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ORIGINAL ARTICLE

Evaluation the Effects of Complement Terminal Component (C5b-9) on Immunothrombosis in Severe and Critical COVID-19 Infections Compared to Mild Cases

Abdul Q. W. Muhammed*, Hadi R. Hassan

Department of Clinical Laboratories, College of Applied Medical Sciences, University of Kerbala, Iraq

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Corresponding author:

Abdul Q. W. Muhammed

Email: abdwasfi4877@gmail.com

Department of Clinical Laboratories, College of Applied Medical Sciences, University of Kerbala, Iraq

ABSTRACT

Background: The study aimed to evaluate the association between the complement terminal component (C5b-9) and immuno-thrombosis in severe and critical COVID-19 patients compared to mild cases.

Materials and Methods: A cross-sectional study was conducted. From October 1, 2021, to May 30, 2022, 82 COVID-19 patients were recruited at Imam AL-Hussein Medical City in Kerbala, consisting of 44 males and 38 females, and their ages ranged from 25 to 85 years old. All patients tested positive for severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) and were hospitalized. Twenty-seven of those were diagnosed with mild COVID-19 and 27 with severe diseases, while 28 patients had critical diseases.

Results: C5b-9 level significantly elevated with the severity of COVID-19.

Conclusion: There is a significant correlation between complement terminal component (C5b-9) elevation and COVID-19 severity.

INTRODUCTION

The novel coronavirus disease 2019 (COVID-19), the most significant pandemic in the past century, remains a significant threat to public health. As of March 15, 2022, COVID-19 has taken nearly 6 million lives and infected more than 535 million people, and the number of cases continues to increase worldwide (WHO, 2022).

Even though lung infections are the most common symptom of coronavirus disease, the infection is frequently made worse by coagulopathy, and thrombo-embolic events can be observed in many affected individuals.[1] Dehydration, acute inflammatory conditions, diabetes, obesity, hypertension, prior ischemic stroke, peripheral artery disease, and other conditions are frequently present in hospitalized COVID-19 subjects and may increase the risk of thrombo-embolic events. However, other possible causes can still be found, such as increased synthesis of adhesion molecules that might cause endothelial activation and vascular inflammation.[1]

COVID-19 induces a systemic inflammatory response involving several inflammatory cells' dysregulation and misexpression. The activation and recruitment of inflammatory cells require the expression of some types of inflammatory mediators, such as cytokines (for example, platelet-activating factor [PAF]), adhesion molecules, and chemokines.[2]

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Pathological evidence of venous thrombo-embolism, direct viral infection of the endothelial cells, and diffuse endothelial inflammation have been reported in recent studies.[3] It was discovered that critical illness is related to parameters that indicate an activated coagulation process, specifically higher D-dimer and fibrinogen levels. On the other hand, relatively minimal alterations were identified in prothrombin time, and platelet counts. In addition, a series of autopsies conducted on deceased COVID-19 patients described many instances of thrombosis. Accordingly, the vascular micro-thrombotic disease is likely the predominant contributor to mortality in critically ill COVID-19 patients.[4]

The mechanisms underlying increased thrombotic events are not fully understood; however, mounting evidence suggests that endothelial and platelet activation that leads to thrombosis (Immunothrombosis) plays a critical role. Because viral inclusions have been identified in endothelial cells, damage and activation of endothelial cells could be the driving force behind platelet activation and the resultant coagulopathy. Hence, understanding the involvement of platelets in COVID-19 critical sickness is essential for both comprehending the biology of SARS-CoV-2 infection and locating potential therapy methods.[5]

Recent articles have reported the presence of severe endothelial injury and widespread pulmonary micro-thromboses along with increased angiogenesis in the lungs of deceased patients who had been infected with the coronavirus COVID-19. These results support other recent publications from various centers reporting the presence of increased coagulation markers and micro-thromboses in lungs and other organs of patients with COVID-19.[6]

