

Study of Some Hematological, and Biochemical Parameters in Patients with SARS-CoV-2 in Kirkuk City/Iraq

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ABSTRACT

Background: Coronavirus disease 2019 (COVID-19) or so-called SARS-CoV-2 (severe acute respiratory syndrome coronavirus 2), has been implicated in cases of mild to severe diseases of the respiratory system. The World Health Organization (WHO) declared that SARS-CoV-2 infection is a pandemic infection.

Objectives: Our study aimed at the detection of the relationship of some hematological and biochemical markers with SARS-CoV-2 infection in Kirkuk City/Iraq.

Methods: A cross-sectional study performed at Kirkuk general hospital (May 15 to, August 25 /2020). It was done on a total of (84) patients (50 male and 34 female), who were aging between (16-70) years old. They already diagnosed with SARS-CoV-2 infection through having chest computerized tomography scan (CT-Scan) from (Simens Co.), and 60 non-infected with SARS-CoV-2, who used as a control group at similar age group. Blood samples taken from the patients and control group to test for; serum ferritin, D-dimer, blood urea, serum creatinine, lymphocytes and total white blood cells count.

Results: The study revealed the highest rate (72%, and 42 %) of decreased white blood cells (WBCs) in male and female patients respectively; this was also true for the lymphocytes count where the patients showed the highest rate of decreased lymphocytes count. The majority of the patients showed increased levels of serum ferritin, D-dimer, blood urea, and serum creatinine. Most of the patients were from blood group A.

Conclusions: Infection with SARS-CoV-2 may decrease the levels of WBCs and lymphocytes, while having reverse impact on the serum ferritin, D-dimer, blood urea and serum creatinine levels which increases them. Most of the patients from Kirkuk were from blood type A.

Keywords: SARS-CoV-2, WBCs, Lymphocyte, D-dimer, Ferritin, Blood urea, Creatinine.

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INTRODUCTION

Coronavirus disease 2019 (COVID-19) or so-called SARS-CoV-2 (severe acute respiratory syndrome coronavirus 2) [1, 2], identified as an enveloped RNA beta *Coronavirus* that belongs to *Coronaviridae* family. It has been implicated in cases of mild to severe diseases of the respiratory system [1,3]. The World Health Organization (WHO) declared that it is emerging infectious agent, which causes global public health emergency [4].

SARS-CoV- 2 has developed to a pandemic phenomenon too fast, where it was only just an epidemic outbreak when first found in Wuhan, China [5]. On 24 /2/2020, in AL-Najaf-Iraq, the first patient identified with SARS-CoV-2 in Iraq, later then more cases were formerly recorded [6].

There is about 80% similarity between Covid-19 and SARS-CoV in the way of infecting human cells. SARS-CoV-2 binds to a special receptor called Angiotensin-converting enzyme 2 (ACE 2) [5]. Despite the documentation that showed that SARS-CoV-2 manifested as a respiratory infection in the first place, but new data indicated that it must be regarded it as a systemic disease infecting numerous systems, such as; gastrointestinal, immune, respiratory, hematopoietic, and cardiovascular system [7,8].

The mortality rates of SARS-CoV and Middle East respiratory syndrome (MERS) are higher than SARS-CoV-2; [9] however, SARS-CoV-2 is more fatal than seasonal flu. Immunocompromised patients with chronic diseases and older people are at higher risk of death from SARS-

CoV-2, but younger patients who have no major underlying infections and diseases might also presented with potentially fatal complications like disseminated intravascular coagulopathy and fulminant myocarditis[10,11].

Hereby, we presented some biochemical and hematological findings in patients with SARS-CoV-2 and reveal the relationship of these findings with the infection.

MATERIALS AND METHODS

A cross-sectional study performed at Kirkuk general hospital (May 15 to, August 25 /2020). It was done on a total of (84) patients (50 male and 34 female), who were aging between (16-70) years old. They already diagnosed with SARS-CoV-2 infection through having chest computerized tomography scan (CT-Scan) from (Simens Co.), and 60 non-infected with SARS-CoV-2, who were used as a control group at similar age group.

Patients: Each infected patient and each individual of the control group was tested for a total white blood cells count, lymphocyte count, serum Ferritin, blood urea, serum creatinine, D-dimer, and blood group, by withdrawing (10 ml) of a peripheral venous blood. The patients and the control group showed up at Kirkuk general hospital. The infected patient diagnosed already having SARS-CoV-2 by using CT-Scan) from (Simens Co.). The control group were also had CT-Scan which came out with no evidence of SARS-CoV-2.

