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Respiratory and gastrointestinal infections at the 2017 Grand Magal de Touba, Senegal: A prospective cohort survey

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ABSTRACT

Background: The Grand Magal of Touba is the largest Muslim pilgrimage in Senegal with a potential for infectious disease transmission.
Methods: Clinical follow-up, adherence to preventive measures and qPCR-based respiratory and gastrointestinal pathogens carriage pre- and post-Magal, were assessed.
Results: 110 pilgrims from South Senegal were included. The duration of stay in Touba was 3 days. 41.8% and 14.5% pilgrims reported respiratory and gastrointestinal symptoms. Most individuals having the onset of symptoms during their stay in Touba, or soon after returning. The acquisition of rhinoviruses, coronaviruses and adenovirus was 13.0, 16.7 and 4.6% respectively and that of *Streptococcus pneumoniae* and *Haemophilus influenzae* was 3.7% and 26.9%.

Acquisition of gastrointestinal viruses and parasites was low, while bacterial acquisition ranged from 2.2% for *Campylobacter jejuni* to 33.0% for enteropathogenic *Escherichia coli*.
Conclusion: This preliminary study confirms that Grand Magal pilgrims are likely to be exposed to communicable disease risk as observed in other pilgrimage settings. Further study including larger numbers of pilgrims are needed to investigate potential risk factors for respiratory and gastrointestinal infections at the Grand Magal.

1. Introduction

The Grand Magal is at once a religious ceremony and a festival. About 4–5 million Mourid participate to the celebration each year, coming from across Senegal and the surrounding countries, as well as from countries outside Africa, making this event the largest Muslim religious mass gathering in West-Africa. The two central events of the Magal are visits to the Great Mosque of Touba, which involves a partial circumambulation of the mosque, and to the mausoleum of Cheikh Ahmadou Bamba. Pilgrims also visit the mausoleums of several other important Mouride leaders who were descendants of the Cheikh. The Mosque area is a secluded place surrounded by walls, within the city. The access to the Mosque area is allowed to everyone (including tourists) but flows are regulated by security staffs. In addition, pilgrims visit places in Touba that are associated with the holy life of the Cheikh, including the "Well of Mercy," a spring which sacred water is reputed to be able to heal all sorts of illnesses and misfortune. A visit to the central library of Touba which contains the writings of the Cheikh and other influential Mouride theologians, and to the Mouride University are among the other places visited by pilgrims. Finally, pilgrims visit their personal Mouride spiritual guides, or Marabouts who receive their followers in their personal residence in the city. Recitation of the Koran and spiritual introspection complete these religious activities. During the Grand Magal period, in addition to the religious celebrations, intense cultural activity takes place including conferences, seminaries, and debates between representatives of various Muslims communities

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from Senegal and from the diaspora. Recitation of poems by Ahmadou Bamba, and collective hymn-singing sessions are organized at night, in the streets, under large open tents. Informally, pilgrims also visit Touba's large temporary marketplace, one of the biggest in the country where a wide range of products can be found with prices lower than those of any other market in Senegal.

During this period, pilgrims are accommodated in private housing structures since there are no hotels in Touba. These houses can be residences of local inhabitants, where family members living in different parts of Senegal or who have emigrated outside the country, come together during the Magal period, or Marabout houses with a capacity that can be up to hundreds of individuals. Many pilgrims sleep on carpets, on the floor, into the houses or on outside terraces.

Food is prepared collectively by family members or by Marabout followers. Slaughtering of animals is mostly done in the streets, in front of housing structures. Different community associations of Mouride disciples (dahiras) are responsible for feeding the pilgrims through free provision of food in the streets and along the roads to Touba. The Senegalese government also provides some technical support.

Medical care is provided free of charge during the Grand Magal. Most of the available local public medical infrastructures are involved in the medical preparation, surveillance and response during the event [1]. As an example, three tertiary care and four secondary care hospitals (total bed capacity of 481) and about 300 primary healthcare centers and 50 private structures in Touba and surrounding cities were mobilized in 2015. The vast majority of the medical staffs taking care of patients during the Grand Magal are nurses, midwives and volunteer community health agents. Overall, 78 medical doctors were mobilized during the Grand Magal 2015, many of them coming from Dakar to temporarily reinforce the local staff.

Based on the registers of consultations carried out in the public health structures of the region, we evaluated the number and the profile of morbidity observed during the pilgrimage of 2015 among 32,229 in and out-patients [1]. The most frequent complaints were fatigue (12%) followed by trauma (11%) and heatstroke (11%). Infectious diseases have also been highly reported, including febrile systemic illnesses (5%) and rapid-test confirmed malaria (3%), diarrheal diseases (5%), respiratory tract infection symptoms (5%) and ear, nose and throat infection symptoms (5%). Microbiological investigation of these infections was not conducted in most cases.

The objective of the present preliminary descriptive study was to evaluate the prevalence of respiratory and gastrointestinal symptoms and the carriage of pathogens commonly associated with these symptoms among pilgrims before and after their stay in Touba.

