects on virus transmission and pathogenicity.

Our results revealed a low prevalence among Ivorian pilgrims. However, the epidemiological and sanitary stakes merit particular attention. Pilgrims' participation in an international gathering such as the Hajj exposes them to various viral strains circulating in other countries. This could encourage the introduction of new variants of the virus into Côte d'Ivoire, increasing the risk of spread and

new epidemic waves. Increased vigilance is therefore required to limit this potential impact. These results underline the importance of reinforcing detection measures, particularly at points of entry to the national territory (airports, ports and border posts). Systematic surveillance following pilgrims' return would enable positive cases to be identified rapidly and community transmission to be limited. Vaccination remains an essential tool for limiting viral transmission and reducing the severity of infections. It is crucial to make pilgrims aware of the importance of a complete vaccination regimen prior to travel, accompanied by barrier measures (wearing a mask, hand hygiene, avoiding crowds). The results of this study corroborate those of other research conducted on infectious diseases associated with mass gatherings. For example, [6] also reported an increased risk of spreading new variants at international events such as the Hajj, justifying the implementation of enhanced surveillance strategies. Similarly, an analysis by [27] highlighted the importance of preventive measures, including vaccination, in reducing the transmission of respiratory diseases. That study, like the present analysis, highlights the impact of international travel on public health and the imperative of effective health preparedness. Finally, [28] emphasized the need for post-Hajj follow-up to better understand the evolution of pilgrimage-acquired infections, which echoes the limitations of our study with regard to the lack of longitudinal follow-up of positive cases. Nevertheless, the study may not accurately reflect all pilgrims due to limited or non-random sampling. The absence of longitudinal follow-up prevents an assessment of the long-term impact of the infection, particularly in terms of complications and secondary transmission.

Conclusion

The return of Ivorian Hajj pilgrims from June to July 2024 in a context of resurgence of COVID-19 in West Africa, was not alarming due to the low infection rates observed. Female sex and the over-65 age group were the most important socio-demographic factors, and should be the most closely monitored for future Hajj. Our results also show the limits of the effectiveness of COVID-19 vaccine. Omicron remains the main variant of SARS-CoV-2, with numerous sub-variants identified. The study also highlighted the lack of vaccination for some Hajj pilgrims. It would therefore be advisable recommend vaccination for people taking part in mass gatherings outside the country, including the Hajj, in order to prevent outbreaks. The implementation of a post-Hajj respiratory disease surveillance protocol to ensure effective epidemiological surveillance after the return of pilgrims, enabling the rapid identification of any abnormal increase in cases of respiratory disease. In-depth research into the impact of travel on viral transmission is also essential, including further analysis of the impact of the Hajj on seasonal epidemics and pilgrims' immune resistance. These actions will enable us to anticipate and better control the health risks associated with international gatherings, while safeguarding public health in Côte d'Ivoire.

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