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Role of total antioxidant capacity, white blood cell and platelets to lymphocyte ratio with severity of corona virus -19 disease

A graduation project submitted to the faculty of nursing university of Babylon as part of the requirements for obtaining bachelor's degree in nursing by

Zahraa Jassim Owaid

Zahraa Saleh Hadi

Zahraa Mohammed Hassoun

Zainab Saleh Hadi

supervisor

Dr. Ameera AL-Aaraji

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بسم الله الرحمن الرحيم

((فَلْيَنْظُرِ الْإِنْسَانُ إِلَىٰ طَعَامِهِ ﴿٢٤﴾ أَنَّا صَبَبْنَا الْمَاءَ صَبًّا ﴿٢٥﴾
ثُمَّ شَقَقْنَا الْأَرْضَ شَقًّا ﴿٢٦﴾ فَأَنْبَتْنَا فِيهَا حَبًّا ﴿٢٧﴾
وَعِنَبًا وَقَضْبًا ﴿٢٨﴾ وَزَيْتُونًا وَنَخْلًا ﴿٢٩﴾
وَحَدَائِقَ غُلْبًا ﴿٣٠﴾ وَفَاكِهَةً وَأَبًّا ﴿٣١﴾ مَتَاعًا لَكُمْ وَلِأَنْعَامِكُمْ ﴿٣٢﴾))

سورة عبس من الآية 24 إلى 32

الإهداء

إلى من جرع الكأس فارغاً ليسقيني قطرة حب

إلى من كلّت أنامله ليقدم لنا لحظة سعادة

إلى من حصد الأشواك عن دربي ليمهد لي طريق العلم

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إلى الروح التي سكنت روحي

الآن تفتح الأشرعة وترفع المرساة لتنطلق السفينة في عرض بحر واسع مظلم هو بحر الحياة وفي هذه الظلمة لا يضيء إلا قنديل الذكريات ذكريات الأخوة البعيدة إلى الذين أحببتهم وأحبوني (أصدقائي)

الشكر والتقدير

الشكر والتقدير لا يسعنا بعد الانتهاء من اعداد هذا البحث الا ان نتقدم بجزيل الشكر وعظيم

الامتنان الى الدكتورة الفاضلة

(.د اميره جاسم)

التي تفضلت بالأشراف على هذا البحث حيث

قدمت لنا كل النصح والدعم والارشاد طيلة فترة الإعداد فلها منا كل الشكر والتقدير .

وكذلك نتقدم بالشكر الجزيل د. منقذ الجنابي لمساعدته بجمع العينات من مرضى

ولا ننسى المرضى اللذين ساهموا في انجاح هذا المشروع متمنين الهم الصحة والعافية

وكما نشكر كل من ساعدنا من قريب أو بعيد ولو بكلمة أو دعوة صالحه .

Abstract:-

Background ; Coronavirus disease 2019 (COVID-19) is a rapidly emerging disease caused by a highly contagious virus called severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), The significance of the study lies in the need to evaluate the levels of antioxidants, white blood cell (WBC) as well as platelets to lymphocyte ratio (PLR) among mild and sever COVID-19 patients so that the outcome can imply a more robust and holistic approach to combating the devastating disease.

Objectives :To evaluate role of total antioxidant capacity, WBC, PLR with disease severity of COVID-19**.**

Patients and Methods: A purposive study was carried out in this study, which includes 60 individuals. These subjects were divided into two groups: The first group includes 30 sever COVID-19 patients and the second group includes 30mild COVID-19 patients . Blood tests were performed using routine methods for the detection of WBC, Lymphocyte, and platelets measurement . The total antioxidant capacity measurement by manual method.

Results: In this study, total antioxidant capacity, WBC, and PLR were measured in sever and mild COVID-19 patients. Results demonstrated significant alterations in total antioxidant capacity, WBC as well as PLR between two groups. Contribution of those factors with severity of the disease has been shown.

Conclusions:

-Total Antioxidant capacity is the significant variable relates with severity of COVID-19 disease.