One of the expected mechanisms for immuno-thrombosis formation is thought to be caused by the activation of the complement cascade, which is also detected in coronavirus disease 19 (COVID-19). This cascade comprises over fifty plasma proteins that sense and responds to invading pathogens.[7] This component of the innate immune response connects the innate and adaptive immune responses. Activation can happen through one of three pathways: the classical, lectin, or alternative pathways. These different routes ultimately converge at the same location, the source of the C5 convertase and a number of anaphylatoxins. C5 convertase is responsible for producing C5b-9, the complement terminal complex. C5b-9 induces the formation of cytotoxic membrane channels, ultimately resulting in the death of cells; yet, even sublytic doses have important immunomodulatory effects.[8] Excessive and unregulated complement activation may promote the formation of a systemic pro-inflammatory, pro-oxidant, and pro-coagulant state characterized by multi-organ dysfunction and an increased risk of severe clinical outcomes.[9,10] The complement system is a central mediator of innate immune defense, and it, together with the coagulation system, helps the peri-and intravascular elimination of invading microorganisms in a process termed immuno-thrombosis. Some studies suggest that the pathophysiology of COVID-19 involves dysregulation of microvascular thromboinflammatory pathways, the most prominent of which is the complement system.[11]

MATERIALS AND METHODS

In accordance with the WHO, RT- PCR and CT scans were used to diagnose COVID-19 in all patients. For this study, a total of 82 COVID-19 patients were admitted to AL-Hussein Medical City in Kerbala, consisting of 44 males and 38 females. Their ages ranged anywhere from 25 to 85 years old, on average.

These patients are divided into three groups: mild (27 patients: 14 males and 13 females); severe (27 patients: 14 males and 13 females); and critical (28 patients: 17 males and 11 females). Patients were chosen at random from the local community, taking into consideration their ages and genders.

Blood samples were taken from all participants, and each sample was divided into three parts:

Part one- placed in EDTA tube for hematological tests (Complete blood count (CBC)).

Part two- transferred into sodium citrate tube to perform (D-dimer, PT, PTT).

Part three – put in a gel tube for biochemical tests (C. reactive protein, Blood urea, and Serum creatinine) and immunological test (C5b-9).

Statistical Analysis

The data was entered into a Specific Software Statistical Package for the Social Sciences (SPSS) version 21 for Windows computers for statistical analysis. Results were presented as mean, and standard deviation (mean ± SD). A p-value less than 0.05 was deemed statistically significant; a *p*-value less than 0.001 was considered highly significant. In addition, the Pearson correlation (*r*-value) was used to explain the relation of C5b-9 levels with hematological, biochemical, and coagulation parameters.

RESULTS AND DISCUSSION

Demographic Data of the Studied Groups

The current study included 82 patients with COVID–19 determined by SARS-COV-2 specific real-time PCR on nasopharyngeal (NP) swab specimens.

According to SpO2 percentage and respiratory rate (RR), COVID-19 patients were classified into mild (SpO2 $\geq$ 94, RR $\leq$ 22), severe (SpO2 $\leq$ 93, RR $\geq$ 30), and critical which they were the same as severe but required mechanical support such as ventilator and Continuous positive airway pressure (CPAP) therapy. Computerized topography (CT) percentage has been considered, and it is elevated with the severity of the disease. As a result, the following patient groups were chosen (n = 27 mild, n = 27 severe, and n = 28 critical). There were 44 (53.7%) males and 38 (46.3%) females among them. The average age was 59.9 years, ranging from 25 to 85 years.

