THE EVALUATION OF PREVALENCE AND HEMATOLOGICAL PARAMETERS FOR ASSESSMENT OF SEVERITY OF CORONAVIRUS INFECTION

Abstract

Objectives: We determined the prevalence of coronavirus (COVID-19 virus) infection on the basis of age, sex, clinical presentation and risk of ABO Blood Groups antigens with COVID-19 virus. We also highlight the Hematological Parameters such as neutrophils to lymphocytes ratio and thrombocytopenia to assess severity of COVID-19 viral infection.

Methodology: A descriptive study was conducted from May 2020 to May 2021 at rural and urban areas of District Shaheed BenazirAbad in the homes of patients diagnosed with Covid-19 viral infection where they were present in quarantine. A total of 351 patients with their ages ranging between 12 and 68 including 329 males and 21 females present in isolation within their homes with diagnosis of coronavirus infection were included in this study. The clinical features of all these patients were noted and their 5 ml blood samples mixed with EDTA were sent to the diagnostic and research pathology laboratory of PUMHS Nawabshah for determination of Blood Groups and complete blood count showing hematological parameters.

Results: The mean age of these patients was 40 ± 28 years while male to female ratio was 15.6:1. Out of the 351 patients, persons bearing Blood Group A was present in 169 patient (45.3%) followed by Blood Group B in 117 patient (33.8%), Blood Group AB in 50 patient (11.3%) and O Blood Group in 26 patients (8.6%) respectively were founded. In this study, the mean value of neutrophil to lymphocyte ratio was found to be 4.7 ±1.7 while the mean value of platelets count was 210000 ± 95000/ mm^3 in these patients.

Conclusion: The ratio of coronavirus infection among the patients with Blood Group A was higher than the B, AB and O Blood Group in the males at the age of 12 to 68 years. The increased NLR ratio and thrombocytopenia indicate bad prognosis as well as more severe disease requiring urgent treatment.

Keyword: Prevalence, Coronavirus Infection (COVID-19), Blood Groups, hematological parameters Assessment.

Comment [ON1]: Rephrase this statement to read as:
"The clinical features of the patients were noted and 5 ml of blood was collected in an EDTA bottle from each patient for determination of blood group and complete blood account at the diagnostic and research pathology laboratory of PUMHS, Nawabshah.

Comment [ON2]: Rephrase this statement as: "169 (45.3%) were blood group A, 117 (33.8%) were blood group B, 50 (11.3%) were blood group AB and 26 (8.6%) were blood group O."

Comment [ON3]: Expunge "In this study," and start the sentence with "The mean..

INTRODUCTION

The word corona is derived from Italian language that means crown, therefore spike-like glycoprotein on the surface of the virus gives rise crown appearance to the corona virus so it is called corona virus that belongs to be family of coronaviridae and sub-family coronaviridae divided into alpha-beta, Gamma and Delta coronavirus[1]. Corona Viruses are enveloped viruses with a large plus-strand RNA genome being 27-32kb in size capped and polyadenylated and range from 80 to 160 nm in diameter. The genome of coronavirus composed of 5 and 3 untranslated region (UTR) and open reading frame (ORF) la/b structural genes present at 3 terminus encodes for the structural portents including spike (s) envelope (E) membrane (m) and nucleocapsid which are common features to all Coronavirus [2]. When the corona virus transmitted in to the human or animal, the lifecycle of the virus are the started and the ACE2 receptor is an intra-membrane receptor on the II pneumocystis, where the virus is able to replicate after getting endocytosis with in the cytoplasm as the viral load increases within the alveolar epithelial that will burst releasing the newly replicated viral RNA[3]. Then the virus enters into the another alveolar epithelial cells causes wide destruction of cells and set up the inflammatory reaction with engorgement of blood vessels and release of inflammatory mediators by the damaged epithelial cells that causes neutrophils, macrophages and lymphocytes along with inflammatory exudate that accumulated in alveolar spaces of lungs leading into consolidation or pneumonia[4] . The inflammation within the lung progress to systemic inflammatory response syndrome and the patient with covid 19 will develop symptoms and the incubation period from onset of symptoms of disease to death ranged from 6 to 41 days and during this period, patients presents with Fever, dry cough, shortness of breath, myalgia of fatigue, nasal congestion, chills, night sweats, headache, rhinorrhea, chest pain, nausea and vomiting[5]. Accurate molecular diagnostic tests such as polychromatic chain reaction for detection of ribonucleic acid (R N A) of covid 19 is necessary for confirming a diagnosis of coronavirus disease.[6] Among ABO Blood groups antigens, blood groups A persons are more liable to develop COVID infection then the other blood group antigens such as B, AB and O groups because receptor binding domain of SARS-CoV-2 sequence similarity percent on the blood group A persons of respiratory epithelial cells, leading to SARS-CoV-2 infection in these person [7,8.] The common hematological parameters in COVID-19 infection neutrophilia with lymphopenia, neutrophilia is caused by release of neutrophil-chemoattractant elements and the resulting recruitment of neutrophils are a global host response to viral infection[9]. The COVID-19 infection causes lymphopenia due to the exaggerated inflammatory response known as cytokine storm that is composed of interleukin (IL)-2, IL-7, interferon-γ and tumor necrosis factor-α that may promote destruction of the lymphocytes[10]. From the above discussion our study was designed to evaluate prevalence of COVID infection on the basis of age, sex, clinic-laboratory findings and association of blood group antigens with COVID infection. We also highlighted the hemotologypatmeter of COVID-19 infection to assess the severity of disease.

