Exploring the Influence of Neutrophil-Lymphocyte Ratio, D-Dimer, C - Reactive protein, and Ferritin on Outcome Prediction of Severely-Ill Patients with COVID-19 in Al-Sader Medical City in Al-Najaf City, Iraq

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ABSTRACT:

BACKGROUND:

Coronaviruses are microorganisms of animal and mankind. The WHO categorized the disease as COVID-19 in February, 2020.

Various laboratory markers can support the identification and follow up of COVID-19 case. Current evidence proposes a prognostic impact of neutrophil-lymphocyte ratio (NLR) in diverse inflammatory syndromes. Hence, it is rationalized to investigate its role as a prognostic biomarker in severe COVID-19 pneumonia.

OBJECTIVE:

The objective is to analyze the effect of NLR, D-dimer, CRP, and ferritin role in the prediction of consequence of inpatients with severe COVID-19.

METHOD OF THE STUDY:

• A retrospective cross-sectional study was conducted on 96 patients aged (18-80) years whom PCR SARS-CoV-2 positive, and admitted to (Al-Sader Medical City) in Al-Najaf City, Iraq between (July to October 2020).

•The absolute number of neutrophil count divided by the absolute number of lymphocyte count is measured on admission to determine the value of (NLR). Parameters like (D-dimer, C-reactive protein, and ferritin) also have been investigated in relation to outcome.

RESULT:

High (NLR) was significantly (p-value = 0.001) linked with lower oxygen concentration and worse prognosis.

CONCLUSION:

Raised (NLR) was a convenient indicator connected with unfortunate outcome in those admitted with COVID-19. High inflammatory markers, like ferritin, CRP, and D-dimer were also associated with doomed prognosis.

KEYWORDS: coronaviruses, neutrophil to lymphocyte ratio, prognosis.

INTRODUCTION:

Coronaviruses are microorganisms of animal and mankind. In the late 2019, recognition of a new coronavirus was established after the appearance of atypical viral pneumonia in Wuhan, capital of Hubei, China. It swiftly spread, causing an epidemic all over China, afterwards, a growing amount of cases in further countries all through the world. The WHO categorized the condition as COVID-19 in February, 2020, that is, coronavirus disease 2019. The WHO proclaimed the illness as international pandemic on March 11, 2020 [(1)]. Symptomatic infection spectrum varies from mild stage to critical; mostly they are not severe [(2)].

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Mild cases when the constitutional symptoms exist, but lacking shortness of breath or hypoxia. In moderate cases, the patient may present breathless along with constitutional symptoms of fever, cough and radiological findings of pneumonia without hypoxia. A severe case defined as the respiratory rate ≥ 30 rpm, SaO_2 of ≤ 93%, or PaO_2/FiO_2 ≤ 300 mmHg, together with respiratory involvement of ≥ 50%. Critical level is likely when if a patient is in respiratory failure that necessities mechanical ventilation, in shock state, or has multi-organ failure [(3)].

The keystone diagnostic test for COVID-19 is by the polymerase chain reaction (PCR) test, to identify the viral genetic material on an upper respiratory swab [(4)].

Lymphopenia is similarly one of the utmost recognized hematologic abnormalities that serve as a great prognostic tool.

Various healthcare centers and professionals used particular parameters to predict outcome for instance neutrophil-to-lymphocyte ratio (NLR)[5]. Neutrophil to lymphocyte ratio is a simple, quantifiable, and cheap biomarker of inflammation in infected patients with COVID-19, thus can be employed on admission as a marker of prognosis [3].

The normal value of (NLR) in a healthy individual has been described as (0.78-3.5) [9].

AIM OF THE STUDY:

The study's objective is to have a better understanding of the role of neutrophil-lymphocyte ratio in the determination of the consequence of COVID-19 and to assist in predicting disease severity.

PATIENTS AND METHODS:

1. PATIENTS:

- A total of 96 patients (male: 44 and female: 52) within age group 18-80 years who verified positive cases for COVID-19 based upon the nucleic genome detected from a nasopharyngeal and oropharyngeal swab using PCR, who were admitted to (Al-Sader Medical City) in Al-Najaf City between (July to October 2020) were enrolled in a retrospective cross-sectional type of study.
- Exclusion: known malignancy, Pregnancy.

2. METHOD:

- The files of 96 COVID-19 patients were reviewed, after granting permission from Al-Sader Medical City administration committee, for patients' medical history, laboratory tests, the treatment regime, and outcomes.
- The absolute number of neutrophil count divided by the absolute number of lymphocyte count on admission to determine the value of (NLR) and parameters like (D-dimer, C-reactive protein, and ferritin) also have been investigated in relation to outcome.

Through the applying of the receiver operating characteristic (ROC) curve, the ultimate threshold values of the continuous variables were calculated.

