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EPIDEMIOLOGICAL, CLINICAL CHARACTERISTICS AND POSSIBLE TREATMENT OF CORONAVIRUS DISEASE (COVID-19) DURING EARLY SITUATION IN POPULATION OF DISTRICT KARAK, KHYBER PAKHTUNKHWA, PAKISTAN



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Abstract

Globally, the coronavirus is a persistent health problem. Numerous risk factors for mortality in COVID-19 have been identified. The purpose of this study was to examine the epidemiological, clinical characteristics and treatment base regimen of hospitalized patients during the COVID-19 outbreak. For the screening clinic, swab specimens of the oropharynx and nasopharyngeal were collected in a single viral-transport medium tube and transported to the Khyber Medical University Laboratory for the quantitative identification by Real-Time PCR. All the data were analyzed through a Microsoft Excel worksheet. In a total of 435 patients, n=156 were COVID-19 positive of which n=110 (71%) were male and a high positivity rate were found in between 30 to 50 years age group. High transmission rates n=69 (44%) was found among the local population. The majority of the patients came with fever n=105 (67%) and n=89 (57%) was suffering from cough as second most common sign. Hypertension was the most common underlying condition as compared to the others. The highest mortality rate was found in female (15%) in age group >71. Azithromycin was the most common treatment regimen as compared to the others. The current study concluded that a high rate of positivity was found in the male in age group between 30 to 50. Contact with the local population, direct contact, contact with a family member, and unknown sources were the main routes of transmission. The most presented clinical sign was fever. Hypertension, diabetes, and cardiac disease were co-morbidities in most of the COVID-19 positive patients. The current study also concluded that azithromycin and hydroxychloroquine were the most commonly used treatment regimen during an early pandemic.

Keywords: Epidemiological, Clinical Characteristics, Real Time PCR, Coronavirus disease, Karak District

INTRODUCTION

Chinese authorities reported an occurrence of pneumonia-like respiratory illness in December 2019 (1). It was recognized as the sixth public health emergency by the World Health Organization (WHO) on January 30, 2020 (2). In March 2020, it was declared a global pandemic (3, 4). The WHO designated this viral pneumonia Corona Virus Disease-19 (COVID-19) on February 11, 2020 (5). The metagenomics study was conducted using Bronchoalveolar lavage samples obtained from infected individuals (6), and by Center of Disease Control and Prevention (CDC) in US called it 2019 novel coronavirus (7). SARS and COVID-19 share about 88% of their genomic sequences. SARS viruses' bat-SL-CoVZXC21 and bat-SL-CoVZC45 were both

coronaviruses originated from bats (8). Both uses similar receptor (9). The new coronavirus has been classified as the coronavirus family's seventh member (10).

In China, the coronavirus was identified on 8th December 2019, according to numerous epidemiological studies (11-13). Later, the disease expanded to other parts of the world, and on 11th March 2020 it was declared a pandemic (14). Pakistan detected first case of coronavirus at the end of February 2020 (15, 16). It is highly spreadable and can easily be transmitted through person-to-person contact (17).

The coronavirus is wrapped in an enormous RNA genome that has positive strands and alters extremely quickly due to defects in RNA (10, 18). Because of its regular alteration, it is extremely infectious and can be detected in a variety of animals (19-21). According to one Indian estimate, COV-19 infections will continue to rise due to increased transmission rates and seasonal frequencies (22, 23). Numerous mathematical models have proposed that by using preventive measures such as social separation, isolation, and contact tracing, the virus's transmission could be slowed (24, 25). It remains asymptomatic in some individuals which can be a source of disease career (26-29).

COV-19 is a member of a virus family that can produce a variety of symptoms including pneumonia, fever, dyspnea, and pulmonary edema (30). Clinical symptoms include fever, cough, discomfort, muscle aches, and fatigue, as well as normal or low WBC levels and radiological signs of pneumonia. Severe cases may result in organ failure or death, but severity depends on age, sexual identity, and co-morbidities. As with COV-19 and highly virulent influenza, SARS-CoV-2 is likely to be transmitted by respiratory droplets (31). In terms of transmission and pandemic risk, the coronavirus was estimated to have an effective reproducibility number (R) (2.9) that is greater than the effective reproducibility number (R) reported for SARS (1.77). COVID-19 is currently believed to have an average incubation period of 7 days, ranging from 2 to 14 days, rather than the 4 or 6 days previously estimated (32).

The symptoms of COVID-19 infection match with those of other viruses, making a clinical diagnosis extremely difficult. For the detection of COVID-19, reverse transcription polymerase chain reaction diagnostic test is used which is mainly carried out for the identification of viral sequences (33). The focus of this research was to demonstrate the epidemiological and clinical characteristics of patients diagnosed with coronavirus after confirmation through RT-PCR.

MATERIALS AND METHODS

STUDY AREA

The research was carried out from March to July, 2020 in the District Head Quarter (DHQ) Hospital, District Karak, KPK, Pakistan. All the data were obtained from DHQ hospital, which offered in-patient treatment for COVID-19 patients. This approved hospital received a significant number of COVID-19 patients from district Karak. Using a predesigned data collection tool, data about gender, age, medical history like cardiac disease, diabetes, stroke, asthma, and stroke were recorded. Patients were admitted on the basis of their clinical signs and symptoms like fever, sore throat, shortening of breath (SOB), malaise and fatigue. Routine care, provision of care according to case management guidelines and clinical outcome (dead or discharged alive) was also recorded. To ensure the privacy and confidentiality of participants, data were kept in a locker and soft data were secure with password in a computer. Additionally, a unique identifier was issued to each participant.