2. Materials and methods

The complementary Fig. 1 details the procedure of this study.

2.1. Study population

A convenience sample of pilgrims participating in the Grand Magal 2017 was surveyed by a longitudinal prospective cohort study that was conducted from November 4th to November 23rd, 2017. Inhabitants of two villages: Dielmo (13°43′ 22.07″ N, 16°24′40.09″ W) and Ndiop (13°41′08.01"N, 16°23′01.01″ W), located in Fatick region, South Senegal were included. Villages are divided in concessions where individuals from a same family live in close contact. The population in the villages was 991 (510 Ndiop and 481 Dielmo) in 2017, including about 50% Mourides, part of whom did not travel to Touba because of limited financial resources. All pilgrims participating to the 2017 Magal in these villages were invited to participate in our study and were identified by the nurses in charge of the primary health care centers in both villages. At inclusion, the pilgrims were questioned using a standardized questionnaire. This questionnaire addressed demographics, chronic medical conditions and vaccination status against influenza. In Touba, pilgrims were housed in different concrete houses; three groups in three family houses and others in different Marabout houses. Health issues occurring during and after the Magal, were recorded by a nurse who travelled with the group of pilgrims. Housing location in Touba and use of preventive measures (face mask, disposable handkerchiefs, hand washing with soap hand disinfectant) were investigated by questionnaires during the stay in Touba. The consumption of antibiotics was also documented through post-travel questionnaires. Influenza like illness (ILI) was defined as the association of cough, sore throat and fever [2]. Diarrhea was defined by at least three loose or liquid stools per day.

2.2. Sample collection

The procedure included systematic throat, nasal and stool swabs, 1–3 days before departing from the villages (pre-Magal specimens) and 3–6 days following return (post-Magal specimens). Respiratory samples were collected with commercial rigid cotton-tipped swab applicators (Medical Wire & Equipment, Wiltshire, UK) that were inserted in the anterior nose or in the oro-pharynx, and then placed in viral transport media (Sigma Virocult®). Participants provided stools samples in sterile containers. Investigators secondary collected small amounts of stools using swab applicators, placed in viral transport media (Sigma Virocult®). This standard procedure was previously explained to the pilgrims by the investigators. Swabs were kept at 4 °C before being transported to the Dakar laboratory for storage in a −80 °C freezer within 48 h of collection and subsequently transferred to Marseille on dry ice for being processed.

2.3. Identification of respiratory and gastrointestinal pathogens

2.3.1. Respiratory specimens

The EZ1 Advanced XL (Qiagen, Hilden, German) with the Virus Mini Kit v2.0 (Qiagen) was used for the DNA and RNA extraction from the respiratory samples according to the recommendation of the manufacturer. All quantitative real-time PCR were performed using a C1000 Touch™ Thermal Cycle (Bio-Rad, Hercules, CA, USA). Positive results of bacteria or virus amplification were defined as those with a cycle threshold (CT) value ≤ 35. Negative control (PCR mix) and positive control (DNA from bacterial strain or RNA from viral strain) were included in each run.

2.3.2. Identification of respiratory virus

Human coronavirus (HCoV) and human para-influenza virus (HPIV) were detected by one-step duplex quantitative RT-PCR amplifications of HCoV/HPIV-R Gene Kit (REF: 71-045, Biomérieux, Marcy l'Etoile, France), according to the manufacturer's recommendations. The Multiplex RNA Virus Master Kit (Roche Diagnostics, France) was used to detect influenza A, influenza B, human rhinovirus, human enterovirus, metapneumovirus, respiratory syncytial virus, adenovirus and internal controls MS2 phage by one-step simplex real-time quantitative RT-PCR amplifications [3].

2.3.3. Identification of respiratory bacteria

Real-time PCR amplifications were carried out by using LightCycler® 480 Probes Master kit (Roche diagnostics, France) according to the manufacturer's recommendations. The SDD gene of *Haemophilus influenzae*, phoE gene of *Klebsiella pneumoniae*, nucA gene of *Staphylococcus aureus*, lytA gene of *Streptococcus pneumoniae* [4], P1 gene of *Mycoplasma pneumoniae* [5], ctrA gene of *Neisseria meningitidis* [6] and Toxin gene of *Bordetella pertussis* [7] were amplified with internal DNA extraction controls T4, as previously described.

