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A Comprehensive Analysis of Electrolytes, Inflammatory Biomarkers and Liver Function Enzymes Alterations in Hospitalized COVID-19 Patients—Insights from a Single Center in Iraq

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Abstract: Studying the status and involvement of laboratory biomarkers in COVID-19 infection has the potential to enhance clinical management and improve the scenario in terms of morbidity and mortality. The aim of this study was to investigate the status of serum electrolytes, inflammatory biomarkers, and liver function enzymes, as well as their correlation with the severity of COVID-19 infection. This observational study was conducted on 310 confirmed adult patients (> 18 years) with mild, moderate or severe symptoms of COVID-19, admitted to Al-Shifa 14 Hospital in Iraq between March and May 2022. The study outcomes were analyzed using one-way ANOVA and Pearson's correlation coefficient. Our data indicated that COVID-19 patients with severe symptoms had notably lower levels of serum calcium (mmol/L) and potassium (mmol/L) compared to mild cases (calcium: 0.88 ± 0.04 vs. 1.13 ± 0.11; potassium: 3.03 ± 0.32 vs. 4.17 ± 0.46), respectively. In the severe group, both electrolytes were observed to be below normal levels. Inflammatory biomarkers, specifically C-reactive protein and procalcitonin, demonstrated statistically significant elevations in severe cases compared to the moderate group, as well as in the moderate group compared to the mild group. Another significant finding was that liver function enzymes were notably elevated in patients with severe COVID-19 compared to those with mild cases, as indicated by statistical analysis. Hypocalcemia, hypokalemia, and elevated liver function enzymes (ALP, AST, and bilirubin) were strongly associated with COVID-19 severity. This research could provide a basis for using these biomarkers as useful assessment tools for COVID-19 infections.

Keywords: Serum Electrolytes; Calcium; Potassium; Inflammatory Biomarkers; liver Function Enzymes; COVID-19 Severity

1. Introduction

The COVID-19 pandemic, triggered by the SARS-CoV-2 virus, has been a substantial global health crisis, rapidly spreading worldwide [1]. Since its occurrence in December 2019 in Wuhan (China), the virus spread to numerous countries and territories, prompting widespread public health responses and measures to control its transmission [2]. The COVID-19 pandemic has profoundly affected global health, economies, and societies, resulting in millions of confirmed cases and deaths worldwide. Vaccination efforts have been essential in controlling both the spread and severity of the disease [3–5].

Symptoms of COVID-19 infection could range from asymptomatic carrier states to mild upper respiratory tract infections, and can progress to moderate or severe acute respiratory distress syndrome [6]. COVID-19 cases with

severe symptoms are usually accompanied by pneumonia, multiorgan dysfunction, and high mortality rates [7]. Accurate diagnosis is essential for effective epidemic control, as misdiagnoses can have far-reaching consequences for both individual patients and public health efforts [8]. The initial documentation of COVID-19 patients at risk of progressing to a more severe stage is essential for mitigating mortality rates [9]. The development of laboratory biomarkers to establish and predict the severity and progression of COVID-19 remains necessary, despite the identification of several laboratory-related risk factors that assess the disease's severity, including C-reactive protein, lymphocytes, and procalcitonin (PCT) [10–13]. In addition to these well-known COVID-19 risk factors, electrolyte disorders, such as calcium and potassium imbalances, have also been reported in patients with serious symptoms [14–16]. Disturbances in electrolyte balance can have serious implications for the management of the disease [15]. It is also noteworthy to mention that severe and critical COVID-19 patients had elevated liver enzyme levels as well as elevated total bilirubin levels [17,18].

In this context, our objective was to investigate the relationship between serum levels of calcium, potassium, sodium, and chloride and the severity of COVID-19. Additionally, we aimed to examine the relationship between liver function enzymes and the seriousness of COVID-19. An exploration of the correlation between laboratory biomarkers and the severity of COVID-19 infection may enhance the clinical management of patients and mitigate disease progression towards a severe manifestation through timely interventions. Furthermore, this research could provide an opportunity to utilize these biomarkers as effective assessment tools for the evaluation of COVID-19.

2. Methods

2.1. Study Setting and Design

This observational cross-sectional study was conducted with a cohort of 310 confirmed cases of COVID-19 patients exhibiting mild, moderate, or severe symptoms who were admitted to Al-Shifa 14 Hospital in Iraq between March and May 2022. Al-Shifa 14 Hospital, with a capacity of 200 beds, was designated exclusively for the admission of COVID-19 patients in accordance with the COVID-19 diagnosis and treatment protocols established by the Iraqi Ministry of Health. The SARS-CoV-2 nucleic acid reverse transcription polymerase chain reaction (RT-PCR) assay was utilized to confirm the diagnosis of COVID-19 by detecting the presence of SARS-CoV-2 in nasopharyngeal swab samples collected during hospitalization. Clinical and laboratory findings were recorded from the patients' medical records at the time of admission.

