Introduction

The severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is a severe viral infection leading to the global pandemic of the coronavirus 2019 (COVID-19) disease. It caused a novel pneumonia emerged in December 2019, which was proved to be a highly contagious disease with rapid human transmissions through close contact with symptomatically or asymptomatically infected individuals or aerosols (1). COVID-19 and seasonal influenza are viral respiratory infections that are mainly transmitted through respiratory droplets or airborne particles through close contact from person to person (2). Based on the WHO reports, more than 272,640,660 cases were confirmed including more than 5,348,891 deaths globally. Of these confirmed cases, 6,165,454 cases including 130,946 deaths were reported in Iran since December 16, 2021 (https://www.worldometers.info/coronavirus). The pressure that COVID-19 has placed on health care systems, economies, and communities around the world is comparable to the 1918 influenza pandemic (3, 4). The COVID-19 pandemic preparedness plan currently in use in many countries is largely based on the experience of several influenza pandemics over the past decades. Therefore, a detailed understanding of the common features and differences between patients with COVID-19 and influenza infections as well as the most effective way to combat these infections will be helpful (2). Symptoms of COVID-19 disease could be difficult to...
distinguish from influenza illness. COVID-19 outbreak shares a coincidence with influenza season, posing double threats to public health and complicating the disease diagnosis and management. The accurate and rapid diagnosis of COVID-19 infection is essential to control the transmission pathway and create suitable treatment methods (5).

Research has demonstrated that the SARS-CoV-2 virus binds to surface cell receptor angiotensin-converting enzyme 2 of epithelial cells in the respiratory and gastrointestinal tracts and may cause organ failure and even death (6). Recent studies suggest that COVID-19 pneumonia is likely to become a chronic influenza-like illness until effective vaccines or therapeutic measures are available. Given the potential for long-term coexistence with humans globally, the current priority is to develop methods for identifying and evaluating infected individuals. The influenza virus is another infectious disease which caused the global pandemics of the H1N1 swine influenza in 2009 and resulted in the death toll up to 757,000 people (7, 8). In the recent century, mutations and the creation of novel strains of influenza led to the outbreak of new epidemics that caused widespread illness, death, and disruption, generally accompanied by serious symptoms, which caused increases in mortality in young adults. The influenza viruses (e.g., influenza A and B viruses) and the novel coronavirus are both infectious and can cause severe respiratory diseases. Abdominal pain, conjunctivitis, cough, fever, headache, myalgia, nausea or vomiting, seizure, and skin rash are common symptoms of influenza disease. However, symptoms such as calf pain, diarrhea, and rhinorrhea are the symptoms that differentiate influenza A and B (7). Infections caused by influenza A and influenza B viruses can have clinical symptoms similar to the COVID-19; therefore, it is difficult to diagnose COVID-19 pneumonia from influenza, especially during the influenza season. Typically, the incubation period of influenza infection is from 1 to 4 days after infection, but in COVID-19 infection symptoms can appear 1 to 14 days after infection, which can be different (9). The most common symptoms of COVID-19 are fever, tiredness and dry cough, skin rash, myalgia, nausea or vomiting, muscle pain, sore throat, and loss of taste or smell (9). Some patients may have aches and pains, nasal congestion, runny nose, sore throat, or diarrhea. COVID-19 was found to indicate a higher mortality rate than influenza. Currently, in outpatients, the rapid differentiation of COVID-19 patients from influenza A or B is the main issue (8).

Symptoms of COVID-19 disease could be difficult to distinguish from influenza illness (5, 10). COVID-19 outbreak shares a coincidence with influenza season, posing double threats to public health and complicating the disease diagnosis and management. The accurate and rapid diagnosis of COVID-19 infection is essential to control the transmission pathway and create suitable treatment methods (11).