As showed in Table 1, there was no significant difference between female and male patients, as agreed by,[2] who claimed that the percentage of men and women among ICU patients

Table 1: Demographic Data of the Studied Groups

Total number			*82*		
Age			Mean (59.9) year Average (25 – 85)yr		
Gender			Male 44 (53.7%) Female 38 (46.3%)		
Groups	Gender		SpO_{2} % Mean ± SD	Respiratory Rate (RR) R per min	(CT%) Computerized topography%
	Male No. (%)	Female No. (%)			
Mild (N = 27)	14 (51.9%)	13 (48.1%)	96% ± 2	22.1 ± 5.4	12% ± 4
Severe (N = 27)	14 (51.9%)	13 (48.1%)	89% ± 4	36.3 ± 6.1	39% ± 14
Critical (N = 28)	17 (60.7%)	11 (39.3%)	81% ± 3	40.8 ± 9.1	57% ± 11
			p = <0.001	*p* = <0.001	*p* = <0.001

and non-ICU patients was the same. Despite the fact that there were no statistically significant differences between gender and illness severity in this specimen, male gender was discovered to be a risk factor for disease severity by,[12,13] who found that the frequency of symptomatic COVID-19 was greater in males than in women.

Oxygen saturation (SpO2) and respiratory rate (RR) were significantly correlated with disease severity. The patients with severe and critical disease had a significantly higher respiratory rate as compared to mild patients, while the SpO2 of those with the severe and critical disease was significantly lower as compared to those with mild disease. All patients with critical illness required mechanical ventilation. These findings were similar to those mentioned by,[14] in which they concluded that patients with the severe and critical disease have significantly higher RR and significantly lower SpO_2 than mild patients.

Age Distribution for COVID-19 Studied Patients

In table 2, the ages of the subjected patients were distributed into three groups; the first young group aged from 25–45 years, and they mostly occurred in mild cases and decreased in severe and critical cases. The second post-young group, aged from 46–65 years old, showed a decrease in mild cases and an increase in both severe and critical cases. The third elderly group ranged in age from 66 to 85 years old, and they mostly presented in critical and severe cases, with a decrease in mild cases. This means that the severity of the disease significantly correlates with aging, and the elderly are more susceptible to severe and critical diseases. This means a significant correlation exists between patient's age and disease severity.

The explanation for this link is that older people frequently have low immunity and are suffering from one or more chronic conditions such as (hypertension, diabetes mellitus, smoking, etc...), resulting in increased complications and illness severity. A comparable set of data was recently published by,[15] which they discovered that older people had the highest percentage of critical COVID-19 cases.

Mortality Distribution Among the Groups of COVID-19 Patients

There were no death cases in the mild group, while three patients were dead within the severe group, and they represented 3.6% of the total patients' groups (Table 3). The mortality number in the critical group was more than three folds from the severe group, there were 11 deaths within the critical group, and they represented 13.4% of total patients. The total mortality percentage was 17%, and all patients who died were between (66-85) years old.

Mortality was significantly increased with disease severity and the patients aging in this study because no death was recorded within mild patients, while three deaths in severe cases and 11 deaths in critical cases were recorded. These outcomes suggest that younger individuals are more prone to have mild symptoms and they are less prone to die, but older individuals are more likely to die and have severe symptoms. This was agreed upon by many other researchers, including,16 who stated that elderly COVID-19 patients had a significant death rate due to a high case fatality rate and symptomatic infection rate. Also, he mentioned that approximately 80% and 90% of deaths have occurred in patients aged > 70 years and ≥ 60 years in Korea and Italy, respectively.

Table 2: Age Distribution for COVID-19 Studied Patients:

Age group (yrs)	*Mild (N = 27) No. (%)*	*Severe (N = 27) No. (%)*	*Critical (N = 28) No. (%)*
(25–45) Young	18 (66.6)	7 (25.9)	3 (10.7)
(46–65) Post young	5 (18.56)	8 (29.7)	5 (17.9)
(66–85) Elderly	4 (14.8)	12 (44.4)	20 (71.4)
Total	27 (100)	27 (100)	28 (100)