2.2. Ethical Considerations

The Ethics Committee of Al-Kitab University, College of Pharmacy, approved this study (approval number: 00364, February 15, 2022) in accordance with the ethical guidelines established by the Declaration of Helsinki. Written informed consent was obtained from patients for access to their medical and laboratory records.

2.3. Inclusion Criteria

Patients included in this study were adults (≥ 18 years old) who were admitted with symptoms indicative of acute respiratory tract infections, such as fever, cough, and dyspnea, without any other identifiable clinical cause. Chest CT scans confirmed the diagnosis of pneumonia in all patients. COVID-19 patients were further subclassified into mild, moderate, or severe categories based on the World Health Organization (WHO) criteria, as outlined in references [19,20]. Our study encompassed all COVID-19 patients classified as having mild, moderate, or severe cases who received treatment and follow-up at the center between March and May 2022 and had complete baseline clinical and laboratory data.

2.4. Exclusion Criteria

The study excluded pregnant or lactating women, cancer patients, immunosuppressed individuals, and subjects with liver cirrhosis (comorbidities). Patients who received calcium or potassium supplements, intravenous electrolyte replacement fluids, or glucocorticoids during admission were excluded from in this study.

2.5. Study Outcomes

The primary study outcomes were the levels of the studied electrolytes and laboratory biomarkers.

2.6. Data Analysis

The variables collected from patients enrolled in the present study were subjected to statistical analysis using R Studio (v1.3.1093, Switzerland). A one-way analysis of variance was performed to compare the multiple groups, and intergroup comparisons were assessed using Tukey's post hoc test. A p-value of less than 0.05 was considered indicative of a statistically significant difference.

3. Results

A total of 310 patients diagnosed with COVID-19 were enrolled in the current study. Among them, 106 (34%) were found to have mild disease, 102 (33%) had moderate disease, while the other 102 (33%) had severe disease upon admission. Based on gender, 159 (51.3%) were males and 151 (48.7%) were females. The study sample was subcategorized into five age categories. The mean age of the participants was 50.25 years (± 10.34), with an age range of 18 to 69 years. Further details are provided in **Table 1**.

Table 1. Demographic Characteristics of the COVID-19 Patients Included in the Study.

Variables		Frequency N (%)			
		Mild (N=106)	Moderate (N=102)	Severe (N=102)	Total (N=310)
Gender	Male	54(33.9%)	52(32.8%)	53(33.3%)	159(100%)
	Female	52(34.4%)	50(33.1%)	49(32.5%)	151(100%)
Age	18-29	20(39.2%)	17(33.3%)	14(27.5%)	51(100%)
	30-39	23(38.9%)	20(33.9%)	16(27.2%)	59(100%)
	40-49	21(33.9%)	20(32.2%)	21(33.9%)	62(100%)
	50-59	20(30.3%)	21(31.8%)	25(37.9%)	66(100%)
	60-69	22(30.6%)	24(33.3%)	26(36.1%)	72(100%)

3.1. Effect of COVID-19 on Serum Electrolytes

Severe COVID-19 patients demonstrate significantly lower mean calcium concentrations (mmol/L) compared to those with moderate or mild cases ($p < 0.01$). The calcium levels were (0.88 ± 0.04), (1.11 ± 0.21), and (1.13 ± 0.11) for the severe, moderate, and mild groups, respectively (**Table 2**). In terms of gender, both male and female patients in the severe group demonstrated significantly lower mean calcium levels when compared statistically to those in the moderate or mild groups (**Table 3**). Similarly, the potassium levels (mmol/L) in patients with severe COVID-19 were significantly lower when compared to those in the mild group ($p < 0.05$), with mean values of 3.03 ± 0.32 and 4.17 ± 0.46, respectively (**Table 2**). It was observed that both calcium and potassium were below the normal range in the severe group. However, a statistically non-significant difference in sodium levels (mmol/L) was observed among the various groups of COVID-19 patients. Furthermore, no statistically significant difference in chloride levels (mmol/L) was found between the different study groups.

Table 2. Levels of Serum Electrolytes, Inflammatory Biomarkers, Liver Function Enzymes and Kidney Function Markers among Studied Groups of Patients with Mild, Moderate, and Severe COVID-19.

