

## Viral respiratory infections at the Hajj: comparison between UK and Saudi pilgrims

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### ABSTRACT

A high incidence of respiratory infection, including influenza, has been reported at the Hajj in Mecca, Saudi Arabia. Reported rates of influenza have been higher among UK than among domestic pilgrims, but this could be explained by methodological differences among studies. Accordingly, the present study compared the frequencies of respiratory viruses among UK and Saudi pilgrims using the same study design. Pilgrims with upper respiratory tract symptoms were recruited from Mecca and the neighbouring valley Mina during the Hajj 2006. Nasal swabs were used for point-of-care influenza testing and real-time RT-PCR (rtRT-PCR) tests for influenza virus, rhinovirus, parainfluenza virus, adenovirus, human metapneumovirus and respiratory syncytial virus. Of 260 pilgrims investigated, 150 were from the UK and 110 were Saudi; of these, 38 (25%) UK pilgrims and 14 (13%) Saudi pilgrims had respiratory infections detectable by rtRT-PCR ( $p < 0.01$ ). In the UK group, there were 19 (13%) cases of rhinovirus infection, 15 (10%) cases of influenza virus infection, two (1%) cases of dual infections with influenza virus and rhinovirus, one (3%) case of parainfluenza virus infection, and one (1%) case of respiratory syncytial virus infection. Fifty-six (37%) UK pilgrims had been vaccinated against influenza virus, with the rates of influenza in the vaccinated and unvaccinated group being 7% and 14%, respectively ( $p < 0.19$ ). In the Saudi group, there were three (3%) cases of rhinovirus infection and 11 (10%) cases of influenza. Only four (4%) Saudi pilgrims had been vaccinated against influenza virus, and none of these was infected with influenza virus. Overall, a significantly higher proportion of the UK pilgrims had detectable respiratory infections (25% vs. 13%,  $p < 0.01$ ). Influenza rates were similar in both groups, but the reported rates of influenza vaccination differed.

**Keywords** Hajj, influenza, pilgrims, respiratory tract infections, vaccination rates, virus infections

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### INTRODUCTION

The Hajj in Mecca, Saudi Arabia, attracts nearly 1.5 million overseas pilgrims from all parts of the

world. With another 0.5 million pilgrims from the host country, the Hajj is one of the most crowded annual mass gatherings on the planet [1,2]. The focal attraction of the pilgrimage is the symbolic house of God, the *Ka'bah*, and its surrounding mountains, and the Holy Mosque (or Grand Mosque) compound, which confines the pilgrims in a semi-closed setting. As part of the rituals, pilgrims circumambulate the *Ka'bah* for hours in successive episodes, and then, in the later part of

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the festival, they move in a body to Mina, a valley on the outskirts of Mecca, where they stay in tents for several nights, usually in groups of 50–100 individuals in a large tent [2]. Prolonged close contact, physical exertion and overcrowding in a semi-closed setting increase the susceptibility of pilgrims to airborne infections, including meningococcal disease and influenza [2–4].

A UK study, based on paired serum samples collected before and after the Hajj, revealed that 38% of returning UK pilgrims suffered from influenza during the Hajj 2003, while in the same year, a Saudi Arabian study used virus culture of pharyngeal swabs collected at the Hajj to reveal that 6% of symptomatic pilgrims (half were domestic pilgrims) had influenza [5,6]. Subsequent surveillance using PCR to investigate nasal samples from symptomatic UK pilgrims indicated a 14% influenza rate during the Hajj 2005, at a time when overall influenza virus activity was low throughout Europe, including the UK [7].

While these studies have consistently revealed higher rates of influenza among UK pilgrims than among Saudi pilgrims, it is unclear whether the differences could be caused by variations in study design, timing of sample collection, type of samples obtained and laboratory procedures applied. In addition, a range of other viruses, e.g., rhinovirus, parainfluenza virus, adenovirus and respiratory syncytial virus (RSV), can cause infections that frequently mimic influenza. Furthermore, human metapneumovirus (hMPV) has been reported to be an important cause of respiratory infections in children, as well as adults, since its discovery in 2001 [8], but its incidence among the Hajj pilgrims is unknown. In this context, the present investigation used a consistent study design, including sample collection and laboratory methods, to compare the rates of influenza virus, rhinovirus, RSV, hMPV, adenovirus and parainfluenza virus infection among UK and Saudi pilgrims suffering from upper respiratory tract infection (URTI) during the Hajj 2006.

