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Determination of COVID-19 prevalence with regards to age range of patients referring to the hospitals located in western Tehran, Iran

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ABSTRACT

We decided to examine suspected samples of pneumonia outbreak caused by the new coronavirus SARS-CoV-2 and provide information about the mortality rate due to this infection in different age groups in Iran. In this descriptive-cross-sectional study, a total of 784 samples of naso/oropharyngeal swabs of suspected patients with COVID-19 symptoms who had referred to Imam Khomeini, Shahid Fayaz-Bakhsh and Modarres hospitals in Tehran from February 24, 2020 to March 24, 2020 were examined by RT-PCR method. The highest incidence of the disease was within the age group of 50–59 years, while the lowest rate was in the 0–9 years age group. The highest rate of positive samples in terms of COVID-19 among suspected individuals was for patients > 80 years of age (89%) and the highest mortality rate was in the age range of 70–79 years (31%) and > 80 years (30), respectively. In terms of recovery, the highest rate was in the 30–39 years age group (65.2%). Statistical analysis showed that mortality significantly increased in the age group of > 60 years old and in fact, mortality was significantly associated with older ages. According to the results of the current study, the prevalence of COVID-19 in lower age (0–9 years old) is lower and mortality rate is higher in older ages as significant increase in mortality was observed in those aged > 60 years old. However, further epidemiological studies on a larger study population in different regions of Iran are needed to explain the prevalence, clinical features, and course of the disease.

1. Introduction

Coronaviruses (CoVs) are enveloped sense-positive RNA viruses belonging to the Coronaviridae family and Nidovirales order. These viruses are widely found in humans and other mammals as well as birds leading to neural, hepatic, enteric, and respiratory disorders. Up to now, seven pathogenic Coronavirus spp. have been identified in humans. Four common CoVs include 229E, OC43, NL63, and HKU1 that usually lead to common cold in healthy individuals (Payne, 2017; Kasmi et al., 2020). The emergence of severe acute respiratory syndrome-coronavirus (SARS-CoV) pandemic in 2002–2003 followed by Middle East respiratory syndrome-coronavirus (MERS-CoV) in 2012 indicates that some of these CoVs are threatening to human health (of the International CSG, 2020). In fact, although most CoV infections are mild in humans, the epidemiology of the two beta-CoVs, SARS-CoV and MERS-CoV, have generally affected 10,000 cases in the last two decades with the mortality rate of about 10% and 37%, respectively. Due to the high prevalence and widespread distribution of CoVs, their genetic diversity, wide distribution of their genomes and the elevated interaction between humans and animals following inter-species infections, there is a possibility of the emergence of novel CoV (nCoV) (Sohrabi et al., 2020; Hassan et al., 2020). In late 2019, an nCoV was identified that led to an outbreak of acute respiratory disease in Wuhan, China. The nCoV also known as SARS-CoV-2 belongs to beta-coronaviruses with 70% genetic similarity to SARS-CoV causing coronavirus disease 2019 (COVID-19). SARS-CoV-2 is different from SARS-CoV and MERS-CoV and is the seventh known member of the coronavirus family to infect humans (Kannan et al., 2020; Guo et al., 2020). Following the identification of SARS-CoV-2 genome, bats were identified as the primary host of the virus, while pangolins are most probably its intermediate host (Wong et al., 2020).

The COVID-19 virus infects people of all ages. However, the evidence to date shows that two groups of people are at higher risk for developing COVID-19. One group of these people are older people (i.e. people over 60 years old). The other group has people with underlying medical conditions (such as cardiovascular disease, diabetes, chronic respiratory disease, and cancer). The risk of developing a severe disease gradually increases with age from about 40 years. It is important to adults in this age range to protect themselves and in turn protect others who may be more vulnerable. From the first reports of SARS-CoV-2,
COVID-19 has been confirmed in > 3 million people worldwide (Organization WH, 2020).

The symptoms of the disease range from mild to acute, and most patients do not show acute symptoms. According to the reports from China, of the 445,500 confirmed patients, 81% showed mild symptoms or were asymptomatic, 14% showed severe symptoms including dyspnea or hypoxia, and 5% had acute symptoms such as respiratory disorder, shock, and organ failure. Death was reported in 2.3% of individuals (Lai et al., 2020).