Parameters	Reference Value	Mild (N=106) Mean ± SD	Moderate (N=102) Mean ± SD	Severe (N=102) Mean ± SD	P-Value	Summary of P-Value
Calcium mmol/L	1.0–1.3	1.13 ± 0.11	1.11 ± 0.21	0.88 ± 0.04	Mi & Mo, 0.71 Mi & S, < 0.01 Mo & S, < 0.01	ns ** **
Potassium mmol/L	3.5–5.1	4.17 ± 0.46	3.83 ± 0.45	3.03 ± 0.32	Mi & Mo, 0.63 Mi & S, 0.0010 Mo & S, 0.057	ns ** ns
Sodium mmol/L	135–145	137.31 ± 3.10	137.1 ± 4.79	136.1 ± 4.98	Mi & Mo, 0.80 Mi & S, 0.43 Mo & S, 0.28	ns ns ns
Chloride mmol/L	95–115	101.08 ± 2.10	96.9 ± 5.9	101.42 ± 6,74	Mi & Mo, 0.35 Mi & S, 0.87 Mo & S, 0.38	ns ns ns
C–reactive protein mg/L	< 5	1.503± 0.37	25.34 ± 17.62	94.78 ± 51.12	Mi & Mo, 0.001 Mi & S, < 0.0001 Mo & S, < 0.0001	** **** ****

Table 2. *Cont.*

Parameters	Reference Value	Mild (N=106) Mean ± SD	Moderate (N=102) Mean ± SD	Severe (N=102) Mean ± SD	P-Value	Summary of P-Value
Procalcitonin ng/mL	< 0.05	0.12 ± 0.07	0.22 ± 0.07	0.26 ± 0.12	Mi & Mo, < 0.001 Mi & S, < 0.001 Mo & S, 0.091	*** *** ns
ALP IU/L	44–147 I	67.40 ± 13.23	103.4 ± 59.95	132.5 ± 95.56	Mi & Mo, 0.062 Mi & S, < 0.0001 Mo & S, 0.107	ns **** ns
AST IU/L	10–40	20.56 ± 4.28	25.16 ± 8.83	28.09 ± 6.30	Mi & Mo, 0.063 Mi & S, 0.0009 Mo & S, 0.137	ns **** ns
Bilirubin mg/dL	0.1–1.2	0.30 ± 0.08	0.53 ± 0.25	0.73 ± 0.64	Mi & Mo, 0.123 Mi & S, 0.0004 Mo & S, 0.117	ns *** ns
ALT IU/L	7–56	21.06 ± 4.97	22.40 ± 8.42	24.65 ± 7.82	Mi & Mo, 0.712 Mi & S, 0.127 Mo & S, 0.369	ns ns ns
Blood Urea mg/dL	7–20	21.31± 7.57	26.59 ± 9.31	28.33 ± 8.03	Mi & Mo, 0.032 Mi & S, 0.0052 Mo & S, 0.699	* ** ns
Creatinine mg/dL	0.6–1.3	0.54 ± 0.10	0.99 ± 0.21	1.13 ± 0.33	Mi & Mo, <0.001 Mi & S, < 0.001 Mo & S, 0.112	*** *** ns

Note: Results were evaluated using a one-way ANOVA with Tukey's multiple comparisons test. The results are presented as means ± SD. A p-value of less than 0.05 was considered as statistically significant. Mi=Mild, Mo=Moderate, S=Severe.

3.2. Effect of COVID-19 on Inflammatory Markers

CRP level (mg/L), which is an inflammatory and infection marker, was statistically higher in severe cases when statistically compared to the moderate group, and in the moderate group when statistically compared to the mild group (**Table 2**). In terms of gender, both male and female patients in the severe cases exhibited statistically higher levels of C-reactive protein (CRP) compared to those in the moderate group. Furthermore, both male and female patients in the moderate group displayed statistically higher CRP levels than their counterparts in the mild group (**Table 3**). The other inflammatory marker, PCT (ng/ml), was also statistically higher in severe cases when compared to the moderate or mild group with values of (**Table 2**). It should be noted that the normal range for PCT is (0–0.5 ng/mL). Both male and female patients in the severe cases exhibited statistically higher PCT levels when compared to the mild group (**Table 3**).

Table 3. Levels of Serum Electrolytes, Inflammatory Biomarkers, Liver Function Enzymes and Kidney Function Markers Based on Gender among Studied Groups of Patients with Mild, Moderate, and Severe COVID-19.

