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## Detection of COVID-19 incidence among attendees of the 2022 Hajj season

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### ABSTRACT

**Objectives:** This study investigated the presence of COVID-19 cases among pilgrims, health care workers (HCWs), and non-HCWs of Hajj 2022.

**Methods:** Nasopharyngeal samples were collected from 491 attendees of Hajj 2022. All participants received three doses of COVID-19 vaccines. Paired (n = 138; 69 participants) and unpaired (n = 422; 422 participants) nasopharyngeal swabs were subjected to reverse transcription–polymerase chain reaction targeting E gene of  $\beta$ -coronaviruses and RNA-dependent RNA polymerase of SARS-CoV-2. The results were linked to the participants' profiles, including role during Hajj, presence of respiratory symptoms or comorbidities, contact with symptomatic individuals, smoking status, and COVID-19 recovery.

**Results:** A total of 20 (20 of 560; 3.6%) samples tested positive for COVID-19. Most cases (18 of 20; 90%) were pilgrims and non-HCWs. Six (30%) samples belonged to participants with previous positive reverse transcription–polymerase chain reaction. A total of 12 (60%) samples belonged to participants with respiratory symptoms. Three (15%) cases were linked to participants who had contact with individuals with respiratory symptoms. All cases belonged to individuals with no comorbidities, apart from a single case who has a chronic sinusitis. Five (25%) cases were smokers. No significant association was found between positive COVID-19 test and participants' profiles.

**Conclusions:** Few COVID-19 cases were detected in this study. Sustainable surveillance of COVID-19 and other respiratory viruses during Hajj seasons remains necessary.

### Introduction

Respiratory viral infections are among the global major concerns due to their rapid transmission through air droplets, aerosols, and close contact with infected individuals [1–3]. The nature of mass gathering (MG) events, whether planned and spontaneous, can worsen the problem [1–3]. MG events lead to accumulation of people at a specific location for a period. The purpose for these gathering events can be planned, such as religious, sporting, or social occasions, or it can be unplanned and spontaneous (e.g. natural disasters) [1,2]. Several religious MG events take place on a regular basis in certain countries. Visitors of these events, however, arrive from all over the world. For instance, the Purna Kumbh

Mela “full Kumbh”), which is the Hindu pilgrimage, is held every 12 years in India. With almost 120 million visitors, the Kumbh Mela held in 2013 is considered the largest religious gathering in the world. The Ardh Kumbh Mela (“half Kumbh”) is held approximately every 6 years. The last Ardh Kumbh Mela was held in 2021 during the COVID-19 pandemic [4–6]. Some reports provided evidence that links this religious event to the massive spread of the B.1.617.2 (Delta) variant of the SARS-CoV-2 virus, which, consequently, severely challenged the Indian health care system [4–6]. Hajj, the Islam pilgrimage, is another planned religious event that requires multi-sectoral coordination and organization [1,2]. It occurs annually and usually gathers more than 2 million domestic and international pilgrims in Makkah and other holy places (e.g. Mina, Muz-

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dalifah, and Arafat) in Saudi Arabia. Due to the nature of Hajj rituals, this MG religious event provides a favorable environment for transmission and recombination of respiratory pathogens [1,2]. Indeed, several reports demonstrated respiratory symptoms as the most common health complaint among pilgrims during Hajj seasons [1,2]. This is particularly true during Hajj seasons held before COVID-19 pandemic (2019 Hajj season or earlier). In many cases, symptomatic pilgrims sought medical care and molecular laboratory investigations of their samples revealed a wide variety of circulating respiratory viral infections [1,7–11]. These include human rhinovirus, influenzas A and B viruses, respiratory syncytial virus, parainfluenza viruses, and human coronaviruses (other than severe acute respiratory syndrome coronavirus and Middle East respiratory syndrome coronavirus). Single infection and co-infection with up to four viruses have been detected [7,12]. Moreover, phylogenetic analysis demonstrated high genetic diversity and co-circulation of many lineages of some respiratory viruses (e.g. human rhinovirus and influenzas A virus) that were originated from different geographic regions [9,13].