Table 3: Mortality Distribution Among the Groups of COVID-19 Patients

Classified groups	*No. of Cases*	*Mortality distribution within age groups No. (%)*			
		Young	*Post young*	*Elderly*	*Total*
Mild	27	0	0	0	0
Severe	27	0	1 (1.2%)	2 (2.4%)	3 (3.6%)
Critical	28	0	1 (1.2%)	10 (12.2%)	11 (13.4%)
Total	82	0	2 (2.4%)	12 (14.6%)	14 (17%)

Hematological Parameters in COVID-19 Patients

According to Table (4) Total white blood cell counts were not significantly elevated but the percentage of neutrophils was significantly elevated in severe and critical in comparison

Table 4: Hematological Parameters in COVID-19 Patients

Variables	*Normal range*	*Mild Mean ± SD*	*Severe Mean± SD*	*Critical Mean ± SD*	*P- value*
White blood cell counts $x10^9$/L	4.0–11.0	11.73 ± 4.77	13.09± 5.92	11.43± 5.12	*P*= 0.253
Neutrophil %	40–75 %	73% ± 12	80% ± 8↑	86% ± 5↑	*P*= < 0.001
Lymphocyte %	20–40 %	↓18% ± 7	↓14% ± 5	8% ± 3 ↓	*P*= < 0.001
Hemoglobin g/L	13.5–17.5	13.8 ± 1.8	↓11.5 ± 1.5	↓10.8 ± 1.8	*p*= 0.001

Table 5: Some Biochemical Parameters in COVID-19 Patients

Variable	*Normal Range*	*Mild Mean ± SD*	*Severe Mean ± SD*	*Critical Mean ± SD*	*p-value*
C-reactive protein (CRP) mg/L	0–5	20.0 ± 9.6↑	47.9 ± 20.4↑	63.4 ± 25.6↑	*p*= < 0.001
Blood urea (BUN) mg/L	15–45	54.5 ± 20.2↑	71.3 ± 34.8↑	121.1 ± 59.5↑	*p*= < 0.001
Serum creatinine (S.creatinine) mg/L	0.2–1.2	1.0 ± 0.4	1.3 ± 0.6↑	1.9 ± 0.8↑	*p* = < 0.001

with mild COVID-19 cases (p = <0.001). In contrast, the percentage of lymphocytes was significantly lower in severe and critical cases, as demonstrated by a significant correlation (p = 0.001). This might imply that the COVID-19 infection is preferentially targeting lymphocytes. Hemoglobin (Hb) levels were slightly lower in all three groups (mild anemia), and this slight reduction was also statistically significant (p = 0.001).

Total WBC counts showed a slight, not significantly increase with the disease severity. While neutrophil percentages were significantly increased in severe and critical COVID -19 patients, this may be due to increased inflammatory process and excessive cytokine release (cytokine storm). This explanation is similar to that reported by,[17] who said that the elevation in neutrophils is associated with increased secretion of some cytokines that include IL-8, IL-6, interferon 10 (IP-10), granulocyte-macrophage colony-stimulating factor (GM-CSF), IL-1b, IL-10, and TNF.

In contrast, lymphocyte percentages decreased significantly with illness severity, which might be due to the functional depletion of lymphocytes such as cytotoxic T lymphocytes (CTLs) and natural killer (NK) cells, which are involved in viral infection management. Zhang C and colleagues have found that the overall number of CTLs and NK cells was significantly reduced in SARS-CoV-2 patients. These results suggest that SRAS-CoV-2 infection is associated with the functional depletion of cytotoxic lymphocytes.[18]