Unfortunately, at present, no specific treatment or effective vaccine has been identified for this disease and the best infection control strategies include rapid diagnosis and isolation of patients. Currently, SARS-CoV-2 genetic sequencing has provided specific detection tests based on real-time RT-PCR (Lan et al., 2020). This technique is used to detect viruses in respiratory tract samples such as nasal swabs and is highly sensitive and can detect low viral loads. In fact, the early characteristic of the viral complete genome led to the designation of about 94 specific primers and specific laboratory protocols for SARS-CoV-2. The first RT-PCR assay that targeted genes coding for RNA-dependent RNA polymerase (RdRp), envelope (E), and nucleocapsid (N) was introduced on January 23, 2020. This assay was designed and validated using synthetic acid nucleic technology (Chan et al., 2020; Corman et al., 2020). The viral E protein is a small global protein that is involved in several aspects of the virus life cycle, such as assembly, germination, and pathogenicity. This protein acts as a viral perinuclear and ion channel and is involved in the interaction between viral and host cell proteins (Schoeman and Fielding, 2019). The RdRp component also known as nsp12, catalyzes viral RNA synthesis and thus plays an important role in SARS-CoV-2 replication and transcription and possibly requires cofactors such as nsp7 and nsp8. This protein is considered to be the main target of viral inhibitors such as Remdesivir (Gao et al., 2020). Finally, the N protein plays an important role during viral assembly and interacts with the viral genome and membrane (M) proteins. These N proteins are involved in viral RNA transcription and replication (Mousavizadeh and Ghasemi, 2020). Therefore, in this study, we aimed at targeting these three genes using real-time RT-PCR method to examine suspected samples of COVID-19 and to determine the mortality rate due to this infection in different age groups in Iran.

2. Materials and methods

2.1. Population study

The current work is a descriptive cross-sectional study. Inclusion criteria in this study was the presentation of COVID-19 symptoms. Most patients showed symptoms including common cold with dry cough. Naso/oropharyngeal swabs were collected from symptomatic patients referring to Imam Khomeini, Shahid Fazy-Bakhsh, and Modarres hospitals, Tehran, Iran from 24 February 2020 to 24 March 2020. A total of 784 samples were transferred to the laboratory using proper specimen transport media in sealed safe packs.

2.2. RNA extraction and real-time RT-PCR

Viral RNA from clinical samples was extracted using QIAamp viral RNA minikit (QIAGEN, Hilden, Germany) according to the protocols provided by the manufacturer. RNA extraction protocol for all clinical samples was the same. A 25 μL reaction volume consisting of 5 μL RNA, 12.5 μL 2× buffer with a single-step RT-PCR Superscript III with Platinum Taq polymerase (Invitrogen, Germany, consisting of 0.4 mM of each dNTP and 3.2 mM MgSO_4), 1 μL Taq reverse transcriptase, 0.4 μL of 50 mM MgSO_4, and 1 μg non-acetylated bovine serum albumin (Roche, Germany) was used for cDNA synthesis. Primers and probes used in this study are presented in Table 1. Thermocycling conditions were as follows: 55 °C for 10 min, 45 cycles of 95 °C for 3 min, 95 °C for 15 s, and 48 °C for 30 s. Gene coding for human RNase P was used as the internal control.

2.3. Statistical analysis

To determine the association between age and recovery rate as well as mortality, chi-square test was performed using SPSS software v. 22.

3. Results

A total of 784 naso/oropharyngeal samples of patients with COVID-19 symptoms referring to Imam Khomeini, Shahid Fazy-Bakhsh, and Modarres hospitals, Tehran, Iran were evaluated. Using SARS-CoV-2 specific primers, the prevalence of COVID-19 in different age groups was confirmed with RT-PCR. One sample was collected from each patient. The number of suspected patients with COVID-19, number of confirmed cases, and number of recovered patients are presented in Table 2. According to the results, the highest and the lowest numbers of suspected COVID-19 cases were within the age range of 50–59 and 0–9 years, respectively. Among 161 suspected cases within the age range of 50–59 years, 102 cases were confirmed positive using RT-PCR, among which 16 (15.6%) died. Also, among 13 suspected cases within the range of 0–9 years, two cases were positive using RT-PCR and no death was reported among them. The highest prevalence of positive COVID-19 cases were among suspected cases within the age range of > 80 years. In addition, the highest mortality rate was within the age range of 70–79 years, among which 7 out of 24 (30%) confirmed cases died. The highest rate of recovery (65.2%) in COVID-19 patients was observed in those within the age range of 30–39 years and overall, a high rate of recovery was indicated (Tables 1 and 2). According to the statistical analysis, mortality rate was significantly high in the age range of > 60 years, indicating a significant relationship between mortality rate and old age (P-value < 0.05). Also, all age ranges showed a significantly high recovery rate (P-value < 0.05) (Table 2).

