



Short Communication

Influenza at the 2021 Grand Magal of Touba and possible spread to rural villages in South Senegal - a genomic epidemiological study



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ABSTRACT

Objectives: Influenza is frequent among pilgrims participating in the Grand Magal de Touba (GMT), in Senegal, with a potential to spread to contacts when they return home.

Methods: Ill pilgrims consulting at a health care center in Mbacké city close to Touba during the 2021 GMT, pilgrims returning to Dielmo and Ndiop villages, and patients who did not travel to Touba and consulted at health care centers in these two villages in 2021 were tested for the influenza virus by polymerase chain reaction on nasopharyngeal samples. Next-generation sequencing and comparative and phylogenetic analyses of influenza A virus genomes were performed.

Results: A total of 62 of 685 patients tested positive for influenza A virus, including 34 of 53 who were consulted in Mbacké in late September, six of 129 pilgrims who returned home in early October, and 20 of 42 villagers from October 3 to 29. A total of 27 genomes were obtained. Four clusters were observed based on the phylogenetic analyses, suggesting that Mbacké patients and returned pilgrims may have shared closely related viral strains with patients inhabiting the villages who did not participate in the GMT.

Conclusions: Villagers in Ndiop and Dielmo may have been infected with viral strains originating from the GMT and possibly imported by pilgrims who returned from the GMT.

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Introduction

The Grand Magal of Touba (GMT) is the largest religious mass gathering in Senegal. An estimated 4–5 million pilgrims participate in the event each year.

Early works showed that respiratory tract infection (RTI) symptoms are among the most common reasons for ill pilgrims to present to health care centers during the GMT [1,2]. In addition, cohort studies conducted in pilgrims from two rural villages in South Senegal evidenced rates of 41% for RTI symptoms and acquisition of respiratory viruses after participating in this event [3–5]. Finally, studies conducted in pilgrims consulting health care cen-

ters for RTI during the GMT allowed evidencing outbreaks of influenza related to this mass gathering, with up to 64% of individuals testing positive for the influenza A virus [6]. Therefore, the present study aimed to test the hypothesis of the introduction of the influenza virus into two villages in South Senegal by pilgrims returning from the September 26, 2021 GMT, with a subsequent spread of the virus.

Materials and methods

Patients and samples

Data were collected in (i) the Mbacké health care center (near Touba) from 53 patients with cough from September 24 to 28 2021, (ii) Dielmo and Ndiop health care centers located in the south of

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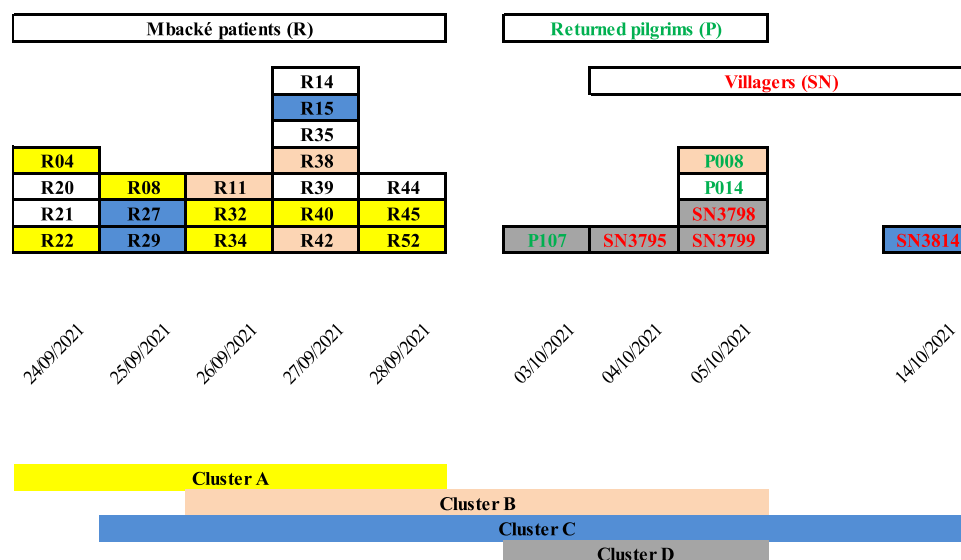


Figure 1. Clusters of influenza cases based on phylogenetic analysis of genomes.