Hb significantly decreased in severe and critical cases in comparison with mild cases in both males and females. The explanation of this is that the inflammation in COVID-19 patients can lead to an alternation of iron hemostasis and reduced intestinal iron absorption, resulting in the reduced availability of the metal for erythropoiesis and the production of hemoglobin (anemia of inflammation). Another cause of Hb decrease is impaired renal function that may occur in severe and critical COVID-19 cases, where the kidneys are the primary organs that regulate erythropoiesis. This is agreed by,[19] who reported that the elevated expression of ferritin during COVID-19 infection leads to alteration in iron homeostasis and retention within macrophages along with reduced iron absorption by the intestine, and this results in reduced metals for erythropoiesis. Also, patients with anemia were older, had reduced renal function, and had significantly higher inflammatory markers such as C- reactive protein or interleukine-6 (IL6). However, another study found that there was no significant correlation between hemoglobin levels and COVID-19 severity.[20]

Some Biochemical Parameters in COVID-19 Patients

The most prominent elevation in the mild cases occurred by C. reactive protein (CRP), where it significantly elevated more than three folds compared to the normal range, and it increased in severe and critical more than mild cases (p = < 0.001). This makes it the most sensitive COVID-19 biomarker predictor, particularly for mild cases. Blood urea (BUN) was only slightly higher in mild cases, while it was significantly elevated in severe and critical cases. Blood urea levels is significantly elevated with disease progression (p =< 0.001). Serum creatinine levels in mild cases were within the normal range (Table 5). However, Serum creatinine levels were significantly elevated in the severe and critical cases in comparison with mild cases (p = < 0.001).

CRP is an acute-phase protein of that increase following interleukin-6 secretion by macrophages and T cells. Therefore, SAR-CoV-2 infection significantly caused CRP secretion from the beginning of the disease, and it was more elevated with COVID-19 severity. This was in agreement with,[21] who stated that although CRP levels are often low in viral infections, adaptive immunity seems to be necessary for the clearance COVID-19 virus, and the macrophage activation syndrome might explain the higher serum CRP levels and lead to increase the severity of the disease. Cytokines, such as IL-6, and TNF-α, stimulate hepatocytes to produce CRP during a cytokine storm that the process of COVID-19 pneumonia can trigger.

Blood urea (BUN) and serum creatinine levels elevation during COVID-19 may indicate early injury of the kidney. One possible reason for the high incidence of kidney involvement is that the systemic immune response to SARS-COV-2 can be harmful in certain individuals, resulting in what is known as a cytokine storm. As a result, the kidney may be a vulnerable target for this novel coronavirus. These were agreed with (20) who demonstrated that proximal tubule cells in the kidneys exhibit high levels of ACE2, which implies that SARS-CoV-2 may target them at an early stage of illness. The spike

Table 6: Some Coagulation Parameters in COVID-19 Patient

Variable	*Normal range*	*Mild Mean ± SD*	*Severe Mean ± SD*	*Critical Mean ± SD*	*P - value*
D. dimer ng/mL	0-500	328.8 ± 114.9	1031.5 ± 305.2	2351.4 ± 1133.8	*p*= <0.001
Platelet count x10^9/L	150 - 450	239.23 ± 70.92	260.3 ± 109.3	231.4 ± 89.2	*p*= 0.422
Prothrombin time (PT)	10 - 14	13.1 ± 1.1	13.1 ± 1.2	13.9 ± 1.4	*p*= 0.053
Partial thromboplastin time(PTT)	30 - 38	32.3 ± 2.6	32.6 ± 4.1	33.5 ± 2.6	*p*= 0.256

glycoprotein of SARS-CoV-2 has an affinity for binding to ACE2, which is considered a major determinant of disease severity.[22]

Some Coagulation Parameters in COVID-19 Patients

According to Table 6 in mild cases, no one of these parameters increased more than the normal range, but in severe and critical cases, only D-dimer levels increased to more than normal. And this elevation was statistically more significant (p = < 0.001). There is no significant elevation in platelets count, prothrombin time, and partial thromboplastin time (p=0.422; p=0.053, p=0. 0.256, respectively). Therefore, D-dimer was considered the single routine marker that indicated the presence of thrombosis (immuno-thrombosis) in COVID-19 patients during this study. All mortalities in the current study were associated with high D-dimer levels. This indicates that D-dimer is significantly correlates with COVID-19 severity, hypercoagulability, that increased the risk of venous thromboembolism (VTE) events, leading to thrombo-inflammation and even death in severe and critical conditions.