Three Hajj seasons (2020–2022) were held since the emergence of SARS-CoV-2. Exceptional protective measures and crisis management plans were applied in these seasons [1,14–16]. For instances, the number of pilgrims allowed was substantially lowered compared with previous years (approximately 2 million pilgrims). Only 10,000 healthy domestic pilgrims, including citizens and residents ranging in age between 18 and 65 years, performed Hajj in 2020 [15]. All pilgrims had to provide a proof of negative COVID-19 polymerase chain reaction (PCR) within 72 hours before arrival to Makkah. Furthermore, all pilgrims were subjected to a mandatory quarantine for 14 days before and after performing the rituals. Strict COVID-19 protective measures (e.g. wearing facial masks and maintaining social distancing) were also implemented. No cases of COVID-19 were reported in this Hajj season (2020) [15]. The number of pilgrims was increased to 58,745 domestic pilgrims in 2021 [17]. With the availability of several COVID-19 vaccines worldwide before this Hajj season, all pilgrims required a proof of receiving COVID-19 vaccinations [14]. Providing negative COVID-19 PCR tests were also required from all pilgrims within 72 hours of commencing Hajj. COVID-19 personal protective measures were also enforced during every stage of the rituals. A total of 41 COVID-19 cases were reported among samples collected before, during, and after the Hajj 2021 season [14]. In Hajj 2022, the number of pilgrims was 899,353, including 119,434 domestic and 779,919 international pilgrims [18]. In addition, 228,721 workers, including health care workers (HCWs) and non-HCWs, attended this Hajj season [18]. With the availability of several COVID-19 vaccines worldwide, proof of vaccination was required from all pilgrims, whereas providing negative PCR test was not necessary. More than 150 health care centers equipped with medical beds and ambulances, isolation rooms, and emergency medical teams were made available for all pilgrims around the clock [18].

To the best of our knowledge, as of now (more than 2 years after the 2022 Hajj concluded), there is a lack of studies addressing the pattern of respiratory and non-respiratory microbial infections in the 2022 Hajj season. Herein, we investigated the presence of SARS-CoV-2 nucleic acid in nasopharyngeal swabs collected from pilgrims, HCWs, and non-HCWs who attended Hajj 2022. Our laboratory findings were linked to the demographic data, medical statuses, and COVID-19 disease and vaccine profiles of the study population.

## Methods

### Ethical statement

This research was approved by the biomedical research ethics committee at Umm Al-Qura University (No. HAPO-02-K-012-2022-06-1142; Data 30-06-2022). Participation in this project was entirely voluntary. Written consent was obtained from all participants.

### Study design and population

We recruited 491 pilgrims, HCWs, and non-HCWs who attended the 2022 Hajj season. All participants completed questionnaires on demographic data, medical statuses, and COVID-19 vaccination profile. Nasopharyngeal swabs were collected from participants and stored at  $-80^{\circ}\text{C}$  until further processing. The study population comprises 69 participants who agreed to provide paired samples ( $n = 138$ ). The first samples were collected upon arrival to the holy places before the beginning of Hajj, and the second samples were collected after the end of the rituals. A total of 422 participants provided single unpaired samples during any day of the rituals.

### Molecular detection of SARS-CoV-2

Viral nucleic acid was extracted from the nasopharyngeal swabs using the ExiPrep 96 Viral DNA/RNA kit (BiONEER, Seoul, South Korea) on the automatic extractor BiONEER, following the manufacturer's instructions. Detection of SARS-CoV-2 was performed by the PowerChek 2019-nCov Real-Time PCR Kit (Seoul, Korea) which, as per the World Health Organization's recommendations, amplifies two target genes separately: E gene of  $\beta$ -coronaviruses and RNA-dependent RNA polymerase of SARS-CoV-2.

### Statistical analysis and data curation

Data were analyzed using GraphPad prism 9.4.1. COVID-19 cases were calculated as the proportion of samples with positive reverse transcription-PCR (RT-PCR) results relative to the total number of samples. The results were presented as actual numbers and percentages. Multivariate logistic regression was used to identify the association between positive COVID-19 acquisition and the characteristics of participants. Adjusted odds ratios with 95% confidence upper and lower limit estimated the magnitude of the association between COVID-19 acquisition and characteristics of participants.  $P \leq 0.05$  was considered statistically significant.