4. Discussion

At present, it seems that all people, including children and pregnant women, are susceptible to COVID-19. According to some studies, older people (mean age > 55 years old) are more susceptible to the virus and frequently show symptoms such as fever, cough and fatigue with runny nose and headache (Novel CPERE, 2020; Peeri et al., 2020; Singhal, 2020). According to the presented symptoms, infections are classified into three groups of mild, severe, and acute. Patients with mild symptoms either showed mild pneumonia or no pneumonia. Severe symptoms include dyspnea, increased respiration rate (30 times per min), blood oxygen saturation (< 93%), relative arterial oxygen pressure, and pulmonary infiltration of > 50% within 24–48 h. Acute symptoms include respiratory failure, septic shock, organ failure, and eventually death (He et al., 2020; Huang et al., 2020). In this study, we aimed to investigate the prevalence of COVID-19, recovery rate and death due to this infection in Iran with regards to age range. For this purpose, a total of 784 samples of naso/oropharyngeal swabs of people who had referred to hospitals in Western Tehran with COVID-19 symptoms were examined (Table 3).

According to the results, the highest and the lowest rates of suspected COVID-19 cases were within the age range of 50–59 and 0–9 years, respectively. The highest prevalence of positive COVID-19 cases were among suspected cases within the age range of > 80 years. In addition, the highest mortality rate was within the age range of 70–79 years, among which about 30% confirmed cases died. Overall, a high rate of recovery was indicated. According to the statistical analysis, mortality rate was significantly high in the age range of > 60 years, indicating a significant relationship between mortality rate and old age (P-value < 0.05). Also, all age ranges showed a significantly high recovery rate (P-value < 0.05).

So far, other epidemiological studies have been reported. For
fever and cough and were divided into two groups of < 50 and > 50 years of age. In patients with > 50 years of age, more areas of the lung were affected. Thirteen of the 51 patients were further examined, showing recovery in pulmonary symptoms in 7 cases and further invasion in 4 cases aged > 50 years. According to this study, it can be concluded that with age, pulmonary involvement increases and the probability of the persistence of this infection increases (Song et al., 2020). This was in accordance with the results of the present study, in which the highest number of deaths and positive cases were reported in people in the age group of > 70 years. In general, according to WHO reports, so far, no mortality has been observed in patients within the age range of 0–9 years old and the mortality rate of 21.9% has been reported in patients > 80 years of age (Battegay et al., 2020).

Several studies have focused on the pediatric population. As the immune system of children is immature, this age range is susceptible to respiratory infections and therefore, a variety of symptoms may occur. As the epidemic spreads, the number of infected children gradually increased. There have been no reports of children being a source of infection. The transmission route of SARS-CoV-2 in children is similar to that of adults, which include contact and respiratory droplets, however, so far, symptomatic infections have rarely been observed (Zeng et al., 2020). In the present study, the lowest rate of COVID-19 infection. The transmission route of SARS-CoV-2 in children is similar to that of adults, which include contact and respiratory droplets, however, so far, symptomatic infections have rarely been observed (Zeng et al., 2020). In the present study, the lowest rate of COVID-19 and fatality was observed in children.

5. Conclusion

The recent outbreak of COVID-19 is one of the most serious health concerns worldwide, and the number of people infected with the SARS-CoV-2 is increasing. According to the results of the present study, it can be stated that the prevalence of COVID-19 infection is lower in younger individuals (0–9 years) and the mortality rate due to this infection increases with age as the mortality rate in the age group of > 60 years significantly increased. However, further studies are required on the epidemiology of this disease with larger study populations in other cities of Iran.

Table 1

| Target gene | Oligonucleotides | Sequence (5’→3’) | Reference |
|-------------|------------------|------------------|-----------|
| RdRP        | RdRp, SARSr-F    | GTGARATGCTATGTCGGCGG | (Chan et al., 2020) |
| RdRP        | RdRps, SARSr-P   | FAM-CAGGTGAACACCCTCACGAGATG-BBQ | |
| RdRP        | RdRps, SARSr-P1  | FAM-CCAGGGWACRRCTCAACMGTTAGTG-BBQ | |
| RdRP        | RdRps, SARSr-R   | CARATGTAAASACACTATAGGTA | (Chan et al., 2020) |
| E           | E, Sarbeco F     | ACAGTGCACCAATCGAGTGGG | |
| E           | E, Sarbeco_P1    | FAM-ACACTAGCACCTCCTACTGCCGCTTGG-BBQ | (Chan et al., 2020) |
| N           | N, Sarbeco_F, F  | ATATGGCAGGACGGCCACACACA | |
| N           | N, Sarbeco_P, R  | FAM-ACCCTGCAAHAGACACTGGA-CGA | (Chan et al., 2020) |
| RNase P     | RNaseP-F         | GAGGAAGAGAAAGGAGCTTTG | (Emery et al., 2004) |
| RNase P     | RNaseP-R         | GAGGAGCTGTTCGCAAGAT | |
| RNase P prob|                  | Fam-TTCTGACGAAAGGCTCTGCGG-BHQ1 | |