In mild COVID-19 cases, D-dimer levels were slightly elevated in the patients but remained within high normal range. However, D-dimer levels in severe and critical cases were significantly elevated compared to mild cases. These results agreed by,[20] they claimed that D-dimer, ferritin, and LDH levels were significantly increased in patients with the critical disease.

D-dimer is a fibrin breakdown product that has a mechanistic role in COVID-19 thrombo-inflammation. As a result, D-dimer can be employed as an essential coagulation biomarker that can assist in establishing patient screening, therapy options, and prognosis management.[23]

Patients with D-dimer levels >1000 ng/mL have a 20-fold higher mortality risk than those with lower D-dimer levels. Therefore, D-dimer is a screening test for VTE in COVID-19 patients, and changing therapeutic anticoagulant dosages based on D-dimer elevation is more useful to patients than preventive doses.[24]

Platelet counts showed no significant correlation with disease progression. This may be due to the early and good prognosis of the COVID-19 cases in this study because the platelets (PLTs) at the time of admission were not affected and the number of PLTs was normal. This was agreed by,[20] who reported that platelet count did not show a statistically significant association with disease severity. Researchers have a consensus that COVID-19-related thrombocytopenia is a delayed event during infection, and thrombocytopenia in COVID-19 patients occurs in the intensive care unit (ICU) and threatens the life of severe COVID-19 cases.[25]

Table 7: The levels of C5b-9 in COVID-19 patients

	Normal Range	*Mild Mean ± SD*	*Severe Mean ± SD*	*Critical Mean ± SD*	*p-value*
C5b-9 ng/mL	< 250	483.8 ± 201.0↑	759.1 ± 371.8↑	797.6 ± 360.8↑	*p* = 0.047

Prothrombin time (PT) is normal or near-normal in most COVID-19 patients, with just 5% who have extended PT. Results like these are also agreed by.[23] who said that prothrombin time did not show a significant increase in COVID-19 progression

In the current study, partial thromboplastin time (PTT) also did not show statistical significance with disease severity. PTT is frequently normal in patients with COVID-19 infection, with only 6% developing PTT prolongation, and the average length of PTT appears to be similar in COVID-19 critically ill and non-critically ill patients, with no significant link to disease severity or death. As a result, PTT does not seem to be a good predictor of COVID-19 development.[26]

The Levels of C5b-9 in COVID -19 Patients

C5b-9 level significantly elevated with COVID-19 severity (Table 7).

C5b-9 elevation indicates COVID-19-induced complement activation that mainly occurs in the lungs and kidneys. This was agreed by,[27] who have found that acute renal failure associated with tubular necrosis and abundant complement deposition develops in a significant percentage of patients with severe COVID-19. The complement system can be activated by SARS-CoV-2 either directly through the lectin, classical or alternative pathways or cause endotheliopathy (endothelial cell injury and dysfunction) and thromboinflammation (inflammation associated with coagulation and thrombosis), which in turn activate the complement system and leads to endothelial injuries by the complement terminal component C5b-9 that binds to cells and causes cell lysis by forming pores in the plasma membrane. Therefore, high serum levels of C5b-9 were significantly associated with higher COVID-19 severity and mortality.[28,29]

The Correlation of Serum Levels of C5b-9 with Hematological Parameters in Mild, Severe, and Critical COVID-19 Patients

As showed in table 8 there is a negative statistically significant correlation between C5b-9 and hemoglobin levels in critical cases, and this correlation (r = 0.423, p = 0.019).