Parameters	Gender	Mild (N=106) Mean ± SD	Moderate (N=102) Mean ± SD	Severe (N=102) Mean ± SD	P-Value Male	Summary of P-Value	P-Value Female	Summary of P-Value
Calcium mmol/L	M F	1.14 ± 0.12 1.08 ± 0.08	1.14 ± 0.11 1.12 ± 0.05	0.88 ± 0.04 0.89 ± 0.05	Mi & Mo, 0.98 Mi & S, <0.001 Mo & S, <0.001	ns ** **	Mi & Mo, 0.71 Mi & S, < 0.001 Mo & S, < 0.001	ns ** **
Potassium mmol/L	M F	4.20 ± 0.50 4.17 ± 0.54	3.99 ± 0.43 3.90 ± 0.46	3.86 ± 0.44 3.88 ± 0.43	Mi & Mo, 0.281 Mi & S, 0.013 Mo & S, 0.59	ns * ns	Mi & Mo, 0.21 Mi & S, 0.024 Mo & S, 0.51	ns * ns
Sodium mmol/L	M F	138.23 ± 3.58 136.61 ± 4.25	137.58 ± 3.90 136.88 ± 3.66	136.7 ± 4.86 136.44 ± 3.3	Mi & Mo, 0.70 Mi & S, 0.65 Mo & S, 0.93	ns ns ns	Mi & Mo, 0.86 Mi & S, 0.81 Mo & S, 0.93	ns ns ns
Chloride mmol/L	M F	101.12 ± 2.1 101.1 ± 1.8	99.54 ± 5.71 99.66 ± 5.7	99.87 ± 6.92 103.4 ± 6.10	Mi & Mo, 0.36 Mi & S, 0.24 Mo & S, 0.45	ns ns ns	Mi & Mo, 0.67 Mi & S, 0.47 Mo & S, 0.11	ns ns ns
C–reactive protein mg/L	M F	1.53 ± 0.40 1.45 ± 0.34	23.06 ± 13.30 28.16 ± 21.81	95.50 ± 53.47 92.27 ± 44.27	Mi & Mo, 0.041 Mi & S, < 0.0001 Mo & S, < 0.0001	* **** ****	Mi & Mo, 0.0065 Mi & S, < 0.0001 Mo & S, < 0.0001	** **** ****
Procalcitonin ng/mL	M F	0.136 ± 0.08 0.111 ± 0.09	0.25 ± 0.05 0.167 ± 0.06	0.29 ± 0.134 0.21 ± 0.14	Mi & Mo, 0.002 Mi & S, 0.002 Mo & S, 0.315	** *** ns	Mi & Mo, 0.095 Mi & S, 0.0002 Mo & S, 0.29	ns *** ns

Table 3. *Cont.*

Parameters	Gender	Mild (N=106) Mean ± SD	Moderate (N=102) Mean ± SD	Severe (N=102) Mean ± SD	P-Value Male	Summary of P-Value	P-Value Female	Summary of P-Value
ALP IU/L	M F	67.20 ± 12.15 67.73 ± 15.3	109.2 ± 66.84 89.9 ± 26.33	152.9 ± 113.6 101.6 ± 44.57	Mi & Mo, 0.14 Mi & S, 0.0003 Mo & S, 0.07	ns *** ns	Mi & Mo, 0.27 Mi & S, 0.012 Mo & S, 0.58	ns * ns
AST IU/L	M F	21.3 ± 4.76 19.33 ± 3.57	26.7 ± 7.29 23.92 ± 9.87	28.13 ± 6.83 27.17 ± 5.33	Mi & Mo, 0.10 Mi & S, 0.019 Mo & S, 0.74	ns * ns	Mi & Mo, 0.28 Mi & S, 0.042 Mo & S, 0.37	ns * ns
Bilirubin mg/dL	M F	0.30 ± 0.09 0.32 ± 0.08	0.53 ± 0.33 0.56 ± 0.21	0.72 ± 0.77 0.77 ± 0.4	Mi & Mo, 0.24 Mi & S, 0.007 Mo & S, 0.29	ns ** ns	Mi & Mo, 0.11 Mi & S, <0.001 Mo & S, 0.15	ns *** ns
ALT IU/L	M F	21.67 ± 4.92 19.87 ± 5.19	22.21 ± 10.06 22.40 ± 6.53	24.29 ± 8.31 24.09 ± 4.25	Mi & Mo, 0.97 Mi & S, 0.53 Mo & S, 0.62	ns ns ns	Mi & Mo, 0.37 Mi & S, 0.16 Mo & S, 0.69	ns ns ns
Blood Urea mg/dL	M F	20.57 ± 6.8 22.73 ± 9.06	26.09 ± 11.16 27.50 ± 4.58	29.07 ± 7.14 32.27 ± 6.7	Mi & Mo, 0.08 Mi & S, 0.004 Mo & S, 0.24	ns *** ns	Mi & Mo, 0.23 Mi & S, 0.003 Mo & S, 0.19	ns ** ns
Creatinine mg/dL	M F	0.55 ± 0.10 0.58 ± 0.13	1.01 ± 0.21 0.97 ± 0.22	1.11 ± 0.35 1.09 ± 0.39	Mi & Mo, <0.001 Mi & S, <0.001 Mo & S, 0.31	*** *** ns	Mi & Mo, < 0.001 Mi & S, < 0.001 Mo & S, 0.402	*** *** ns

Note: Results were evaluated using a one-way ANOVA with Tukey's multiple comparisons test. The results are presented as means ± SD. A p-value of less than 0.05 was considered as statistically significant. Mi=Mild, Mo=Moderate, S=Severe.