Table 1
Characteristics of the participants.

	Patients with respiratory tract infection symptoms at Mbacké health care center during the GMT N = 53	Pilgrims from villages before the GMT N = 129	Pilgrims from villages after the GMT N = 129	Patients with fever in the villages (January–December 2021) N = 374
Median age (years)	13	23	23	NA
Male gender	37 (69.8)	61 (47.3)	61 (47.3)	NA
Respiratory symptoms	53 (100.0)	17 (13.2)	51 (39.5)	NA
Cough	53 (100.0)	11 (8.5)	36 (27.9)	NA
Rhinitis	44 (83.0)	6 (4.7)	21 (16.3)	NA
Fever	30 (56.6)	0 (0)	13 (10.1)	374 (100.0)
Positive for influenza A by PCR	34 (64.2)	0 (0)	8 (6.2)	20 (5.3)
Positive for influenza B by PCR	0 (0)	0 (0)	0 (0)	0 (0)
Number of positive samples that underwent virus genome sequencing	20/34	0/0	3/8	4/20

GMT, Grand Magal de Touba; PCR, polymerase chain reaction.

Senegal from January 1 to December 19, 2021 for 374 patients who presented with fever $\geq 37.5^{\circ}\text{C}$ to detect the first cases of influenza in the villages; and (iii) 129 GMT pilgrims from these two villages who were systematically sampled before leaving for Touba (September 14 to 18, 2021) and upon return (October 5 to 8, 2021).

Virological analyses (Supplementary material)

The samples were tested for influenza A and B viruses by polymerase chain reaction (PCR), and next-generation sequencing of the influenza A virus genomes was performed on RNA extracts, with a PCR cycle threshold value below 32.

Results

Prevalence of respiratory symptoms and influenza virus infections

In the patients with cough who presented to the Mbacké health care center, the median age was 13 years (range 0–75 years) and 70% were men (Table 1). Apart from cough (100%), the most frequent symptoms were rhinitis (83%) and fever (57%). A total of 34 of 53 (62%) patients were positive for the influenza A virus and none for the influenza B virus. In pilgrims returning from the GMT to the villages, the median age was 23 years (range 0–76 years) and 47% were men. A total of 51 of 129 (40%) patients reported respiratory symptoms, including cough (36 [28%]) and rhinitis (21 [16%]). Eight of 129 (6%) pilgrims were positive for the influenza A virus

and none for the influenza B virus. Finally, in patients with fever who presented to the health care center in the villages, 20 of 374 (5%) patients tested positive for the influenza A virus and none for the influenza B virus.

Chronology of positivity of samples for influenza A virus by polymerase chain reaction

From January 1 October 1, 2021, no influenza case was detected in ill people in the villages of Dielmo and Ndiop. In the 34 of 53 patients positive for influenza A at the Mbacké health care center, 10 of 16, three of six, seven of nine, nine of 13, and five of nine were diagnosed on September 24, 25, 26, 27, and 28, respectively. The first case of influenza A in the village was detected on October 2, 2021 in a pilgrim returning from the GMT (Supplementary Figure 1). Further cases were observed in other returned pilgrims on October 2, 3, and 4 and in villagers who did not travel to Touba from October 3–29. Of note, one case was also observed in a returned pilgrim on October 28.

Analysis of influenza A virus genomes

A total of 27 full-length or near full-length genomes with the eight segments were obtained. They were generated from 20 patients who participated in the GMT and were diagnosed with influenza A virus at the Mbacké health care center, from three patients who participated in the GMT and were pilgrims diagnosed

with influenza A virus when returning in Ndiop and Dielmo villages, and from four patients from the villages who did not participate in the GMT. All genomes were classified in the same influenza A/H1N1 virus lineage 6B.1A.5a.1 (Supplementary results). Four clusters (A, B, C, and D) could be identified based on phylogenetic analyses (Figure 1), suggesting that patients who were infected in Touba may have shared closely related viral strains with patients inhabiting the Ndiop village who did not participate in the GMT.