DATA COLLECTION

For the screening clinic, under transmission-based precaution, combined swab specimens of their oropharynx and nasopharyngeal were collected in a single viral-transport medium tube. Using a nylon-flocked swab, swabs were collected at least three times from each tonsil region of the posterior pharynx, omitting the tongue. The samples were instantly placed in a sterile tube having 2-3 ml of viral transport media. All samples were packed and transported to the Khyber Medical University Laboratory following the standard protocol.

LABORATORY TESTS

For the quantitative detection of COVID-19, nasal and oropharyngeal swabs were collected from suspected patients. A real time RT-PCR was then carried out as per manufacture guidelines. It includes primers, control material, and manual nucleic acid extraction. RdRp was also targeted with oligonucleotide primers and probes to detect SARS-CoV-2. cDNA was synthesized by reverse transcription of RNA from oropharynx and nasopharyngeal, real-time PCR was used to amplification of the result in accordance with the recommended temperature. The amount of whole-genome RNA of SARC-COV-2 was 0.33 copies/ μ l. Within six to eight hours of collection, samples were processed, and reports were sent. The accuracy of the RT-PCR test results was double-checked by running an internal control alongside each patient sample, as well as one replicate of each positive and negative control. A positive test result was one with a Ct-value less than 37, while a negative test result was one with a Ct-value greater than 40. Retesting was required for confirmation of a medium load, which was defined as a Ct value of 37 to less than 40. Isolation and amplification/detection of nucleic acids were performed on each control sample. The two SARS-CoV-2 targets, as well as the internal control target, all had control results (detection cycles or Ct). An acceptable run required acceptable results for the SARS-CoV-2 as well as the internal control.

STATISTICAL ANALYSIS

Percentages and counts were used to summarize categorical variables. Only descriptive statistics are considered. Microsoft excel worksheet was used for all analyses.

RESULTS

A total of 435 patients in the month of March-July, 2021 were tested for COVID-19 RT-PCR test because of their suspected sign and symptoms like fever, sore throat, cough, shortening of breath (SOB), diarrhea, fatigue, and others in DHQ, Hospital, Karak, Pakistan. Out of 435 subjects, n=156 were SARC-COV positive of which n=110 (71%) were male and n=46 (29%) subjects were female ($P < 0.01$) (Fig. 1).

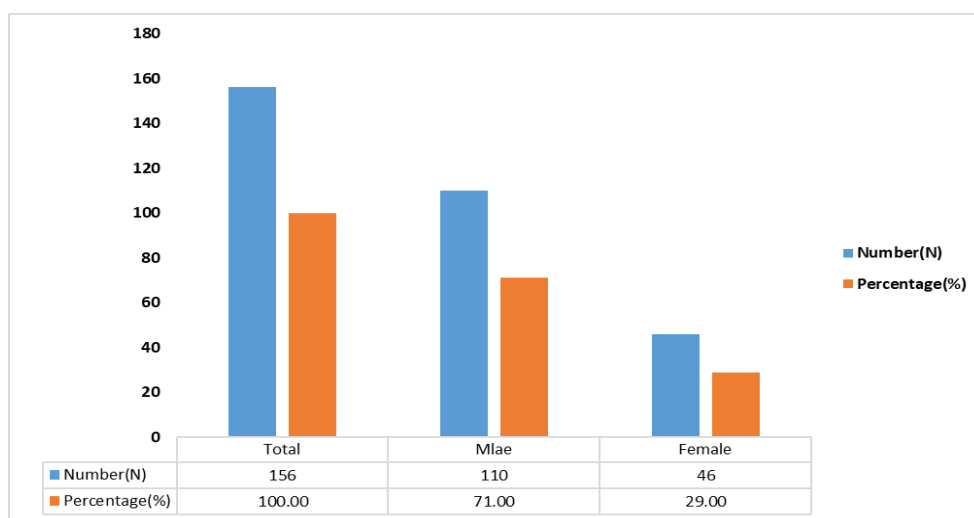


Fig. 1. Gender wise distribution of COVID-19 positivity rate

COVID-19 positivity rate was also checked among different age groups of patients. Age was categories into seven groups: <19 (n=7), 19-29 (n=31), 30-40(n=47), 41-50 (n=27), 51-60 (n=19), 61-70 (n=15) and >71 (n=10). High positivity rate (30%) was found in age group 30-40 followed by in age group of 19-29 (20%), 17% in 41-50, 12% in 51-60, 10% in 61-70, 7% in >71 and 4% in <19 age group (Fig. 2).

In current research work, the transmission rate of COVID-19 was also studied. In total of 156 COV-19 positive patients, the high transmission rate of 44% (n=69) was found among the local population as compared to another source of transmission; unknown source 29% (n=45), direct contact 19% (n=30) and family member contact 8% (n=12) (Fig. 3).