2.3.4. Gastrointestinal specimens

Stool swabs were centrifuged at 4000 × g for 10 min. The RNA were extracted from 200 μL clear supernatant using the EZ1 Advanced XL

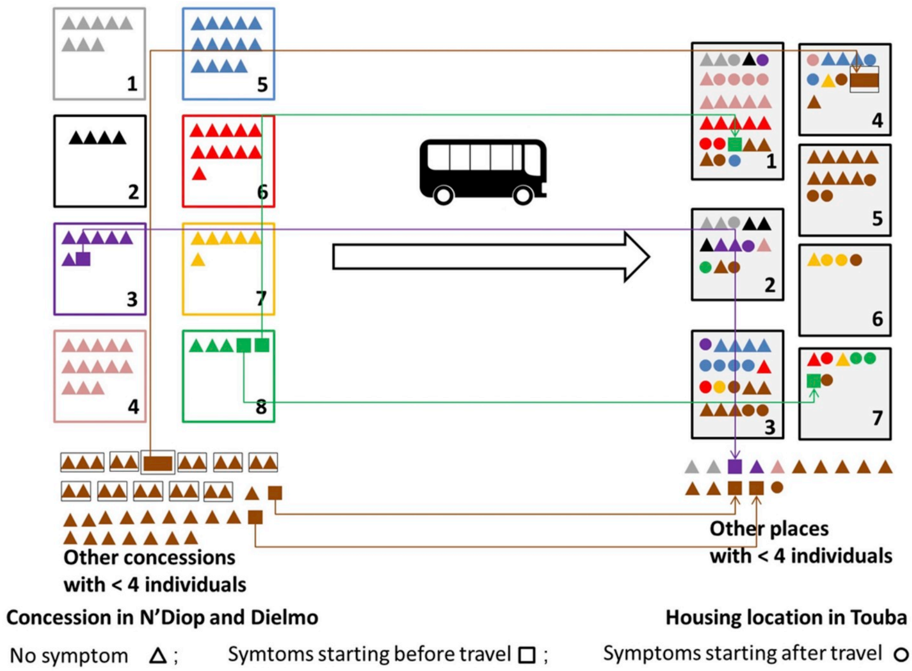

Fig. 1. Distribution of pilgrims according to concession in villages and to housing location in Touba.

(Qiagen, Hilden, German) with the Virus Mini Kit v2.0 (Qiagen) according to the manufacturer's recommendation.

The DNA was extracted by semi-automated extraction. 200 µL of rectal swabs was added to 350 µL of G2 lysis buffer (Qiagen) and glass powder in a tube, then disrupted in a FastPrep BIO 101 apparatus (Qbiogene, Strasbourg, France) at maximum power for 40s. The incubation was at 100 °C for 10 min to allow for complete lysis. Then, the tubes were centrifuged at 10,000 g for 1 min. Subsequently, 200 µL of supernatant was collected in other tubes and enzymatically digested using 20 µL of proteinase K (20 mg/mL, Qiagen), and incubated overnight at 56 °C. The automated procedure using the EZ1 Advanced XL (Qiagen, Hilden, German) with the DNA Tissue Kit (Qiagen) according to the manufacturer's recommendation [8].

2.3.5. Identification of gastrointestinal virus

The Multiplex RNA Virus Master Kit (Roche Diagnosis, France) was used for detecting hepatitis A and E virus, adenovirus, rotavirus, norovirus, astrovirus and internal controls MS2 phage by one-step simplex real-time quantitative RT-PCR amplifications [9–11].

2.3.6. Identification of gastrointestinal bacteria and parasites

Real-time PCR amplifications were carried out using the LightCycler® 480 Probes Master kit (Roche diagnostics, France) according to the manufacturer's recommendations. The mapA gene of *Campylobacter jejuni*, ipaH gene of *Shigella spp*/EIEC *(enteroinvasive Escherichia coli)* [12], gene invA of *Salmonella spp*, gene stx1 and stx2 of enterohemorrhagic *E. coli,* EAF and eae gene of enteropathogenic *E. coli* (EPEC) and pCVD432 gene of enteroaggregative *E. coli* (EAEC) [13] were amplified with internal DNA extraction controls TISS. The 18S gene was used to detect *Entamoeba histolytica* and *Giardia lamblia*. Finally, gene Hsp70 was amplified to test *Cryptosporidium spp.* [8].

All quantitative real-time PCR to detect respiratory and gastrointestinal pathogens were performed using a C1000 Touch™ Thermal Cycle (Bio-Rad, Hercules, CA, USA). Negative control (PCR mix) and positive control (DNA from bacterial strain or RNA from viral strain) were included in each run. A cycle threshold (CT) value ≤ 35 was used to assess positive results of bacteria or virus amplification.

2.4. Statistical analysis

Pearson's chi-square or Fisher's exact tests were used for statistical analyses. The acquisition of respiratory and gastrointestinal pathogens was defined as negative before travel and positive when returning from Touba. Prevalence of respiratory and gastrointestinal pathogens carriage before and after the Grand Magal, were compared using the McNemar's Test. Percentages and comparisons were carried out using STATA 11.1 (Copyright 2009 StataCorp LP, http://www.stata.com). A p value of ≤0.05 was considered significant.

2.5. Ethics

Pilgrims were invited to participate on a voluntary basis. Participants (or their parents when minors) were asked to sign a written consent form. The protocol was approved by the National Ethic Committee for Health Research in Senegal (SEN17/62). It was performed in accordance with the good clinical practices recommended by the Declaration of Helsinki and its amendments.