The aim of this study was to determine the prevalence of influenza and the COVID-19 with the onset of the COVID-19 pandemic and before the onset of the COVID-19 pandemic and also to compare their disease symptoms in samples sent to the laboratory of Hormozgan Health Center and gathered from patients with the acute respiratory syndrome.

Materials and Methods

Data Collection

This is a descriptive, cross-sectional study, and the study population included patients with COVID-19 and influenza symptoms referred to medical centers in Hormozgan province from October 2019 to December 2021. In this study, total samples were taken from 229,456 patients with symptoms of the acute respiratory syndrome and were characterized to the reference laboratory of Bandar-E-Abbas, Iran. Further, samples were categorized into two groups: COVID19 and influenza (type A and B). The clinical data including age, gender, fever, cough, shortness of breath, nausea, dizziness, headache, body pain, and diarrhea were recorded for all patients. Then, patients suspected of having influenza or COVID-19 were sampled from the nasopharynx with a special swap.

Ribonucleic Acid Extraction and Reverse Transcription Polymerase Chain Reaction

Two nasopharyngeal and oropharyngeal throat swab samples were collected and tested for SARS-CoV-2 and influenza A and B for each patient with the onset of the COVID-19 pandemic. Viral RNA was extracted from 140 μL of each clinical sample using the High Pure Viral Nucleic Acid Kit (RNJia Virus Kit, Yazd, Iran) according to the manufacturer’s instructions. The RNA was immediately kept at -70°C. For 229,456 of the samples, real-time RT-PCR assays were performed separately including influenza A and B and COVID-19 viruses. We used primer/probe sets in SARS-CoV-2 and influenza A and B that are listed in Table 1 (12-14). The result of SARS-CoV-2 was reported as positive when the cycle threshold value for the E gene was ≤ 40. Likewise, the result of influenza was reported as positive when the cycle threshold value for the HA gene was ≤ 40.

Statistical Analysis

All statistical analyses were performed using SPSS 20.0 software (IBM, Armonk, NY, USA) and GraphPad Prism 5.0 software (GraphPad Software, Inc., San Diego, CA). Demographic, clinical, and outcome variables were compared between the two groups of patients using a t test and chi-square test, and P values less than 0.05 were considered statistically significant.
Results

General Characteristics of COVID-19 Patients

In the present study, which was conducted during 2019-2021 before and after the onset of the COVID-19 pandemic (Figure 1), out of a total of 229,456 samples of patients with symptoms of acute respiratory syndrome who were sent to the central health laboratory from different areas of Hormozgan province, 71,142 (30%) cases were diagnosed with positive COVID-19 and 527 (0.22%) cases with positive influenza. The average age of patients was 46.52 ± 20.3 years. As indicated in Figure 2, among 71,142 cases of COVID-19 which were reported from October 2019 to December 2021, 37,598 (52.85%) and 33,544 (47.15%) cases were male and female, respectively. For patients with influenza, 310 cases (59%) were male and 217 cases (41%) were female. The results of this study indicated that from October 2019 to February 2020, before the onset of the COVID-19 pandemic in Iran, 298 influenza patients were diagnosed. In March 2020, after the onset of COVID-19, this number reduced to 34 cases, of which 33 cases were influenza B and one case was influenza A, while 107 cases of positive COVID-19 patients were reported. During the onset of the COVID-19 pandemic in Iran, during April 2020-September 2020, 7790 cases of COVID-19 were reported, while no positive case of influenza was detected. After the onset of the COVID-19, from October 2020 to March 2021, 13,902 cases of patients with positive COVID-19 were diagnosed. During this time, two cases of patients with positive influenza B were documented. Similarly, from April 2021 to September 2021, 46082 cases of COVID-19 were reported, while no positive case of influenza was detected. The number of positive influenza cases increased to 192 cases from October 2021 to December 2021, while the positive cases of COVID-19 decreased, and only 3261 cases were detected in that period.