Clinical signs and symptoms were also observed in COVID-19 positive patients. Most of the patients were presented with fever n=105 (67%) and cough n=89 (57%) followed by shortening of breath (SOB) n=75 (48%), sore-throat n=60 (38%), fatigue n=44 (28%) and diarrhea n=21 (13%) (Fig. 4).

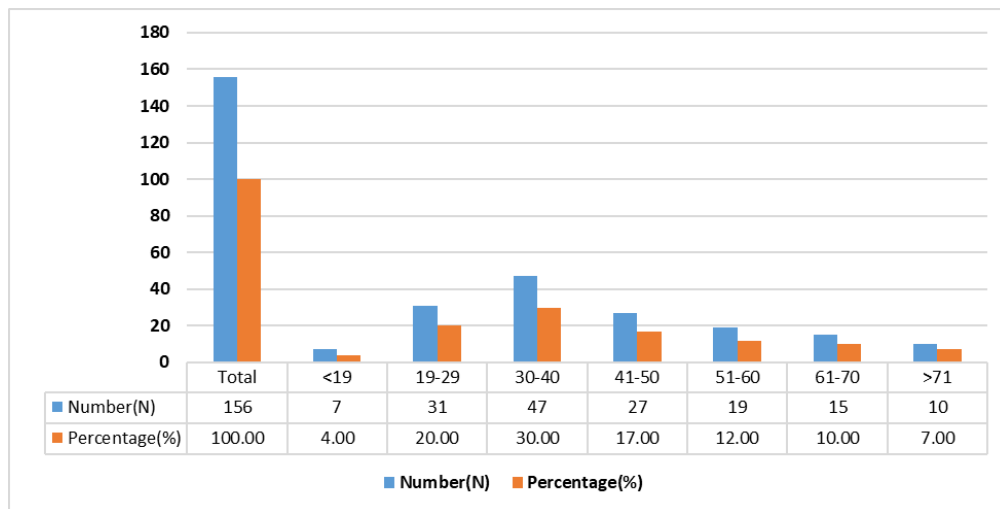


Fig. 2. COVID-19 positivity rate in different age groups

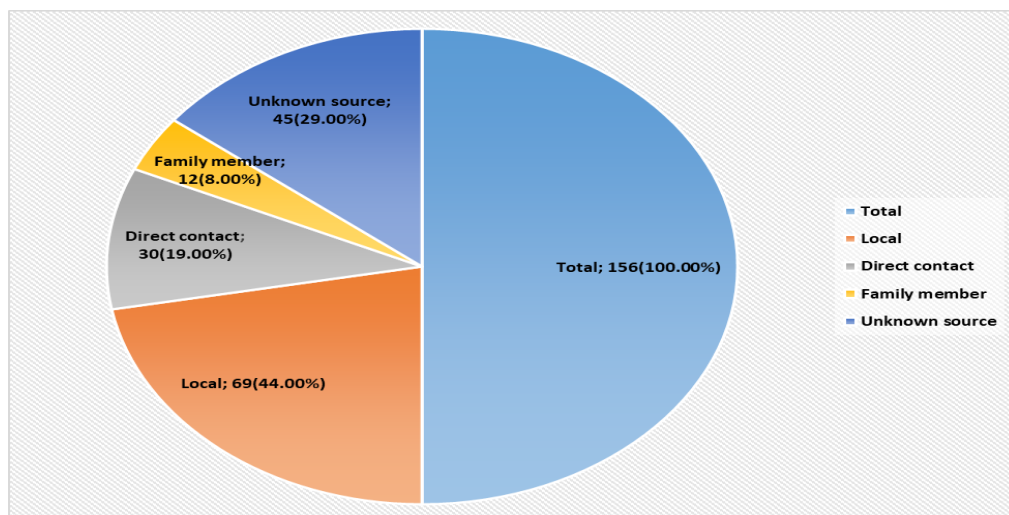


Fig. 3. Different sources of COVID-19 transmission

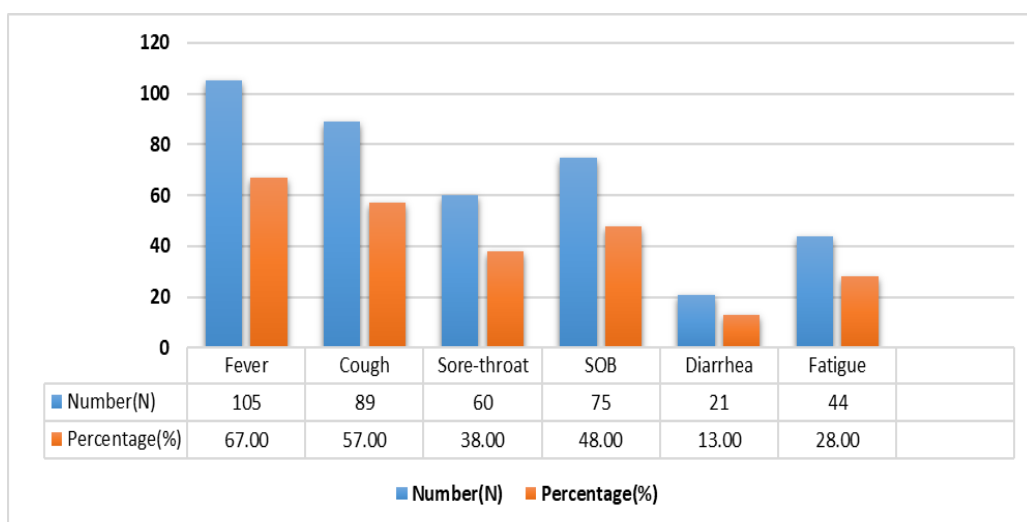


Fig. 4. Percentage of clinical signs and symptoms of COVID-19 positive patients

In the current study, medical history was also taken from COV-19 positive patients. Out of 156 patients, n=90 patients reported medical history of hypertension, diabetes, cardiac disease, asthma, and others during/at the time of admission. Hypertension n=30 (34%) was the most presented underlying condition as compared to diabetes n=23 (26%), cardiac disease n=16 (17%), asthma n=12 (13%) and others n=9 (10%) (Fig. 5).

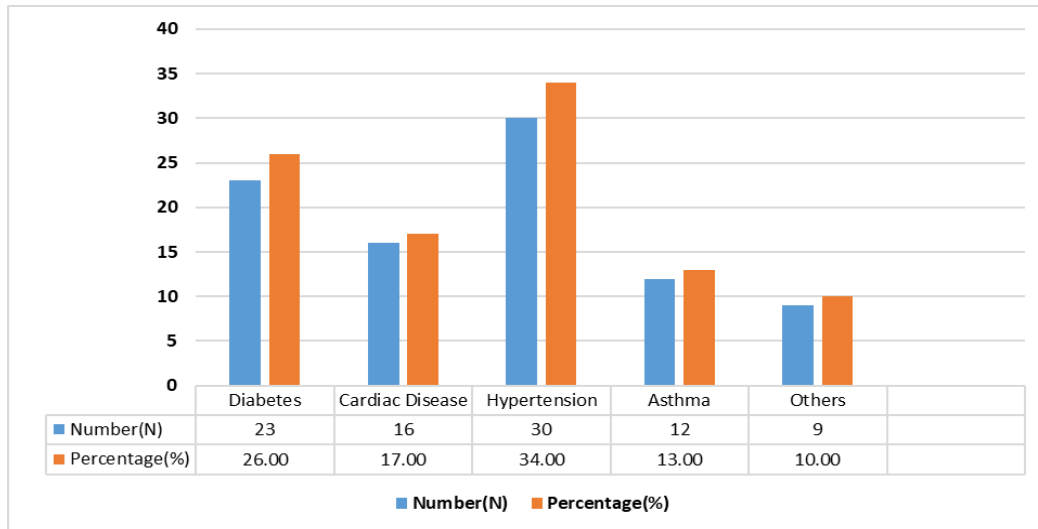


Fig. 5. History of underlying medical conditions

Mortality rates were also studied among gender and different age groups during the treatment of COV-19 positive patients. Out of n=156 patients, n=23 (15%) patients died during the treatment in which mortality rate was higher i.e., n=7 (15%) out of n=46 (29%) in female and n=16 (14%) out of n=110 (71%) in male patients. Most of the patients n=7 (30%) died were from the age group of >71 years followed by n=6 (26%) in 61-70 years as compared to all other age groups (Table I).

Table I. Mortality rate among different age groups and genders

Age Group	Male		Female	
	Cases (%)	Death (%)	Cases (%)	Death (%)
Total	110 (71)	16 (14)	46 (29)	7 (15)
<19	5 (71)	-	2 (29)	-
19-29	20 (65)	-	11 (35)	-
30-40	35 (74)	1 (3)	12 (26)	-
41-50	18 (67)	3 (17)	9 (33)	1 (11)
51-60	12 (63)	3 (25)	7 (37)	2 (28)
61-70	12 (80)	5 (42)	3 (20)	1 (33)
>71	5 (50)	4 (80)	5 (50)	3 (60)

Most frequently prescribed treatment regimens (Azithromycin, Salbutamol, Hydroxy-Chloroquine, Oxygen, Steroid, and other regimen) were also studied in the current study. Azithromycin was the most frequently prescribed medicine (prescribed to n=132, 85% patients) followed by hydroxy-chloroquine (prescribed to n=90, 58% patients), oxygen was given to n=80 (51%), steroid n=72 (46%), other regimens n=66 (42%) and salbutamol n=57 (37%) patients (Table II).

Table II. Treatment Regimen during early Covid-19 pandemic

Treatment	Regimen N (%)	Alive N (%)	Dead N (%)
Azithromycin	123(93)	9(7)	132(85)
Salbutamol	55 (96)	2 (4)	57 (37)
Hydroxy-Chloroquine	88 (98)	2 (2)	90 (58)
Oxygen	75 (94)	5 (6)	80 (51)
Steroid	68 (94)	4 (6)	72 (46)
Other Regimen	65 (98)	1 (2)	66 (42)