3. Results

3.1. Study population

A total of 123 pilgrims agreed to participate to the survey. Among them, 110 participated to the Grand Magal and fully completed the study and 13 participants cancelled their trip to Touba after inclusion in the study. Most participants (61.8%) were from family groups from 8 different main concessions including 4 or more persons per concession participating to the Magal, while 38.2% remaining participants travelled in small groups from a same concession with 3 persons (2.7%) or 2 persons (18.2%) or alone (17.3%) (Fig. 1). Forty-eight pilgrims were male (43.6%) and 62 female (56.4%) with a gender ratio of 0.77. The median age was 20 years (min = 8 months, max = 75 years, interquartile (15, 31 years)). Twenty-nine (26.4%) participants were children < 15 years and 5 (4.5%) pilgrims were over 60 years of age. Most participants were from N'Diop village (90/110, 81.8%). Chronic respiratory disease was the most common comorbidity (8.2%) (Table 1).

The duration of the stay in Touba was short (3 days) and most of

Table 1
Characteristics of the population.

Variables		n (N = 110)	%
Village	Dielmo	20	18.2
	Ndiop	90	81.8
Concession in village	1	8	7.3
	2	4	3.6
	3	7	6.4
	4	13	11.8
	5	14	12.7
	6	11	10.0
	7	6	5.5
	8	5	4.5
	Others[a]	42	38.2
Median duration of stay in Touba (interquartile) (days)		3 (3, 4)	
Gender	Female	62	56.4
	Male	48	43.6
Age	Median age (interquartile) (years)	20 (15, 31)	
	Age ≤ 15 years old	29	26.4
	Age > 15–45 years old	67	60.9
	Age > 45–60 years old	9	8.2
	Age > 60 years old	5	4.5
Chronic diseases	Diabetes mellitus	1	0.9
	Hypertension	2	1.8
	Chronic respiratory disease	9	8.2
	Chronic heart disease	2	1.8
	Others	14	12.7
Housing location in Touba	1	28	25.5
	2	13	11.8
	3	20	18.2
	4	11	10.0
	5	12	10.9
	6	4	3.6
	7	7	6.4
	Others[a]	15	13.6

[a] Less than 4 individuals per concession in villages or housing location in Touba.

pilgrims (86.4%) were housed in 7 different locations with some overlapping between concessions (Fig. 1).

Regarding preventive measures, 46.4% of pilgrims reported washing their hands more often than usual during the Grand Magal, 63.6% used hand gel frequently. 32.3% and 2.8% pilgrims reported using frequently disposable handkerchiefs and face mask, respectively during the pilgrimage. Only one pilgrim was vaccinated against influenza (Table 2).

3.2. Clinical features and detection of respiratory and gastrointestinal pathogens

3.2.1. Respiratory symptoms and pathogens

During the study period 46 (41.8%) pilgrims reported at least one respiratory symptom with most individuals having the onset of symptoms during their stay in Touba (20.0%) or soon after returning (10.9%). Only 7 participants (6.4%) reported suffering from respiratory symptoms before traveling to Touba. Median time between arrival in Touba and onset of symptoms was 3 days. Rhinitis was the most frequent symptom (30.0%), followed by cough (24.6%), sore throat (10.0%), expectoration (4.6%) and voice failure (4.6%). However, 10.0% ill pilgrims reported fever and 1.8% ILI. Only 2.7% received antibiotics for respiratory tract infection and none of the participants was hospitalized. A total of 42/46 (91.3%) individuals were still symptomatic on returning to Ndiop and Dielmo.

Pre- and post-Magal nasopharyngeal swabs were collected from all 110 and 108 (98.2%) participants, respectively. The most common virus detected was human rhinovirus (18.2%) and human coronavirus (18.2%), followed by adenovirus (7.3%) and influenza B virus (2.7%) (Table 4). The global carriage of bacteria was high, ranging from 34.5% for *S. pneumoniae* to 77.3% for *H. influenzae*. The prevalence of rhinoviruses, coronavirus and adenovirus increased post-Magal with acquisition of 13.0, 16.7 and 4.6% respectively. This increase was statistically significant for coronaviruses, $p = 0.0001$. Carriage of *S. pneumoniae, S. aureus* and *K. pneumoniae* decreased following a participation to the Magal, while that of *H. influenzae* significantly increased, $p = 0.03$. Despite the overall decrease in bacterial carriage, the acquisition of bacteria ranged from 3.7% for *S. pneumoniae* to 26.9% for *H. influenzae* (Table 3). We found no significant difference in the prevalence of respiratory symptoms and in the acquisition of respiratory pathogens according to concession and housing location in Touba (Supplementary Tables S1 and S2).

3.2.2. Gastrointestinal symptoms and pathogens

At total of 16 (14.6%) pilgrims reported gastrointestinal symptoms mostly during their stay in Touba (7.3%) or soon after returning (3.6%). Only 4 participants (3.6%) reported suffering gastrointestinal symptoms before traveling to Touba. Vomiting was the most frequent symptoms (9.1%) followed by diarrhea (4.6%), constipation (1.8%) and nausea (0.9%). None of participants was hospitalized and only 2/110 (1.8%) received antibiotic for gastrointestinal symptoms. A total of 7/16 (43.8%) of the pilgrims were still symptomatic upon their return to the villages.

