# Low prevalence of community-acquired influenza coinfections among COVID-19 patients in Al-Madinah, Saudi Arabia: A retrospective cohort study

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## ARTICLE INFO

## ABSTRACT

*Background:* Coinfections with respiratory viruses among SARS CoV-2 patients have been reported by several studies during the current COVID-19 pandemic. Most of these studies designated these coinfections as being hospital-acquired infections; however, there is inadequate knowledge about community-acquired respiratory coinfections among SARS CoV-2 patients.

*Methods:* In this retrospective cohort study, we investigated the seroprevalence of influenza A, influenza B, and parainfluenza-2 among newly hospitalized patients with confirmed COVID-19 infections (n = 163). The study was conducted during the early phase of the COVID-19 pandemic in Saudi Arabia (from April to October 2020). The patients' serum samples were subjected to commercial immunoglobulin M (IgM) antibody tests against the three aforementioned viruses.

*Results:* Seropositivity for influenza A and B and parainfluenza-2 occurred only in 4.2% (7/163) of COVID-19 patients, indicating simultaneous acute infections of these three viruses with SARS CoV-2 infection. All coinfection cases were mild and misdiagnosed during the care period in the hospital.

*Conclusion:* This study highlights the low prevalence of community-acquired respiratory infections among COVID-19 patients in the current pandemic and we discussed the possible factors for this finding. During newly emerging epidemics or pandemics, considering other respiratory viruses circulating in the community is essential to avoid their misdiagnosis and account for their possible negative effects on pandemic disease management and prognosis.

## 1. Introduction

The severe acute respiratory syndrome coronavirus 2 (SARS CoV-2) is a newly emerged, enveloped RNA virus that belongs to the β-coronavirus family [1]. The virus exhibits the ability to be transmitted via different modes such as direct/indirect contact as well as droplet and airborne routes, making it highly contagious [2]. SARS CoV-2 can cause illnesses ranging from mild respiratory infection to severe pneumonia and acute respiratory distress syndrome (ARDS). Fever, dry cough, and fatigue are the most common symptoms, while loss of taste or smell, headache, diarrhea, nasal congestion, and red eyes are less common [3–5].

SARS CoV-2 has become a significant challenge and concern for global public health since its emergence in Wuhan, China, in December 2019 [6]. The first case of COVID-19 infection in Saudi Arabia was reported on March 2, 2020. Since then, the number of cases has exceeded 632,736, with 8912 deaths as of January 19, 2022 [7].

Beyond SARS-CoV-2 pathogenesis, the incidence of infections of other respiratory viruses along with COVID-19 has been observed in different studies worldwide. The impact of viral coinfection on COVID-19 patients is not fully understood but respiratory viruses, in general, can damage the airway epithelium, decrease mucociliary clearance, and trigger immune system disorders [8–10]. Therefore, coinfections have clinical significance, being related to a prolonged hospital stay with a higher required level of care and impacting the disease prognosis by possibly leading to ARDS and other complications [11].

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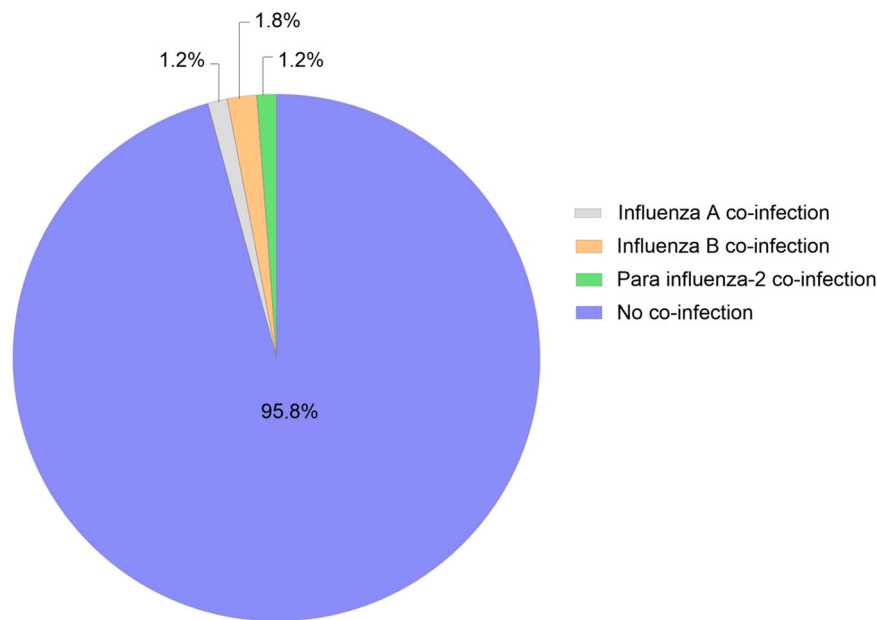*E-mail address:* shoufie@taibahu.edu.sa (S.T. Alhoufie).