DISCUSSION

The outbreak began in Pakistan, as many people from Iran, Saudi Arabia, and Europe came in Pakistan with positive COVID-19 and spread the disease throughout the country. On 26-Feb-2020; Pakistan confirmed the first positive case, involving a patient travelling from Iran to Karachi, Pakistan, with his family. A total of 156 COV-19 positive patients were admitted to District Head Quarter Hospital (DHQ), Karak in this study during the months of June-August 2020 after confirmation of positive RT-PCR results. The current study showed a high positivity rate of COV-19 in males n=110 (71%) compared to females n=46 (29%). Age-wise prevalence rates were also observed in a current study showing a high positivity rate of 30% in the age group 30-40 years followed by 20% in 19-29 years age group. Our results are consistent with the results of a previous study (34). Our findings are consistent with a study conducted in China, where the percentage of infected females was lower than the percentage of infected males (31). According to some research, the majority of patients were between the age of 25-89 years (35), with fewer cases observed in children and newborns too (36). A recent study conducted by Ul-Haq et al., (2020) showed a high positivity rate of COV-19 in males n=150 (83.8%) as compared to females n=29 (16.2%) and a high positivity rate of n=60 (33.90%) was found in the age group of 40 years or below (37). According to the 1998 census, the country's population demographics indicate that almost 40% of the population is adults, while 53% of the population is under the age of 19 and 5.54% of the population is over the age of 60 years (38).

Source of transmission of COV-19 were also observed in a current study showing the highest transmission rate 69(44%) through the local population as compared to other sources of transmission; unknown source 45 (29%), direct contact 30 (19%) and family member contact 12(8%). The first source of transmission as entering of first COV-19 positive patient from Iran to Pakistan and its contact with local population became a source of COV-19 pandemic in Pakistan (39). According to Ahmad et al., 2020, viral spread inside Punjab province as high through the local population as compared to other sources of transmission (40). Another study also shows high transmission rate through human to human in the local population which show similar findings to our results (41).

Clinically, most of the patients were presented with fever 105(67%), cough 89(57%) were second most common sign symptoms followed by shortening of breath (SOB) 75(48%), sore-throat 60(38%), fatigue 44(28%) and diarrhea 21(13%). Clinically, most of the patients were presented with fever 105(67%), cough 89(57%) were second most common sign symptoms followed by shortening of breath (SOB) 75(48%), sore-throat 60(38%), fatigue 44(28%) and diarrhea 21(13%). A recent study also shows most frequent signs and symptoms in COV-19 positive patients were fever, cough, fatigue, dyspnea, and gastrointestinal discomfort (42-45). Another study conducted in Karachi hospital showed clinical signs symptoms of enrolled patients included dry cough, fever, lethargy, fatigue, vomiting and diarrhea (46). A study conducted by Ahmad et al., 2021 also show consistent results with our findings (39).

In the current study medical history were also studied among COV-19 positive patients, Hypertension 30(34%) were the most presented underlying condition as compared to diabetes 23 (26%), cardiac disease 16(17%), asthma 12(13%) and others 9(10%). Several studies observed a similar pattern, with hypertension being the most prevalent comorbidity, followed by diabetic, cardiovascular disease, and respiratory problems (47-49). Several other investigations confirmed the pattern described previously, with hypertension, diabetes, and heart disease serving as the primary underlying condition (35, 50).

Mortality rates were also studied among gender and different age groups during the treatment of COV-19 positive patients. A total 23 (15%) patients died during the treatment in which the mortality rate was higher 7(15%) out of 46 (29%) in female and 16(14%) out of 110 (71%) in male patients. In different countries of the world, the rate of mortality in female as compared to male and old-age mortality from COV-19 were higher (51-55).

In the current study, Azithromycin 132(85%) were the most frequently prescribed medicine followed by hydroxy-chloroquine 90(58%), oxygen, steroid, other regimen, and salbutamol. During the early stages of

the COVID-19 epidemic in Pakistan, paracetamol, chloroquine, and cetirizine were the most frequently recommended medications (56-59). Some other studies also show similar findings to our study (60, 61).

CONCLUSION

Globally, the Coronavirus is a persistent health problem that has altered the course of human history in unimaginable ways. It is rapidly spreading around the world, including Pakistan, and is associated with a range of symptoms. The current study conclude that the COV-19 positivity rate was higher in male as compared to female because of their outdoor activity. In the current study majority of patients were reported with clinical sign symptoms like fever, cough, shortness of breath (SOB), fatigue and diarrhea. The current study also concluded that hypertension, diabetes, cardiac disease, Asthma were the most common co-morbidities. High mortality rate was observed in female and in patients with age groups between 60 to >71 years. Azithromycin is the most commonly used treatment regimen followed by Hydroxy-chloroquine, oxygen, steroid and salbutamol. To prevent broad transmission throughout the community, close surveillance and large-scale isolation characterized by high methods will be required. Additional tests, specimens, and procedures may be explored for the COVID-19 epidemic.

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Conflict of Interest:

Authors found no conflict of interest during the study.