Sixty (54.5%) participants provided stool sample pre- and 69 (62.7%) post-Magal, with 45 (40.9%) paired specimens collected. Most common viruses were adenovirus (9.5%) and norovirus (2.4%). 10.7% individuals tested positive for *G. lamblia*. The global carriage of bacteria was high ranging from 1.2% for *C. jejuni* to 78.6% for EAEC. Of note, 8.3% of all participants were positive for *T. whipplei*. Acquisition of gastrointestinal viruses and parasites was low, while bacterial acquisition ranged from 2.2% for *C. jejuni* to 33.3% for EPEC (Table 4).

Table 2
Prevalence of adherence with preventive measures during the stay in Touba.

Preventives measures		n (N = 110)	%
Influenza vaccination		1	0.9
Face mask	No	104	94.6
	Sometimes	3	2.7
	Often	3	2.7
Hand washing	As usual	59	53.6
	Much more	51	46.4
Disinfectant gel	No	10	9.1
	Sometimes	30	27.3
	Often	70	63.6
Disposable handkerchiefs	No	44	40.0
	Sometimes	30	27.3
	Often	36	32.7

4. Discussion

Religious mass gatherings like the Hajj pilgrimage in Mecca, Saudi Arabia have been associated with outbreaks of gastrointestinal infections, respiratory infections and meningitis [14], mostly due to crowding conditions favoring the inter-human transmission of pathogens. The Grand Magal pilgrimage in Touba Senegal qualifies for the WHO mass gathering definition [15]. This is the first report addressing the morbidity in a cohort of pilgrims at the Grand Magal. Publications about Grand Magal associated diseases are scarce. A cholera outbreak involving 12,135 patients in the Grand Magal area occurred in 2004–2006, with a total of 29,556 cases reported over an 18-month in the country [16]. In 2018, a dengue outbreak was associated to the Grand Magal [17,18]. As described in the introduction, our group provided overall figures of syndromic surveillance data during the 2015 Grand Magal. The most frequent reasons for consultation were trauma, followed by fatigue and heatstroke. Infectious diseases were also prevalent with, notably, a high rate of febrile systemic illnesses and malaria, diarrheal diseases, and respiratory tract infections [1].

Table 3
Prevalence of respiratory pathogens.

Respiratory pathogens	Pre- and/or post-travel N = 110		Acquisition N = 108		Pre-travel N = 110		Post-travel N = 108		P[a] (Post-travel versus pre-travel)
	n	%	n	%	n	%	n	%	
Respiratory virus									
Influenza A	0	0	0	0	0	0	0	0	NA
Influenza B	3	2.7	1	0.9	2	1.8	1	0.9	0.56
Metapneumovirus	0	0	0	0	0	0	0	0	NA
Respiratory syncytial virus	0	0	0	0	0	0	0	0	NA
Human rhinovirus	20	18.2	14	13.0	6	5.5	14	13.0	0.07
Enterovirus	0	0	0	0	0	0	0	0	NA
Adenovirus	8	7.3	5	4.6	3	2.7	6	5.6	0.25
Coronavirus	20	18.2	18	16.7	2	1.8	19	17.6	0.0001
Coronavirus HKU1	0	0	0	0	0	0	0	0	NA
Coronavirus 229E	4	3.6	3	2.8	1	0.9	3	2.8	0.32
Coronavirus NL63	10	9.1	9	8.3	1	0.9	10	9.3	0.003
Coronavirus OC43	9	8.2	9	8.3	0	0	9	8.3	0.003
Parainfluenza virus	0	0	0	0	0	0	0	0	NA
Respiratory bacteria									
Staphylococcus aureus	80	72.7	15	13.9	65	59.1	50	46.3	0.03
Streptococcus pneumoniae	38	34.5	4	3.7	34	30.9	22	20.4	0.01
Haemophilus influenzae	85	77.3	29	26.9	56	50.9	69	63.9	0.03
Klebsiella pneumoniae	66	60.0	7	6.5	59	53.6	30	27.8	< 0.0001
Neisseria meningitidis	0	0	0	0	0	0	0	0	NA
Bordetella pertussis	0	0	0	0	0	0	0	0	NA
Mycoplasma pneumoniae	0	0	0	0	0	0	0	0	NA

[a] *McNemar's test on 108 paired samples.*

In our study, the population was characterized by the young age of participants with 26.4% children and only few elderly pilgrims, in marked contrast with the characteristics of Hajj pilgrims [19]. Indeed, the Grand Magal is a familial event and many adult pilgrims travel with their children. As a consequence of the young age of participants, the prevalence of chronic conditions was low, as opposed to Hajj pilgrims [19–23]. Also, in comparison with the Hajj pilgrimage, where pilgrims stay in Saudi Arabia for around 3 weeks [19], we note that the duration of stay in Touba during the Grand Magal is shorter (3 days) (Table 5).