METHODS AND MATERIALS

A descriptive study was conducted from May 2020 to May 2021 at rural and urban areas of District Shaheed BenazirAbad in the homes of patients with Covid-19 virus where they were present in quarantine. Total 351 patients with their ages ranging between 12 and 68 including 309 males and 41 females present in isolation within their homes with diagnosis of coronavirus infection were included in this study. Patients with diabetes mellitus, hypertension, malaria,

Comment [ON4]: The introduction section dwelt on pathogenesis/pathophysiology of COVID-19. What was already known about the research topic was not highlighted at this section. The section was therefore clear on the aim of the study but not the statement of problem and its justification. The author should

Comment [ON5]: Paraphrase this statement for clarity.

Comment [ON6]: There are so many grammatical errors

Comment [ON7]: Paraphrase this statement as follows:
"The common hematological abnormalities in COVID-19 infection include neutrophilia and lymphopenia. Neutrophilia is caused by the release of neutrophil-chemoattractant elements and the resulting recruitment of neutrophils is a host response to viral infection.[9]

Comment [ON8]: Expunge "From the above discussion our" and replace with "This"

Comment [ON9]: Change clinic-laboratory to clinic-pathological

Comment [ON10]: What is "hemotologypatmeter"?

Comment [ON11]: What is the justification of this aim in a population COVID-19 patients?

Comment [ON12]: The Methods and Materials section must be well-structured into the various sub-sections for easy readability. The sub-sections include:
a)Study site: Where was this study conducted?
b) Study design: What type of study design did you use to carry out this study?
c) Study Population: population size and selection criteria (inclusion and exclusion criteria)- What is the sample size (include the formula with reference). How did you select your sample/study population?
d)Survey Procedure?
e)Specimen collection and laboratory methods
f)Data Management or Statistical Analysis

Comment [ON13]: The numbers of male and female participants from your study do not give the sum total of 351 which was your study population size.
Also, the gender distribution pattern you gave here is different from what you have in your Abstract. Kindly reconcile!

dengue fever, viral hepatitis. pregnant women, and patients who were not giving permission for sampling were excluded from this study.
The clinical features of all these patients such as running nose, fever, dry cough, dyspnea and sore throat were noted. All these patients were already diagnosed by PCR test for coronavirus infections.

5 ml blood samples of the COVID-19 patients were received and out of each 5ml, 3ml was mixed in a bottle containing EDTA while 2ml blood was placed in a plain bottle for detection of blood group. The hematological parameters were detected by hematology analyzer (Nihon Kohden) and ESR was detected by Westergren method; ABO blood groups were done by forward and reverse methods. In the forward method the patient's saline-washed red cells were mixed with known commercially prepared antiserum of anti-A, anti B and anti-D in a test tube; the mixture was incubated at room temperature followed by centrifugation. For the reverse method, the patient's serum was mixed with reagent red cells of groups of A, B and O (available commercially), incubated at room temperature followed by centrifugation. Then a red cell button observed at the bottom of the tube was examined for agglutination.