**Fig. 1.** The prevalence of community-acquired influenza virus's coinfections amongst 163 SARS-CoV-2 patients.

Coinfections of circulating respiratory viruses have also been reported in previous respiratory virus outbreaks. For instance, during the Middle East respiratory syndrome coronavirus (MERS-CoV) epidemic in Saudi Arabia in 2017, Alfaraj and colleagues reported influenza virus coinfections among MERS-CoV patients [12]. During the current COVID-19 pandemic, several studies have reported coinfections of respiratory viruses among COVID-19 patients. Early observations (February 2020) by Wang and colleagues in Wuhan, China, revealed that coinfections occurred in 5.8% of COVID-19 patients and that the median age of the patients was 51 years [13]. Another study in the United States found that 20.7% of COVID-19-positive patients were coinfected with one or more pathogens, the most common of which included respiratory viruses such as non-SARS-CoV-2 Coronaviridae, respiratory syncytial virus, enteroviruses, and rhinovirus [14].

Several studies have detected coinfections of influenza viruses among COVID-19 patients with mild to severe infections. For instance, in the early COVID-19 pandemic in Spain, Elena and colleagues reported influenza A and B coinfections among critically ill COVID-19 patients [15]. On the other hand, studies in China and Turkey reported influenza virus coinfections among patients with COVID-19 of mild to moderate severity who recovered without complications [16,17].

Despite the increasing number of reports on influenza virus coinfections among COVID-19 patients, there is limited knowledge about whether these coinfections are community- or hospital-acquired. Therefore, we performed this retrospective cohort study to investigate the prevalence of community-acquired influenza virus coinfections among COVID-19 patients in Al-Madinah, Saudi Arabia.

## 2. Methodology

### 2.1. Study design and sample collection

This was a single-center, retrospective cohort study. A total of 163 serum samples were collected from randomly selected confirmed SARS-CoV-2 patients during their visit to the emergency department at King Salman Medical City in Al-Madinah, Saudi Arabia, during the early stage of the pandemic from April to October 2020. Demographic data, including age, gender, and symptoms at the time of hospital admission, were extracted from the patients' medical records. Serum samples were collected from the hospital's clinical laboratory and aliquoted, then stored promptly at − 20 °C for retrospective investigation. Ethical approval was obtained from the institutional review board of the General Directorate of Health Affairs in Al-Madinah (approval no. 9–2021).

### 2.2. Serological detection of influenza virus coinfections

Immunoglobulin M (IgM) antibodies against influenza A, influenza B, and parainfluenza-2 were screened for via indirect ELISA using Vircell IgM ELISA kits (Vircell, Granada, Spain) according to the manufacturer's instructions. All sample wells were treated with IgG sorbent to avoid interference from rheumatoid factor and IgG antibodies. Sera and controls provided in the kits were incubated in microplates for 45 min at 37 °C. Then, wells were washed and incubated with the IgM conjugate for 30 min at 37 °C. After the second round of washing, tetramethylbenzidine (TMB) substrate was added and the plates were incubated for 20 min at room temperature, protected from light. Thereafter, the stop solution was added immediately to all wells, and the absorbance was measured at 450 nm. The antibody index values were calculated as the sample optical density value divided by the mean of the optical density values of the cutoff controls, multiplied by 10. An antibody index of > 11 was considered a positive result.