3.3. Effect of COVID-19 on Liver Function Enzymes

The levels of liver function enzymes, specifically ALP and AST, both measured in IU/L, along with bilirubin levels quantified in mg/dL, were found to be statistically significantly elevated in the severe COVID-19 group compared to the mild group ($p < 0.001$) (**Table 2**). In terms of gender differences, both male and female patients in the severe COVID-19 cohort exhibited statistically higher levels of ALP, AST, and bilirubin compared to the mild group (**Table 3**). Conversely, no statistically significant difference in ALT levels (IU/L) was observed among the various COVID-19 patient groups (**Tables 2** and **3**).

3.4. Effect of COVID-19 on Blood urea and creatinine

Blood urea (mg/dL) and creatinine (mg/dL) are essential biomarkers for evaluating renal function and overall health status. When compared to the mild group, patients with severe and moderate COVID-19 exhibited statistically significant elevations in these two markers (**Table 2**). Specifically, the mean blood urea levels were 28.33 ± 8.03, 26.59 ± 9.31, and 21.31 ± 7.57 for the severe, moderate, and mild groups, respectively. The mean creatinine levels were 1.13 ± 0.33, 0.99 ± 0.21, and 0.54 ± 0.10 for the severe, moderate, and mild groups, respectively (**Table 2**). Furthermore, both male and female patients in the severe group demonstrated statistically higher levels of blood urea and creatinine compared to those in the mild group (**Table 3**).

3.5. Correlation between laboratory biomarkers and the severity of COVID-19 infection

Then, we sought to determine the correlation between laboratory biomarkers in patients with COVID-19 infection. To illustrate our findings, we developed circular plots to depict the correlations among various variables across different patient groups using R Studio (**Figures 1**, **2**, and **3**). Our analysis revealed no significant correlation between laboratory biomarkers in patients with mild COVID-19 (**Figure 1**). However, a strong positive correlation was observed between calcium and potassium levels in the moderate (r value = 0.7) and severe groups (r value = 0.6), as both electrolytes decreased in a similar pattern in these two groups (**Figures 2** and **3**). Consistent with our previous findings, which showed reduced calcium and potassium levels and elevated inflammatory marker CRP in moderate and severe COVID-19 patients, calcium and potassium showed a strong negative correlation with CRP (r value= -0.8 and -0.7), (r value= -0.7 and -0.8), respectively, (Table S1). In the moderate group, liver function enzymes ALT, AST, and bilirubin had a negative correlation with calcium and potassium, and a positive correlation with CRP and PCT (**Figures 2** and **3**) (Table S1). However, in the severe group, liver function enzymes ALP, ALT, and AST exhibited a negative correlation with calcium and potassium, and a positive correlation with CRP and PCT (Table

S1). Kidney function markers (blood urea and creatinine) also showed a positive correlation with the inflammatory marker CRP in the severe group (**Figure 3**).

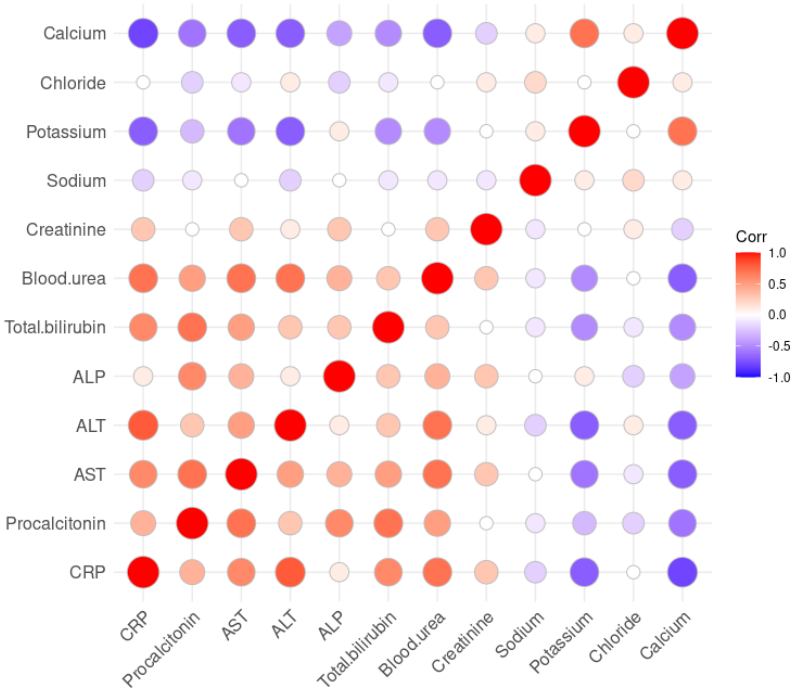

Figure 1. Correlation between laboratory biomarkers in mild COVID-19 patients, the correlation was identified using the Pearson correlation coefficient (R value).

