



Gender and Age Differences in COVID-19 in Iraqi Patient Detected By Real-Time PCR

Zainab Fadhil Abbas¹ and Baqer J. Hasan^{1*}

¹*Department of Laboratory Analysis, College of Health and Medical Technology, University of Uruk, Iraq.*

Authors' contributions

This work was carried out in collaboration between both authors. Author ZFA designed the study, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. Author BJH managed the analyses of the study. Both authors read and approved the final manuscript.

Article Information

DOI: 10.9734/AJMAH/2021/v19i830357

Editor(s):

- (1) Dr. Mohamed Salem Nasr Allah, Weill Cornell Medical College, Qatar.
- (2) Dr. P. Veera Muthumari, V. V. Vanniaperumal College for Women, India.
- (3) Dr. Ashish Anand, GV Montgomery Veteran Affairs Medical Center, USA.

Reviewers:

- (1) Ghanshyam Gahlot, Rajasthan University of Health Sciences, India.
- (2) Tânia Mara Pinto Dabés Guimarães, Federal University of Minas Gerais, Brazil.
- (3) Khoo Lay See, Hospital Kuala Lumpur, Malaysia.

Complete Peer review History: <https://www.sdiarticle4.com/review-history/70408>

Original Research Article

Received 01 May 2021
Accepted 05 July 2021
Published 06 August 2021

ABSTRACT

Since the SARS-CoV-2 epidemic started, it became clear that the impact of the infection incidence and fatality rate were nearly related to the population structure. The outbreak of COVID-19 have severe impacts on the population, lead to different affection in women and men. epidemic in Iraq as elsewhere penetrate rapidly across the country, identifying vulnerable groups and gender analysis and also understanding the communities help to make an effective decisions. Our study Aimed to found the differences of gender and age in Iraqi people from the coronavirus disease 2019. 21545 cases were extracted from Razi Medical center in Baghdad from October 12, 2020 to March 3, 2021, 488 where positive result was according the performance of real-time reverse transcription-PCR (RT-PCR) assays used to targe different gene of SARS-CoV-2 based on kit manuscript. For each group, we calculated cumulative positive rates stratified age and gender for corresponding risks for men vs. women. the positive 358 patients (73.4 %) were men and 130 patients (26.6%) were women. The major part of cases was in the age group of 40-60 years of old and the male to female ratio was 2.75:1. The findings of the study represented higher positive cases rate in men

*Corresponding author: E-mail: baqer.hikma.iq@gmail.com;

than women. so Male gender and older age notably associated with the risk of positive COVID-19 patients. paying special attention to male elderly patient one of the most important strategies against spreading COVID-19.

Keywords: COVID-19; real-time PCR; gender and age.

1. INTRODUCTION

After the first case report of undiagnosed pneumonia in Wuhan, China of COVID-19 in 2019 the disease soon spread across the world and became a pandemic. The disease agent identified as the Severe Acute Respiratory Syndrome Coronavirus-2 (SARS-CoV-2) [1,2]. The disease caused by the virus was named coronavirus disease 2019 (COVID-19) chosen by WHO, leading to considerable morbidity and mortality, COVID-19 is the first virus of the coronavirus family to cause a pandemic. This pandemic disease was because of rapidly spreading the disease resulting in an, followed by an increasing number of cases in other countries throughout the world. In Iraq, as elsewhere, the coronavirus pandemic started in February 24, 2020 when a traveler identified positive and after that total confirmed cases reached more than 1,136,917 cases to this date [3]. Sequencing of the virus revealed that a novel β -coronavirus SARS-CoV-2 present with acute onset of fever, myalgia, cough, dyspnea, and radiological evidence of ground-glass lung opacities compatible with atypical pneumonia [4,5]. Because sex and age differences impact the outcome of COVID-19 patients Most studies worthwhile focused on age and gender varying susceptibility to infection by SARS-CoV-2 [6]. Recognizing the differences with higher susceptibility help minimizing the epidemic adverse effects. Based on the data collected from different research showed that age and gender are two most important factors that influence the severity of COVID-19.

According to several research done on global rates of infection with SARS-CoV-2 data showed almost identical gender ratio, despite this data hospitalizations and mortality are higher in men than in women. Females may be less susceptible to COVID-19 due to sex-based differences. Frequently diseases manifest differently in men and women. These differences of symptoms, diagnosis, severity, duration of the disease involve biological sex (genetic, anatomy, hormones and physiology) as well psychological and cultural behavior (ethnicity, social, and religious background, environmental and

lifestyle) changing throughout the life course [7]. The World Health Organization (WHO) pays particular attention on gender differences and their impact on diagnosis to shed light on the risk factors causing disease in men and women and address the corresponding specific treatments [6,8].