## Results

Nasopharyngeal swabs ( $n = 560$ ) collected from pilgrims, HCWs, and non-HCWs during the 2022 Hajj season were screened for the presence of SARS-CoV-2 RNA using commercially available Food and Drug Administration-approved RT-PCR kit. Two target genes E gene of  $\beta$ -coronaviruses (screening) and RNA-dependent RNA polymerase of SARS-CoV-2 (confirmatory) were considered. Samples that were tested positive for both genes were considered positive. The results of RT-PCR were linked to data obtained from the study participants who were required to complete questionnaires about the purpose of attending the Hajj, current health status, presence of respiratory symptoms, contact with symptomatic individuals, smoking habits, nationality, and awareness of previous COVID-19 diagnosis. The number of pilgrims were 290 and each one of them provided a single nasopharyngeal swab during any day of the Hajj season. The remaining 270 samples were collected from HCWs and non-HCWs. Some of these samples ( $n = 138$ ) were collected from 69 participants as paired samples. The first samples were collected upon arrivals to the holy places, and the second samples were collected after the Hajj rituals were concluded. The study population included Saudi and non-Saudi citizens, smokers and non-smokers, individuals with and without comorbidities, individuals with and without respiratory symptoms, and individuals with and without contact with symptomatic individuals. With regards to COVID-19 profile, all participants received three doses of COVID-19 vaccines, with a subset of them previously having positive COVID-19 RT-PCR results. Table 1 shows the detailed characteristics of study population.

Among the 560 samples collected, 20 samples tested positive, with an overall rate of 3.6%. The majority of cases (90%; 18 of 20) were

**Table 1**  
Baseline characteristics for the study population.

Study population		Overall (n = 560; 491 participants)		Non-paired samples (n = 422; 422 participants)		Paired samples (n = 138; 69 participants)	
		n	%	n	%	n	%
Type of participant	<b>Pilgrims</b>	290	51.8	290	68.7	0	0.0
	<b>HCW</b>	98	17.5	50	11.8	n = 48; 24 participants	34.8
	<b>Non-HCW</b>	172	30.7	82	19.4	n = 90; 45 participants	65.2
Country of residence	<b>Saudi</b>	200	35.7	146	34.6	n = 54; 27 participants	39.1
	<b>Non-Saudi</b>	360	64.3	276	65.4	n = 84; 42 participants	60.9
Comorbidities	<b>Yes</b>	60	10.7	60	14.2	0	0.0
	<b>No</b>	500	89.3	362	85.8	n = 138; 69 participants	100.0
Previous COVID-19 reverse transcription–polymerase chain reaction	<b>Yes</b>	184	32.9	138	32.7	n = 46; 23 participants	33.3
	<b>No</b>	376	67.1	284	67.3	n = 92; 46 participants	66.7
Contact with individuals with respiratory symptoms	<b>Yes</b>	109	10.5	99	23.5	n = 10; seven participants <sup>a</sup>	7.2
	<b>No</b>	451	89.5	323	76.5	n = 128; 62 participants <sup>a</sup>	92.8
Respiratory symptoms	<b>Yes</b>	246	43.9	235	55.7	n = 11; 10 participants <sup>b</sup>	8.0
	<b>No</b>	314	56.1	187	44.3	n = 127; 59 participants <sup>b</sup>	92.0
Smokers	<b>Yes</b>	120	21.4	102	24.2	n = 18; nine participants	13.0
	<b>No</b>	440	78.6	320	75.8	n = 120; 60 participants	87.0

HCW, health care worker.

<sup>a</sup> Some participants have been exposed to individuals with respiratory symptoms before the collection of first samples, whereas others were exposed afterward.

<sup>b</sup> With exception of a single participant, all experienced respiratory symptoms at the collection of either the first or second sample.