All data was analyzed using microsoft excel software. All quantitative variables were presented as mean and standard deviation while the qualitative variables were presented as frequency and proportions.

RESULTS

The mean age of these patients was 40 ± 28 years while male to female ratio was 7.5:1. The mean value of hemoglobin, erythrocytes sedimentation rate , total leukocyte count , differential leukocyte count such as percentages of neutrophils, lymphocytes, neutrophil to lymphocyte ratio and platelet count were 12.1 ± 0.1 gm/dl, 50 ± 10 /hr., 13000 ± 1000/cumm, Neutrophils 80 ± 5%, Lymphocyte 10 ± 2%, 4.7 ± 1.7, 210000 ± 95000/cumm respectively. These patients were mildly anemic and their ESR as well as total leukocytes counts were significantly increased that showing infection. There was neutrophilia with lymphopenia and raised neutrophil to lymphocyte ratio that also showed severity of covid infection while there was thrombocytopenia and these values indicate predictors of prognosis of covid infection.
Out of a total of the 351 study participants, 169 (45.3%) were Blood Group A, 117 (33.8%) Blood Group B while 50 (11.3%) and 26 (8.6%) were Blood Groups AB and O respectively. Therefore persons bearing A group were more liable to risk of covid infection then the persons bearing Group O.

Table 1.

DEMOGRAPHY OF CORONAVIRUS INFECTION

N=351

MEAN AGE:	**SEX:**	**Male/Female: Ratio**
40+28	**MALE 309** **FEMALE 42**	**7.5: 1**
CLINICAL PRESENTATION	**Hematological parameters**	**Values**
RUNNING NOSE **HEADACHE 341(97.1%)**	**Hemoglobin** **ESR**	**12.1 ± 0.1 gm/dl** **50 ± 10 /hr.**

Comment [ON14]: ? button or bottle

Comment [ON15]: This was not the M:F ratio shown in your abstract.

Comment [ON16]: How did you test your statistical significance here?
Also, you do not make logical clinical impression while describing the result of a study.

Comment [ON17]: Expunge this. This statement is appropriate at the Discussion session.

Comment [ON18]: Expunge this. This statement is appropriate at the Discussion session.

Comment [ON19]: Table 1 entitled "Demography of Coronavirus infection" should be rearranged. The Age, Gender, Clinical Presentation and Haematological Parameters of study Population should have separate rows and columns on the same table.
Example is shown below:
Table 1:
Characteristics ...

Comment [ON20]:
Characteristics Values (n)
1.Age
2.Gender
3.Clinical Presentation
4.Haematological Parameters

As shown below

FEVER AND BODYACHE 250 (71.2%)	**Total leukocyte count**	**13000 ± 1000/cumm**
DRY COUGH 210(59.8%)	**Differential leukocyte count**	**Neutrophils 80 ± 5 % Lymphocyte 10 ± 2 %**
DYSPNEA 115(32.7%)	**Neutrophil to lymphocyte ratio**	**4.7 ± 1.7**
SORE THROAT 280(79.7%)	**Platelet count**	**210000 ± 95000/cumm**

Characteristics	**Values**
Age	**Mean ± Standard Deviation**
Gender • **Male** • **Female**	
Clinical Presentations	
Haematological Parameters	

TABLE.2

THE DISTRIBUTION OF BLOOD GROUPS AMONG THE PATIENTS WITH CORONAVIRUS INFECTIONS
N=351

BLOOD GROUPS	**NUMBER OF PATIENT**	**PERCENTAGE**
A+VE	**169**	**45.6%**
B+VE	**117**	**33.8%**
AB+VE	**50**	**11.3%**
O+VE	**26**	**8.6%**

Comment [ON21]: This table should stratify the ABO blood groups among the gender categories (Male and Female) and test for
Blood Groups Male Freq Female Freq
As shown below.