To make an appropriate decision for patient with COVID-19 and to identifying the epidemiological characteristics of this disease, understanding gender differences exist in the response to public health becomes important for prevention of COVID-19. different studies revealed epidemiological features of COVID-19, however these studies were based on small sample sizes. The aim of this study was to characterize the age and gender differences of COVID-19 in Iraq as a country with a population of more than 39 million people in the middle East [9]. Here we reported the result of analyzing data of all cases that came to Razi center, in Bagdad, Iraq.

2. MATERIALS AND METHODS

2.1 Data Collection

To analyze the association between gender and age the of COVID-19, Data collected from Razi medical center. The data collection methods and the time period covered differed for each region with the earliest date between 12 October 2020 to 3 March 2021. A total of 21545 suspected COVID-19 patients conducted in this study, We grouped the total population different age groups (Table1).

2.2 COVID-19 Real-time RT-PCR Assays

The Real-Time Fluorescent RT-PCR Kit for Detecting SARS-CoV-2 is a molecular in vitro diagnostic test that aids in the detection and diagnosis of SARS-CoV-2 and is based on widely used nucleic acid amplification technology. The product contains primers, labeled oligonucleotide probes, and material that control the quality of test that used in real-time RT-PCR for the in vitro qualitative detection of SARS-CoV-2 RNA in respiratory specimens.

Step	Temperature	Time	Cycle
Revers transcription	50 °C	20 min	1
Initial activation	95 °C	3 min	1
denaturation	95 °C	3 s	40
Annealing	55 °C	20 s	
extension	60 °C	20s	

Fig. 1. Temperature profile of real time PCR for Detecting SARS-CoV-2

Viral RNA was first extracted from the biological specimen collected from the nasal or nasopharyngeal swabs and is purified. Sacace kit (Sacace Biotechnologies, Italy) a diagnostics molecular biology kit used for RNA Extraction (procedure done according to the manufacturer's instructions). Primer and probe sets targeting different gene regions. Real-time RT-PCR assays for SARS-CoV-2 RNA detection were performed using Rotor Gene Q (Qiagen) real-time PCR system (Roche, Basel, Switzerland). Biotech rabbit GmbH (berlin, Germany) PCR kit used for detection the different target. Fig. 1.

2.3 Statistical Analysis

To comparing the performance of the assays Fisher's exact test was used. A *P* value of <0.05 was considered statistically significant.

3. RESULTS AND DISCUSSION

Real-time RT-PCR assays used to target different Genes of SARS-CoV-2. Result Analyzed after the positive and negative controls have been examined and determined to be valid and acceptable. sigmoidal amplification curves in both FAM and VIC/HEX channels appeared. The Ct values in these three channels should not higher than 37 and 35 respectively. If there is no

sigmoidal curve in the FAM channel it will confirm the negative of specimen.

We detected the major differences in the age and gender certain rates of positive from SARS-CoV-2. Table 1 show Association among the composition of age and gender of COVID-19.

For each group, we calculated cumulative positive rates ranked by sex and age comparable relative risks for men vs. women. In this study, 358 patients (73.4%) were men and 130 patients (26.6%) were women. The major part of cases was in the age group of 40 to 60 years of old and male female ratio was 2.7:1.

For determining the population significant risk of SARS-CoV-2 it is important to identifying age and sex related differences to understand the diseases surveillance for taking a better medical intervention to provide insights into geographic, genetic, or cultural factors that could influence the spread of pandemic disease among different population [4,5]. To control the pandemic of COVID-19 it is important to determine the impact of sex and gender on the risk of infection. Despite similar global rates of infection with SARS-CoV-2 hospitalization and death rates are elevated in men vs women.

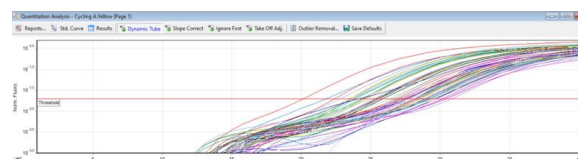


Fig. 2. To ensure PCR reaction and technical errors accuracy internal control curve present in each assay

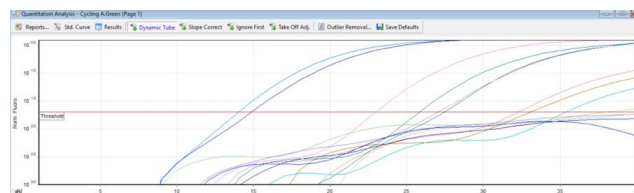


Fig. 3. Ct of target gene in the Real-Time Fluorescent RT-PCR for Detecting SARS-CoV-2