The vaccination rate against influenza was dramatically low. The use of face masks was rare, while 2/3 of pilgrims declared using hand sanitizer. The overall prevalence of respiratory symptoms was high, affecting 42% participants and a 4.8 fold increase was observed following the participation to the Grand Magal. This result is in line with studies conducted at the Hajj, where 50–93% pilgrims presented respiratory symptoms [24]. Gastrointestinal symptoms were less frequent, affecting 15% individuals, but three times more frequent after participation in the Grand Magal. Overall, these symptoms were relatively mild leading to a low proportion of antibiotic consumption (12.5% ill pilgrims) and no pilgrim was hospitalized. However, 91.3% and 43.8% of pilgrims still had respiratory and gastrointestinal symptoms, respectively when they returned home, which can be a source of infection for their family members. Acquisition of respiratory viruses and *H. influenzae* at the Grand Magal was common, as observed in the setting of the Hajj pilgrimage [24]. We found no evidence for increased transmission of respiratory infections between individuals from a same concession or from a same housing location in Touba which suggests that respiratory infections may have resulted from contacts with the

Table 4
Prevalence of gastrointestinal pathogens.

Gastrointestinal pathogens	Pre- and/or post-travel N = 84		Acquisition N = 45		Pre-travel N = 60		Post-travel N = 69		P* (Post-travel versus pre-travel)
	n	%	n	%	n	%	n	%	
Gastrointestinal virus									
Hepatitis A virus	0	0	0	0	0	0	0	0	NA
Hepatitis C virus	0	0	0	0	0	0	0	0	NA
Adenovirus	8	9.5	2	4.4	6	10.0	3	4.3	0.56
Astrovirus	0	0	0	0	0	0	0	0	NA
Rotavirus	0	0	0	0	0	0	0	0	NA
Norovirus	2	2.4	1	2.2	0	0	2	2.9	0.32
Gastrointestinal bacteria									
Entamoeba histolytica	0	0	0	0	0	0	0	0	NA
Giardia lamblia	9	10.7	1	2.2	8	13.3	2	2.9	0.18
Cryptosporidium spp	0	0	0	0	0	0	0	0	NA
Salmonella	5	6.0	1	2.2	1	1.5	4	5.8	1
Shigella/EIEC	10	11.9	2	4.4	5	8.3	9	13.0	0.16
EHEC	29	34.5	8	17.8	10	16.7	25	36.2	0.13
EPEC	52	61.9	15	33.3	25	41.7	38	55.1	0.22
EAEC	66	78.6	11	24.4	40	66.7	50	72.5	0.13
Campylobacter jejuni	1	1.2	1	2.2	0	0	1	1.5	0.32
Tropheryma whipplei	7	8.3	0	0	6	10.0	6	8.7	1

(*) McNemar's test on 45 paired sample.

Table 5
Main characteristics of the Grand Magal in comparison with the Hajj pilgrimage [1,18–23].

	The Grand Magal pilgrimage	The Hajj pilgrimage
Number of attendees in recent years	4-5 Million/year	2-3 million/year (10 million/year if including the Umrah)
Frequency	Annual	Annual
Geographical location	Touba, Senegal, Africa	Mecca, Mina and Arafat, Saudi Arabia, Middle-East
Duration of the event	1 day (18th of Safar)	5 days (8th to 12th of Dhu al-Hijjah)
Main religious sites visited by attendees for praying and main rituals	Touba Great Mosque, mausoleums, Wall of Mercy spring, Central library, Mouride University. Animal sacrifice.	Mecca Great Mosque (Kaaba circumnanbulation and walking between the hills of Safa and Marwah), Zam-Zam spring, Mount Arafat, Muzdalifah (gathering of pebbles), Mina plain and Jamarat pillars (throwing of pebbles). Animal sacrifice. Shaving head (mens)
Satellite activities of attendees	Visit to personal Marabout, family meeting, cultural activities, hymn-singing sessions, visit to the market	Visit of the Mosque and of the Prophet tomb in Medina
Origin countries of attendees	Mostly Senegal and surrounding countries (Côte-d'Ivoire, Gabon, Guinée-Bissau and Mauritania). Diaspora from France, Belgium, Italy, Spain, Portugal, US and Canada	More than 180 countries in the Middle-East, South-East Asia, Europe, Africa, America and Australia.
Demographics of attendees	Mostly young population with children	Mostly middle aged and elderly population
Comorbidities of attendees	Rare	Frequent (diabetes, cardiovascular and chronic respiratory diseases, walking disability)
International published official health recommendations	No	Yes (pre-travel health regulation and advice about vaccination, health check and specific vaccination at the port of entry, vaccination requirements for issuance of travel visas)
Local media communication and health education campaigns during the event	Yes	Yes
Recent and current common reported public health complications	Last cholera outbreak (2004-2006 – 12135 persons), dengue outbreak (2018), diarrhea, respiratory tract infections, heat stress, road accidents Malaria endemic.	Last cholera outbreak (1989–102 persons), invasive meningococcal disease (1987 and 2000–2001), diarrhea, respiratory tract infections, heat stress, stampedes. Malaria at elimination stage since 2008.
Estimated mortality	0.2/100.000 in 2015	2/100.000 in 2013 among Indonesian pilgrims 1.2/100.000 in 2016 among Indian pilgrims
Housing	Private houses and marabout houses in Touba and surrounding cities and villages	Hotels in Mecca (and Medica), tent camps in Mina
Food	House food and free street food	Restaurants and street vendors
Sanitary conditions (safe water and food provision, waste management; vector control)	Control of quality of drinking water and food items, inspection of main kitchens and food areas, treatment of sewage water. Cleaning and disinfection of areas highly frequented by pilgrims. Disinfection of garbage sites. Insecticide spraying of religious sites and houses of main Marabouts.	Strict microbiological control of quality of drinking water sources and food items, microbiological testing of kitchen workers and kitchen environments, treatment of sewage water. Prohibiting of bringing fresh food or agricultural products from outside the country. Routine and widespread insecticide spraying included aerially.
Number of hospitals and beds available during the event	7 hospitals, 481 beds	25 hospitals, 5000 beds including 500 for critical care
Information technology and surveillance	Paper-based consultation registers	Web-based electronic health surveillance systems for rapid communication
WHO center for mass gathering medicine in the hosting country	No	Yes
Number of papers according to PubMed.gov over the last 5 years (key words = Magal [tiab] and Hajj [tiab]), search done on 25/04/2019)	4	287