## MATERIALS AND METHODS

### Case definitions

A UK pilgrim was defined as a traveller from the UK who suffered from URTI during the Hajj 2006. A Saudi pilgrim was defined as a native or expatriate resident pilgrim from Saudi Arabia who suffered from URTI during the Hajj 2006.

### Setting

A clinic was established with the British Hajj Delegation (BHD) team at a hotel close to the Holy Mosque in Mecca in January 2006. The BHD team is composed of several UK general practitioners who provide 'walk-in' primary-care services to UK pilgrims. Leaflets containing the address of the BHD clinic were distributed to the pilgrims during pre-travel seminars in the UK. Clinics were also established subsequently in the British block of tents in Mina. Pilgrims suffering from symptoms of URTI, e.g., cough, sore throat, rhinorrhoea and fever, were asked to participate in the study.

The Saudi cohort was recruited from the National Guard Health Affairs clinic, which is a temporary military base hospital that provides free primary-care and emergency-care services in Mina for pilgrims in general, but particularly Saudi (military as well as civilian) pilgrims. Saudi pilgrims who attended the National Guard Health Affairs clinic with symptoms of URTI were recruited to the study.

Invitation letters in English or Arabic, as appropriate, were handed to the pilgrims, and queries concerning the study were answered by multilingual research team members. Demographical details and other clinical and risk-factor information were recorded, including presenting symptoms, past medical, tobacco smoking, contact and vaccination histories, and axillary temperatures. Two nasal swabs were taken from each pilgrim. The first was used for point-of-care testing for influenza virus (QuickVue influenza test; Quidel Corp., San Diego, CA, USA), and the second was immersed in lysis buffer for subsequent transport via a specialist courier service under optimum conditions to the UK for real-time RT-PCR (rtRT-PCR) assays. Nasal swabs were chosen, rather than nasal washings or throat swabs, because they are quicker and more acceptable to pilgrims, thereby allowing more sampling in the limited time available. Individuals who yielded positive results according to the QuickVue test were offered a course of oseltamivir if symptomatic for <48 h. The study was approved by the Multicentre Research Ethics Committee, UK (MREC), ref. No. MREC/02/12.

### Molecular diagnosis

rtRT-PCR assays were performed at the London South Specialist Virology Centre, Health Protection Agency, UK. RNA was extracted in duplicate from 140 µL of each sample using a QIAamp viral RNA mini kit (Qiagen, Hilden, Germany), to yield, in total, 160 µL of purified RNA extract. The RNA extract was then divided into 20-µL aliquots and frozen at –80°C until analysed by rtRT-PCR. One-step rtRT-PCR analysis was performed using the iScript One-Step RT-PCR kit for probes on a Bio-Rad iCycler iQ system real-time thermal cycler (Bio-Rad, Hemel Hempstead, UK). Extracts were tested for rhinovirus and RSV A and B (multiplex 1), parainfluenza virus 1, 2 and 3 (multiplex 2), and hMPV and influenza virus A and B (multiplex 3). The gene targets were nucleocapsid protein (RSV A and B), 5'-UTR (rhinovirus), the HN gene (parainfluenza virus 1, 2 and 3) and the NS gene (influenza virus A and B). Details of primers, probes and RT-PCR conditions have been described previously [9,10].

### Statistical analysis

Data were analysed using SPSS v.14 software (SPSS Inc., Chicago, IL, USA). Chi-square tests were used to compare proportions.

## RESULTS

In total, 260 pilgrims were recruited to the study, of whom 150 (58%) were from the UK, and 110 (42%) were Saudi Arabian residents. Their demographical and other characteristics are summarised in Table 1.

The 150 UK pilgrims were recruited between 2 and 62 (median 16) days after travelling from the UK. Pilgrims came from throughout the UK, but large proportions were from London (18%), Birmingham (8%), Dewsbury (8%) and Batley (6%). Table 2 summarises the ethnicity and age distribution of the pilgrims. Fifty-six (37%) pilgrims had received influenza vaccine

**Table 1.** Demographical characteristics and real-time RT-PCR virus detection results for pilgrims to the Hajj 2006

	UK, n (%)	Saudi, n (%)	p value
Sample size	150	110	
Median age, years	41 (range 14–81)	30 (range 16–85)	
Male:female	138:12	109:1	0.01
Individuals with chronic diseases	30 (20)	12 (11)	0.05
At-risk individuals	38 (25)	15 (14)	0.02
Vaccinated against influenza	56 (37)	4 (4)	<0.001
Smokers of tobacco	16 (11)	30 (27)	0.001
Influenza virus-positive	15 (10)	11 (10)	1.00
Rhinovirus-positive	19 (13)	3 (3)	0.004
RSV-positive	1 (1)	0 (0)	0.39
Parainfluenza virus-positive	1 (1)	0 (0)	0.39
Dual infections (rhinovirus plus influenza virus)	2 (1)	0 (0)	0.22

RSV, respiratory syncytial virus.