The clinical symptoms studied in this survey for differentiation between COVID-19 and influenza were fever, cough, body pain, headache, shortness of breath, nausea, dizziness, and diarrhea. The result indicated that fever, cough, and body pain were the most prevalent symptoms in influenza patients (85%, 100%, and 75%, respectively). The most common symptoms in COVID-19 patients were cough and headache (65% and 64%, respectively). As Table 1 illustrates, the cough was a common symptom for the disease in both groups (65% in COVID-19 patients and 100% in influenza patients). Body pain was present in more people with influenza.

Table 1. Primers and Probes Used for RT-PCR test of InfA, InfB, and SARS-CoV-2

|        | Forward Primer | Reversed Primer | Probe |
|--------|----------------|----------------|-------|
| InfA   | SwfluA         | GCA CGG TCA GCA CTT ATY | CTR AG |
|        | GCA ACT GG     | GGC AGG CTG GTG TTC ATT TGG TC |
|        | 5’ Fam-CYA CTG CAA GCC CAX ACA CAC AAG CAG GCA-Pho-3’ |
|        | X = BHQ-1-Dt   |  |
|        | Inf (H1N1)     | SwH1           | GAC AAA ATA ACA AAC GAA | GCA ACT GG |
|        | GCA ACT GG     | GGC AGG CTG GTG TTC ATT TGG TC |
|        | 5’ Fam-GCA TTC GCA AY’G GAA AGA AAT GCT GG-3’ + + + + |
|        | Labeled at a modified “Y” with BHQ1 and, + + + + With a terminal phosphate at the 3’-end to prevent probe extension by DNA polymerase |
| SARS-CoV-2 | E_Sarbeco   | ACA GGT ACG TTC ATA GTT ATG AGC GT | ATATCTG CAG CAG TAC GCA CAC A |
|        | FAM-ACA CTA GCC ATC CT TCT ACT GCG CTT CG BHQ1 |

Note. RT-PCR: Reverse transcription polymerase chain reaction; InfA: Influenza A; InfB: Influenza B; SARS-CoV-2: Severe acute respiratory syndrome coronavirus 2.

Figure 1. The Flowchart of Prevalence of Influenza (a) and COVID-19 (b) During 2019-2021 in Hormozgan Province. Note. COVID-19: Coronavirus disease 2019.
but it was infrequent in those with COVID-19 (75% vs. 17.2%, \( P < 0.001 \)). Overall, 85% of patients with influenza and 31% of patients with SARS-CoV2 infection had fever symptoms. The temperature of fever in COVID-19 patients and influenza patients was in the range of 36-40.5°C and 36.2–39°C, respectively.

Results indicated that in 2019 (before the onset of the COVID-19), influenza patients were more hospitalized; however, after the onset of the COVID-19, patients with COVID-19 symptoms were more common.

According to Figure 2, no differences was observed between males and females.

**Discussion**

With the onset of the COVID-19 pandemic, the prevalence of many infectious diseases changed (15). In particular, the prevalence of respiratory diseases such as influenza decreased due to social constraints imposed by governments. In this study, we compared the prevalence of influenza and COVID-19 disease between 2019 and 2021 among patients with acute respiratory symptoms referred to the central laboratory of Hormozgan province. The results indicated that out of a total of 229,456 samples of patients, 71142 (30%) cases were diagnosed with positive COVID-19 and 527 (.22%) cases with positive influenza. As shown in Figure 1, prior to the COVID-19 pandemic, there was a wave of seasonal influenza, leading to the identification of 298 patients with influenza symptoms. The number of positive cases of influenza after the onset of the COVID-19 pandemic was 34 in March 2020. The prevalence of influenza virus reached zero in four weeks after the onset of the COVID-19 pandemic and the beginning of restrictions in Hormozgan province 6 months from October to March 2020. Out of all cases of suspected seasonal influenza, only two cases were positive, while there were 205 cases of influenza in this province in the same period the previous year. In a pattern similar to our study, a report released by the CDC in the United States showed that influenza activity declined in March 2020 and remained stable from October 2020 through May 2021. According to this study, from 2017 to 2021, the peak of influenza was observed every year from October to late December, but no influenza peak was observed in 2020 for the COVID-19 pandemic (16). This decline has also been observed in other countries such as Puerto Rico, Taiwan, Korea, Hong Kong, and Singapore (17-20). Since influenza virus, similar to SARS-CoV-2, is spread by droplets, low transmissibility of seasonal influenza virus (R0 = 1.28), compared to SARS-CoV-2 (R0 = 2-3.5), and implementation of social restrictions probably have contributed to the reduction of influenza transmission (18).