- PLR is inflammatory marker, has been suggested to predict the severity of COVID-19 patients and may be considered as cardinal laboratory findings, with prognostic potential. WBC counts increased significantly as the disease severity increased.

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ABBREVATION	Meaning
ARDS	Acute respiratory distress syndrome
BMI	Body mass index
CBC	Complete Blood Count.
COVID -19	coronavirus -2019.
H2O2	Hydrogen peroxide.
HIV	human immunodeficiency virus
MODS	Multiple Organ Dysfunction Syndrome.
NADPH	Nicotinamide adenine dinucleotide phosphate.
NOX	Nitrogen Oxides.
OS	Oxidative stress.
PLR	Platelet-to-Lymphocyte Ratio.
ROS	Reactive oxygen species .
RT-PCR	reverse transcription-polymerase chain reaction

SARS-CoV-2	severe acute respiratory syndrome coronavirus 2.
SOD	Superoxide dismutase
WBC	White blood cells.

CHAPTER ONE
INTRODUCTION AND LITRETURE REVIEW

INTRODUCTION AND LITRETURE REVIEW

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) was responsible for Coronavirus disease 2019 (COVID-19), the first cases having been reported in Wuhan ,China, in December 2019 [1]. Due to fast transmission and pathogenicity, the coronavirus has spread across all countries, provoking a pandemic [2].

The COVID-19 is a pandemic caused by SARS-CoV-2 which has infected over 74 million people, killing more than 1,600,000 million people globally as of 17th December 2020[3].

Coronaviruses are a set of enveloped, single-stranded, positive-sense RNA genome viruses that are capable of infecting humans and promoting respiratory and gastroenterological infections. There are four subfamilies of coronaviruses: alpha, beta, gamma, and delta. The SARSCoV-2 belongs to the beta family, along with other coronavirus linked to severe diseases related to the respiratory tract [4].

COVID-19 is characterized by aberrant host immune response, leading to excessive inflammatory responses (or cytokine storm)—as evidenced by high blood levels of cytokines, chemokines and C-reactive protein—and is associated with severe damage to the respiratory system and multi-organ failure, contributing to fatal outcomes of infected patients [5]. Patients typically present with fever and cough; gastrointestinal tract manifestations such as diarrhea, vomiting and abdominal pain; and more recently [6] .

Immunological studies have shown that high levels of pro inflammatory cytokines, known as a cytokine storm, are the hallmark characteristic of severe COVID-19 cases. This extreme elevation of cytokines causes a massive pro inflammatory response resulting in Multiple Organ Dysfunction Syndrome (MODS) and ARDS, which subsequently leads to mortality in COVID-19 patients [7].Therefore, in theory, inflammatory markers can be used to assess the severity and mortality risk of COVID-19 patients.

Moreover, hematological parameter and indexes such as the platelet-to lymphocyte ratio were investigated as potential indicator of the severity of the COVID-19 [8]. Complete blood counts (CBCs), play an essential role when dealing with infectious diseases. Recently, it has been reported that hematological parameters and based on blood cell analysis had an important predictive value for the prognosis of infections, and many other diseases [9] .

In a large number of pathologies, inflammation is known to be closely related to oxidative stress, one process being easily induced by the other [10]. Oxidative stress (OS) is defined as an imbalance between toxic reactive oxygen species (ROS) and antioxidants in favor of oxidants, leading to a disruption of redox signaling and/or irreversible oxidative damage to lipids, deoxyribonucleic acid (DNA) or proteins [11] .

Aim :

To evaluate association the total antioxidant capacity , WBC , PLR with severity of COVID-19 disease .

CHAPTER TWO
METHODOLO

METHODOLOGY

Setting of the Study:

This study was carried out on patients attended to respiratory clinic in Merjan Teaching Hospital in Babylon Province. These patients with a positive COVID-19 RT-PCR test, and identified based on World Health Organization interim guidelines [12].