Table 1. Patients features

Patients 488	
Age, years	
0-10	18 (3.7 %)
10-25	60 (12.3 %)
25-40	153 (31.4 %)
40-60	183 (37.5 %)
60-80	61 (12.5 %)
80+	2 (0.4 %)
No sample	11 (2.3 %)
Sex	
Female	130 (26.6 %)
Male	358 (73.4 %)

For providing protection from viral infection in both sex and gender presentation accurate testing and medical care are urgently important in the response of SARS-CoV-2. approaches that will consider both sex and gender should provide understanding in biological attributes and socially constructed roles, behaviors, expressions, identities. provide accurate testing and medical care in case of gender and age differences are importunately needed and urgent against COVID-19 [4]. The present study on 21545 cases between October 2020 to 3 March 2021, gives information about outbreak in Iraq. As in shown in Table1 individual under 25 years old are have least risk to COVID-19, also we observed that males have higher incidence rate of positive result than Females. Ludwig study report a higher mortality in males. Karlberg et al observed higher mortality in younger males -44 years), compared to age group 45-74 [8]. Age is the most essential factor in severity of the disease [10,11]. The death rate for COVID-19 is higher in men and at nearly all ages. According to Global Health data, death rate in the number of COVID-19 confirmed cases due to the disease are high among men in different countries [9,12].

Research done later showed that mortality of males in hospital higher than that of females, also similar report indicate similar pattern of higher mortality in males [13]. Similarly, the study by Alghamdi et al. demonstrate the fatality case rate in males was twice than females in MERS (52 vs. 23%) [14]. Gender difference in the immune response has been suggested as a possible factor in research done later [15]. There are studies that say that the reason men are affected more than women and the cause of death in men more, is due to the reason for the presence of the estrogen hormone in women, as the estrogen hormone protects women from complications of the disease COVID-19 [16].

study done by Zirui Tay et al. revealed that there might be alleles on the X-chromosome that confer resistance to COVID-19 this may explain lower mortality among females therefore hormonal immunoregulation effect the severity of disease [17]. Channappanavar et al. report increased mortality in the SARS-CoV-2 infection in male mice signifying a protective effect of estrogen receptor signaling [18]. Berghöfer et al. report the [18]. Berghöfer et al. report in vitro study on TLR7 ligands result increased production of interferon- α (IFN α) in female cells against to the male's cells [19]. P. CONTI and A. YOUNES study considered gender differences in the immune response to COVID19 infection and inflammatory diseases. In another study Brenner SR. reported that male prevalence phenomena of COVID-19 may be due to the androgen driven pathogen of SARS-CoV-2. Despite this study estrogen could increase IFN- γ promoter's activity, which participates in the responses to viral pathogens [20]. Most of the study done determine that women less susceptible to viral infections based on a different innate immunity, steroid hormones and factors related to sex chromosomes. Linda Juel Ehrenfeld confirmed a higher mortality in European men than European women in almost all age groups by study 10 European regions [21].

Study performed on suspected cases that not confirmed cases on gender differences revealed that children less frequently and less severely affected than adults (2.1 - 2.4% in China, 1.3% in Italy, 2.8% in Australia, and 7.0% in Korea). this might because less susceptible to the infection and less likely to be symptomatic or develop severe symptoms, therefore there is no obvious reason for children pattern of SARS-CoV-2 sickness. The data from the Korea MERS-CoV2 epidemic reported as of 11 March 2020 show that the numbers of people infected and deaths

due to COVID-19 indicate large differences in age and specific gender (40:60 male to female ratio) [22]. The number of associated infections in females was higher than that of males, and in study done show that the number of deaths in males was higher than that of females. This is may be due to immunological differences in biological of male and female. The Reason is still unclear behind the gender disparity in the incidence of COVID-19. Symptoms of the disease in men are stronger than in women, this is may be due to the reason for smoking and the work environment to which they are exposed. Differences between age and gender are closely related in places where health protection measures are applied.

4. CONCLUSION

Recognition vulnerable individual to COVID-19 is crucial to minimizing the impact of this pandemic and control the epidemic of the virus. Properly confirmation the correlation between age and sex in COVID-19 larger individual data recommended it.

CONSENT

It is not applicable.