Statistical Analysis

The statistics were scrutinized by utilizing SPSS program version {24}. The continuous variables were displayed via a mean and standard deviation; and were investigated using a t-test. Categories variables presented as number and percent, these variables underwent chi-square test to study the significance among variables. Via application of the receiver operating curve (ROC) analysis, the ideal cut-off values of the continuous variables were calculated. To exclude the effect of the confounding factors on the outcome (age, gender, chronic diseases), we conducted binary logistic regression analysis. The test was considered significant when P-value ≤ 0.05.

RESULTS:

Table 1: Epidemiological data.

`Variable	Results No.
Age in year (mean ±SD)	58.3± 13.8
Gender	M: 44, F: 52
Average stay in the hospital (days)	11.64 ±4.6
Total death	30
Diabetes	36
Heart failure	10
Obstructive airways disease	12
Chronic kidney disease	7
Hypertension	60
Average oxygen saturation at admission	78.33 ± 6.2%

Table 2: The relationship between comorbid diseases and oxygenation saturation in COVID-19 patients on presentation.

Variable		O_2 saturation (⩾80%) No. (52)	O_2 saturation (<80%) No. (44)	P value
Age (years)	20-60 (N: 51)	30	17	0.8
	> 60 (N: 45)	22	27	
Gender	M (N: 44)	23	21	0.7
	F (N: 52)	29	23	
DM*	Yes 36	20	16	0.3
	No 60	32	28	
HT**	Yes 60	33	27	0.4
	No 36	19	17	
HF***	Yes 10	3	7	0.7
	No 86	49	37	
OAD****	Yes 12	4	8	0.5
	No 84	48	36	
CKD*****	Yes 7	1	6	0.03
	No 89	51	38	

*Diabetes mellitus, **Hypertension, ***Heart failure,
****Obstructive airways disease, *****Chronic kidney disease.

Table 3: The correlation of laboratory tests and O_2 saturation on admission.

parameter	Mean ±SD	Oxygen saturation	No.	p-value
NLR	9.85±3.427	>80	52	0.001
	15.93±3.26	<80	44	
HB (g/dl)	12.1±1.434	>80	52	0.43
	11.89±2.082	<80	44	
Platelet /mm^3	220880.3±108441.4	>80	52	0.61
	236410.1±146497.5	<80	44	
CRP (mg/L)	71.26±22.211	>80	52	0.001
	94.15±19.96	<80	44	
Ferritin (ng/ml)	755.82±450.33	>80	52	0.001
	1336.4±497.491	<80	44	
D-dimer (µg/ml)	2.89±3.13	>80	52	0.002
	6.443±4.855	<80	44	

Table 4: The influence of clinical background on the consequence of patients with COVID-19.

Variables		Survivor	Death	p-value
Age (years)	20-60 (N:47)	38	9	0.02
	> 60 (N:49)	28	21	
Gender	M (N: 44)	26	18	0.07
	F (N: 52)	40	12	
DM*	Yes (N: 36)	24	12	0.4
	No (N:60)	42	18	
HT**	Yes (N: 60)	44	16	0.3
	No (N: 36)	22	14	
HF***	Yes (N:10)	7	3	0.7
	No (N:86)	59	27	
OAD****	Yes (N:12)	5	7	0.04
	No (N:84)	61	23	
CKD*****	Yes (N:7)	3	4	0.2
	No (N:89)	63	26	

*Diabetes mellitus, **Hypertension, ***Heart failure,
****Obstructive airways disease, *****Chronic kidney disease.

Table 5: The association between laboratory variables and the outcome in COVID-19 patients.

Lab. variable	Mean ±SD	outcome	p-value
NLR	18.01±1.626	Death	0.001
	10.20±2.9	survivor	
HB (g/dl)	12.05±1.97	Death	0.9
	11.99±1.651	survivor	
Platelet /mm^3	207670.2±129829.3	Death	0.2
	237240.2±125358.1	survivor	
CRP (mg/L)	101.2±18.62	Death	0.001
	73.02±20.32	survivor	
Ferritin (ng/ml)	1435.3±489.83	Death	0.002
	834.1±474.16	survivor	
D-dimer (µg/ml)	8.71±5.135	Death	0.003
	2.6188±2.10525	survivor	

Table 6: Calculation of the odd ratio of the comorbid conditions and laboratory parameters using logistic regression analysis.