overall population in Touba in overcrowding areas, in and around the Mosque rather than from intra-familiar or intra-domiciliar transmission. Our survey has several limitations, including its small sample size and the fact that most pilgrims came from a single village. Also, the short time between inclusion and post-Magal questionnaire administration and testing makes it difficult to establish a formal link between symptoms and pathogen acquisition and pilgrimage participation. Another limitation is that we did not investigate *Mycobacterium tuberculosis* prevalence in our study. However, given the long incubation time of tuberculosis and the short duration of stay of pilgrims in Touba, PCR-based detection of *M. tuberculosis* in relation to the event would not have been feasible. Finally, the proportion of participant who provided paired samples of stool specimen was low. Further study including larger numbers of pilgrims are needed to investigate potential risk factors for respiratory and gastrointestinal infections at the Grand Magal. Nevertheless, this preliminary study confirms that Grand Magal pilgrims are likely to be exposed to communicable disease risk, as observed in other pilgrimage settings, despite the relatively short duration of the event.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.tmaid.2019.04.010.

References

[1] Sokhna C, Mboup BM, Sow PG, Camara G, Dieng M, Sylla M, et al. Communicable and non-communicable disease risks at the Grand Magal of Touba: the largest mass gathering in Senegal. Trav Med Infect Dis 2017;19:56–60https://doi.org/10.1016/j.tmaid.2017.08.005.

[2] Rashid H, Shaf S, El Bashir H, Haworth E, Memish ZA, Ali KA, et al. Influenza and the Hajj: defining influenza-like illness clinically. Int J Infect Dis 2008;12(1):102–3.

https://doi.org/10.1016/j.ijid.2007.03.009. Epub 2007 May 30. PMID: 17540601.

[3] Ninove L, Nougairede A, Gazin C, Thirion L, Delogu I, Zandotti C, Charrel RN, De Lamballerie X. RNA and DNA bacteriophages as molecular diagnosis controls in clinical virology: a comprehensive study of more than 45,000 routine PCR tests. PLoS One 2011;6:e16142https://doi:10.1371/journal.pone.0016142.

[4] Memish ZA, Assiri A, Turkestani A, Yezli S, Al Masri M, Charrel R, et al. Mass gathering and globalization of respiratory pathogens during the 2013 Hajj. Clin Microbiol Infect 2015;21:571. e1-8 https://doi:10.1016/j.cmi.2015.02.008. Epub 2015 Feb 17.

[5] Cohen-Bacrie S, Ninove L, Nougairède A, Charrel R, Richet H, Minodier P, et al. Revolutionizing clinical microbiology laboratory organization in hospitals with in situ point-of-care. PLoS One 2011;6:e22403.https://doi:10.1371/journal.pone.0022403. Epub 2011 Jul 19.

[6] Vuong J, Collard JM, Whaley MJ, Bassira I, Seidou I, Diarra S, et al. Development of real-time PCR methods for the detection of bacterial meningitis pathogens without DNA extraction. PLoS One 2016;11:e0147765https://doi:10.1371/journal.pone.0147765.

[7] Appelt S, Armougom F, Le Bailly M, Robert C, Drancourt M. Polyphasic analysis of a middle ages coprolite microbiota, Belgium. PLoS One 2014;9:e88376.https://doi:10.1371/journal.pone.0088376. eCollection 2014.

[8] Menu E, Mary C, Toga I, Raoult D, Ranque S, Bittar F. Evaluation of two DNA extraction methods for the PCR-based detection of eukaryotic enteric pathogens in fecal samples. BMC Res Notes 2018;11:206https://doi:10.1186/s13104-018-3300-2.