Study Population

A purposive study was carried out in this study which includes 60 individuals. These subjects were divided into two groups: The first group includes 30sever COVID-19 patients and the second group includes 30 mild COVID-19 patients.

Data Collection

The inclusion and exclusion criteria for this study are as follows:

Inclusion criteria

The participants in the present study were mild and sever COVID- 19 patient, those who were accepted to participate in the current study.

Exclusion criteria

Any Subjects with a history of diabetes, hypertension, cancers and autoimmune disorders were excluded from the studied groups. Also, subjects were excluded if they had took antioxidant supplement.

Study Instruments

Questionnaire

The sociodemographic characteristics composed of age, weight, length , duration , and medical history ,CBC . while the length and weight of the participants were measured by measuring tape and electronic balance, respectively. The calculation of body mass index (BMI) was measured by dividing the weight (kg) on the square of height (m) BMI = kg/m^2 .

Samples Collection

Samples were collected, during the period from January 2021 to March 2021 , from the visitors of respiratory clinic in Merjan Medical City; verbal consent agreement from all subjects was obtained before the collection of samples. Three ml of venous blood sample was aspirated by 5 ml disposable syringe, then the blood was kept in gel tube for serum separation and sodium citrate tube for blood tests were performed using routine methods for the detection of WBC, Lymphocyte , and platelets measurement . The total antioxidant capacity measurement by apak(apak et al 2005) [13].

Statistical Analysis

Numerical variables were expressed as mean ± standard deviation (SD). Student's t-test was used to determine the difference in means between control and COVID-19 groups for numerical variables using SPSS version 20 software (SPSS Inc.). In statistical analysis, the level of significant (P-value) was <0.05.

CHAPTER THREE

Results

Results

In the present study,60 individuals have participated comprised of 30 confirmed sever COVID-19 patients and 30 confirmed mild COVID-19 patients. The study shows significant changes (p value <0.05) between sever and mild COVID-19 positive patients parameters (Total antioxidant capacity, WBC and PLR) as shown in Table 1

Table (1): sever and mild COVID-19 patients parameters

Parameters	Groups	No.	Mean± SD	P value
Total antioxidant capacity	Sever	30	612.72±130.8	<0.05
	Mild	30	1237.37±54.29	
WBC	Sever	30	11.75±3.5	<0.05
	Mild	30	7.07±1.5	
PLR	Sever	30	1413±300	<0.05
	Mild	30	821±167	

Figure 1: The comparison between the mild and sever COVID-19 patients in terms of the average total antioxidant capacity is presented here. These data showed that the differences between the two groups .Total antioxidant capacity in mild group is higher than sever group .

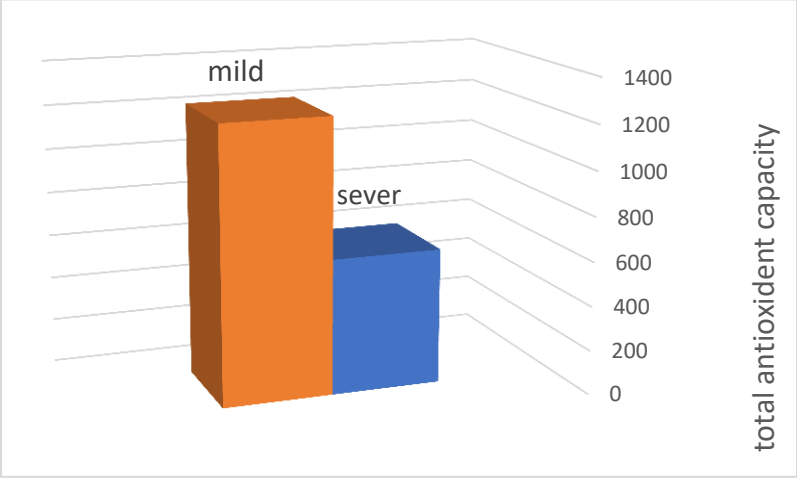

Figure (1): total antioxidant capacity comparison between the mild and sever COVID-19 patients