In mild cases, where hemoglobin (Hb) levels began to

Table 8: Correlation of Serum Levels of C5b-9 with Hematological Parameters in Mild, Severe, and Critical COVID-19 Patients

Variables		C5b-9 (483.8 ± 201.0) ng/mL	Correlation
Mild	White blood cell counts x 10^9/L	11.73 ± 4.77	r = -0.186 p = 0.473
	Neutrophils%	73% ± 12	r = 0.154 p = 0.554
	Lymphocyte%	18% ± 7	r = 0.031 P = 0.905
	Hemoglobin g/l	13.3 ± 1.6	r = -0.098 P = 0.706
Variables		C5b-9 (759.1 ± 371.8) ng/mL	Correlation
Severe	White blood cell counts, x10^9/L	13.1 ± 5.92	r = -0.129 p = 0.529
	Neutrophils%	80 % ± 8	r = 0.297 p = 0.140
	Lymphocyte%	14 % ± 5	r = -0.204 p = 0.316
	Hemoglobin g/L	11.5 ± 1.5	r = 0.252 p = 0.214
Variables		C5b-9 (797.6 ± 360.8) ng/mL	Correlation
Critical	White blood cell counts x 109/L	11.4 ± 5.1	r = -0.061 = 0.751 p
	Neutrophils%	86 % ± 5	r = 0.221 p = 0.249
	Lymphocyte%	8 % ± 3	r = -0.198 p = 0.303
	Hemoglobin g/l	10.8 ± 1.8	r = -0.431 p = 0.019

decrease but remain within low normal range. While in severe and critical cases, Hb down below the normal range because of the inflammation in COVID-19 patients can lead to an alternation of iron hemostasis and reduced intestinal iron absorption resulting in intravenous hemolysis and, in turn, triggers platelets activation. Such data were published by.[30]

In critical cases, the significant correlation between C5b-9 and Hb may be due to an increased rate of hemolysis that causes complement system activation, leading to increased C5b-9 levels. These were in agreement with[31] who reported that heme and red blood cell degradation products are triggers to complement activation, and increased C5b-9 levels are important evidence of complement activation.

The Correlations of Serum Levels of C5b-9 with Biochemical Parameters in Mild, Severe, and Critical COVID-19 Patients

According to table 9 a positive statistically significant correlation occurred between C5b-9 and C-reactive protein levels in critical cases (r = 0.388, p = 0.037).

CRP acts as a complement system activator, whereas it binds to C1q and activates the classical pathway of the

Table 9: Correlation of Serum Levels of C5b-9 with Biochemical Parameters in Mild, Severe, and Critical COVID-19 Patients

Variables		*C5b-9 (483.8 ± 201.0) ng/ml*	*Correlation*
Mild	C-reactive protein mg/l	20.0 ± 9.6	r = -0.197 p = 0.447
	Blood urea mg/L	54.5 ± 20.2	r = 0.334 p = 0.189
	S.creatinine mg/L	1.0 ± 0.4	r = 0.297 p = 0.246
Variables		C5b-9 (759.1 ± 371.8) ng/ml	Correlation
Severe	C-reactive protein mg/L	47.9 ± 20.4	r = -0.067 p = 0.742
	Blood urea mg/L	71.3 ± 34.8	r = 0.541 p = 0.125
	S.creatinine mg/L	1.3 ± 0.6	r = 0.137 p = 0.502
Variables		C5b-9 (797.6 ± 360.8) ng/ml	Correlation
Critical	C-reactive protein mg/L	63.4 ± 25.6	r = 0.388 p = 0.037
	Blood urea mg/L	121.1 ± 59.5	r = 0.131 p = 0.498
	S.creatinine mg/L	1.9± 0.8	r = -0.198 p = 0.302

complement system, leading to increased C5b-9 levels. This is the explanation of the significant correlation between C5b-5 and CRP.[32]

Ethical Approval

The approval of the ethics was obtained from the Kerbala Health Directorate. In addition, prior to taking the sample, verbal consent from the patients and/or their parents was brought before the procedure was carried out. When conducting the sampling, we followed all appropriate safety and health protocols.

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