References:

1. Li Q, Guan X, Wu P, Wang X, Zhou L, Tong Y, Ren R, Leung KS, Lau EH, Wong JY, Xing X. Early transmission dynamics in Wuhan, China, of novel coronavirus–infected pneumonia. *New England Journal of Medicine*. 2020;382(13):1199-1207.
2. Bilgin S, Kurtkulagi O, Kahveci GB, Duman TT, Meryem B. Millennium pandemic: A review of coronavirus disease (COVID-19). *Experimental Biomedical Research*. 2020;3(2):117-26.
3. Cucinotta D, Vanelli MJABMAP. WHO declares COVID-19 a pandemic? *Acta Bio Medica: Atenei Parmensis*. 2020;91(1):157.
4. Tam PC, Ly KM, Kernich ML, Spurrier N, Lawrence D, Gordon DL, Tucker EC. Detectable severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in human breast milk of a mildly symptomatic patient with coronavirus disease 2019 (COVID-19). *Clinical Infectious Diseases*. 2021;72(1):128-30.
5. Rodriguez-Morales AJPPAM. Katterine Bonilla-Aldana D., Tiwari R., Sah R., Rabaan AA, Dhama K. Covid-19, an emerging coronavirus infection: current scenario and recent developments-an overview. *Journal of Pure and Applied Microbiology*. 2020;14(1):5-12.
6. Wu Y, Wang T, Guo C, Zhang D, Ge X, Huang Z, Zhou X, Li Y, Peng Q, Li J. Plasminogen improves lung lesions and hypoxemia in patients with COVID-19. *An International Journal of Medicine*. 2020;113(8):539-45.
7. Paules CI, Marston HD, Fauci AS. Coronavirus infections—more than just the common cold. *Jama*. 2020 Feb 25;323(8):707-8.
8. Lu R, Zhao X, Li J, Niu P, Yang B, Wu H, Wang W, Song H, Huang B, Zhu N, Bi Y. Genomic characterization, and epidemiology of 2019 novel coronavirus: implications for virus origins and receptor binding. *The lancet*. 2020;395(10224):565-74.
9. Kumar A, Narayan RK, Kumari C, Faiq MA, Kulandhasamy M, Kant K, Pareek V. SARS-CoV-2 cell entry receptor ACE2 mediated endothelial dysfunction leads to vascular thrombosis in COVID-19 patients. *Medical hypotheses*. 2020; 145:110320.
10. Barua R, Datta S, Roychowdhury A, Datta P. The study of the epidemiology and clinical features of the novel coronavirus (COVID-19). *Epidemiological Research Applications for Public Health Measurement and Intervention: IGI Global*. 2021;25-39.

11. Covid CD, Team R, COVID C, Team R, COVID C, Team R, Bialek S, Boundy E, Bowen V, Chow N, Cohn A. Severe outcomes among patients with corona virus disease 2019 (COVID-19)—United States, February 12–March 16, 2020. *Morbidity and mortality weekly report*. 2020;69(12):343.
12. Khan S, Siddique R, Bai Q, Liu Y, Xue M, Nabi G, Liu J. Coronaviruses disease 2019 (COVID-19): causative agent, mental health concerns, and potential management options. *Journal of infection and public health*. 2020;13(12):1840-4.
13. Waris A, Atta UK, Ali M, Asmat A, Baset AJ. COVID-19 outbreak: current scenario of Pakistan. *New Microbes and New Infections*. 2020;35:100681.
14. Weber A, Ianelli F, Goncalves S. Trend analysis of the COVID-19 pandemic in China and the rest of the world. *arXiv preprint arXiv:2003.09032*. 2020 Mar 19.
15. Mehmood K, Bao Y, Abrar MM, Petropoulos GP, Soban A, Saud S, Khan ZA, Khan SM, Fahad S. Spatiotemporal variability of COVID-19 pandemic in relation to air pollution, climate and socioeconomic factors in Pakistan. *Chemosphere*. 2021;271:129584.
16. Baloch GM, Sundarasan S, Chinna K, Nurunnabi M, Kamaludin K, Khoshaim HB, Hossain SF, AlSukayt A. COVID-19: exploring impacts of the pandemic and lockdown on mental health of Pakistani students. *PeerJ*. 2021;9:e10612.
17. Pongpirul WA, Pongpirul K, Ratnarathon AC, Prasithsirikul W. Journey of a Thai taxi driver and novel coronavirus. *New England Journal of Medicine*. 2020;382(11):1067-8.
18. Drexler JF, Gloza-Rausch F, Glende J, Corman VM, Muth D, Goettsche M, Seebens A, Niedrig M, Pfefferle S, Yordanov S, Zhelyazkov L. Genomic characterization of SARS-related coronavirus in European bats and classification of coronaviruses based on partial RNA-dependent RNA polymerase gene sequences. *Journal of virology*. 2010;84(21):11336-49.
19. Shi J, Wen Z, Zhong G, Yang H, Wang C, Huang B, Liu R, He X, Shuai L, Sun Z, Zhao Y. Susceptibility of ferrets, cats, dogs, and other domesticated animals to SARS–coronavirus 2. *Science*. 2020;368(6494):1016-20.
20. Liu H, Zhang Q, Wei P, Chen Z, Aviszus K, Yang J, Downing W, Jiang C, Liang B, Reynoso L, Downey GP. The basis of a more contagious 501Y. V1 variant of SARS-CoV-2. *Cell Research*. 2021;31(6):720-2.
21. Zahradník J, Marciano S, Shemesh M, Zoler E, Harari D, Chiaravalli J, Meyer B, Rudich Y, Li C, Marton I, Dym O. SARS-CoV-2 variant prediction and antiviral drug design are enabled by RBD in vitro evolution. *Nature microbiology*. 2021;6(9):1188-98
22. Khajanchi S, Sarkar K. Forecasting the daily and cumulative number of cases for the COVID-19 pandemic in India. *Chaos: An interdisciplinary journal of nonlinear science*. 2020;30(7):071101.
23. Khajanchi S, Sarkar K, Mondal J, Nisar KS, Abdelwahab SF. Mathematical modeling of the COVID-19 pandemic with intervention strategies. *Results in Physics*. 2021;25:104285.
24. Samui P, Mondal J, Khajanchi S. A mathematical model for COVID-19 transmission dynamics with a case study of India. *Chaos, Solitons & Fractals*. 2020;140:110173.
25. Sarkar K, Khajanchi S, Nieto JJ. Modeling and forecasting the COVID-19 pandemic in India. *Chaos, Solitons & Fractals*. 2020;139:110049.
26. Yang X, Yu Y, Xu J, Shu H, Liu H, Wu Y, Zhang L, Yu Z, Fang M, Yu T, Wang Y. Clinical course and outcomes of critically ill patients with SARS-CoV-2 pneumonia in Wuhan, China: a single-centered, retrospective, observational study. *The Lancet Respiratory Medicine*. 2020;8(5):475-81.
27. Al-Qahtani M, AlAli S, AbdulRahman A, Alsayyad AS, Otoom S, Atkin SL. The prevalence of asymptomatic and symptomatic COVID-19 in a cohort of quarantined subjects. *International Journal of Infectious Diseases*. 2021;102:285-8.
28. Smalley DL, Cisarik PM, Grantham J, Cloud W, Neil RB, DePriest P. Impact of pool testing in detection of asymptomatic patients with COVID-19. *Laboratory Medicine*. 2021;52(1):e15-6.
29. Andreas Kronbichler M, Daniela Kresse M, Sojung Yoon B, Keum Hwa Lee M. Original Research Asymptomatic patients as a source of COVID-19 infections: A systematic review and meta-analysis. *International Journal of Infectious Diseases*. 2020;98:180-186.
30. Yue XG, Shao XF, Li RY, Crabbe MJ, Mi L, Hu S, Baker JS, Liang G. Risk management analysis for novel coronavirus in Wuhan, China. *Journal of Risk and Financial Management*. 2020;13(2):22.
31. Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, Zhang L, Fan G, Xu J, Gu X, Cheng Z. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *The lancet*. 2020;395(10223):497-506.
32. Wu A, Peng Y, Huang B, Ding X, Wang X, Niu P, Meng J, Zhu Z, Zhang Z, Wang J, Sheng J. Genome composition and divergence of the novel coronavirus (2019-nCoV) originating in China. *Cell host & microbe*. 2020;27(3):325-8.