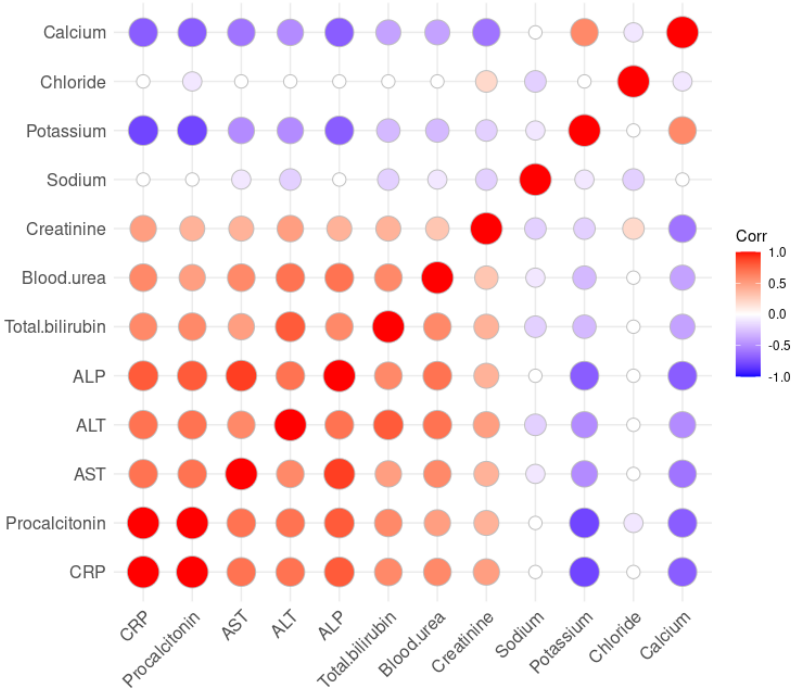

Figure 2. Correlation between laboratory biomarkers in moderate COVID-19 patients, the correlation was identified using the Pearson correlation coefficient (R value).

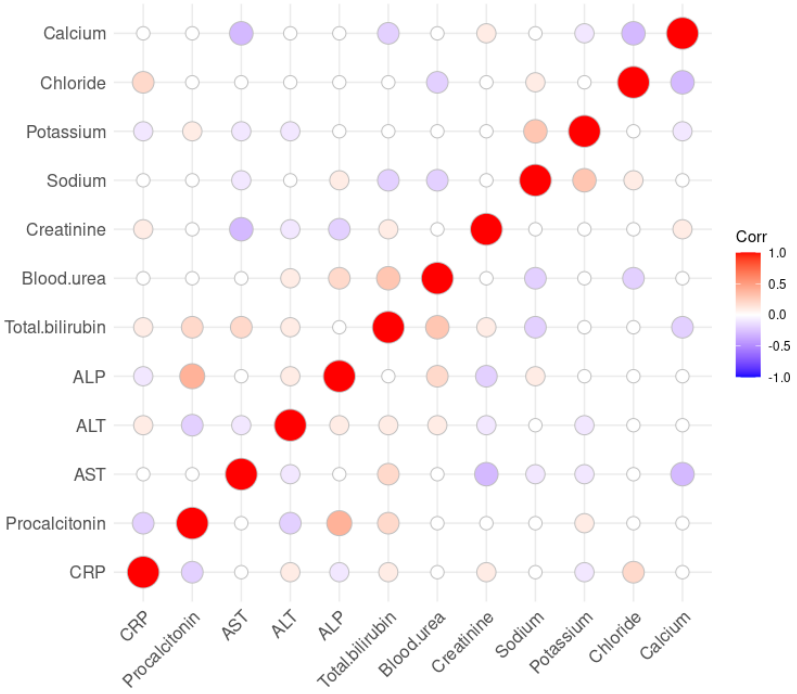

Figure 3. Correlation between laboratory biomarkers in severe COVID-19 patients, the correlation was identified using the Pearson correlation coefficient (R value).

4. Discussion

In the present study, we examined the association between biomarkers and COVID-19 disease severity in 310 patients admitted to Al-Shifa 14 hospital between March and May 2022. Identification of lab biomarkers that can assess COVID-19 severity will assist physicians in identifying patients who may develop severe disease. Our data indicates that individuals who were 50 years of age or older were at increased risk of developing into severe disease. In the severe cases, 50% of the patients were 50 years or older, while 39% of the mild cases were in the same age group. This association between age and disease severity has been observed in various studies conducted in the United States, Europe and China [21–23]. Similar findings have also been reported in a study conducted in Africa [24]. This could be attributed to weakened immune defense mechanisms and underlying health conditions that are more common in older individuals, thereby increasing their susceptibility to severe illness and adverse consequences.