**Table 2**  
Results related to COVID-19 reverse transcription–polymerase chain reaction test.

Study population		Overall (n = 20 of 560; 3.6%)		Non-paired samples (n = 16 of 422; 3.8%)		Paired samples (n = four of 138; 2.9%) <sup>a</sup>	
		n	%	n	%	n	%
Type of participant	<b>Pilgrims</b>	9	45	9	56.25	0	0
	<b>HCW</b>	2	10	2	12.50	0	0
	<b>Non-HCW</b>	9	45	5	31.25	4	100
Country of residence	<b>Saudi</b>	8	40	4	25	4	100
	<b>Non-Saudi</b>	12	60	12	75	0	0
Comorbidities	<b>Yes</b>	1	5	1	6.25	0	0
	<b>No</b>	19	95	15	93.75	4	100
Previous COVID-19 reverse transcription–polymerase chain reaction	<b>Yes</b>	6	30	2	12.50	4	100
	<b>No</b>	14	70	14	87.50	0	0
Contact with individuals with respiratory symptoms	<b>Yes</b>	3	15	3	18.75	0	0
	<b>No</b>	17	85	13	81.25	4	100
Respiratory symptoms	<b>Yes</b>	12	60	12	75	0	0
	<b>No</b>	8	40	4	25	4	100
Smokers	<b>Yes</b>	5	25	5	31.25	0	0
	<b>No</b>	15	75	11	68.75	4	100

HCW, health care worker.

<sup>a</sup> The four samples belonged to four participants. Therefore, only one of the paired samples tested positive. None of the participants had both samples positive for COVID-19.

pilgrims and non-HCWs. Cases belonged to Saudi residents comprised 40% (eight of 20) of the total number of cases. Although 14 (70%) samples never had a positive RT-PCR result, six (30%) samples were likely cases of re-infection. Twelve (60%) samples belonged to participants who were experiencing respiratory symptoms, such as coughing, rhinitis, nasal congestion, and fever, at the time of sample collection. Three (15%) cases belonged to participants who had contact with individuals with respiratory symptoms. A single case of COVID-19 belonged to a participant with chronic sinusitis. All other cases were individuals with no known comorbidities. Fifteen (75%) cases were obtained from non-smokers. The analysis of paired samples revealed four samples with positive COVID-19. These four samples belonged to four participants. None of the participants had both paired samples positive for COVID-19 (Table 2). Interestingly, the purpose of attending the Hajj season, the presence of respiratory symptoms and other comorbidities, being in contact with symptomatic individuals, history of previous COVID-19 infection, nationality, and smoking did not seem to increase the risk of acquiring the infection (Table 3).

## Discussion

At the global level, the COVID-19 pandemic had major socio-economic and health impacts. As an infectious control strategy, many countries decided to cancel or minimize social, sporting, and religious events. Saudi Arabia was among those countries that implemented proactive plans (e.g. suspension of flights, activation of distance learning, and imposing curfew) even before detecting the first COVID-19 case in the country on March 2, 2020 [19]. One of the main historical events in Saudi Arabia was, indeed, the suspension of Umrah (minor Islam pilgrimage) and closure of the holy mosque on March 5, 2020 [20]. This was followed by substantially minimizing the number of pilgrims conducting the 2020 Hajj (major Islam pilgrimage) which occurred in July and August 2020 [16]. Owing to the introduction of COVID-19 vaccination, the number of pilgrims was subsequently increased in the next two seasons (2021 and 2022) [17,18]. Unlike the 2020 and 2021 Hajj, the local pilgrims and workers of the 2022 season were not obligated to provide negative PCR results for COVID-19. Thus, there

**Table 3**  
Association between participant characteristics and testing positive for COVID-19.

Variable (yes/no)	COVID-19 reverse transcription–polymerase chain reaction results		Coefficient	Odds ratio	95% confidence interval Lower	95% confidence interval Upper	P-value
	Positive	Negative					
Attended Hajj as a health care worker	2/18	96/444	−1.212	0.297	−2.913	0.488	0.162
Participant is Saudi	8/12	192/348	0.727	2.068	−0.309	1.762	0.169
History of chronic disease	1/19	59/481	−1.001	0.367	−3.071	1.068	0.343
Participants had previous positive COVID-19 reverse transcription–polymerase chain reaction	6/14	178/362	−0.07	0.933	−1.078	0.938	0.892
Contact with individual who has respiratory symptoms	3/17	243/297	−0.264	0.768	−1.6	1.071	0.698
Participants have respiratory symptoms	12/8	234/306	0.083	1.087	−0.943	1.11	0.874
Participants are smokers	5/15	115/425	0.24	1.272	−0.818	1.299	0.656

is a lack of information about COVID-19 cases during the 2022 Hajj season.