Figure (2) revealed WBC in sever COVID -19 patients is higher than mild COVID -19 patients

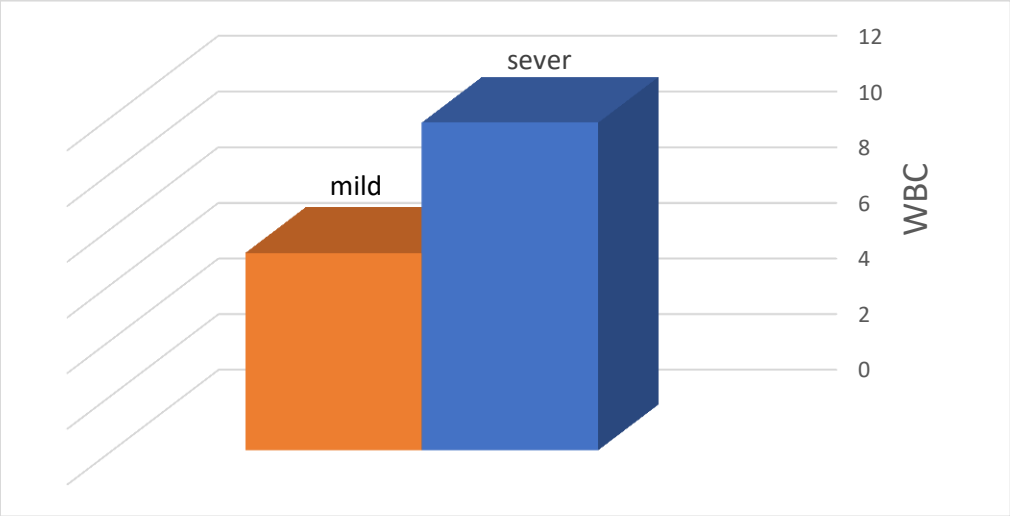

Figure (2):WBC comparison between the mild and sever COVID-19 patient

The ratio between platelet and lymphocytes (PLR) was also considerably higher in the severCOVID-19 patients than other group as in figure (3)

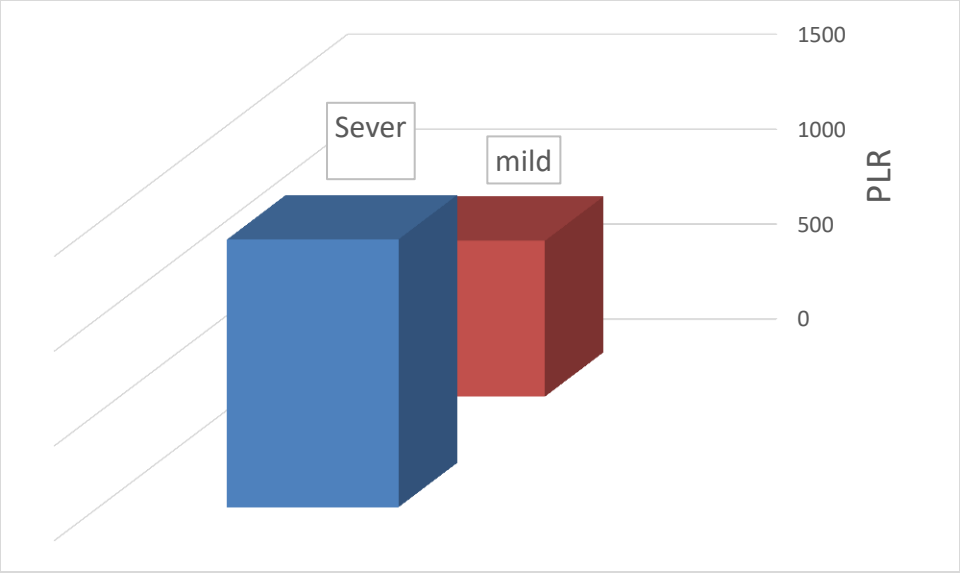

Figure (2): PLR comparison between the mild and sever COVID-19 patient

CHAPTER FOUR
DISCUSSION

DISCUSSION

The COVID-19 pandemic has hit the world, there have been various research activities to understand the etiology, pathogenesis, disease course and progression of the disease and to find a suitable antiviral drug, and vaccines to combat this virus. COVID-19 earlier thought of as a respiratory tract infection, is now considered a systemic disease involving respiratory, gastrointestinal, neurological, cardiovascular, hematopoietic and other systems [14].