## 3. Results

Of the 163 randomly selected confirmed SARS-CoV-2 patients in this study, approximately 4.2% showed seropositivity for influenza A and B and parainfluenza-2 (Fig. 1). IgM seropositivity for influenza A occurred in two patients (50-year-old man and 31-year-old woman). Three patients (two women and one man) showed influenza B seropositivity, all of whom were over 60 years old. Parainfluenza-2 seropositivity occurred in two male patients with age ≤ 40 years. The patients included 76 (55.5%) men and 61 women (44.5%) aged 22–95 years, with an average age of 50 years. As shown in Table 1, 76.6% of patients presented with fever, 34.3% with dyspnea, and 46% with cough. In addition, all patients recovered within a few days of admission without complications and were discharged from the hospital according to their medical records.

**Table 1**
Patient's demographic data and diagnostic criteria.

| | Number | Percentage |
|---|---|---|
| 1- Gender distribution | | |
| Male | 89 | 54.6% |
| Female | 74 | 45.3% |
| Total | 163 | |
| 2- Age groups | | |
| 20–39 | 47 | 28.8% |
| 40–59 | 67 | 41.1% |
| ≥ 60 | 49 | 30% |
| Total | 163 | |
| 3- Clinical signs | | |
| Fever ≥ 37.8 °C | 123 | 75.5% |
| Dyspnea | 47 | 28.8% |
| Cough | 62 | 38% |
| 4- Positive IgM detection | | |
| influenza A | 2 | 1.2% |
| influenza B | 3 | 1.8% |
| parainfluenza 2 | 2 | 1.2% |
| Total | 7 | 4.2 |

## 4. Discussion

In this work, we conducted a retrospective cohort study to investigate the prevalence of community-acquired influenza infections (influenza A and B and parainfluenza-2) among randomly selected SARS-CoV-2-positive patients examined at a single hospital that was designated by the health authority of the Al-Madinah region to provide medical care for SARS-CoV-2 patients during the early phase of the pandemic in Saudi Arabia from April 1 to October 31, 2020.

Among the cohort of SARS-CoV-2 patients observed in this study, retrospective detection of influenza viruses revealed IgM seropositivity for influenza A in two patients, influenza B in three patients, and parainfluenza-2 in two patients, indicating their incidence in only 4.2% (7/163) of SARS-CoV-2 patients (Fig. 1). In addition, all patients recovered within a few days of hospital admission without complications and were discharged according to their medical records. The low prevalence and mild coinfections of influenza viruses noted here are in agreement with previous reports that observed influenza virus coinfections among SARS-CoV-2 patients in China and Turkey [16,17]. Conversely, a recent study in Saudi Arabia reported influenza virus coinfections among SARS-CoV-2 patients in an intensive care unit; however, it is unclear whether the patients' deterioration was due to the influenza coinfection or COVID-19 [18]. Determination of the etiological agent in such cases is still a challenging point because of the similarities in clinical manifestations, radiography, and laboratory results between influenza viruses and SARS-CoV-2 infections.

It is worth noting that a remarkable decrease in the rate of influenza infections has been globally reported during the current COVID-19 pandemic. For instance, in New Zealand, after the implementation of a nationwide lockdown before the influenza season, multiple surveillance systems revealed no confirmed laboratory detections of influenza outbreaks throughout 2020 winter; additionally, a significant reduction in other respiratory infections including those caused by adenovirus, enterovirus, rhinovirus, respiratory syncytial virus, parainfluenza virus types 1–3, and human metapneumovirus was noted post-lockdown compared with the corresponding period in 2019 [19]. Moreover, a study in Japan comparing the incidence rate of influenza infections during the 2019/2020 season with the previous four seasons revealed significant reductions in the influenza infection rate during the COVID-19 pandemic [20].