Discussion

National or international spread of infectious diseases triggered by mass gathering events has been exemplified by the globalization of cholera or meningococcal meningitis after epidemics occurring at the Hajj pilgrimage in Saudi Arabia [7]. Amplification of SARS-CoV-2 circulation at mass gatherings has also been a concern during the COVID-19 pandemic [8]. Surveillance data on the incidence of influenza-related hospitalizations have suggested that the carnival may have played a role in the spread of influenza virus in the Netherlands in 2018 [9]. However, to the best of our knowledge, no study based on virus genomic analysis has been conducted to challenge the hypothesis that a mass gathering could play a role in the spread of the influenza virus.

The present study identified influenza A/H1N1 virus of clade 6B.1A.5a.1 as responsible for the influenza cases diagnosed during the GMT, as well as in two Senegalese villages over a 1-month period. This lineage was present since at least August 2020 in various countries, then considerably decreased (or almost vanished) at the end of 2022. Interestingly, the analyses of viral genomes allowed the identification of four clusters of the influenza A virus, indicating that people who participated in the GMT and others who did not participate in the GMT and inhabit the Ndiop and Dielmo villages may have shared closely genetically related viral strains. In addition, they could indicate that people in the villages may have been infected with viral strains originating from the GMT and possibly imported from villagers in Ndiop and Dielmo when returning from the GMT. These hypotheses are also supported by the chronology of influenza A virus cases diagnosed by PCR because the incubation for influenza is no longer than 2 days and the duration of symptoms is most often no longer than 7 days. Nevertheless, interpretation of phylogeny reconstructions should be cautious. Indeed, the genome sequences analyzed here were obtained from samples collected during the same and short period of time, in the same country, and even the same epidemiological settings for some of them. Hence, they displayed small numbers of nucleotide differences between each other, which hampered providing robustly supported genetic clusters in phylogeny reconstructions. Topologies of phylogenetic trees at least partly differed according to the segment, and sequences from the present study were also clustered with some sequences from the Global Initiative on Sharing All Influenza Data database obtained from patients sampled in Senegal in 2021, for which detailed epidemiological data were not available (Supplementary material, Results, Tables 1 and 2, and Figures 2–9). Thus, the strains detected here may only be representative of those that circulated during the study period and that could have been transmitted from one place to another, including from Touba to the Ndiop and Dielmo villages.

Before the COVID-19 pandemic, influenza epidemiology in Senegal was characterized by a continuous circulation of influenza virus throughout the year, with two distinct seasonal peaks, the first around March and the second during the rainy season, which is around August. This epidemiological pattern was also observed in 2000; however, in 2021, the March seasonal peak was completely absent, whereas the August seasonal peak started between the September 13 and 19 and lasted until end of November [10]. In-

terestingly, although the influenza virus was circulating in Senegal since mid-September 2021, the first cases in Ndiop and Dielmo were diagnosed in early October in pilgrims returning from the GMT and then in villagers who did not travel to Touba.

Taken together, present data suggest that mass gatherings, such as the GMT, may play a role in the spread of influenza viruses at the country level through infected participants returning to areas where the virus was not yet circulating. Genomic surveillance of communicable diseases could help identifying the geographical spread of respiratory viruses in mass gathering settings.

Author contributions

CS and PG designed the study. NG, FSD, MB, HB, MS, LT and PC analyzed the data. NG, PG, CS and PC wrote the manuscript. MD, AD and CS supervised the data collection. All authors read and approved the final manuscript.

Declarations of competing interest

The authors have no competing interests to declare.

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Ethical approval

The project was approved by the National Ethics Committee for Health Research in Senegal, authorization number N°. 00087 MSAS/ DGS/ DS/ CNERS for the surveillance among villagers and N°. SEN17/62 for the surveillance among pilgrims. It was performed in accordance with the good clinical practices recommended by the Declaration of Helsinki and its amendments.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at [doi:10.1016/j.ijid.2024.01.019](https://doi.org/10.1016/j.ijid.2024.01.019).

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