Similarly, in Hormozgan province, Iran, with the reduction of social restrictions due to the injection of the COVID-19 vaccine, the number of positive influenza cases increased from October to December 2021. In April 2021, the first case of H3N2 influenza was identified in Hormozgan province, Iran. The start of this wave was predictable due to the widespread vaccination of COVID-19 and the reduction of social restrictions. The current increase could represent a return to pre-pandemic seasonality. Since the prevalence of the influenza virus in last autumn and winter was very low, it can be concluded that social restrictions have reduced the activity of influenza. These social restrictions can be applied to reduce the spread of future influenza pandemics, especially in populations at higher risk for severe illness or side effects. However, influenza vaccination is still the best way to prevent the disease for all people over 6 months.
of age and is especially effective in autumn and winter when SARS-CoV-2 and the influenza virus are likely to spread simultaneously. Furthermore, Liang et al proposed that using masks provided a significant protective effect (21). Some reports warned that influenza prevalence will return to pre-pandemic circulation patterns even more severely, and the reduced circulation of influenza viruses during the past year might affect the severity of the upcoming influenza season given the prolonged absence of ongoing natural exposure to influenza viruses. For this reason, clinicians are advised to follow preventive health protocols and get vaccinated (16, 22).

Fever, cough, and headache were the most common symptoms among both groups, but body pain was less common in patients with COVID-19, while it was common in patients with influenza. However, most signs and symptoms were similar between the two groups, making clinical distinction unreliable. Laris-González et al compared clinical characteristics of patients with influenza with COVID-19 in Mexico City, and in agreement with our study, fever and cough were reported as the most common clinical manifestations in both groups (23). In another study, in a retrospective cohort, Song et al found that fever, gastrointestinal symptoms, headache, and myalgia were the most common symptoms in patients hospitalized with COVID-19 than in patients with influenza (24). In sum, influenza and COVID-19 have a similar manifestation in patients, which makes diagnostic tests necessary for proper diagnosis and management.

The study of a single center as well as the frequency of comorbidities among participants may limit generalization of findings. Some data on patients with influenza before the onset of the COVID-19 pandemic was collected retrospectively from clinical records while the COVID-19 cohort was followed-up prospectively. Differences in periods between the two studies may be due to differences in diagnostic and therapeutic approaches.

Conclusion
It can be concluded that social restrictions and adherence to health protocols can significantly reduce the incidence of seasonal influenza, and even after controlling the COVID-19 pandemic, the incidence of seasonal influenza can be controlled by adhering to health protocols along with vaccination. On the other hand, influenza and COVID-19 have similar symptoms in patients, so diagnostic tests are necessary for proper diagnosis and management.

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Authors’ Contribution
HG, KA, and MH designed the study, KA wrote the manuscript. EE and MH analyzed and interpreted the data. KA, EE, PD, and MM performed the revision of the manuscript. All authors read and approved the final manuscript.

Availability of Data and Materials
Not applicable.

Conflict of Interest Disclosures
The authors declare that they have no competing interests.

Ethical Statement
The current study was approved by the Ethics Committee of the Hormozgan University of Medical Sciences, Bandar Abbas, Iran [IR.HUMS.REC.1398.470]. There was no need for informed consent because this study was retrospective without posing any potential risk to patients.

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Informed Consent
Not applicable.

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