The reduction in influenza incidence rates can be attributed to the implementation of COVID-19 control measures such as hand washing / rubbing, wearing masks, social distancing, lockdowns, and travel bans in most countries during the current pandemic [21].

COVID-19 and influenza viruses have comparable transmission routes, including transmission via airborne droplets and direct contact (human-to-human transmission) [22], which might be affected by these control measures and lead to a decline in transmission rates and infections in the community.

Despite the similarities in the transmission routes of COVID-19 and influenza viruses and the strict control measures in New Zealand, Japan, and other countries, COVID-19 incidence did not decline as much as influenza virus incidence, and cases continue to be identified [19,20]. The differences in the duration of survival, asymptomatic transmission, and transmission dynamics of these viruses may play a role in this regard. For instance, data from many influenza seasons revealed a median primary reproduction number ($R_0$) of 1.28 [23]. In contrast, recent work conducted in the United States and some European countries found a COVID-19 $R_0$ ranging from 3.6 to 6.1 [24] that might explain the continued incidence of COVID-19 compared with influenza viruses despite the implementation of control measures.

Another speculative reason for the reduced rate of influenza virus infections during the pandemic may involve virus-virus interactions. For example, during the H1N1 pandemic in 2009, studies in Sweden and France suggested that rhinovirus circulation in the community might reduce or delay emerging influenza virus activity [25,26]. Other observations found that the increased circulation of influenza A virus in the community affected rhinovirus prevalence via an interferon-mediated mechanism [27]. Furthermore, a study in Wuhan reported on the effect of the dramatic increase in COVID-19 incidence in January 2020, which interfered with the influenza epidemic in the 2019–2020 season and resulted in a sharp decline in influenza A and B incidence that might be a reflection of virus-virus interactions [28]. This phenomenon raises the question: has COVID-19 affected influenza virus prevalence during the pandemic, and how? Further prospective studies and investigations are necessary to elucidate the possible positive and negative interspecific interactions among COVID-19, influenza, and other respiratory viruses on the scale of the individual patient and community populations.

Moreover, underestimating the circulation of respiratory viruses such as influenza during the newly emerging pandemic may have reduced the reported incidence of flu and other respiratory viruses. For instance, during the early phase of the influenza A(H1N1)pdm09 virus pandemic in Australia in 2009, Ratnamohan and colleagues reportedly misdiagnosed other clinically meaningful co-circulating respiratory agents including influenza A and B viruses, respiratory syncytial virus, human metapneumovirus, and enteroviruses [29], a finding similar to our results in this study, wherein the detected influenza viruses were misdiagnosed during the patient's hospitalization and only detected retrospectively. All the above-mentioned factors that are proposed to have reduced influenza infection rates during the current COVID-19 pandemic are illustrated in Fig. 2.

The present study has some limitations. This study was conducted on patients from a single center. Additionally, we could not find in the patient's records whether they recently received influenza vaccines or not, which might influence coinfection severity. However, the study sheds light on the prevalence of community-acquired influenza virus confections among SARS-CoV-2 patients in Saudi Arabia.

## 5. Recommendations and concluding Remarks

In this study, we documented the low prevalence and mild coinfections of community-acquired influenza viruses (influenza A and B and parainfluenza-2) among SARS-CoV-2 patients during the early phase of the COVID-19 pandemic in Al-Medinah, Saudi Arabia.