Procedures: The blood samples divided into two parts; one part included (7.5 ml) of clotted blood that

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centrifuged for about 15 minutes to get a clear serum for the biochemical tests, and (2.5 ml) of the whole blood were added into EDTA tube for the hematological tests. The blood urea, serum creatinine, serum Ferritin, and D-dimer examined automatically through using of Spinreact full-automated machine made in Spain by TC matrix. The white blood cells and lymphocyte counting conducted through using of full-automated Celltac by Nihone made in Japan. The blood grouping done on the patients by having three drops of whole blood against anti-A, anti-B, and anti-D antigens using slide method. All the kits of the biochemical tests were from Spinreact, made in Spain.

Statistical Analysis:

The statistical analysis done through using of computerized statistical method called SPSS (Statistical Package for Science Services) to find out the probability (P value) and Chi-square (X^2). If the probability value was greater than 0.05 the results considered non-significant statistically, while the P value was statistically significant if it was lower than or equal to 0.05. The P value was considered highly significant statistically if its value was lower than 0.01.

RESULTS

Relation of white blood cells count with SARS -CoV-2

Table one showed highly significant results regarding the relation of SARS-CoV-2 infection with white blood cells count in which the highest rate of the male patients (72%) showed decreased white blood cells count this was also true for the female patients who exhibited 42% of decreased white blood cells count.

Relation of lymphocyte count with SARS -CoV-2 infection

The highest rate of infected female patients 100% showed decreased lymphocyte count, and the male patient showed the highest ratio of decreased lymphocyte count 80%, only 20% of infected male patients showed normal lymphocyte count while the female patients revealed none. The results were significant as revealed in table 2.

Relation of ABO blood group with SARS-CoV-2 infection

As shown below in table (3) the highest rate for both male and female patients (80%), (100%) respectively were belong to blood group (A). Whereas blood groups AB shows zero infected patients for both sexes. The result was non-significant.

Relation of Serum Ferritin levels with SARS-CoV-2 infection

The highest rate (54%) of infected male patients shows increased serum Ferritin level. The results were highly significant since the P value was lower than 0.01 as shown in table 4.

Relation of Blood Urea levels with SARS-CoV-2 infection

The highest rate (59%) of infected female patients showed increased blood urea level while the lowest rate (20%) of infected male patients showed increased blood urea level as shown below in table (5) and the results were highly significant.

Relation of SARS-CoV-2 infection with Serum Creatinine Levels

The highest rate (71 %) of infected female patients exhibited increased serum creatinine level. The results were significant, as presented in table 6 below.

D-dimer levels in infected and control group (non-infected)

The highest rate (60%) of infected patients showed increased D-dimer level, while the highest rate (80%) of the non-infected patients showed normal levels of D-dimer. The results were highly significant. P value lower than 0.01. Refer to table 7.

The Biochemical parameters in the control group

Table 8 shows the biochemical parameters in the control group non-infected individuals. The highest rate of blood urea, serum creatinine and serum Ferritin were normal, and the results were non-significant as revealed in table 8.

The Hematological Parameters in control group

Table 9 shows the relation of control group with white blood cells count and lymphocyte count in which the control group show the highest rate of normal white blood cells and lymphocyte count. The results were non-significant.

Table 1. Relation of White blood cells count with SARS-CoV-2 infection

White blood cells count	Patients with SARS-CoV-2			
	Male		Female	
	No.	%	No.	%
Normal	10	20	8	23
Increased	4	8	12	35
Decreased	36	72	14	42
Total	50	100	34	100
$X^2 = 11.263$ P = 0.0035832 P < 0.01 Highly Significant				

Normal Range=4.5-11.0 x10³/μL

Table 2. Relation of Lymphocyte count with SARS-CoV-2 infection

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Lymphocyte count	Patients with SARS- CoV-2			
	Male		Female	
	No.	%	No.	%
Normal	10	20	0	0
Increased	0	0	0	0
Decreased	40	80	34	100
Total	50	100	34	100
$X^2 = 7.719 P = 0.02107854 P < 0.05$ significant				

Normal range: 20-40%

Table 3. Relation of ABO blood group with SARS-CoV-2 infection

ABO Blood Group	Patients with SARS- CoV-2			
	Male		Female	
	No.	%	No.	%
A	40	80	34	100
B	2	4	0	0
o	8	16	0	0
AB	0	0	0	0
Total	50	100	34	100
$X^2 = 7.719 P = 0.05219053 P > 0.05$ non-significant				