**Table 2.** Ethnicity and age distribution of pilgrims with influenza virus and rhinovirus infections

Ethnicity	UK pilgrims			Saudi pilgrims		
	Total	Influenza, n (%)	Rhinovirus, n (%)	Total	Influenza, n (%)	Rhinovirus, n (%)
Pakistani	55	11 (20)	11 (20)	4	1 (25)	0 (0)
Bangladeshi	34	3 (9)	6 (18)	3	1 (33)	0 (0)
Indian	32	3 (9)	2 (6)	1	0 (0)	0 (0)
Saudi	1	0 (0)	0 (0)	68	5 (7)	2 (3)
Sudanese	0	0 (0)	0 (0)	13	2 (15)	0 (0)
Egyptian	0	0 (0)	0 (0)	10	2 (20)	0 (0)
Moroccan	0	0 (0)	0 (0)	7	0 (0)	0 (0)
Others	13	0 (0)	0 (0)	3	0 (0)	1 (33)
Unknown	15	0 (0)	2 (13)	1	0 (0)	0 (0)
Age group, years						
<16	1	0 (0)	0 (0)	1	0 (0)	0 (0)
16–25	13	1 (8)	1 (8)	34	3 (9)	2 (6)
26–35	35	4 (11)	3 (9)	31	4 (13)	0 (0)
36–45	34	6 (18)	7 (21)	26	3 (12)	0 (0)
46–55	36	3 (8)	7 (19)	9	1 (11)	0 (0)
56–65	13	2 (15)	2 (15)	3	0 (0)	0 (0)
>65	11	0 (0)	0 (0)	4	0 (0)	0 (0)
Unknown	7	1 (14)	1 (14)	3	0 (0)	1 (33)

1–3 months before the Hajj, 91 (61%) had definitely not been vaccinated, and three (2%) were not sure of their vaccination status.

Of 110 Saudi pilgrims, 36 (32.7%) came from Mecca and its neighbouring port city Jeddah, and 22 (20%) came from the Saudi capital Riyadh. Their ethnicity and age distribution are summarised in Table 2. Four (4%) pilgrims had received influenza vaccine, 103 (94%) had definitely not been vaccinated, and three (3%) were not sure of their vaccination status.

### Epidemiology

The most common presenting symptoms of the UK pilgrims were sore throat (72%), cough (68%), rhinorrhoea (52%) and fever (41%), with positive predictive values for influenza (not adjusted for dual/non-influenza infections) of 65%, 76%, 53% and 53%, respectively. Forty-three (29%) patients had additional symptoms, e.g., diarrhoea, rash, abdominal pain, myalgia and headache.

Two (1%) UK pilgrims were positive for influenza virus according to the QuickVue test, with both being confirmed by rtRT-PCR. Thirty-eight (25%) UK pilgrims were positive for at least one virus according to rtRT-PCR, i.e., 19 (13%) rhinovirus, 15 (10%) influenza virus, one (1%) RSV A, one (1%) parainfluenza virus type 3, and two (1%) dual infections with influenza virus and rhinovirus. Of the total of 17 (11%) cases of influenza virus infection, 13 were caused by influenza A and four by influenza B.

All except one patient (who was diagnosed with influenza and rhinovirus and presented within 2 days of arrival) presented  $\geq 7$  days after reaching Mecca. The proportion of rhinovirus in pilgrims recruited from Mecca (BHD) was higher than that recruited from Mina (24% vs. 10%,  $p$  0.03). The proportion of influenza virus was also higher among recruits from Mecca (17% vs. 9%), but this difference did not reach significance ( $p$  0.2). Twenty-nine (19%) pilgrims reported contact with individuals with influenza-like illness, of whom three had influenza and two had rhinovirus infection. Sixty-three (42%) pilgrims denied any such contact, six of whom had influenza virus, five rhinovirus, one parainfluenza virus and one RSV A infection. Fifty-eight (39%) pilgrims were unsure whether they had had any such previous contact, six of whom had influenza virus infection, 12 rhinovirus infection

and two dual infections (influenza virus and rhinovirus).

Common presenting symptoms of Saudi pilgrims were sore throat (86%), rhinorrhoea (72%), cough (66%), myalgia (46%) and fever (43%), with positive predictive values for influenza (not adjusted for dual/non-influenza infections) of 81%, 64%, 72%, 72% and 91%, respectively. Nine (8%) pilgrims complained of diarrhoea, of whom one had influenza virus A infection.