[9] Jiang Y, Fang L, Shi X, Zhang H, Li Y, Lin Y, et al. Simultaneous detection of five enteric viruses associated with gastroenteritis by use of a PCR assay: a single real-time multiplex reaction and its clinical application. J Clin Microbiol 2014;52:1266–8https://doi:10.1128/JCM.00245-14.

[10] Gerber Priscilla F, Xiao Chao-Ting, Cao Dianjun, Meng Xiang-Jin, Opriessnig Tanja. Comparison of real-time reverse transcriptase PCR assays for detection of swine hepatitis E virus in fecal samples. J Clin Microbiol 2014;52:1045–51https://doi:10.1128/JCM.03118-13.

[11] Jothikumar N, Cromeans TL, Sobsey MD, Robertson BH. Development and evaluation of a broadly reactive TaqMan assay for rapid detection of hepatitis a virus. Appl Environ Microbiol 2005;71:3359–63https://doi:10.1128/AEM.71.6.3359-3363.

[12] Van Lint P, De Witte E, De Henau H, De Muynck A, Verstraeten L, Van Herendael B, et al. Evaluation of a real-time multiplex PCR for the simultaneous detection of *Campylobacter jejuni, Salmonella spp.*, *Shigella spp.*/EIEC, and *Yersinia enterocolitica* in fecal samples. Eur J Clin Microbiol Infect Dis 2015;34:535–42.https://doi:10.1007/s10096-014-2257-x. Epub 2014 Oct 19.

[13] Hardegen C, Messler S, Henrich B, Pfeffer K, Würthner J, MacKenzie CR. A set of novel multiplex Taqman real-time PCRs for the detection of diarrhoeagenic *Escherichia coli* and its use in determining the prevalence of EPEC and EAEC in a university hospital. Ann Clin Microbiol Antimicrob 2010;9:5https://doi:10.1186/1476-0711-9-5.

[14] Abubakar I, Gautret P, Brunette GW, Blumberg L, Johnson D, Poumerol G, et al. Global perspectives for prevention of infectious diseases associated with mass gatherings. Lancet Infect Dis 2012;12:66–74https://doi:10.1016/S1473-3099(11)70246-8.

[15] World Health Organization. Public health for mass gatherings: key considerations. Geneva: WHO; 2015http://apps.who.int/iris/bitstream/handle/10665/162109/WHO_HSE_GCR_2015.5_eng.pdf;jsessionid=92D12CF6BBA72B7635B2BC5AB159FB1E?sequence=1, Accessed date: 8 November 2018.

[16] Manga NM, Ndour CT, Diop SA, Dia NM, Ka-Sall R, Diop BM, et al. Cholera in Senegal from 2004 to 2006: lessons learned from successive outbreaks. Med Trop 2008 Dec;68(6):589–92. PMID: 19639824.

[17] Diagne CT, Barry MA, Ba Y, Faye O, Sall AA. Dengue epidemic in Touba, Senegal: implications for the Grand magal pilgrimage for travelers. J Travel Med 2018. https://doi.org/10.1093/jtm/tay123.

[18] Sokhna C, Goumballa N, Gautret P. The Grand Magal of Touba in the time of a dengue outbreak in Senegal. Trav Med Infect Dis 2019https://doi.org/10.1016/j.tmaid.2018.11.002.

[19] Memish ZA, Zumla Alimuddin, Alhakeem Rafat F. Hajj: infectious disease surveillance and control. Lancet 2014;383:2073–82.https://doi:10.1016/S0140-6736(14)60381-0. Epub 2014 May 20.

[20] Pane M, Imari S, Alwi Q, Nyoman Kandun I, Cook AR, Samaan G. Causes of mortality for Indonesian Hajj Pilgrims: comparison between routine death certificate and verbal autopsy findings. PLoS One 2013 Aug 21;8(8):e73243https://doi.org/10.1371/journal.pone.0073243.

[21] Khan ID, Khan SA, Asima B, Hussaini SB, Zakiuddin M, Faisal FA. Morbidity and mortality amongst Indian Hajj pilgrims: a 3-year experience of Indian Hajj medical mission in mass-gathering medicine. J Infect Public Health 2018;11:165–70https://doi.org/10.1016/j.jiph.2017.06.004.

[22] Ahmed QA, Arabi YM, Memish ZA. Health risks at the Hajj. Lancet 2006;367(9515):1008–15. https://doi.org/10.1016/S0140-6736(06)68429-8. PMID:16564364.

[23] Al-Tawfiq JA, Gautret P, Memish ZA. Expected immunizations and health protection for Hajj and Umrah 2018 -An overview. Trav Med Infect Dis 2017;19:2–7.

[24] Hoang VT, Gautret P. Infectious diseases and mass gatherings. Curr Infect Dis Rep 2018 Aug 28;20(11):44. PMID: 30155747 https://doi:10.1007/s11908-018-0650-9.