Herein, we tested 560 nasopharyngeal swabs collected from 491 pilgrims, HCWs, and non-HCWs who attended Hajj 2022. The prevalence rate was 3.6% (20 of 560), with no significant association identified between participant characteristics and testing positive for COVID-19. The majority of cases (90%; 18 of 20) were pilgrims and non-HCWs. Although one may expect that HCWs are more likely to get exposed to infections, the awareness of HCWs and the adherence to personal protective equipment might explain this finding. In fact, even at early stages of the COVID-19 pandemic and before the introduction of vaccines, many studies demonstrated a low sero-prevalence rate of SARS-CoV-2 antibodies among HCWs as an indication of minimal exposure to the virus [21,22].

Approximately one-third of cases (30%; six of 20) were due to re-infection, as indicated by the presence of previous positive RT-PCR. This is not uncommon given that COVID-19 vaccine breakthrough infections have been repeatedly reported [23–25]. Of note, we cannot comprehensively conclude the rate of re-infection because some of the participants may have had an asymptomatic infection or even a symptomatic infection that had not been properly diagnosed before.

Of the 20 COVID-19 cases, eight (40%) cases did not experience respiratory symptoms. Vaccine-induced neutralizing immunity likely played a role in this finding, although asymptomatic (silent) COVID-19 cases are also well-documented possibly as a result of host innate immunity [21,22,26,27]. The remaining 12 cases exhibited respiratory symptoms including sore throat, cough, rhinorrhea or rhinitis, nasal congestion, and fever. Cases of co-infection, with up to four respiratory viruses, during the Hajj seasons have been previously reported [7,28]. Our current data cannot determine whether the presence of respiratory symptoms is due to a solo COVID-19 infection or due to simultaneous infection with multiple respiratory pathogens. Another possibility that may explain the presence of symptoms in the infected vaccinated individuals is the lack of efficient vaccine-induced neutralizing immunity. Indeed, some vaccine effectiveness studies demonstrated association between some health conditions (e.g. obesity and rheumatic diseases) and the lack of protective immunity upon vaccination [23,25,29]. The mechanisms underlying these findings can be failure to mount neutralizing antibodies or rapid waning of these antibodies [23,25,29]. Serum samples were not available and, hence, assessing the presence and activity of anti-SARS-CoV-2 antibodies by immunoassays (e.g. enzyme linked immunosorbent assay, clinical laboratory improvement amendments, and micro-neutralization assay) could not be performed.

In addition, it is important to note that RT-PCR cannot differentiate between the presence of remnant viral nucleic acids and replication-competent viruses [30]. Therefore, our data cannot conclude whether these cases are new cases or due to remaining viral RNA from previous infections. Moreover, our data cannot determine the contagiousness

status of the detected COVID-19 cases. Viral isolation, which was not available to us at the time of study, would be necessary to determine SARS-CoV-2 infectivity. In addition, it would have been informative to sequence the positive samples and identify SARS-CoV-2 variants. A high genetic diversity of circulating respiratory viruses (e.g. rhinovirus and influenza viruses) among pilgrims during Hajj seasons has been reported in previous works [9,13]. Indeed, MG events in general offers a favorable environment for viral transmission and recombination.

## Conclusion

It is now, as of May 2024, almost 20 months since the conclusions of Hajj 2022. Hajj-related COVID-19 outbreaks were neither reported during the seasons nor after it. Indeed, the success of Hajj organization during COVID-19 era was evident, particularly, with regards to infection control and disease management. In this study, few COVID-19 cases were detected by molecular assays. They could be cases of active infections or remnant viral RNA from previous infections.

## Declarations of competing interest

The authors have no competing interests to declare.

## Recommendations

To the best of our knowledge, this is the first study that investigated COVID-19 among the 2022 Hajj attendees including pilgrims and non-pilgrims. Sustainable surveillance and continuous investigations on the pattern of circulating respiratory viruses during the past and upcoming Hajj seasons always remain of great importance.

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## Ethical approval

This research was approved by the biomedical research ethics committee at Umm Al-Qura University (No. HAPO-02-K-012-2022-06-1142; Data 30-06-2022).

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## Consent

Participation in this project was entirely voluntary. Written consent was obtained from all participants.

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