Seven (6%) Saudi pilgrims were positive for influenza virus according to the QuickVue test, of whom three were confirmed by rtRT-PCR. Fourteen (13%) patients were positive for at least one virus according to rtRT-PCR, i.e., 11 (10%) influenza virus and three (3%) rhinovirus; no dual infections were detected. Of 11 influenza viruses identified, nine (81%) were influenza A and two (19%) were influenza B.

Eight (7%) Saudi pilgrims reported contact with an individual suffering from influenza-like illness, one of whom had influenza. Ninety-six (87%) pilgrims denied any such contact, ten of whom had influenza and three had rhinovirus infections.

### Risk-group analyses

Thirty-eight (25%) UK pilgrims were considered to be 'at risk' for influenza, 12 because they were aged  $\geq 65$  years, regardless of underlying medical condition, and the others because of pre-existing medical conditions, e.g., diabetes (62%) or lung (23%), heart (12%) and kidney (4%) disease. The rates of influenza vaccination among 'at risk' and 'not at risk' pilgrims were 63% (24/38) and 29% (32/112), respectively ( $p < 0.001$ ), and the corresponding rates of influenza infection were 8% (3/38) and 13% (14/112), respectively ( $p 0.44$ ). Seven pilgrims were known to suffer from bronchial asthma, but none had influenza or rhinovirus infections. Overall, the rates of influenza among vaccinated and unvaccinated UK pilgrims were 7% (4/56) and 14% (13/91), respectively ( $p 0.19$ ). Four (24%) UK pilgrims with confirmed influenza stated that they had received the vaccine 1–2 months before journeying to the Hajj. Sixteen (11%) UK pilgrims were tobacco smokers and 103 (69%) were non-smokers; the proportion of influenza virus and rhinovirus infections among smokers and non-smokers was similar.

Fifteen (14%) Saudi pilgrims were considered to be 'at risk' for influenza, three because they were aged  $\geq 65$  years, and the others because of chronic diseases, e.g., diabetes (33.3%) and heart (33.3%) and lung (33.3%) disease. None of the 'at risk' Saudi pilgrims was vaccinated. The rates of influenza among 'at risk' and 'not at risk' Saudi pilgrims were 13% (2/15) and 9% (9/95), respectively ( $p 0.64$ ). The rates of influenza among unvaccinated and vaccinated Saudi pilgrims were 10% (10/103) and 0% (0/4), respectively ( $p 0.51$ ). Thirty (27%) Saudi pilgrims were tobacco smokers, and 79 (72%) were non-smokers; the rate of influenza virus infection among smokers and non-smokers was similar.

### DISCUSSION

UK pilgrims (except one) with PCR-confirmed infections presented  $>1$  week after reaching the Hajj, thereby indicating that the infections were acquired at the Hajj, since the incubation period for infections caused by influenza virus, rhinovirus, RSV and parainfluenza virus is  $<7$  days. To have been in compliance with Hajj rites, Saudi pilgrims must have joined the congregation at least 2–3 days before they were recruited into the study. Considering the short incubation period for influenza, it is reasonable to suppose that the Saudi pilgrims also acquired the infection at the Hajj.

In this study of a predominantly male population, a significantly higher proportion of infections was detected among UK pilgrims than among their Saudi counterparts (25% vs. 13%,  $p 0.01$ ). In the UK cohort, rhinovirus was the most common virus detected (14%), followed by influenza virus, while influenza virus was the commonest infectious agent among the Saudi cohort, followed by a small proportion of rhinovirus (3%). However, the attack rate of influenza virus was similar (*c.* 10%) in both groups, with influenza A predominating. Thus, it appears that influenza virus was circulating widely at the Hajj, while rhinovirus was circulating selectively among the UK pilgrims, particularly those recruited in Mecca City.

The attack rate of influenza virus among UK pilgrims in the present study was lower (but not significantly so) than in 2005 (11% vs. 14% [7]), possibly because of annual variation, although this may relate to a higher overall vaccine cover-