Our results indicate that serum calcium and potassium levels are significantly associated with the severity of COVID-19. A statistically significant decrease in serum calcium and potassium levels was observed in patients with severe COVID-19. This study suggests that patients with lower serum electrolyte levels of calcium and potassium are at an increased risk of developing severe COVID-19. These findings align with a retrospective study conducted on 127 COVID-19 patients at Tongji Hospital in China [25]. Regardless of disease severity, there were no significant differences in serum sodium and chloride levels among the mild, moderate, and severe groups. Electrolyte imbalances may occur in any disease state due to renal damage or the pharmacological effects of medications used in treatment. Furthermore, reduced activity of ACE2, which functions as a receptor for the SARS-CoV-2 virus, may also contribute to these imbalances [26]. Electrolyte imbalances can disrupt the function of various organ systems in the body, compromising the body's ability to respond appropriately to the stress caused by the disease. This can ultimately lead to more severe illness and complications [14].

CRP is a protein synthesized in the liver, with its production primarily induced by interleukin-6 (IL-6). CRP functions as a sensitive and early biomarker for detecting inflammation and infection [27]. While the expression level of CRP is generally low, it exhibits a rapid and substantial increase in response to infection [28,29]. Elevated levels of CRP, either alone or in conjunction with other markers, can provide insights into the presence of infections.

In our study, we examined the correlation between CRP and other biomarkers in patients diagnosed with COVID-19. The levels of CRP were found to be statistically significantly higher in the moderate group compared to the mild group, as well as in the severe group compared to the moderate group. These findings align with the concept of a "cytokine storm," which posits that inflammatory factors play a pivotal role in the progression from mild to moderate and from moderate to severe manifestations of the disease. The inflammatory response plays a potentially life-threatening role in COVID-19, with an increase in inflammatory cytokines correlating with the severity of cases [30, 31]. Wan et al. [32] identified that cytokine storm is directly associated with the progression of COVID-19 and can lead to severe complications and mortality. The Diagnosis and Treatment Guidelines for COVID-19 (5th edition), published by the National Health Commission of China, recommend monitoring cytokine levels to improve treatment efficacy and decrease mortality rates [13].

PCT is a precursor of calcitonin, comprising approximately 116 amino acids, and is synthesized by the parafollicular cells of the thyroid gland [33]. Normally, PCT levels in the bloodstream are low or undetectable. However, bacterial infections can cause an increase in PCT levels, while viral infections generally result in relatively low levels. This indicates that PCT levels can be utilized to distinguish between bacterial and viral infections [34]. The findings from the moderate and severe groups showed higher PCT levels than the normal range, suggesting that some COVID-19 patients from these two groups may also have bacterial infections.

Consistent with a recent study conducted in Turkey in 2024 [35], our analyses revealed that higher levels of liver function enzymes, particularly ALP and AST, are associated with a greater risk of severe disease. The mild group had significantly lower levels of ALP, AST, and bilirubin when statistically compared to those with severe diseases. Various mechanisms indicate that the SARS-CoV-2 virus may directly affect hepatocytes. At the same time, the liver may also sustain indirect injury due to an exacerbated inflammatory response associated with elevated immune markers and the toxicity of pharmacological agents employed to treat or mitigate disease progression. Consequently, this liver damage leads to an elevation of liver enzyme levels [36].

Electrolyte imbalances and alterations in liver function enzyme levels are critical factors in the pathophysiology of severe COVID-19, as they may indicate underlying organ dysfunction and contribute to the disease's severity. Hypokalemia, or low potassium levels, is thought to stem from increased urinary potassium loss, a consequence of the interaction between the COVID-19 virus and the renin-angiotensin-aldosterone system (RAAS). The COVID-19 virus gains entry into host cells through the angiotensin-converting enzyme 2 (ACE2) receptor, potentially disrupting the RAAS and resulting in excessive secretion of aldosterone. This dysregulation leads to potassium loss and hypokalemia, which can lead to cardiac arrhythmias, muscular weakness, and respiratory failure in severely affected patients [37, 38]. Moreover, research has demonstrated that hypocalcemia, or low calcium levels, contributes to cardiovascular complications, such as arrhythmias, and exacerbates inflammatory responses. Hypocalcemia has also been associated with increased mortality rates among patients with severe COVID-19 [39].

Additionally, liver dysfunction and elevated liver enzymes, such as ALP and AST, correlate with COVID-19 severity. Increases in liver function enzymes may result from various factors, including direct viral infection of hepatocytes, cytokine storms, hypoxia due to severe respiratory failure, or drug toxicity. Elevated ALT and AST serve as biomarkers of liver damage [40, 41].