Health service providers should take coinfections among COVID-19 patients into account, whether community-acquired or hospital-acquired, in order to prevent their progressive impact on treatment
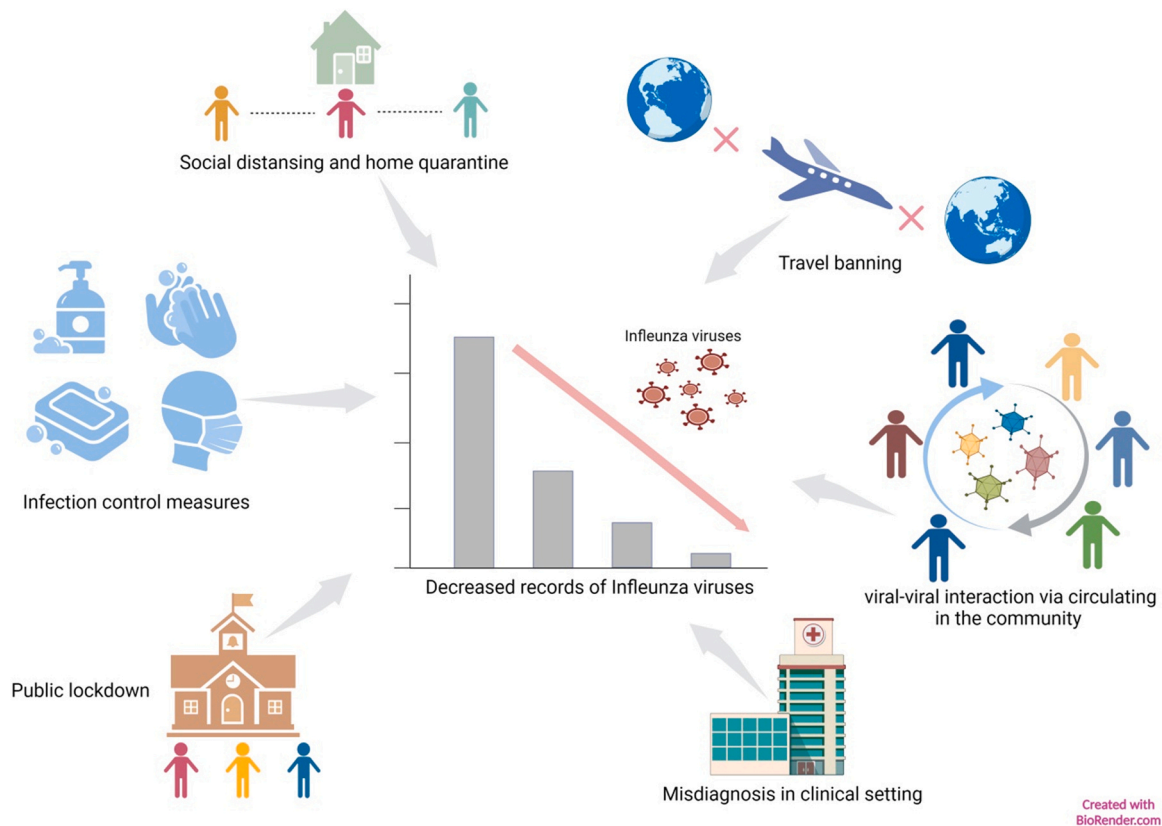
**Fig. 2.** Shows potential factors that might decrease the incidence records for influenza virus infections during the early phases of the Covid-19 pandemic.

and management approaches. In addition, implementing broad range diagnostic tests such as respiratory panels multiplex Polymerase Chine Reaction (PCR) for COVID-19 patients would be helpful in minimizing the possibility of misdiagnosing COVID-19 coinfections as well as providing more indicators of the pathogens cycling in the community and hospitals. Prospective studies and investigations to understand co-circulating pathogens are necessary and should not be neglected during epidemics and pandemics.

### Ethical approval

Written informed consent was obtained via Institutional Review Board from the General directorate of health in Madinah (Approval No.: 9–2021) for support of the use of patient information for the publication of this report.

### Funding

### CRediT authorship contribution statement

STA conceived of the study, designed the experiments, and drafted the manuscript. KOA, HMM, and NAI participated intellectually in the design and framing of the manuscript. STA and NAI provided substantial intellectual direction and participated in experimental design, data analysis, and manuscript drafting. All authors read and approved the final manuscript.

### Data Availability

All data and materials generated during the current study are available from the corresponding author on reasonable request.

### Conflict of interest

The authors declare that they have no known competing financial interests that could have influenced the work reported in this paper.

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