age, i.e., 28% in 2005 compared with 37% in 2006 ( $p$  0.06). Immunisation uptake was low in both years [11]. The rate of influenza among vaccinated pilgrims was lower than that among unvaccinated pilgrims (7% vs. 14%), but this difference was not statistically significant, which is consistent with findings in previous studies [5,7]. There are two possible reasons for the failure of influenza vaccine to make a significant difference: (i) a major proportion of vaccinated pilgrims were 'at risk' individuals who are naturally more susceptible to influenza virus (and in whom vaccine is less immunogenic) than the unvaccinated group, most of whom were otherwise healthy and hence less vulnerable to infection; and (ii) there was a possible mismatch between the vaccine strains and circulating strains. Thus, during the Hajj 2004 (in March), the commonest circulating strain of influenza virus was influenza B Sichuan [12], but the corresponding vaccine strain was influenza B Hong Kong for both hemispheres [13,14]. Other examples of vaccine failure and mismatch among non-Hajj travellers have been reported [15,16]. It is open to debate whether the same influenza vaccine is fully protective in both hemispheres of the world, particularly in travellers with poor immunity. Therefore, antiviral drugs should be considered as an adjunct to vaccination for post-exposure prophylaxis at the Hajj [17].

More worryingly, despite official Saudi policy, the reported uptake of influenza vaccine among 'at risk' Saudi pilgrims was nil. Only four (4%) Saudi 'not at risk' pilgrims had received the vaccine, with no improvement in uptake since 2003, when Balkhy *et al.* [6] reported 4.4% coverage among 500 international pilgrims, of whom a major fraction were Saudi. A recent survey also revealed a very low uptake (5.9%) of influenza vaccine among Saudi healthcare workers employed at the Hajj [18]. It seems that a vigorous campaign is required to improve influenza vaccine coverage among domestic pilgrims and healthcare workers at the Hajj.

This is the first study to diagnose rhinovirus infection from respiratory specimens taken at the Hajj, although El-Sheikh *et al.* [19] used serological methods to diagnose five (7%) cases of infection with picorna virus (without species categorisation) among 76 serum samples taken at the Hajj in 1991 and 1992, which presumably were rhinoviruses. However, these viruses were not detected in virus culture of the throat sam-

ples. Seasonal variations are the likely explanations for the varying results in these two studies. Scorching summer temperatures during the Hajj in 1991 and 1992 might have resulted in poor detection of the virus in the study by El-Sheikh *et al.* [19], while the lower temperatures during the Hajj 2006 might have resulted in greater virus yields [20]. The frequency of rhinovirus was significantly higher in the Mecca City area than in Mina, indicating that rhinovirus was predominantly circulating selectively in the Grand Mosque area among certain groups of worshippers (e.g., UK pilgrims), who later introduced it to other individuals in their respective blocks of tents in Mina. Unlike influenza virus, rhinovirus spreads mainly via close personal contact [21]. The spread of rhinovirus specifically among the UK pilgrims may have been accelerated by the crowded conditions at the Grand Mosque during circumambulation and other prayers, and in local hotels at least 2 weeks before the domestic pilgrims attended the Hajj. Choudhry *et al.* [22] demonstrated that attendance at a large Mosque (i.e., Namera mosque in Arafat) increases the risk of acquiring URTI among Hajj pilgrims. During the Hajj, the Grand Mosque is more crowded than any other mosque in the world, facilitating the spread of respiratory viruses in Mecca City.

Maintenance of good hand hygiene is crucial in reducing rhinovirus infections, but pilgrims may not accept alcoholic hand rubs and scented soaps/detergents, since alcohol is generally forbidden in Islam and scented substances are avoided during the Hajj [23]. Viricidal hand treatments containing non-alcoholic organic acids, which have shown excellent results in preventing rhinovirus transmission among volunteers, may be acceptable once they become widely available [24].

Detection rates for RSV (1%) and parainfluenza virus (1%) were low compared to previous studies [6,7,19], and adenovirus and hMPV were not detected. Other important viruses that could cause influenza-like illness during the Hajj are coronavirus, enterovirus and human bocavirus (a newly identified parvovirus) [25,26]. To date, the highest incidence of the latter virus (18.3%) has been reported in Jordan [27], a neighbour of Saudi Arabia, from which thousands of pilgrims attend the Hajj. Continuing surveillance of respiratory infections at the Hajj should include these novel viruses and H5N1 influenza virus, to

protect not only the pilgrims, but also the global community. Practical issues dictated the descriptive nature of the present study, and some patient characteristics differed significantly between the two groups, despite the use of an identical cross-sectional study design (Table 1). Future studies will consider the advantages of matching cases as much as possible before comparing infection rates.

In summary, both UK and Saudi pilgrims at the Hajj are at considerable risk of influenza virus infection. In addition, UK pilgrims develop other viral infections. Preventive strategies, e.g., vaccination and respiratory hygiene, need to be enhanced, and antiviral influenza prophylaxis should be considered. Ongoing surveillance of influenza and newer respiratory viruses is needed to better understand the epidemiology of respiratory infections at the Hajj.

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