Variables	OR (95% CI)	p-value
Hypertension	1.02 (0.07-2.1)	0.2
Diabetes	1.58 (0.65–1.84)	0.3
Heart failure	1.2 (0.9–2.4)	0.1
Obstructive airways disease	1.4 (1.03–1.6)	0.04
Chronic kidney diseases	1.09 (0.2–1.32)	0.8
NLR at admission	1.626 (1.359–1.946)	0.001
CRP at admission	1.45 (0.671–1.84)	0.1
D-dimer at admission	1.32 (1.08–1.390)	0.001

DISCUSSION:

In this study, the average age of the patients was 58.3± 13.8. The male were about 45.8% and females were 54.2%. While a study by Burugu. H. who recorded male incidence of 74% in his sample of patients [12].

Many patients in the study had comorbidities, such as hypertension in 62.5%, diabetes in 37.5%, obstructive airway disease in 12.5 %, heart failure 10.4%, and renal impairment in 7.2%. In accordance to study by Ai-Ping Yang whose found the hypertension presented in 66%, diabetes 54%, heart failure 37.%, and kidney dysfunction in 33.3%. Patients with chronic diseases were showed high frequency of severe disease presentation [v].

In this study, mortality rate was (31.25%); which is higher than worldwide mortality; this can be attributed to the fact that the study was conducted only on the severe cases that necessities hospital admission.

Likewise, the mean of (NLR) for the dead and recovered patients was significantly different (0.001). The mean of NLR in non-survivor patients was (18.01±1.626) which is higher than that for the recovered patients (10.20±2.9). Additionally, a statistical difference between the two clusters in serum ferritin, CRP, and D-dimer levels (0.002, 0.001, and 0.003, respectively). However, there were no statistical differences in hemoglobin levels and platelet counts (0.9, 0.2 respectively). The result illustrates the independent influences for progression of COVID-19 disease after excluding the effect of gender, age, and co-morbidities. With the aid of multivariate regression analysis, our study reported NLR was independent discriminator for the disease progression as higher levels on admission linked with inferior O_2 saturation and an unfortunate outcome. These results are consistent with other studies that reported the incorporation of raised NLR as a predictive factor that could result in poor prognosis [7]. The study results were steady with the aforementioned studies concerning the association of NLR and the outcome of further contagious illnesses [8].

Many reasons may be behind these results. Neutrophils are considered as a main component of the leukocytes gathering nearly (50-70%) which act and migrate from the venous circulation to the immune organs and further systems, such as the lungs. Neutrophils discharge a huge amount of reactive oxygen species (ROS) which stimulate cellular DNA damaging and viral releasing from cells. Through this action, the virus becomes free and destroyed via antibody mediated immunity. Furthermore, neutrophils interact with a distinctive cell population and release numerous types of cytokines and growth vascular endothelial growth factors that lead to hyper-inflammation and more damage in the alveolar lining that is unconnected to the viral deleterious effect [7].

Furthermore, the immunological reaction towards a viral infection occurs mostly via lymphocytes[9]. Systematic inflammation considerably reduces cell immunity, which meaningfully depresses CD4+ T lymphocyte [8].

Thus, as a result of the migration of neutrophils and the drop in lymphocytes, the inflammation that triggered via the virus elevates NLR. This increase in NLR encourages progression of COVID-19 disease. Additionally, the clinical features become severe and escalate the possibilities of ICU admission or invasive ventilation [7].

Meanwhile, a different research by Qin et al. described that the NLR was significantly elevated in sufferers with severe COVID-19 illness in a group of hospitalized patients [10].

A study of Zhang et al, found the neutrophil was much higher in severe cases and patients of poor outcome such as patients hooked on mechanical ventilation or those admitted in an ICU. They were concluded increase neutrophil count might be due to excess inflammation response [11]. When recovered, in almost all cases, lymphocyte counts return back to normal levels [12].

Besides, a meta-analysis study that encompasses six studies with more than 800 patients exhibited higher NLR serves as a worthy prognosticator of clinical severity and outcome [13].

There was a rise in serum ferritin levels in non-survivor (1435.3±489.83) more than patients who survivor survived (834.1±474.16) in accord with study by Burugu et al, who revealed the amplified levels of serum ferritin were detected in most patients with COVID-19, it was highly raised in dead patients with COVID-19 when compared to the improved patients [12].

The study also revealed that CRP was significantly raised in deceased patients (101.2±18.62) than those who ultimately recovered (73.02±20.32), which correlates with the degree of inflammation. This comes along with study by Matsumoto that exhibited higher CRP values in severe COVID-19 infection [14].

An additional outcome for the study is a statistically significant difference between the values of D-dimer (0.003) was found between the recovered and the dead. D-dimer levels were high in the non-survivor group, which are consistent with Hai-Han Yu study [15].

CONCLUSION:

The study had established that elevated NLR at admission was a useful prognostic marker linked with poor outcome in patients admitted for COVID-19 pneumonia. Other inflammatory markers, such as ferritin, CRP, and D-dimer were also associated with critical illness and increased mortality from COVID-19 disease.

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