W: A/T; R: G/A; M: A/C; S: G/C. FAM: 6-carboxyfluorescein; BBQ: blackberry quencher.

Table 2

Results of the evaluation of patients with COVID-19 symptoms in various age ranges from 24 February 2020 to 24 March 2020 in the three studied hospitals.

| Age range (years old) | Suspected cases (No.) | Positive cases (No.) | Recovered cases (No.) | Death (No.) |
|-----------------------|-----------------------|----------------------|-----------------------|-------------|
| 0–9                   | 13                    | 2                    | 1                     | 0           |
| 10–19                 | 34                    | 16                   | 9                     | 0           |
| 20–29                 | 96                    | 69                   | 32                    | 3           |
| 30–39                 | 124                   | 95                   | 62                    | 5           |
| 40–49                 | 153                   | 118                  | 54                    | 13          |
| 50–59                 | 161                   | 102                  | 57                    | 16          |
| 60–69                 | 126                   | 89                   | 41                    | 23          |
| 70–79                 | 56                    | 42                   | 22                    | 13          |
| > 80                  | 27                    | 24                   | 10                    | 7           |

Table 3

Statistical analysis of COVID-19 patients in different age groups.

| Age range (years old) | Infection rate (Standard error ± mean) (%) | Recovery rate (Standard error ± mean) (%) | Death rate (Standard error ± mean) (%) |
|-----------------------|------------------------------------------|------------------------------------------|----------------------------------------|
| 0–9                   | 15.31 ± 0.29                             | 50.02 ± 0.25                             | 0                                      |
| 10–19                 | 47.24 ± 1.25                             | 56.2 ± 0.17                              | 0                                      |
| 20–29                 | 72.56 ± 2.01                             | 46.3 ± 0.06                              | 4.3 ± 0.11                             |
| 30–39                 | 76.04 ± 0.65                             | 65.2 ± 0.87                              | 5.2 ± 0.05                             |
| 40–49                 | 77.13 ± 1.89                             | 45.7 ± 0.03                              | 11.21 ± 0.49                           |
| 50–59                 | 63.21 ± 2.01                             | 55.9 ± 0.64                              | 15.6 ± 0.25                            |
| 60–69                 | 25.74 ± 0.50                             | 46.2 ± 0.41                              | 25.8 ± 0.91                            |
| 70–79                 | 75.18 ± 0.74                             | 52.3 ± 0.05                              | 31.06 ± 0.45                           |
| > 80                  | 89.62 ± 0.79                             | 41.6 ± 0.31                              | 29.17 ± 0.41                           |

*p-Value < 0.05.
**p-Value < 0.01.

example, in a study on 41 patients with a mean 0–49 years of age, referring with fever and cough, only 13 cases were hospitalized and 6 deaths were reported (He et al., 2020). Their results were consistent with the results of the present study as the mortality rate and the need for hospitalization in people under 50 years of age were significantly lower. In a study in China on 73,2314 cases, 62% cases were positive by RT-PCR, and in terms of age distribution, 1% were under 10 years old, 1% were between 10 and 19 years old, 8% were between 20 and 29 years old, 87% were between 30 and 79 years old, and 3% were > 80 years old. In terms of mortality, the highest mortality rate was related to patients > 80 years of age (14.8%), while patients aged 70–79 showed a mortality rate of 0.8% (Wu and McGoogan, 2020). This is consistent with the results of the current study as the highest mortality rate was in patients > 80 years of age.

In another study on 51 patients, all cases presented symptoms of fever and cough and were divided into two groups of < 50 and > 50 years of age. In patients with > 50 years of age, more areas of the lung were affected. Thirteen of the 51 patients were further examined, showing recovery in pulmonary symptoms in 7 cases and further invasion in 4 cases aged > 50 years. According to this study, it can be concluded that with age, pulmonary involvement increases and the probability of the persistence of this infection increases (Song et al., 2020). This was in accordance with the results of the present study, in which the highest number of deaths and positive cases were reported in people in the age group of > 70 years. In general, according to WHO reports, so far, no mortality has been observed in patients within the age range of 0–9 years old and the mortality rate of 21.9% has been reported in patients > 80 years of age (Battegay et al., 2020).