33. Jeong JH, Kim KH, Jeong SH, Park JW, Lee SM, Seo YH. Comparison of sputum and nasopharyngeal swabs for detection of respiratory viruses. *Journal of medical virology*. 2014;86(12):2122-7.
34. Wang D, Hu B, Hu C, Zhu F, Liu X, Zhang J, Wang B, Xiang H, Cheng Z, Xiong Y, Zhao Y. Clinical characteristics of 138 hospitalized patients with 2019 novel coronavirus–infected pneumonia in Wuhan, China. *Jama*. 2020;323(11):1061-9.
35. Guan WJ, Ni ZY, Hu Y, Liang WH, Ou CQ, He JX, Liu L, Shan H, Lei CL, Hui DS, Du B. Clinical characteristics of 2019 novel coronavirus infection in China. *New England Journal of Medicine*. 2020;382(18):1708-1720.
36. Mahalmani VM, Mahendru D, Semwal A, Kaur S, Kaur H, Sarma P, Prakash A, Medhi B. COVID-19 pandemic: A review based on current evidence. *Indian journal of pharmacology*. 2020;52(2):117.
37. Wang C, Wang X. Prevalence, nosocomial infection, and psychological prevention of novel coronavirus infection. *Chin General Pract Nurs*. 2020;18:2-3.
38. UIHaq Z, Shahzad M, Khattak MI, Fazid S, Ullah N, Shireen A, Ulhaq N, Izhar A, Farooq U, Darwesh NM, Asim M. Clinical characteristics, mortality, and associated risk factors in COVID-19 patients reported in ten major hospitals of Khyber Pakhtunkhwa, Pakistan. *Journal of Ayub Medical College, Abbottabad: JAMC*. 2020;32(4): S633-9.
39. Ahmad D, Afzal M, Imtiaz A. Effect of socioeconomic factors on malnutrition among children in Pakistan. *Future Business Journal*. 2020;6(1):1-1.
40. Tuite AR, Bogoch II, Sherbo R, Watts A, Fisman D, Khan K. Estimation of coronavirus disease 2019 (COVID-19) burden and potential for international dissemination of infection from Iran. *Annals of Internal Medicine*. 2020;172(10):699-701.
41. Ahmad M, Beg BM, Majeed A, Areej S, Riffat S, Rasheed MA, Mahmood S, Mushtaq RM, Hafeez MA. Epidemiological and clinical characteristics of covid-19: A retrospective multi-center study in pakistan. *Frontiers in Public Health*. 2021;9:342.
42. Shereen MA, Khan S, Kazmi A, Bashir N, Siddique R. COVID-19 infection: Emergence, transmission, and characteristics of human coronaviruses. *Journal of advanced research*. 2020;24:91-8.
43. Schettino M, Pellegrini L, Picascia D, Saibeni S, Bezzio C, Bini F, Omazzi BF, Devani M, Arena I, Bongiovanni M, Manes G. Clinical characteristics of COVID-19 patients with gastrointestinal symptoms in Northern Italy: a single-center cohort study. *Official journal of the American College of Gastroenterology | ACG*. 2021;116(2):306-10.
44. Qian GQ, Yang NB, Ding F, Ma AH, Wang ZY, Shen YF, Shi CW, Lian X, Chu JG, Chen L, Wang ZY. Epidemiologic and clinical characteristics of 91 hospitalized patients with COVID-19 in Zhejiang, China: a retrospective, multi-centre case series. *QJM: An International Journal of Medicine*. 2020;113(7):474-81.
45. Hassan SA, Sheikh FN, Jamal S, Ezeh JK, Akhtar A. Coronavirus (COVID-19): a review of clinical features, diagnosis, and treatment. *Cureus*. 2020;12(3).
46. Weng LM, Su X, Wang XQ. Pain symptoms in patients with coronavirus disease (COVID-19): A literature review. *Journal of Pain Research*. 2021;14:147.
47. Asghar MS, Kazmi SJ, Khan NA, Akram M, Khan SA, Rasheed U, Hassan M, Memon GM. Clinical profiles, characteristics, and outcomes of the first 100 admitted COVID-19 patients in Pakistan: a single-center retrospective study in a tertiary care hospital of Karachi. *Cureus*. 2020;12(6).
48. Yang J, Zheng YA, Gou X, Pu K, Chen Z, Guo Q, Ji R, Wang H, Wang Y, Zhou Y. Prevalence of comorbidities and its effects in patients infected with SARS-CoV-2: a systematic review and meta-analysis. *International journal of infectious diseases*. 2020;94:91-5.
49. Naveed M, Naeem M, ur Rahman M, Hilal MG, Kakakhel MA, Ali G, Hassan A. Review of potential risk groups for coronavirus disease 2019 (COVID-19). *New Microbes and New Infections*. 2021;41:100849.
50. Emami A, Javanmardi F, Akbari A, Kojuri J, Bakhtiari H, Rezaei T, Keshavarzi A, Falahati F. Survival rate in hypertensive patients with COVID-19. *Clinical and experimental hypertension*. 2021;43(1):77-80.
51. Zhang JJ, Dong X, Cao YY, Yuan YD, Yang YB, Yan YQ, Akdis CA, Gao YD. Clinical characteristics of 140 patients infected with SARS-CoV-2 in Wuhan, China. *Allergy*. 2020;75(7):1730-41.
52. Zhou F, Yu T, Du R, Fan G, Liu Y, Liu Z, Xiang J, Wang Y, Song B, Gu X, Guan L. Clinical course, and risk factors for mortality of adult in patients with COVID-19 in Wuhan, China: a retrospective cohort study. *The lancet*. 2020;395(10229):1054-62.
53. Li H, Wang S, Zhong F, Bao W, Li Y, Liu L, Wang H, He Y. Age-dependent risks of incidence and mortality of COVID-19 in Hubei Province and other parts of China. *Frontiers in medicine*. 2020; 7:190.