Blood Group	Male	Freq (n)	Female	Freq (n)	p-Value

DISCUSSION

The prevalence on the basis of age, sex, clinical presentation and hematological parameters as well as risk of blood groups antigens among the patients with COVID-19 infection were conducted by the international and local studies. According to World Health Organization report in July 2021, about 185 million people were infected by Coronavirus out of which 69.3 million people recovered, 4 million died; while 12 million people were active cases of coronavirus infection. The National command and control system reported that in Pakistan 966000 people were infected with coronavirus, 22460 died, 910000 people recovered and 34,000 active cases remained in Pakistan.[11]

According to Huang, C. et al report in Wuhan city of China, COVID-19 caused viral pneumonia in 41 patients including 30 male and 11 female with their age ranged between 19 and 80 years diagnosed by PCR test and they presented with fever, cough, myalgia and dyspnea.[12] They also reported that the disease is transmitted from humans and other mammals including camels and bats. Chen N et al noted Covid-19 virus infection such as pneumonia among the 99 cases diagnosed by PCR test including 67 male and 32 female with their ages ranging between 42 and 68 years present with similar clinical features reported in above study.[13]

Diagnosed Covid-19 infected cases of 452 patients including 279 males and 173 females with their age ranged between 44 and 72 years, admitted to five leading hospitals in different cities of the Pakistan, were included in the study conducted by Saadia Omer et al.[14] They reported that these patients were suffering from fever, followed by cough, dyspnea, fatigueness, myalgias, generalized body aches, vomiting, headache, diarrhea, sore throat, sputum, production, nausea, loss of taste, rhinorrhea, anosmia and nasal congestion. A total of 412 patients of more than 60% male with their ages ranging between 41 and 42 years who were residents of East Karachi and tested positive for SARS-CoV-2 as reported by Shumaila T et al. [15] recorded three most common symptoms such as fever, cough and running nose.

The study conducted by Noor A et al at Rawalpindi stated that ABO blood grouping was done by test tube method among the 326 patients infected with Covid-19 virus diagnosed by PCR test. They noted that the blood group A was found to be 122 (37.4%) having significant association with COVID infection and mortality but blood group O had least prevalence 71 (21.8%) with COVID-19 infection.[16]

A total of 78 patients with a mean age 45 years including 41 females and 37 males was studied by Yang H.[17] The neutrophilia with lymphopenia and higher neutrophil to lymphocytes ratio were detected in these patients with severe illness caused by Covid-19 infection. Similar studies conducted by various laboratory workers showed elevated levels of neutrophil to lymphocyte ratio among the older males of the Asian and Pakistan populations as compared to their younger age group.[18,19] This ratio was used to assess severity of Covid-19 virus infection that is higher in the western population.[20] Forty-nine (27.5%) out of 178 patients were diagnosed with severe disease of Covid 19 infection, and 129 patients with non-severe disease and the severe disease groups had significantly lower platelet count as reported by Changqian Bao et al.[20]

What are your study strengths and limitations?

Comment [ON22]: Results were not discussed from multiple angle especially from the demographic perspective of COVID-19 bringing to fore age, gender, clinical presentation and haematological indices.
This discussion would have benefited from additional references.
Consider the following:
Nwabuko OC, Nnaji TO. Epidemiology in the News – The case of COVID-19 pandemic disease. *IOSR-JDMS. 2020;* 19(5):35-38

CONCLUSION

We concluded that the ratio of coronavirus infection among the patients with Blood Group A was higher than the B, AB and O Blood Group in the males at the age of 12 to 68 years required prevention of the disease in Group A population to reduce the mortality rate in these patients. Hematological parameters such as neutrophil to lymphocyte ratio and thrombocytopenia were detected in few patients requiring emergency treatment. Other complications of Covid-19 virus infection would be detected to inform the physicians to treat this dangerous disease. Preventive measures and anti viral drugs would likely be used by the physicians. Screening of Covid infection would be necessary especially in those persons who were coming from foreign countries.

Comment [ON24]: What were your study strengths and limitations? Any Recommendation?

Comment [ON25]: Paraphrase this conclusion and include major findings in this result. There is additional need to correct some grammatical errors

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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