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References

Battegay, M., Kuehl, R., Tschudin-Sutter, S., Hirsch, H.H., Widmer, A.F., Neher, R.A., 2020. 2019-novel Coronavirus (2019-nCoV): estimating the case fatality rate–a word of caution. Swiss Med. Wkly. 150 (0506).
Chan, J.F.-W., Yip, C.C.-Y., To, K.K.-W., Tang, T.H.-C., Wong, S.C.-Y., Leung, K.-H., et al., 2020. Gene Reports 21 (2020) 100910.
2020. Improved molecular diagnosis of COVID-19 by the novel, highly sensitive and specific COVID-19-RdRp/HeL real-time reverse transcription-PCR assay validated in vitro and with clinical specimens. J. Clin. Microbiol. (5), 58.

Corman, V.M., Landt, O., Kaiser, M., Molenkamp, R., Meijer, A., Chu, D.K., et al., 2020. Detection of 2019 novel coronavirus (2019-nCoV) by real-time RT-PCR. Eurosurveillance. 25 (3), 2000045.

Emery, S.L., Erdman, D.D., Bowen, M.D., Newton, B.R., Winchell, J.M., Meyer, R.F., et al., 2004. Real-time reverse transcription-polymerase chain reaction assay for SARS-associated coronavirus. Emerg. Infect. Dis. 10 (2), 311.

Gao, Y., Yan, L., Huang, Y., Liu, F., Zhao, Y., Cao, L., et al., 2020. Structure of the RNA-dependent RNA polymerase from COVID-19 virus. Science 368 (6492), 779–782.

Guo, Y.-R., Cao, Q.-D., Hong, Z.-S., Tan, Y.-Y., Chen, S.-D., Jin, H.-J., et al., 2020. Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) and coronavirus disease-2019 (COVID-19) outbreak—an update on the status. Mil. Med. Res. 7 (1), 1–10.

Hassan, S.A., Sheikh, F.N., Jamal, S., Ezeh, J.K., Akhtar, A., 2020. Coronavirus (COVID-19): a review of clinical features, diagnosis, and treatment. Cureus 12 (3).

He, F., Deng, Y., Li, W., 2020. Coronavirus disease 2019: what we know? J. Med. Virol. 92 (7), 719–725.

Huang, C., Wang, Y., Li, X., Ren, L., Zhao, J., Hu, Y., et al., 2020. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet 395 (10223), 497–506.

Kannan, S., Ali, P.S.S., Sheeza, A., Hemalatha, K., 2020. COVID-19 (Novel Coronavirus 2019)-recent trends. Eur. Rev. Med. Pharmacol. Sci. 24 (4), 2006–2011.

Kasmi, Y., Khatayb, K., Soxiri, A., Emnaji, M.M., 2020. Coronavirus: 100,000 years of emergence and reemergence. In: Emerging and Reemerging Viral Pathogens. Elsevier, pp. 127–149.

Lai, C.-C., Shih, T.-P., Ko, W.-C., Tang, H.-J., Hsueh, P.-R., 2020. Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) and corona virus disease-2019 (COVID-19): the epidemic and the challenges. Int. J. Antimicrob. Agents 55 (3), 145–151 105924.

Mousavizadedeh, H., Ghasemi, S., 2020. Genotype and phenotype of COVID-19: their roles in pathogenesis. J. Microbiol. Immunol. Infect. 4 (7), 214–219. https://doi.org/10.1016/j.jmii.2020.03.022.

Organization WH, 2020. Coronavirus Disease 2019 (COVID-19): Situation Report. pp. 51.

Peeri, N.C., Shrestha, N., Rahman, M.S., Zaki, R., Tan, Z., Bibi, S., et al., 2020. The SARS, MERS and novel coronavirus (COVID-19) epidemics, the newest and biggest global health threats: what lessons have we learned? Int. J. Epidemid. 49 (3), 717–726.

Wong, M.C., Cregeen, S.J.J., Ajami, N.J., Petrosino, J.F., 2020. Evidence of recombination in coronaviruses implicating pangolin origins of nCoV-2019. BioRxiv. https://doi.org/10.1101/2020.02.07.939207.