Our present study found that sever COVID-19 patients had an increased risk of developing severity in patients with COVID-19. This indicates that sever COVID-19 patients have lower values of antioxidants

In accordance with previous studies, an imbalance in the production of reactive species and the body's inability to detoxify these reactive species is referred to oxidative stress [15-16]. It is known that oxidative stress is triggered by a wide variety of viral infections, including HIV 1, viral hepatitis B, C and D viruses, herpes viruses, respiratory viruses, such as corona viruses [17] .

As mentioned in previous studies, hypoxia induced by lung injury can be due to mitochondrial dysfunction. Mitochondrial dysfunction leads to a relative decrease in oxygen and energy production and increase in ROS production. In this regard, superoxide, H_2O_2 and other reactive species are mainly produced by the mitochondrial respiratory chain. Hydrogen peroxide causes the expression of many genes that activate pro inflammatory cytokines in macrophages, neutrophils and endothelial cells through NADPH oxidase (NOx) to produce more superoxide and H_2O_2 [18-19].

Reported that SARS-COV-2 infection activates the phagocytic cells which cause ROS to be excessively produced, while the antioxidant capacity are insufficiently present leading to a weakened antioxidant system in the face of increased ROS production [20].

The study has also demonstrated that decreases in the activities of these antioxidant vitamins in patients with viral disease , and partial reduction of O_2 in oxidative processes generates superoxide which is acted upon by SOD converting

it to hydrogen peroxide (H_2O_2)which may subsequently react forming hydroxyls (OH) [21].

low glutathione has been associated with abnormalities in the lung surfactant system, while normal levels of intracellular glutathione may exert a critical negative control on the elaboration of pro inflammatory cytokines.[22]

Although the etiopathogenesis of the disease is still undergoing research, some studies have shown that, during the incubation period and the early phase of the disease, The virus causes changes in peripheral blood leukocyte and lymphocyte counts resulting in normal to low count [23].

This architecture is essential to the development of functional leucocytes through the critical interactions within the immune cells. These tissue structure changes alter leucocyte populations' distributions, lymphocyte activity, and immune defenses and are linked to a compromised response of innate lymphoid cells [24].

In this respect, the number of lymphocytes count is directly proportional with the severity of COVID-19 infection and the high survival rate of the disease is associated with the ability to renovate lymphocyte cells, particularly T lymphocytes which are crucial for destroying the infected viral particles [25]. During disease severity, remarkable thrombocytopenia was observed and confirmed by Lippi and his colleagues that revealed a reduction of platelet count among severe and died patients with COVID19 supporting that thrombocytopenia could consider as an exacerbating indicator during the progression of the disease [26].

PLR was initially suggested as an excellent candidate marker for determining the severity and mortality of COVID-19. First, PLR is an established marker of inflammation[27].Inflammation plays a considerable role in the pathophysiology of COVID-19, with cytokine storm as a hallmark condition in severe disease and poorer prognosis [28]. Thus, elevated PLR value suggests an overactive inflammatory response and subsequently, worse prognosis. Second, PLR is sensitive to natural and acquired immune response .Third, PLR is an inexpensive and readily available measurement that can be used in resource-limited settings[29].

Our additional lower platelet count in non survivors, are also in line with the results of a recent meta-analysis, which showed that thrombocytopenia is common in severe COVID-19 patients and in other patients suffering critical illness [30].

Thrombocytopenia is another pathological finding that could be detected in a complete blood count. Thrombocytopenia was detected in our study, consistent with the previous study; furthermore, PLR was significantly high. The platelet count, dynamic changes during treatment, and PLR were a source of in severe COVID-19 pneumonia cases. It was interpreted that PLR could serve as a novel indicator of the degree of cytokine storm[31].