54. Chen L, Yu J, He W, Chen L, Yuan G, Dong F, Chen W, Cao Y, Yang J, Cai L, Wu D. Risk factors for death in 1859 subjects with COVID-19. *Leukemia*. 2020;34(8):2173-83.
55. Albitar O, Ballouze R, Ooi JP, Ghadzi SM. Risk factors for mortality among COVID-19 patients. *Diabetes research and clinical practice*. 2020; 166:108293.
56. Wise J. Covid-19: Known risk factors fail to explain the increased risk of death among people from ethnic minorities. *BMJ (Clinical Research ed.)*. 2020;369:m1873.
57. Hassan SM, Sohail H, Iftikhar A, Rathore R, Fatima Q. Pharmacologic agents for the management of COVID-19: a review. *Int J Commun Med Public Health*. 2021;8(446):10-8203.
58. Singh AK, Singh A, Shaikh A, Singh R, Misra A. Chloroquine, and hydroxychloroquine in the treatment of COVID-19 with or without diabetes: A systematic search and a narrative review with a special reference to India and other developing countries. *Diabetes & Metabolic Syndrome: Clinical Research & Reviews*. 2020;14(3):241-6.
59. Platel R, Vaure L, Palleau E, Raffy S, Guerin F, Lagarde D, Cours R, Marcelot C, Warot-Fonrose B, Nayral C, Delpech F. Synthesis of hybrid colloidal nanoparticles for a generic approach to 3D electrostatic directed assembly: application to anti-counterfeiting. *Journal of Colloid and Interface Science*. 2021; 582:1243-50.
60. Faria CG, Weiner L, Petriguet J, Hingray C, Santamaria ÁR, Villoutreix BO, Beaune P, Leboyer M, Javelot H. Antihistamine and cationic amphiphilic drugs, old molecules as new tools against the COVID-19? *Medical hypotheses*. 2021;148:110508.
61. Day M. Covid-19: ibuprofen should not be used for managing symptoms, say doctors and scientists. *BMJ (Clinical Research ed.)*. 2020;368:m1086.