Table 4. Relation of Serum Ferritin levels with SARS-CoV-2 infection

Serum Ferritin levels	Patients with SARS –CoV-2			
	Male		Female	
	No.	%	No.	%
Normal	8	16	20	59
Increased	42	54	14	41
Decreased	0	0	0	0
Total	50	100	34	100
$X^2 = 16.701 P = 0.00023628 P < 0.01$ Highly significant				

Normal range based on the company standards:

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Male 20-110 µg/dl
Female 20-100 µg/dl

Table 5. Relation of Blood Urea levels with SARS-CoV-2 infection

Blood Urea Levels	Patients with SARS-CoV-2			
	Male		Female	
	No.	%	No.	%
Normal *	40	80	14	41
Increased**	10	20	20	59
Decreased***	0	0	0	0
Total	50	100	34	100
X² = 13.286 P= 0.00130311 P<0.01 Highly significant				

Normal range=15-45 mg/dl

Table 6. Relation of SARS-CoV-2 infection with Serum Creatinine Levels

Serum Creatinine Levels	Patients with SARS-CoV-2			
	Male		Female	
	No.	%	No.	%
Normal	30	60	10	29
Increased	20	40	24	71
Decreased	0	0	0	0
Total	50	100	34	100
X² = 7.591 P = 0.02247167 P <0.05 significant				

Normal range: 0.7-1.4 mg/dl

Table 7. D-dimer levels in infected and control group (non-infected)

D-Dimer Levels	Patients		Control group	
	No.	%	No.	%
	Normal *	34	40	50
Increased**	50	60	10	20
Decreased***	0	0	0	0
Total	84	100	60	100

$\chi^2 = 26.449$ $P = 0.00000181$ $P < 0.01$ Highly significant

Normal range < 250 $\mu\text{g/ml}$

Table 8. The Biochemical parameters in the control group

Results	Biochemical parameters					
	Urea		Creatinine		Ferritin	
	No.	%	No.	%	No.	%
Normal *	55	91	50	80	55	91
Increased**	5	9	10	20	4	7
Decreased***	0	0	0	0	1	2
Total	60	100	60	100	60	100

$\chi^2 = 5.576$ $P = 0.23312935$ $P > 0.05$ non-significant

Table 9. The Hematological Parameters in control group

Results	Hematological Parameters			
	White Blood Cells count		Lymphocyte count	
	No.	%	No.	%
Normal *	40	66	51	85
Increased**	3	5	1	2
Decreased***	17	29	8	13
Total	60	100	60	100

$\chi^2 = 5.57$ $P = 0.06172909$ $P > 0.05$ non-significant

DISCUSSION

COVID-19 or what so called SARS-CoV-2, has spread too quickly all over China and some other countries around the world. [12] The government of China of the national health commission has established the diagnosis and treatment plan of SARS-CoV-2. [13]

This study disclosed the ratio of decreased white blood cells count was the highest 72% and 42% among male and female patients with SARS-CoV-2 respectively and the results were highly significant as uncovered in table (1). This agreed with a study conducted in china and Switzerland where showed leukopenia occurred in most patients [14, 15], but disagreed with studies in Baghdad/Iraq and Japan, which showed highest rate of patients with COVID-19 have normal white blood cells count [16, 19]. The variation of the results may be associated with the kinetics of white blood cells response

to the virus during the infections, plus these differences may also attributed to the viral circulation rate and accessibility in the community. Furthermore, the occurrence of these abnormalities in the white blood cells count attributed to the severity of the disease. Therefore, the patients, who suffered from mild disease, tend to have less abnormality than those with sever disease [17]. In addition to what mentioned before, there was no similar study performed in Kirkuk city to compare the results. Regarding the relationship of the lymphocyte count with the infection, the study showed significant differences among the patients with COVID-19 and the highest rate of the patients were with low lymphocyte count (lymphopenia) as noticed above in table 2, this was agreed with a study conducted in Baghdad/ Iraq, in Indonesia, and in Japan that showed lymphopenia was associated with SARS-CoV-2 infection [16,18,19]. Many

factors might play a critical role in the pathophysiology of lymphocytopenia. First, it has found that angiotensin-converting enzyme 2 receptors (ACE2 receptors) are likely the cell receptor of SARS-CoV-2, which was also the receptor for SARS-CoV [20]. For this reason, cells that express ACE2 receptors are vulnerable to SARS-CoV-2 infection. A study done by Xu *et al.* showed that ACE2 receptors were expressed in lymphocytes in digestive system, lungs and oral mucosa [21]. Therefore, the direct effect of SARS-CoV-2 on the lymphocytes that could lead to their lysis, does not look unfounded. In addition to that, SARS-CoV-2 infection induces the production of cytokines like IL-7 IL-2, IL-6, TNF-a and interferon Gamma, which are recognized as a cytokine storm, which lead to not only apoptosis of lymphocytes' but also atrophy of lymphoid organs' (e.x. the spleen) [22]. ACE2 organizes blood pressure. [23].