One study noted that patients with a higher total WBC on admission had a poorer prognosis, while low total WBC levels were found to be a protective factor. Higher total WBC values are probably due mainly to increased levels of neutrophils . In support of this idea, higher neutrophil counts also "predisposed" patients to unfavorable disease outcomes [32].

Conclusions:

-Total Antioxidant capacity is the significant variable relates with severity of COVID-19 disease.

-PLR is inflammatory marker, has been suggested to predict the severity of COVID-19 patients and may be considered as cardinal laboratory findings, with prognostic potential. WBC counts increased significantly as the disease severity increased.

Recommendation:

-people with sever COVID-19 patients typically need higher amounts of antioxidant to maintain adequate levels and/or to correct a deficiency.

-further investigations are needed to stablish an association between severity of COVID-19 disease and antioxidant, hematological parameters and clinical outcomes in this specific population.

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الملخص-:

خلفية؛ يعد مرض فيروس كورونا 2019 (COVID-19) من الأمراض الناشئة بسرعة بسبب فيروس شديد العدوى يسمى فيروس كورونا 2 (SARS-CoV-2) ، وتكمن أهمية الدراسة في الحاجة إلى تقييم مستويات مضادات الأكسدة ، خلايا الدم البيضاء (WBC) وكذلك الصفائح الدموية إلى نسبة الخلايا الليمفاوية (PLR) بين مرضى COVID-19 المعتدل والحاد بحيث يمكن أن تنطوي النتيجة على نهج أكثر قوة وشمولية لمكافحة المرض المدمر.

الأهداف: لتقييم دور القدرة الكلية المضادة للأكسدة ، WBC، PLRمع شدة مرض-COVID 19المرضى.

الطرق: أجريت دراسة هادفة في هذه الدراسة شملت 60 فردًا. تم تقسيم هذه الموضوعات إلى مجموعتين: المجموعة الأولى تضم 30 مريضًا مصابًا بفيروس كورونا المستجد ، والمجموعة الثانية تضم 30 مريضًا خفيفًا من COVID-19 تم إجراء اختبارات الدم باستخدام الطرق الروتينية للكشف عن WBC ، الخلايا الليمفاوية ، وقياس الصفائح الدموية. قياس سعة مضادات الأكسدة الكلية بالطريقة اليدوية.

النتائج: في هذه الدراسة ، تم قياس إجمالي سعة مضادات الأكسدة ، WBC، و PLR في مرضى COVID-19الخفيف والحاد. أظهرت النتائج تغيرات كبيرة في إجمالي سعة مضادات الأكسدة ، WBC وكذلك PLR بين مجموعتين. وقد تم عرض مساهمة تلك العوامل في شدة المرض.

الاستنتاجات:

-السعة الإجمالية لمضادات الأكسدة هي المتغير الهام المرتبط بشدة مرض كوفيد -19.

- PLR هو علامة التهابية ، وقد تم اقتراحه للتنبؤ بشدة مرضى COVID-19 ويمكن اعتباره نتائج معملية أساسية ، مع إمكانية الإنذار. زاد عدد كرات الدم البيضاء بشكل ملحوظ مع زيادة شدة المرض.

جمهورية العراق

وزارة التعليم العالي والبحث العلمي

جامعة بابل

كلية التمريض

دور السعة الكلية لمضادات الأكسدة ، خلايا الدم البيضاء ونسبه الصفائح الدموية الى الخلايا الليمفاوية مع شدة مرض فايروس كورونا -19

قسم العلوم الطبية الأساسية

مشروع تخرج مقدم لكلية التمريض جامعة بابل ضمن متطلبات الحصول على بكالوريوس التمريض من قبل

زهراء جاسم عويد

زهراء صالح هادي

زهراء محمد حسون

زينب صالح هادي

تحت إشراف

د. أميرة الأعرجي

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