ETHICAL APPROVAL

As per international standard or university standard written ethical approval has been collected and preserved by the authors.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Novel Coronavirus Pneumonia Emergency Response Epidemiology Team. The epidemiological characteristics of an outbreak of 2019 novel coronavirus diseases (COVID-19)—China, 2020. *China CDC Weekly*. 2020;2:113–22.
2. Dingtao Hu, Xiaoqi Lou. Influence of age and gender on the epidemic of COVID-19. *Wien Klin Wochenschr*. 2021;133:321–330.
3. Epidemiological characteristics of coronavirus disease 2019 (COVID-19) patients in IRAN: A single center study. Mohamad Nikpouraghdam, Alireza Jalali Farahani. 2020;127:104378.
4. Jasper Fuk-Woo Chan, Cyril Chik-Yan Yip. 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. *Journal of clinical microbiology*; 2020. DOI: 10.1128/JCM.00310-20
5. Sex and age differences in COVID-19 mortality in Europe. Linda Juel Ahrenfeldt, Martina Otavova, Springer-Verlag GmbH Austria, part of Springer Nature. Published online; 2020.
6. Jian-Min Jin, Peng Bai. Gender differences in patients with COVID-19: Focus on severity and mortality. *Front. Public Health*; 2020.
7. Guan WJ, Ni ZY, Hu Y, Liang WH, Ou CQ, He JX, et al. Clinical characteristics of coronavirus disease 2019 in China. *N Engl J Med*. 2020;382(18):1708–20.
8. Joseph P. Dudley and Nam Taek Lee. Disparities in age-specific morbidity and mortality from SARS-CoV-2 in China and the Republic of Korea. *Clinical Infectious Diseases*; 2020.
9. Tomáš Sobotka, Zuzanna Brzozowska. Age, gender and COVID-19 infections. medRxiv preprint; 2020. DOI: <https://doi.org/10.1101/2020.05.24.20111765>
10. Novel Coronavirus Pneumonia Emergency Response Epidemiology Team. The epidemiological characteristics of an outbreak of 2019 novel coronavirus diseases (COVID-19)—China, 2020. *China CDC Weekly*. 2020;2:113–22.
11. Ivan Arisi, Elide Mantuano. Age and gender distribution of COVID-19 infected cases in Italian population. *Research Square*. Rome, Italy. 2020;295:00161.
12. Drosten C, Gunther S, Preiser W, van der Werf S, Brodt HR, Becker S, et al. Identification of a novel coronavirus in patients with severe acute respiratory syndrome. *N Engl J Med*. 2003;348:1967–76. DOI: 10.1056/NEJMoa 030747
13. Zaki AM, van Boheemen S, Bestebroer TM, Osterhaus AD, Fouchier RA. Isolation of a novel coronavirus from a man with pneumonia in Saudi Arabia. *N Engl J Med*. 2012;367:1814–20.

- DOI: 10.1056/NEJMoa 1211721
14. Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, et al. A novel coronavirus from patients with Pneumonia in China, 2019. *N Engl J Med.* 2020;382:727– 33.
DOI: 10.1056/NEJMoa2001017
 15. Chan JF, Kok KH, Zhu Z, Chu H, To KK, Yuan S, Yuen KY. Genomic characterization of the 2019 novel human-pathogenic coronavirus isolated from a patient with atypical pneumonia after visiting Wuhan. *Emerg Microbes Infect.* 2020;9:221–236.
Available:<https://doi.org/10.1080/22221751.2020.1719902>
 16. Tahamtan A, Ardebili A. Real-time RT-PCR in COVID-19 detection: issues affecting the results. *Expert review of molecular diagnostics.* 2020;20(5):453-454.
 17. Zitek T. The appropriate use of testing for COVID-19. *Western Journal of Emergency Medicine.* 2020;21(3):470.
 18. Zarulli V, Barthold Jones JA, Oksuzyan A, Lindahl-Jacobsen R, Christensen K, Vaupel JW. Women live longer than men even during severe famines and epidemics. *Proc Natl AcadSciU SA.* 2018;115(4):E832–E40.
 19. Karlberg J, Chong DS, Lai WY. Do men have a higher case fatality rate of severe acute respiratory syndrome than women do? *Am J Epidemiol.* 2004;159:229–31.
DOI: 10.1093/aje/kwh056
 20. Zhou F, Yu T, Du R, et al. Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. *Lancet.* 2020;395(10229):1054–62.
Available:[https://doi.org/10.1016/S0140-6736\(20\)30566-3](https://doi.org/10.1016/S0140-6736(20)30566-3). Erratum in: *Lancet.* 2020 March 28;395(10229):1038. Erratum in: *Lancet.* 2020 March 28;395(10229):1038.
 21. Du RH, Liang LR, Yang CQ, et al. Predictors of mortality for patients with COVID-19 pneumonia caused by SARS-CoV-2: A prospective cohort study. *Eur Respir J.* 2020;55(5):2000524.
Available:<https://doi.org/10.1183/13993003.00524-2020>.
 22. Promislow DEL. A geroscience perspective on COVID-19 mortality. *J Gerontol A Biol Sci Med Sci.* 2020;75(9):e30–3.
Available:<https://doi.org/10.1093/gerona/glaa094>

© 2021 Abbas and Hasan; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:

The peer review history for this paper can be accessed here:
<https://www.sdiarticle4.com/review-history/70408>