In table3, as it shown above, the highest rate of infected patients was from blood group A, and this was true for both male and female. This agreed with a study conducted in Baghdad/Iraq where it showed the highest rate among infected patients were from blood group A, while it showed disagreement with another study done in Iran where they revealed the highest rate of patient susceptible to the infection were from blood group AB. [24,25]. These differences may be due to the biologic difference among these populations [26]. New studies have also revealed the importance of blood group O in lowering SARS-CoV-2 risk [27,28,29]. However, other research showed the importance of the other blood groups A, B, and AB in raising the infection's risk. In the study done by Zhao *et al* they found that blood group A showed higher risk of death.[28]. It is clear now that, these outcomes suggested that ABO antigens might have significant role in the pathogenesis of SARS-CoV-2; however, the kinetic by which these particles confer protection or susceptibility to the infection is submitted to speculation.

From table (4) as exhibited above, we noticed that the highest rate of the infected male patients with increased levels of serum Ferritin, and the result was highly significant, this was matched to a study performed at al Najaf/Iraq, Kurdistan/Iraq, Indonesia, and Wuhan/China, where they detected high levels of serum Ferritin in patients with COVID-19 [30,31,32, 34]. Ferritin has a significant role in immune dysregulation, particularly when their levels are high in blood, they have impact on the suppression of the immune system and on pro-inflammatory action, that take in part of the cytokine storming [33]. Elevated serum Ferritin levels indirectly connected to acute respiratory distress syndrome and sever SARS-CoV-2 infection. This might have led to the concept of existence of secondary hemophagocytic lymphohistocytosis (sHLH) in SARS-CoV-2 [35]. Which is a state of profuse inflammation in which a cytokine storming is a clear characteristic that may lead to the death [36].

Regarding the association of SARS-CoV-2 infection with renal function tests (blood Urea and serum Creatinine), the results were highly significant in regards with blood urea and significant in regards with serum creatinine levels as shown above in table 5 and 6 respectively, and it was noticed that most of the patients with COVID-19 infection showed increased levels of blood urea and serum creatinine The highest rate of increased blood urea and serum creatinine levels found in female patients with COVID-19 infection, while the highest rate of infected

male patients showed normal levels of blood urea and serum creatinine levels respectively. This agreed with Cheng Y, *et al* [37] who detected high levels of blood urea and serum creatinine in patients with SARS-CoV-2 infection. This also agreed with a study done in Iran [38], in which a huge group of patients with SARS-CoV-2 pneumonia had symptoms of renal disease, like increased levels of blood urea and serum creatinine which might justified with different pathophysiologies occurred in SARS-CoV-2 pneumonia. These results disclose, that COVID-19 may get into the peripheral blood and resides in kidney tissues because of the increased expression of ACE2 in kidney cells and then demolish the resident kidney cells [39]. Potential inflammatory status in chronic kidney disease patients may make them susceptible to SARS-CoV-2 pneumonia due to pro-inflammatory status with defect in function in innate and adaptive immune cells. [40]. that raises the risk of upper respiratory tract infection and pneumonia [41].

Table 7 reveals highly significant results in regards with the relationship of D-Dimer levels with SARS-CoV-2 in which the highest rate of the infected patients showed increased levels of D-Dimer. This showed an agreement with studies done in Indonesia, Japan , Greece and Wuhan/China where they found an association between the severity of SARS-CoV-2 infection and increased levels of D-dimer [32, 34, 42, 43]. These may be due to the majority of the patients with SARS-CoV-2 who suffers from thrombocytopenia showed increased levels of and D-dimer levels leading to impaired coagulation time with fatal outcome because of (DIC) or disseminated intravascular coagulation [44]. Patients with COVID-19 infection may develop sepsis which considered as a major cause of disseminated intravascular coagulation. The progression of DIC happens when platelets, leukocytes, and endothelial cells are activated to induce dysregulation of formation of which happens both locally and systemically effecting the lungs of patients with severe pneumonia. The circulation of thrombin freely, without any control by natural anticoagulants, could make platelets activated and lead to fibrinolysis [45]. Regarding the control group tables 8 and 9 show non-significant relation in regards with the biochemical and hematological markers. From these findings we came to a point that SARS-CoV-2 infections may affect the white blood cells count and specially lymphocyte and affect the levels of Ferritin, blood urea, serum creatinine and D-dimer.

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