Our findings regarding electrolyte disturbances and liver enzyme alterations in COVID-19 patients are consistent with some studies worldwide. For example, a large multicenter study conducted in New York, USA, found that 18.3% of hospitalized COVID-19 patients had hypocalcemia, particularly those with poor outcomes [42]. Similarly, a case-control study conducted in France observed a high incidence of hypokalemia, which was associated significantly with disease severity [43]. In Asia, Chen et al. [37] reported that nearly 50% of COVID-19 patients in China had hypokalemia, which correlated with disease progression and impaired renal function.

Regarding alterations in liver enzymes, our findings are consistent with those reported in Northern Italy, where 62.4% of hospitalized COVID-19 patients exhibited elevated levels of ALT and GGT. These elevations were correlated with increased mortality and a greater necessity for intensive care [43]. Additionally, studies in Southeast Asia reported similar patterns: Indonesian patients with severe COVID-19 had significantly high levels of AST, ALT, and total bilirubin, suggesting a higher incidence of liver injury [44]. These comparisons highlight the global consistency of electrolyte and liver enzyme disturbances in COVID-19 patients and support the clinical relevance of our findings despite the limitations of our single-center study setting.

Our findings suggest that biomarkers such as serum calcium, potassium, CRP, and PCT may serve as early in-

dicators of disease severity in COVID-19. However, integrating these biomarkers into clinical practice presents several challenges. The availability of rapid and reliable testing for certain biomarkers, especially PCT, may be limited by factors such as high costs, inadequate equipment, and a shortage of trained personnel. In contrast, CRP and serum electrolytes, including potassium, are more widely accessible and cost-effective, making them more suitable for initial triage in primary care and emergency departments. Finally, the implementation of biomarker-based protocols must be tailored to local infrastructure, and their effectiveness should be validated through studies across diverse healthcare settings. Future research should also explore cost-benefit analyses and operational strategies to ensure access to these diagnostic tools.

5. Limitations and Strengths of the Study

Future research incorporating gene expression, protein-level analyses, novel or emerging indicators, and/or considering longitudinal data would complement our findings by offering novel mechanistic insights and a more nuanced understanding of the biological processes underlying the observed associations, including the ability to examine dynamic changes in biomarkers over time. Such approaches would be critical for validating and extending our results, providing a deeper exploration of the molecular mechanisms at play, and potentially identifying new therapeutic or prognostic targets. Another limitation is that it was conducted at a COVID-19 hospital in Iraq. This may restrict the generalizability of the results to other populations or healthcare systems with different demographic, clinical, or infrastructural characteristics.

One of the key strengths of our study lies in the comprehensive analysis of a broad spectrum of biomarkers, including serum electrolytes, liver function enzymes, renal function markers, and inflammatory biomarkers, all examined within a single study. This approach contrasts with the majority of previous literature, which predominantly focused on isolated groups of biomarkers. By implementing this comprehensive strategy, we were able to examine the potential interactions among various biochemical parameters and their relationships with disease severity and progression in patients with COVID-19. Integrating these diverse markers enhances our understanding of the disease's pathophysiology and aids in identifying potential prognostic indicators.

6. Conclusion

Low serum calcium and potassium electrolyte levels were associated with COVID-19 infection. We found that decreased levels of serum electrolytes calcium and potassium, as well as elevated liver function enzymes ALP, AST, and bilirubin, were significant predictors of developing COVID-19 disease severity. Additionally, inflammatory biomarker CRP was significantly associated with disease severity. Therefore, conducting an early assessment and monitoring of these laboratory biomarkers can be a beneficial tool for controlling disease severity.

Supplementary Materials

The supporting information can be downloaded at https://ojs.ukscip.com/files/TI-1179-Revised+Manuscript15052025_-_Done[1](2)-for-typesetting.docx.

Author Contributions

Conceptualization, O.D. and M.H.M.; methodology, O.D. and M.H.M.; software, O.D. and M.H.M.; validation, O.D. and M.H.M..; formal analysis, O.D. and M.H.M.; investigation, O.D. and M.H.M.; resources, O.D. and M.H.M.; data curation, O.D. and M.H.M.; writing—original draft preparation, O.D. and M.H.M.; writing—review and editing, O.D. and M.H.M.; visualization, O.D. and M.H.M.; supervision, O.D. and M.H.M.; project administration, O.D. and M.H.M.; funding acquisition, O.D. and M.H.M.. All authors have read and agreed to the published version of the manuscript.

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Institutional Review Board Statement

The ethics committee of Al-Kitab University, College of Pharmacy (approval number: 00364, February 15th, 2022).

Informed Consent Statement

Informed consent was acquired from all individual subjects involved in the study.

Data availability statement

Upon request, the corresponding author can provide the data that substantiates the findings of this study.

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Conflict of Interest

The authors declare no competing interests.

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